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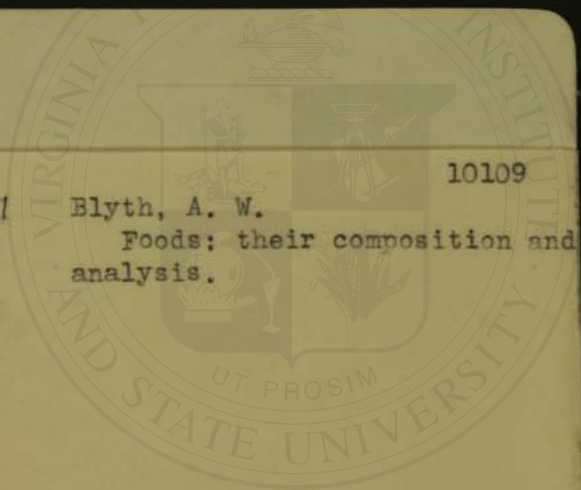
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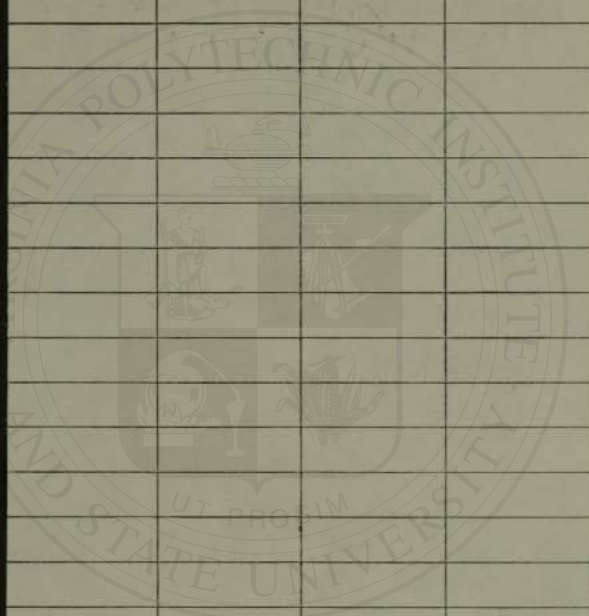
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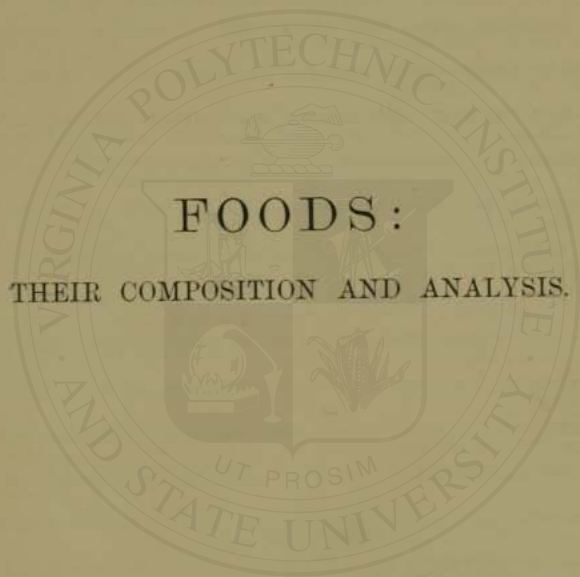
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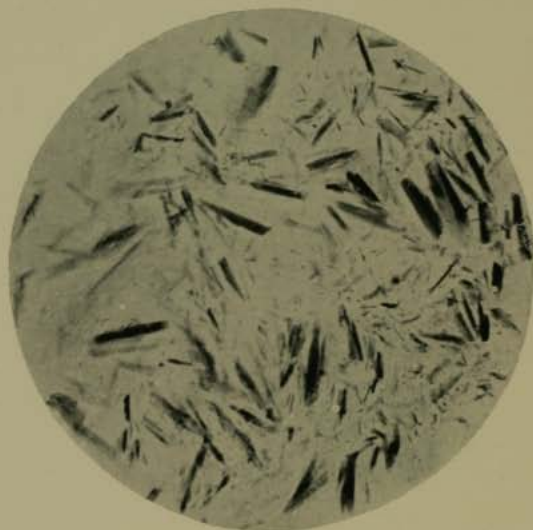
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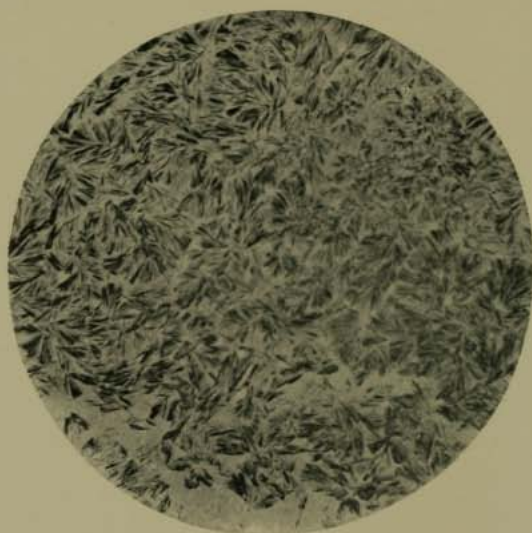
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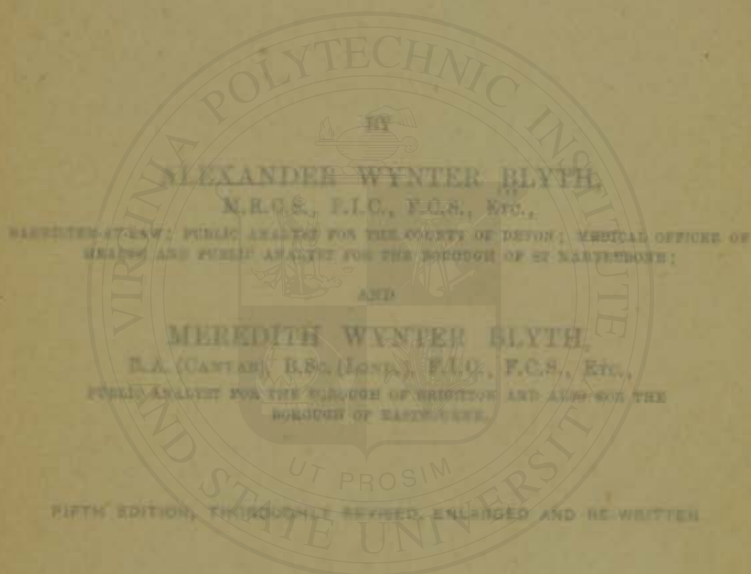
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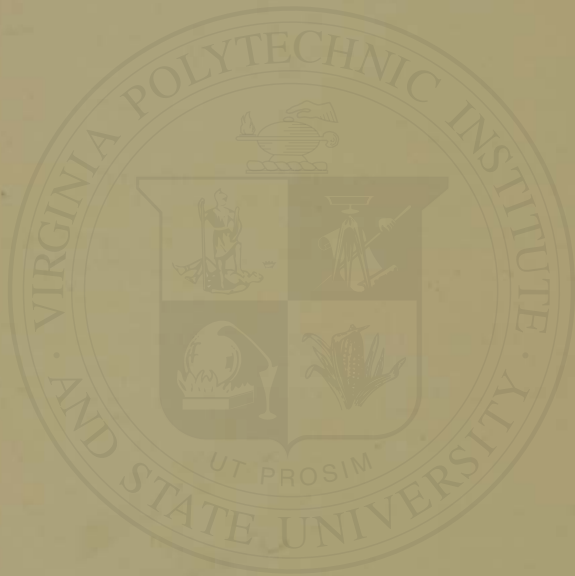


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PREFACE.

SINCE the last edition of this work, the investigations of Parliamentary Committees have resulted in fresh and more exact legislation on the adulteration of Food; standards or limits of purity have been fixed for milk, butter, and margarine; and by the work of both foreign and British scientists many new analytical processes have been devised. The mass poisoning in the North and Midlands by arsenical beer has brought to public notice vast possibilities of danger in the contamination of common articles of diet by injurious substances, and has directed the attention of skilled chemists to the perfection of old processes for the discovery and estimation of minute quantities of arsenic. It is therefore no matter for surprise that the Authors found on commencing the preparation of a new edition that so many changes were necessitated that the work had, to a great extent, to be rewritten. It was at the same time necessary to keep the size within the limits of the last editions; this has been fairly well accomplished by omitting the bibliographies, condensing certain portions, enlarging the page and omitting much which experience showed to be of no great utility. In this way room has been given for the chapters on the detection and estimation of arsenic, the composition and analysis of spices, the bacteriology of water, and so forth. Many new Tables and Diagrams have also been inserted.

The Authors have endeavoured to acknowledge in every instance the source from which they have drawn their information, so as to enable the reader to refer when necessary to the original work or paper.

In its new form the Authors believe that the work will be found useful to analytical chemists, and to those who desire to possess a handy book of reference on the subject.

A. WYNTER BLYTH.
 MEREDITH WYNTER BLYTH.

LONDON, 1903.

MAY 4 1911

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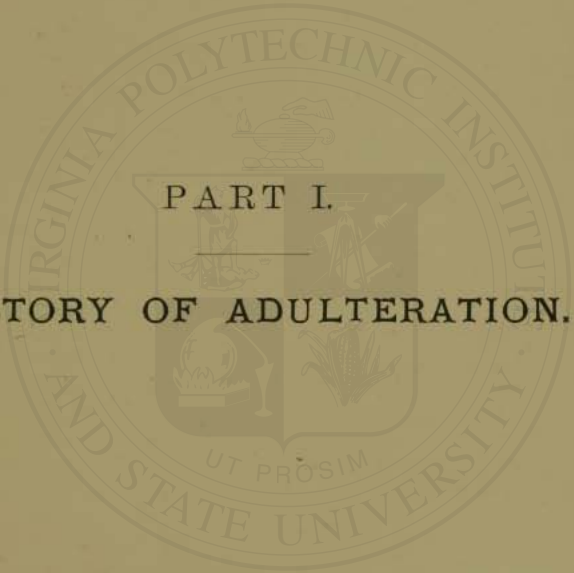
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PART I.

HISTORY OF ADULTERATION.

FOODS: THEIR COMPOSITION AND ANALYSIS.

PART I.—HISTORY OF ADULTERATION.

I.—EARLY NOTICES OF ADULTERATION, ESPECIALLY IN ENGLAND.

§ 1. BEFORE adulteration commences, commerce must develop. In primitive states of society,¹ there may be knavish tricks, ignorant bartering, substitutions of bad for good, falseness and meanness of all kinds, but no systematic sophistication is possible. Again, in the semi-pastoral state (as it existed in some parts of Scotland a century ago), in which the food of a family is raised from the soil on which they dwell, and clothing produced from their own sheep and spun into textile garments at their own fireside, commercial frauds are unknown or undeveloped.

There are several notices of ancient sophistications practised by the Greek and Roman traders; but it is from the Middle Ages that the most copious and interesting materials for a history of adulteration are obtained—a page of history but little explored, yet abounding with curious facts more or less illustrative of the manners of the times.

The mixing, or, rather, alloying of gold or silver with the baser metals may be justly considered of the nature of adulteration, and has prevailed contemporaneously with the art of coinage. The well-worn tale of the detection of the base metal in the crown of Hiero by Archimedes, some two and a half centuries before Christ, may be accepted as probably the earliest scientific detection of adulteration. The process used by the philosopher of Syracuse, when discharging the duties of a *public analyst*, for finding the specific gravity, is quantitative as well as qualitative,

¹ "High things begin low. Astronomy began as astrology, and when trade began, there must have been even more trickery about it than there is now. Conceive a world made up of nomadic tribes engaged in perpetual warfare. It is a commerce of killing. If a tribe desire the richer soil or larger possessions of another, the method is to exterminate that other; but at last there rises a tribe too weak or too peaceful to exterminate, and it prefers to barter, it challenges its neighbours to a contest of arts. They try to get the advantage of each other in bargains, they haggle and cheat; it is not heroic at all, but it is the beginning of commerce and peace."—*Demonology*, by Maurice Conway, M.A.

and, though purely physical, is used daily by all engaged in practical chemistry.

Vitruvius,¹ in his work on architecture, describes the adulteration of minium with lime. He also gives a simple process for its detection: heat to redness on a sheet of iron; if pure, it will blacken, but on cooling return to its former hue.

Dioscorides alludes to the adulteration of opium with gum and with the milky juice of glaucium and lactuca. The test for distinguishing the pure from the false was primitive: the opium was to be burnt; if pure, the flame was clear and brilliant, but the adulterated burnt with difficulty. The quality of the opium was also to be judged by its behaviour when exposed to the rays of the sun; when opium of good quality liquefies, it looks as if it had just come from the plant.² He also specifies the adulterations of several drugs; as, for example, the mixing of styrax resin with styrax sawdust.

Pliny, among other matters, alludes to the frauds practised by bakers; for they added to the bread a white earth, soft to the touch and sweet to the taste, which was obtained from a hill called '*Leucogee*,' situated between Pozzuoli and Naples. It has been suggested that the white earth was carbonate of magnesia; this is doubtful.³

He also speaks of the adulteration of aerugo (under which name was confounded both the acetate and sulphate of copper) with shoemakers' black *atramenta sutorium*, and gives a true chemical method for its discovery. Paper is to be soaked in the juice of galls; if the aerugo is pure, it will not turn the paper black. Another method was to put the substance on a sheet of red-hot iron; if sulphate of iron had been added, it became covered with spots.⁴

The adulteration of wine in Athens necessitated the appointment of a special inspector, whose duty it was to detect and stop these practices. Greek history has handed down the name of one *Canthare*, who excelled in ingenious mixtures, and knew how to impart the flavours of age and maturity to new wines. His ingenuity was such that it was commemorated in the proverb, "Artificial as Canthare."

In Rome, also, wine was much tampered with; even the rich, according to Pliny, could not obtain the natural wines of Falerno, for they were adulterated in the cellars; and certain wines from Gaul had an artificial colour given to them by means of aloes and other drugs.⁵

§ 2. In our own country, and in Europe generally, from the eleventh century onwards, the bakers, the brewers, the 'pepperers,' and the vintners, were most frequently accused of corrupt practices. We must not, however, judge too harshly of the tradespeople of that epoch, for morality was generally low, and adulteration an innocent pastime when compared with the frequency and magnitude of midday highroad robbery and midnight violence.

In the latter part of the twelfth century, that which would now be called crime became the favourite amusement of the principal citizens, "who would sally forth by night, in bands of a hundred or more, for an attack upon the houses of their neighbours. They killed without mercy every man who came in their way, and vied with each other in brutality. . . . False weights, false measures, false pretences of all kinds were the instruments of commerce most generally in use. No buyer would trust

¹ Vitruvius, i. ix. c. 13.

² Dioscorides, iv. 65; Pliny, xx. 76.

³ Pliny, xviii. 29.

⁴ Pliny, xxxiv.

⁵ *Op. cit.*

the word of a seller, and there was hardly any class in which a man might not with reason suspect that his neighbour intended to rob or even to murder him."¹

ASSIZES OF BREAD—BAKERS.

§ 3. The sale of bread was regulated in England as early as the fourth year of the reign of John, by what was called the 'Assize of Bread,' the original object of which was to regulate the price of bread by limiting the profit of the baker on each quarter of wheat, so that the price of the loaf should bear a certain proportion to the price of the quarter of wheat. The assize of John's reign continued in force until 1286, when it was repealed by '*The Statute of Assize.*'

There were various modifications of these assizes, and they were finally abolished in 1815. The 'Assize of Bread' in its influence was probably the exact reverse of what was intended. On the one hand, the development of trade was restricted injudiciously, and, on the other, the bakers often suffered unjustly, and, therefore, had a direct inducement to recover their losses by nefarious practices. Although, at the institution of the assize, adulteration with foreign substances was not the main object of the regulations, yet, as time went on, and the sins of the bakers, both male and female,² accumulated, clauses with regard to the adulteration of bread were inserted, and the later ones may be considered collectively as the ancient English 'Sale of Food Act.' The assize of 1582³ contained the following:—"If there be any that by false meanes useth to sell meale: for the first time he shall be grievously punished, the second tyme he shall lose his meale: the III tyme he shall foreswere the towne, and so likewise the bakers that offende. Also, bouchers that sell mesell porke or mozen flesche: for the first time they shall be grievously amerced, for the second tyme so offendinge they shall have the judgement of the pillory, for the third tyme they shall be comytted to pryson until ransomed, and the fourth tyme they shall forswere the towne, and thus ought other transgressors to be punished, as cooks, forestallers, regrators of the markets when the cookes serve, roste, bake, or any otherwyse dresse, fysche or flesche unwholesome for man's body."

The assize of 1634 had some stringent regulations with regard to musty meal:—"If there be any manner of person or persons, which shall by any false wayes or meanes, sell any meale unto the kinge's subjects, either by mixing it deceitfully or [sell any] musty or corrupted meal, which may be to the hurte and infection of man's body, or use any false weight, or any deceitful wayes or meanes, and so deceive the subject, for the first offence he shall be grievously punished, the second he shall lose his meale, for the third offence he shall suffer the judgement of the pillory, and the fourth time he shall forswere the towne wherein he dwelleth."

These extracts give some idea of the punishments inflicted on dishonest bakers during the Middle Ages in England. First offences were often visited by corporal chastisement and exposure in the pillory (generally with a rope and a loaf round the neck); fourth, and even third, convictions

¹ *A History of Crime*, by Luke Owen Pike.

² The bakers, as well as the brewers, were of both sexes.

³ The title runs: "Here beginneth the boke named the assyse of bread, what it ought to weye, after the pryce of a quarter of wheat, also the assize of ale, with all manner of wood and cole, lath, bolside, and tymber, and the weight of butter and cheese. Imprinted, by Thomas Wyatt, 1582."

were considered so heinous that it was thought better to cast the man forth from the city to earn his livelihood elsewhere.

In the curious paper entitled "A Quip for an Upstart Courtier,"¹ there is a powerful and quaint expostulation with the different traders:—"And for you Goodman baker, you that love to be seen in the open market-place upon the pillory, the world cries out on your wiliness: you crave but one deere yeare to make your daughter a gentlewoman. You buy your corne at the best hand, and yet will not be content to make your bread weight by many ounces. You put in yeaste and salt to make it heavie: and yet all your policie cannot make it. The poore crie out, the riche find fault, and the lord maior and the sheriffs, like honourable and worshipful maistrates, every daie walk abroad and weigh your bread, and yet all will not serve to make you honest men. But were extremities used and the statutes put in the highest degree in practice, you would have as few eares on your heade as the collyer."²

The manner of adulteration seems to have varied.³ Sometimes the bread was made altogether of 'putrid and rotten materials,' sometimes it was good outside and bad within, and as for the addition of alum or mineral matters, such was only detected when in considerable quantity and coarsely done. The more artful mixtures required for their detection the application of a chemical science not then possessed. The following may serve as examples of a few of the earlier instances:—

One "Alan de Lyndseye, baker, was sentenced to the pillory because he had been convicted of baking *pain demayn* that was found to be of bad dough within and good dough without, and because such falsity redounds much to the deception of the people who buy such bread."

The same baker seems a few days afterwards to have been again in trouble, for "Alan de Lyndseye, baker, and Thos. de Patimere, baker, were taken and brought before the Mayor and Aldermen, and sentenced to the pillory for selling bread made of false, putrid, and rotten materials, through which who bought bread were deceived, and might be killed."⁴

A similar fraud is recorded at perhaps an earlier date (A.D. 1311), for "the bread taken from William de Somersete, baker, on the Thursday next before the Feast of St. Lawrence (Aug. 10) in the 5th year of the reign of King Edward II. was examined and adjudged upon. . . . Because it was found that such bread was putrid and altogether rotten, and made of putrid wheat, so that persons eating that bread would be poisoned and choked, the Sheriff was ordered to take him and have him here on Friday next after the Feast of St. Lawrence, then to receive judgment for the same."⁵

Mr. Pike, in his *History of Crime*,⁶ speaks of loaves being adulterated in the Middle Ages by lumps of iron, probably referring to the following case:—"On Wednesday next after the Feast of St. Matthew the Apostle

¹ The "Quip for an Upstart Courtier" was written in 1592. The original is in black letter.

² The sanitary state of the bakehouses in the fifteenth and sixteenth centuries was, as a rule, bad. According to Mr. Pike, they appear to have been the favourite receptacle for dead bodies after a murder had been committed. *A History of Crime*, by Luke Owen Pike, M.A., Vol. I. p. 256.

³ In the reign of Edward I. it was enacted at one of the 'Hallmots,' that no bread should be coated with bran, or so as to be found worse when broken than it was on the outside.

⁴ *Memorials of London*, by H. T. Riley, pp. 120, 121.

⁵ *Op. cit.*

⁶ Vol. I. p. 237.

(11th of Sept., A.D. 1387), in the 11th year, Richard Porter, servant of John Gibbe, baker, of Stratforde, was brought here before Nicholas Extone, Mayor of the said city, John Hafle, and other Aldermen, and questioned for that when the same Mayor on that day went into chepe to make assay there of bread, according to the custom of the city, he, the said Robert, knowing that the bread of his master in a certain cart there was not of full weight, took a penny loaf, and in it falsely and fraudulently inserted a piece of iron weighing about 6s. 8d. (4 oz.), with intent to make the said loaf weigh more. . . . He was sentenced to the pillory with the loaf and iron round his neck, and the cause of the punishment was proclaimed by the Sheriffs.¹ But the placing of a mass of metal in a loaf under the circumstances recorded is somewhat different from adulteration, for the man slipped in the iron to avoid a conviction for false weights.

BREWERS AND VINTNERS.

§ 4. *Beer*.—The fraudulent practices of the early brewers are thus detailed in the Black-letter Tract before mentioned:—"And you, maister brewer, that growe to be worth forty thousand pounds by selling of soden water, what subtilty have you in making your beere to spare the malt, and put in the more of the hoppe, to make your drinke, be barley never so cheape, not a whit the stronger, and yet never sell a whit the more measure for money. You can when you have taken all the harte of the malt away, then clape on store of water, 'tis cheape enough! and mashe out a turning of small beere, like rennish wine: in your conscience how many barrel draw you out of a quarter of malt? Fie! fie! I conceal your falsehood, least I should be too broad in setting down your faults." Not only the brewer but the retailer of the beer was also condemned.

"Last to you, Tom Tapster, that take your small cannes of beere, if you see your guests begin to be drunke, halfe smal and halfe stronge; you cannot be content to pinch with your small pottes and your ostrie faggots, but have your drugges and draw men on to villany and to bring customers to your house, where you sell a joint of meat for xii. pence that cost you scarce six, and if any chance to go on the skore, you skore him when he is asleep and set up a pot a day more than he hath, to find you drinking pots with your companions. To be short, thou art a knave!"

As early as the reign of Edward the Confessor, we find it recorded in Domesday Book that in the city of Chester a knavish brewer, "*malam cerevisiam faciens, in cathedra ponebatur stercoris*"—in other words, the offender was taken round the town in the cart in which the refuse of the place had been collected, and to this degradation was often added corporal chastisement.

In many towns in the sixteenth century we find 'ale-tasters,' whose duty it was to inspect the beer.

In 1529, for example, the Mayor of Guildford ordered that the brewers make a good useful ale, and that they sell none until it be tasted by the 'ale-taster.' These officials had to take the following oath:—"You are chosen ale-tasters of this town. You shall well and truly serve his Majesty and this town in the same office. You shall at all times try, taste, and assize the beer and ale to be put to sale in this liberty, whether the same be wholesome for man's body, and present those that offend, or refuse to

¹ Riley's *Memorials of London*, p. 493.

suffer you to assay it. You shall give your attendance at all courts, and present from time to time the offenders, and all things else belonging to your office you shall do and execute. So help you God." The ale was not only tasted, but some of it was spilt on a wooden seat, and on the wet place the taster sat, attired in leathern breeches, then common enough. If sugar had been added to the beer, the taster became so adherent that rising was difficult; but if sugar had not been added, it was then considered that the dried extract had no adhesive property. A less coarse, but not dissimilar, method was also applied by the earlier Inspectors to test the purity of milk.

§ 5. *Wine*.—The frauds of the vintners or wine-sellers attracted some share of public attention in the sixteenth and seventeenth centuries, as shown by municipal records, fugitive tracts, and broadsides. In August 1553 a certain Paul Barnardo brought into the port of London some wine, and there is extant an order in council directing the Lord Mayor to find five or six vintners to rack and draw off the said pipes of wine into another vessel, and to certify what drugs or ingredients they found in the said wine or cask to sophisticate the same.¹ At a later date the records of the Common Council contain a certificate from the Lord Mayor to the Lords of the Council, stating that the wines of a certain 'Peter Van Payne' had been drawn off in his presence, and that in eight of the pipes had been found bundles of weeds, in four others some quantities of sulphur, in another a piece of match, and in all of them a kind of gravel mixture sticking to the casks; that they were conceived to be unwholesome and of a nature similar to others formerly condemned and destroyed.² In *The Search after Claret*, by Richard Ames, a thin quarto, the last leaf is occupied by the following advertisement:—"If any vintner, wine-cooper, etc., between Whitechapel and Westminster Abbey, have some tuns or hogsheads of old rich unadulterated claret, and will sell it as the law directs for sixpence a quart, this is to give notice that he shall have more customers than half his profession, and his house be as full from morning to night as a conventicle or Westminster Hall the first day of term."³

Later, the vintners became more scientific in their operations. Addison (in the *Tatler*, No. 131, 1710) alluded to a certain fraternity of chemical operators who wrought underground in holes, caverns, and dark retirements to conceal their mysteries from the eyes and observations of mankind. "These subtle philosophers are daily employed in the transmutation of liquors, and by the power of magical drugs and incantations raise under the streets of London the choicest products of the hills and valleys of France; they squeeze Bordeaux out of the gloe, and draw Champagne from an apple."

SPICES—DRUGS.

§ 6. The London pepperers, or spicers, formed a separate guild, and were under special ordinances. The ordinance, A.D. 1316, in Norman-French, has the following regulations:—"No one of the trade or other person in his name for him, shall mix any manner of wares, that is to say, shall put old things with new, or new things with old, by reason whereof the good

¹ *Remembrancia*, vii. 92.

² *Remembrancia*, viii., 12th July 1635.

³ "The Search after Claret, or a Visitation of the Vintners." A poem in two Cantos. Second ed., London, 1697, 4to.

thing may be impaired by the old, nor yet things of one price, or of one sort, with other things of another sort; also, that no man shall dub any manner of wares, that is to say, by putting in a thing that was in another bale, and then dressing the bale up again in another manner than the former in which it was first bought, so as to make the ends of the bale contain better things than the remainder within the bale, by reason whereof the buyer may be deceived, and so lose his goods. Also that no man shall moisten any manner of merchandise, such as saffron, alum, ginger, cloves, and such manner of things as may admit of being moistened; that is to say, by steeping the ginger, or turning the saffron out of the sack and then anointing it, or bathing it in water; by reason whereof any manner of weight may, or any deterioration, arise to the merchandise.¹

In England the trades of the druggist and the grocer were combined. Drugs and groceries were sold in the same shop, and they were under the same regulations until 1617, when the apothecaries separated themselves from the grocers. Very soon after they had become a distinct body they began to complain of the frauds and artifices of the grocers, from whom they continued to be supplied with many drugs, and therefore established a dispensary for the purpose of compounding the more important preparations themselves. In 1540 the physicians were empowered to search, view, and see the apothecary wares and stuffs, and to destroy such as they found unfit for use. In 1553 very extensive powers were conferred on the College of Physicians for this purpose. "The four censors, or any three of them, shall have authority to examine, survey or govern, correct and punish all and singular physicians and practisers in the faculty of physic, apothecaries, druggists, distillers, and sellers of waters and oils, and preparers of chemical medicines, according to the nature of his or their offences." The great power of the censors was on more than one occasion abused. In 1724, for example, they burnt the drugs of one 'Goodwin,' the drugs not having been examined, and the history of the whole affair showing that the act was merely a gratification of private spite. Goodwin petitioned Parliament, and ultimately, it is said, obtained £600 compensation.

The College of Physicians compiled the first *Pharmacopœia*, and published it in 1613. Subsequent editions bear the dates of 1621, 1632, 1650, etc. As may be expected, the early editions contain lists of very absurd and superstitious remedies, and have no pretensions to a scientific character.²

II.—ADULTERATION IN FRANCE.

§ 7. In France, from very early times, the general supervision of provisions, as to purity and quality, and the inspection of weights and measures, were under the '*police des commissaires*,'³ and various special statutes were enacted from time to time. Thus an ancient statute (1292) of the Paris brewers forbade the adulteration of beer; "whoever put into

¹ Riley's *Memorials of London*, p. 120.

² See *Historical Sketch of the History of Pharmacy*, by Jacob Bell. London, 1861.

³ "*La police des commissaires . . . il est de leurs soins de faire punir le débit des vivres corrompus, alterez, falsifiez, les faux poids et les faux mesures.*"—*Traité de la Police de la Mare*. Tom. i. liv. i. titre xi., chap. vi.

beer baye, pimento or 'poix resine' was to be fined 20 francs, and his *brassins* were to be confiscated, for such things are neither good nor loyal to put in beer, for they are bad for the head and for the body, for the healthy and the sick." A later statute, dated March 16, 1630, among various sanitary provisions, forbade the use of buckwheat, "yvtoye or other bad matters under a penalty of 40 Parisian pounds." Judges were also to examine the materials before use, in order to see that there was nothing in them impure, heated, mouldy or spoiled. If such were found, the materials were to be cast into the river.¹

§ 8. *Flour and Bread.*—There were various special regulations as to flour and bread; by an *Ordonnance* of the Provost of Paris, October 11, 1382, the miller was to grind the corn without mixing it, to increase his fee, with bran, pease, beans, or anything else save that which had been given him to grind.² Later, by a decree dated July 13, 1420, the bakers were forbidden to be millers, it being thought that if they ground the wheat as well as made it into bread, there would be facilities for fraudulent dealing. The punishment of bakers for false bread—whether the falseness were admixture of foreign substances, the use of damaged flour, or simply light weight—was very similar to that of English bakers, except that it partook more of the character of a religious penance. Thus, in 1525, a baker convicted of 'false bread' was condemned by the court to be taken from the Chatelet prison to the cross before the 'Eglise des Carmes,' and thence to the gate of Notre Dame and to other public places in Paris, in his shirt, having the head and feet bare, with small loaves hung from his neck, and holding a large wax candle lighted, and in each of the places enumerated he was to make '*amende honorable*,' and ask mercy and pardon of God, the king, and of justice for his fault.³ False weights were also often punished by corporal punishment. In 1491 the case of three bakers is recorded, who, having been convicted of selling loaves 'too small,' were stripped and beaten with rods through the streets of Paris, and were admonished for the future to sell the three kinds of bread ordered by the law, of the weight and quality they ought to be.⁴ In still later times, we find the practice of the courts remarkably severe. In 1699 a baker named Pasquier was convicted of converting into bread bad and unwholesome flour. Sacks filled with good flour and others filled with bad had been found on his premises, and it was affirmed that he had mixed the two together. He was fined 500 livres, his oven demolished, and his shop closed for six months, with a placard upon it stating the crime and the punishment.⁵

§ 9. *Wine.*—A curious decree of the provost of Paris, in 1371, compelled the tavern-keepers to permit any one who purchased wine, whether to be drunk on the premises or taken away, himself to see the wine drawn from

¹ "*La police des commissaires visitoient les Marchez, et il estoit de leurs soins d'y procurer l'abondance des viures et des autres provisions necessaires à la subsistance des citoyens, ils empechoient qu'il ne s'y commist aucune fraude, soit en la qualité ou au prix, soit au poids ou en la mesure, ils estoient principalement chargés de se donner tous les soins à l'égard des grains, du pain, de la viande, et du vin.*" *Loc. cit.*

² "*Que nul meusnier ne soit si osé ne si hardy sur quanque il se peut mefaire envers le roy, en corps, et en biens, de mesler, mettre ou fair mettre en aucune maniere es farines des blez qu'ils moudront aucune mixtion ou meslec, pour rendre plus grande mouture, comme de bran, d'orge, de pois, de fèves, ou autres choses quelconques, qui ne soit du blé qui leur sera baillé.*"—*Traité de la Police*, t. ij. liv. v. titre ix.

³ *Traité de la Police*, tome ii. livre v. titre xii.

⁴ *Op. cit.*, tome ii. livre v. titre xii.

⁵ *Op. cit.*, t. i. livre iv. titre iv., p. 570.

the cask. The penalty was, for neglect or disobedience to this law, four Parisian pounds, one-fourth of which went to any informer.

An *Ordonnance* of January 30, 1330, forbade the mixing of two wines together; no wine-seller was to give a false name to a wine, or to give a wrong description of its age; the penalty was confiscation of the wine and a fine. Similar edicts were promulgated in 1415, 1635, and 1672. Still the evil did not diminish, and in 1708 two hundred inspectors of wine and drinks were appointed in Paris.

The 'Baillie' of Bergheim, in 1718, had condemned to a month's imprisonment one André, who had falsified his wine with some poisonous plant (probably belladonna), and his wife, who had sold the wine, to a month's imprisonment, and a fine of 130 livres. This wine caused the death of one person, and the illness of several who had partaken of it. The sentence having been annulled on the appeal of the condemned to the Superior Council of Alsace, André and his wife were ultimately ordered to be led by two sergeants for one day through the streets of Bergheim, carrying placards both before and behind, with '*frelateurs de vin*' printed thereon. They had also to pay 30 livres fine, "*pour faire prier Dieu pour le repos de l'âme du défunt*," and the fine of 130 livres, pronounced by the first judge. The council promulgated a very severe decree directed against such practices.

It was also forbidden to adulterate wine with litharge, Indian wood, isinglass, '*raisin de bois*,' or other drugs, or mixtures capable of injuring the health of those who drank the wine, under a penalty of 500 livres and corporal punishment. Even the possession of matters likely to be used for adulteration was an offence. So late as 1710, one Denys Porcher and his wife were convicted of conveying barrels of '*vin de raisin de bois*' into Paris. They were fined 30 livres, the four barrels of wine were spilt on the pavement, and the sentence placarded in Paris and various places around.

§ 10. *Butter*.—An *Ordonnance*¹ of the provost of Paris, dated November 25, 1396, forbade the colouring of butter with 'saucy flowers,' other flowers, herbs, or drugs. Old butter, likewise, was not to be mixed with new, but the sale was to be separate, under penalty of confiscation and fine.

The ancient laws of the merchant butter-sellers and fruiterers, confirmed in 1412, reiterated the above, and also forbade the sale of butter in the same shop in which fish was sold. The retail or sale of butter by spicers, chandlers, apothecaries, and generally by all carrying on offensive trades, was made illegal. A subsequent enactment in 1519 confirmed this law.

§ 11. *Drugs*.—The drug-sellers were also under regulation, and without doubt their practices, with regard to sophistication, were quite on a par with those of other trades. Gargantua in Paris is made to visit the shops of druggists, herbalists, and apothecaries, where he "diligently considered the fruits, roots, leaves, gums, seeds, the grease and ointment of some foreign parts, as also how they did adulterate them—*i.e.*, all the said drugs."²

In the Middle Ages the French apothecaries were at first confounded and amalgamated, as in England, with the merchant spicers; but in 1777 the two trades were separated, and they formed a definite body. In the fifteenth century the shops were little more than open booths, as may be

¹ *Traité de la Police*, tome i. livre iv. titre ix.

² *Rabelais*, cxxiv. p. 53.

seen from a miniature in '*Le Régime des Princes*,' a manuscript of the fifteenth century, preserved in the Arsenal Library, Paris.

Philip VI., as early as 1336, issued a regulation by which no one could be an apothecary unless he was a born or naturalised Frenchman, and a good Catholic. According to the law, neither the spicers nor the apothecaries were permitted to employ in the preparation of their medicines, drugs, confections, conserves, oils, or syrups, any sophisticated or exhausted or corrupted drugs, under penalty of a fine of fifty livres, and the seizure and burning of the merchandise thus adulterated in front of the dwelling in which it had been found.

Charles VIII. released the apothecaries from some of the strict regulations of earlier times, and both he and his successors were the authors of many edicts relative to the apothecaries and spicers; besides which these trades were regulated by local enactments in different towns.

§ 12. *Conseils de Salubrité*.—In 1802 the '*Conseil de Salubrité*' was established in Paris. It originally consisted of only four members, and took cognisance of adulteration, epizootics, unhealthy trades, and a little later of the administration of prisons and public charities. Afterwards the *Conseil* had the direction, generally speaking, of public hygiene.

The *Conseil de Salubrité* of the Seine in its later development was composed of fifteen titular and six supplementary members, including also several honorary members, with others, who, by virtue of their office, were members of the committee. These were, the Dean, the Professors of Hygiene, of Legal Medicine, and of the Faculty of Medicine, a Member of the '*Conseil de Santé des Armées*,' the Director of the School of Pharmacy, the General Secretary of the Préfecture of Police, the Inspector-General of the bridges and causeways, besides engineers, architects, and the chiefs of the police departments. Most of the provinces followed the example of the capital, and established '*Conseils de Salubrité*.' All these boards, whether provincial or Parisian, had one essential feature in common—viz., that the medical, veterinary, and chemical professions were always represented on them. Whatever expert a town possessed would probably have a voice, and find a seat in the '*Conseil de Salubrité*.' From these health boards, or committees, very excellent reports have emanated, and they continue at the present time to do useful work.

III.—ADULTERATION IN GERMANY.

§ 13. If we turn to the records of Germany, we find that all those who adulterated foods or drinks in the Middle Ages were punished severely, with painful and dishonouring penalties, such as public exposure of the fraud and whipping at the gate. The earliest regulations¹ related more to the goodness of the work and the general quality of the goods produced than to adulteration. Every considerable trade was a little corporation, and bad workmanship or falsity in the goods offered was an offence against the guild itself; the member was consequently expelled or punished by

¹ A small work, *Der Kampf gegen die Lebensmittelfälschung von Ausgang des Mittelalters zum Ende des 18. Jahrhunderts*, by L. Wassermann, Mainz, 1879, contains some very interesting particulars with regard to the regulations in practice in the Middle Ages, both as to General Hygiene and the Adulteration of Food.

the officers of the guild. For example, in 1272 the two sworn masters of the bakers' guild at Berlin were held responsible for seeing that good bread was baked. The tailors of Berlin and the bakers of Basil excluded a man for ever from their respective guilds if guilty of bad workmanship (1333). The Berlin weavers, not quite so severe, excommunicated the offender against their regulations for one year (1295). The false butcher at Augsburg (1276) was expelled from the city for a month. In Nuremberg almost everything was regularly inspected; there was a 'Bäckerschau,' a 'Safranschau,' and a 'Schau,' with regard to brandy, drugs, syrup, hops, roses, tobacco, iron, meat, salt-fish, honey, leather, and many other things. It was at a 'Safranschau' in 1444, that one Jobst Fendeker was burnt, together with his false saffron, and in the following year two men and a woman were buried alive there for the same offence.¹

In all the cities of Germany there were copious regulations with regard to three things—bread, wine, and drugs.

§ 14. *Bread.*—In Nuremberg, in the fifteenth century, the baker was not allowed to mix the different kinds of corn, which must be baked separately. In Augsburg, it would appear that there were no less than six different kinds of bread.² The punishments for offending bakers were various. In some places the delinquent was put in a basket at the end of a long pole and ducked in a muddy pool, similar treatment to that which in England befell 'the Scolds.'

§ 15. *Wine.*—According to an old Augsburg chronicle, it was in 1453 that the adulterated wine of the Franks first appeared in that city; but there is abundant evidence to show that wine had been tampered with previously, and in 1390, one Ludwig von Langenhaus was sentenced to be led out of the city with his hands bound and a rope round his neck, because of his practices in the adulteration of wine. In 1400 two wine-sellers were branded and otherwise severely punished, and about the same period a special law enacted forbidding the sulphuring of casks, the colouring of wine, or the addition of sugar, honey, or other sweet things. In the year 1435, says the old chronicle, "were the taverner Christian Corper and his wife put on a cask in which he had sold false wine, and then exposed in the pillory. The punishment was adjudged because they had roasted pears, and put them into new sour wine, in order to sweeten the wine. Some pears were hung round their necks like unto a Pater-noster." It further appears that they narrowly escaped being burnt.

In 1451 the city of Cologne made a strong representation to the governing body at Antwerp on the prevalent adulterations of wine. At Biebrich on the Rhine, in 1482, a falsifier of wine was condemned to drink six quarts of his own wine. He died from the effects.³ In the fifteenth century at Ulm, every tavern keeper had to appear at stated times before the sheriff (*Stadtrechner*⁴) and swear that neither he, his wife, his children, nor any one else in his name, had mixed with the wine, woad, or extract of woad, chalk, mustard-seed, clay, 'Scharlachkraut,' must of apples, lead, mercury or vitriol; no water might be added, and the same wine was to be retailed as bought. The *Stadtrechner* had also to

¹ Henry II. of France enacted that if saffron was adulterated, the offenders should be punished by corporal chastisement, the drugs confiscated and burnt.

² Maurer, *Geschichte der Stadtverfassung in Deutschland*, Bd. III., s. 24.

³ In some places in very early times, the regular legal penalty was capital punishment; for example, in 1269, the law of Ripen punished the seller of false honey and wax with death.

⁴ Perhaps 'Stadtrechner' might be translated 'Mayor.'

see that no sour, ropy, or otherwise bad wine was sold. In the same town an *Ordonnance* of 1499 decreed, that since adulteration was most readily practised by putting substances into the cask, no cask was henceforth to be closed up save by the sworn cooper, who, on finding anything amiss, was to give information; penalty for default, a guilder.

In the fourteenth century Nuremberg was a great centre of the wine trade; consequently in that city there were very many regulations against adulteration of wine, but they were similar to those already mentioned. At Frankfort-on-the-Maine, on false wine being found, the cask was placed on the knacker's cart, and a red flag displayed, with the inscription '*stummer Wein*,' that is, mute or dumb wine. The jailer marched before, the rabble after, and when they came to the river they broke the cask and tumbled the stuff into the stream.¹ About the same date, wine is said to have been sophisticated with the following substances: earth, eggs, albumen, argol, mustard, salt, burnt salt, sweet milk, brandy, almond milk, wheat flour, clay, and several other things.

In the early part of the seventeenth century, a circumstance happened in Wurtemberg which led to some stringent regulations with regard to metallic contamination of wine. A number of people having been seized with colic, paralysis, and other symptoms of poisoning by lead, it was noticed that all those attacked drank one particular species of wine only; and on investigation the epidemic was discovered to be due to the contamination of the wine by the use of metallic fastenings to the casks. The occurrence is related by Gockelius,² who styles the disease '*Weinkrankheit*.'

§ 16. *Drugs*.—Those who sold drugs, roots, spices and the like, were strictly supervised, and in the reign of Frederick the Second of Prussia the examination of drugs was made a special calling, the inspectors being appointed by the king.

In Augsburg, Frankfort, and a few other places, the trade in medicine was taxed. By virtue of this tax the druggists and doctors enjoyed a monopoly, and medicines were forbidden to be sold by other trades.

In the seventeenth century there were committees of doctors, whose duty it was to inspect the druggists, and from the committees—as in London, so on the Continent—originated the pharmacopœias. Thus *Pharmacopœia Antwerpiensis*, 1661; *P. Utrajectina*, 1664; *P. Amstelodamensis*, 1668; *Antidotarium Bononiense*, 1674; *Regia Chemica et Galenica, Geneva*, 1684, etc.³

IV.—HISTORY OF ENGLISH LEGISLATION IN REFERENCE TO THE ADULTERATION OF FOOD.

§ 17. The first General Act in this country was the Act of 1860, previous to which date individual articles, such as tea, coffee, chicory, beer,

¹ The article for sale was sometimes merely forbidden to be sold: this was especially the case with importations from other countries. For example, at Cologne, in 1483, casks of butter imported from England, which were found to contain a mixture of old and new, were not allowed to enter the market. In like manner false oil was excluded from the city.

² *Beschreibung der Weinkrankheit*, 1637.

³ The dukes of Saxony regulated the trade of druggist as early as 1607. J. Guillaume published *Règlements entre les médecins et les apothicaires pour la visite des drogues*, Dijon, 1605. Thomas Bartolin edited the work of Licetti Benanci: *Declaratio fraudum quæ apud pharmacoepos committuntur*, Franc. 1667 and 1671, 8vo.

and wine were legislated for by special statutes, the object of which was, for the most part, to prevent the defrauding of the revenue; the health of the purchaser, and the injury done to him, being somewhat less considered, although not lost sight of.

§ 18. *Bakers—Bread.*—An Act in the reign of George IV. was directed particularly against the use of alum. Bakers, either journeymen or masters, using alum, and convicted on their own confession or on the oath of one or two witnesses, were to forfeit a sum not exceeding £20, and not less than £5; if beyond the environs of London, a sum not exceeding £10, nor less than £3. If within London or its environs, the justices were allowed to publish the names of the offenders. The adulteration of meal or flour was punishable by a like penalty, and loaves made of any other grain than wheat were to be distinguished by a large M. The possession on his premises by a miller, baker, etc., of any ingredient adjudged to have been placed there for the purposes of adulteration was punishable by fine.

An Act passed in 1836 (6 and 7 Will. IV., c. 37) relative to bread may be considered as a modern development of the old 'Assize of Bread.' It repealed the several Acts relating to bread sold out of the city of London and the liberties thereof, and beyond the weekly bills of mortality and ten miles from the Royal Exchange, and provided other regulations for the making and sale of bread, and for preventing the adulteration of meal, flour, and bread beyond the limits aforesaid. This Act made it lawful for the bakers to make bread of "wheaten flour, barley, rye, oats, buckwheat, Indian corn, pease, beans, rice or potatoes, or any of them, and with any common salt, pure water, eggs, milk, barm, leaven, potato or other yeast, and mixed in such proportions as they think fit, and with no other ingredient or matter whatever." They were also permitted to make the bread any weight or size they chose. The bread, however (sect. 4), was to be sold by weight, and in no other manner.

Section 3 provided that no baker or other person within the limits prescribed . . . "shall use any mixture or ingredient whatever in the making of such bread other than, and except as hereinbefore mentioned, on any account or under any colour or pretence whatsoever"; penalty not less than £5 and not more than £10. In default of payment the offender could be imprisoned for a period of not more than six months, with or without hard labour,¹ or unless the penalty was sooner paid, the magistrates could also publish the offender's name, and defray the expense of such publishing out of the fines.

By section 9 no person is to put into any corn, meal, or flour, at the time of grinding, dressing, or manufacturing, any ingredient or mixture whatsoever, not being the real and genuine produce of the corn or grain; nor is flour of one sort or corn to be sold as flour of another sort. Penalty not less than £5 and not more than £20.

Sections 11 and 12 provide that bread made partially or wholly of pease, beans, or potatoes, or of any other sort of corn or grain other than wheat, is to be marked with a large Roman M, under a penalty not exceeding 10s. Justices may issue a search warrant to enter a baker's premises at reasonable hours, and search for adulterated bread or substances used for adulteration. If such substances have been found, the justices may dispose of them as they think fit. The penalty for a first offence is a fine not exceeding £10 and not less than £2; for the second offence £5, and

¹ In the case of *Cobe v. James*, 41 L. J. M. C. 19, it was held that for a conviction under this section guilty knowledge was necessary.

for every subsequent offence £10. In default of payment, imprisonment for not more than six calendar months. It is also lawful for the magistrates to publish the name of the offender. The Act contains various other provisions with regard to obstructing search, offences committed by a journeyman or servant, the baking of bread on the Lord's Day, etc.

§ 19. *Beer and Porter*.—An Act, passed in 1816 (56 Geo. III., c. 58), enacted that no "brewer of, or dealer in, or retailer of beer," shall receive or have in his possession, or use or mix with any worts or beer any molasses, honey, liquorice, vitriol, quassia, cocculus indicus, grains of paradise, guinea pepper, or opium, or any article or preparation whatever for or as a substitute for malt or hops. If any person contravene this provision, the officer of the excise may seize the worts and beer, together with the casks containing the same."—Penalty, £200. By the same Act, druggists who sold colouring materials or malt substitutes to brewers were liable to a penalty of £500. An Act (7 and 8 Geo. IV., c. 52) provided that any brewer, having in possession or in use substitutes for malt or hops, or for darkening the colour of beer, was liable to a penalty of £200, and the ingredients, beer, worts, etc., might be seized. Later on, by the 10 and 11 Vic., c. 5, brewers were allowed to make for their own use a colouring matter out of sugar.¹

§ 20. *Wines*.—An old statute (51 Hen. III., st. 6) forbade the use of unwholesome wine or meat. By the statute 12 Charles II. c. 25, any adulteration of wine was made punishable, with the forfeiture of £100 if done by the wholesale merchant, and £40 if done by the vintner or retail trader. Additional regulations were made by I. William and Mary. All of these Acts, it is scarcely necessary to say, are now obsolete and repealed.

§ 21. *Tea*.—An Act was passed in 1723 (11 Geo. I., c. 30), which enacted that "no dealer in tea, or manufacturer or seller thereof, or pretending to be, shall counterfeit tea, or adulterate tea, or cause or procure the same to be counterfeited or adulterated, or shall either fabricate or manufacture tea with terra japonica, or with any drug or drugs whatsoever, nor shall mix, or cause or procure to be mixed with tea any leaves other than leaves of tea, or other ingredients whatsoever, on pain of forfeiting and losing the tea so counterfeited, adulterated, altered, fabricated, manufactured, or mixed, or any other thing or things whatsoever added thereto, or mixed or used therewith, and also the sum of £100."

Some years afterwards, 1730-31, a further Act was passed (4 Geo. II., c. 14), prescribing a penalty for what is termed in the statute 'sophisticating tea.' It recites "that several ill-disposed persons do frequently dry, fabricate, or manufacture very great quantities of sloe leaves, liquorice leaves, and the leaves of tea that have been before used, or the leaves of other trees, shrubs, or plants in imitation of tea, and do likewise mix, colour, stain, or dye such leaves, and likewise mix tea with terra japonica, sugar, molasses, clay, logwood, and with other ingredients, and do sell and vend the same as true and real tea, to the prejudice of the health of His Majesty's subjects, the diminution of the revenue, and to the ruin of the fair trader." The penalty under this statute was £10 for every pound of tea sophisticated.

The next Act was passed in 1776 (17 Geo. III., c. 29). The preamble asserts that great quantities of sloe leaves and the leaves of the ash, elder, and other trees, shrubs, and plants, were manufactured in imitation of tea,

¹ At the present time it is not illegal to mix bitter beer with wholesome bitters other than hops.

and were then sold to dealers in tea, who, after mixing the leaves with tea, sold it as true and real tea; but as the persons who fabricated or manufactured the leaves were not dealers in tea, they were not punishable by the law then in force. The Act, therefore, rendered any person, whether a dealer or not, who fabricated leaves in imitation of tea, or who mixed tea with other ingredients, or who sold, exposed for sale, or had in his custody fabricated or mixed teas, liable on conviction to a penalty of £5 for each pound of such tea, or in default to imprisonment for not less than six months, nor more than twelve. The officers of the excise were empowered to enter under warrant any premises by day or night and seize the leaves, which, on a further warrant, were to be destroyed.

§ 22. *Coffee*.—The history of the regulations with regard to coffee and chicory is rather curious, inasmuch as coffee appears to have been adulterated almost immediately after its introduction, and legislative interference was soon necessary.

The first Act (5 Geo. I., c. 11, 1718), with regard to coffee, recited that "Whereas divers evil-disposed persons have at the time of, or soon after, the roasting of coffee made use of water, grease, butter, or such-like materials, whereby the same is rendered unwholesome, and greatly increased in weight to the prejudice of His Majesty's revenue, the health of his subjects, and the loss of all honest and fair dealers in that commodity," and went on to enact that "any person or persons whatsoever, who shall at the roasting of any coffee, or before, or at any time afterwards, make use of water, grease, butter, or any other material whatsoever, which shall increase the weight, or damnify or prejudice the said coffee in its goodness, he, she, or they shall forfeit the sum of £20 for every such offence." This penalty was increased to £100 by a subsequent Act (12 Geo. I., c. 30), passed in the year 1724.

An Act passed in 1803 (43 Geo. III., c. 129) ordered that "If any burnt, scorched, or roasted peas, beans, or other grains, or vegetable substance or substances be used, prepared, or manufactured for the purpose of being an imitation of, or in any respect to resemble coffee or cocoa, or to serve as a substitute for coffee or cocoa, or alleged or pretended by the possessor or vendor thereof so to be, shall be made or kept for sale, or shall be found in the custody or possession of any dealer or dealers of coffee or cocoa; or any burnt, scorched, or roasted peas, beans, etc., not being coffee or cocoa, shall be called by its preparer or manufacturer, possessor, or vendor thereof, by the name of coffee, or by the name of American cocoa, or English or British cocoa, or any other name of cocoa, the same respectively shall be forfeited, and, together with packages containing the same, shall or may be seized by the excise." The person convicted was to be fined £100.

A subsequent Act (3 Geo. IV., c. 53) permitted persons, not dealers in coffee, to sell roasted corn, peas, beans, or parsnips whole, but not ground, crushed, or powdered.

In 1832 grocers were allowed by the Excise to keep chicory on their premises, and a Treasury Minute, dated August 4, 1840, allowed the sale of coffee mixed with chicory, a step which no doubt opened the way to wholesale adulteration. This is evident from a meeting of those interested in the coffee trade, held at the London Tavern, on March 10, 1851, in which the chairman¹ explained that although more of what was called coffee was now consumed, yet that there was a less consumption

¹ T. Baring, Esq., M.P.

of *genuine coffee*. "We wish," he continued, "to come to the real question, and we desire that it should be publicly understood that what is coffee be sold as coffee, and that what is not coffee, being a cheaper article, and, if you will, a more nutritious article, and as eligible for consumption, be sold to the consumer at the price at which it can be afforded." A grocer from Shoreditch having produced at the meeting a compound of burnt peas, dog-biscuit, prepared earth, and a substance "which," he said, "I shall not describe, because it is too horrid to mention," went on to affirm that several tons of the same material were in existence, and that it was used as a substitute for chicory and for snuff.¹

The *Lancet* also gave details about the same time of the microscopical examination of thirty-four samples of chicory, nearly one-half of which were mixtures, the substances found being roasted beans, burnt corn, and acorns. It was under the protection of this Minute that Messrs. Duckworth of Liverpool took out a patent for the compression of mixtures of chicory and coffee into the shape of berries. Popular writers have, as usual, made the most of this patent, and the story has been retailed with additions from one book to another as a glaring instance of wholesale fraud; but, although the purity of the manufacturers' intentions may be open to doubt, the fact remains that they did nothing against the existing law. The patent does not appear to have been profitable, and but few of the chicory berries were put in circulation.

The subject of coffee adulteration was not, however, permitted to escape the attention of Parliament, and petitions from planters, growers, and others interested in the sale or production of coffee increased in number. In the Commons, during the course of a long debate (April 14, 1851), Mr. T. Baring stated that it could not be denied that there had been a diminution since 1847 in the consumption of coffee to the extent of six million pounds, the real cause of which was the wholesale admixture of coffee with chicory—this chicory of home growth. In 1840, at the time of the issuing of the Treasury Minute suspending the law as regarded that article, all the chicory used in the country came from abroad, and as an excisable import on which duty was paid, but since the issue of the Minute it had been cultivated largely in England.

Similar statements were made in the House of Lords (July 3, 1851), on the occasion of Lord Wharncliffe's presenting a petition to that House. In the following year, under the pressure of popular feeling on the subject, the objectionable Minute was rescinded, and a new Treasury Minute, dated July 27, 1852 (which was embodied in a subsequent order of the Inland Revenue Commissioners), permitted licensed dealers in coffee to "keep and sell chicory and other vegetable substances prepared to resemble coffee, provided that they be sold unmixed with coffee, in packages sealed or otherwise secured, containing respectively not less than two ounces, and having pasted thereon a printed label with the name of the seller, the exact weight and true description of the article contained therein, and provided that no such article be kept in a loose state, or otherwise than in such packages as aforesaid, in any room entered for the storage or sale of coffee." This regulation was, without doubt, irksome both to traders and consumers, since every one who desired his coffee mixed with chicory could not buy the mixture prepared,

¹ *Adulteration of Coffee. A verbatim report of the proceedings at a public meeting held at the London Tavern.* London, 1851.

but was obliged to purchase the coffee and chicory separately, and compound it himself. Hence many memorials were presented praying "That the sale of a mixture of coffee and chicory be not interfered with, provided each package has legibly printed thereon words plainly indicating such mixture." In consequence of these representations this Minute was also rescinded,¹ and a new one prepared. An order of the Commissioners of Inland Revenue, dated May 13, 1853,² followed, requiring that on every package containing a mixture the words, "This is sold as a mixture of chicory and coffee," be printed in capital letters of Roman character, of at least one-eighth of an inch in height, on the outside of the packages or canisters, on the same side of which there was to be no other printing or writing. On no other part of the package, further, was there to be any other writing save the name and address of the seller.

It would be a great error to suppose that these minutes of the Treasury, and subsequent orders of the Revenue Commissioners, had for their leading object the prevention of adulteration in its reference to the health of the subject. It will at once be noticed that they only touched on 'excisable articles,' and it was entirely a fiscal question. In a word, had the sophistication been of such a nature as to increase instead of diminish the revenue, the Treasury would have let it pass without notice.

§ 23. *The Select Committee of 1855.*—The prelude to legislation on adulteration as a whole was the appointment of 'A Select Parliamentary Committee,' which entered on its labours in 1855. The early appointment of this committee was, without doubt, due to the influence of the late Mr. Wakley, the able and courageous editor of the *Lancet*. In 1850 Mr. Wakley had established, in connection with his powerful journal, '*The Lancet Sanitary Commission*,' of which commission Dr. Hassall was the leading spirit, with Dr. Letheby as occasional coadjutor in matters purely chemical, and (what at that time was of great importance) with the assistance of an artist, who drew microscopical objects with fidelity.

The 'Analytical Sanitary Commission' was commenced in the first number of the *Lancet* for 1851, and the scope of the inquiry, as stated by the editor, was as follows:—"We propose, for the public benefit, to institute an extensive and somewhat rigorous series of investigations into the present condition of the various articles of diet supplied to the inhabitants of this great metropolis and its vicinity. . . . Special features of the inquiry will be that they are all based upon actual inquiry and experiment; the microscope and the test tube will be our constant companions." Notice was also given that at the expiration of three months the names and addresses of the shopkeepers from whom purchases had been made would be given; but at the commencement the street alone was to be indicated. The promise was kept, and hazardous although the experiment most certainly was, yet in April we find the names of large firms freely published, and, so to speak, 'pilloried,' for having sold impure and false goods.

In 1855 Dr. Hassall collected the articles which had been published in the *Lancet* into a volume, entitled *Food and its Adulterations, comprising the reports of the Analytical Sanitary Commission of the 'Lancet' for the years 1851-54*. London, 1855.

In 1855 'The Select Committee on the Adulteration of Food' commenced its labours, and examined as far as possible all those who were

¹ Parl. Paper, No. 165, vol. xcix., sess. 1852-53.

² Parl. Paper, No. 508, sess. 1854-55.

likely to have any special knowledge of the adulterations themselves, the methods necessary to detect them, and their effect on the revenue and on health. Dr. Hassall stated to the committee the results of his inquiries both for the *Lancet* Commission and during the course of his other labours, and gave in detail the frauds practised in regard to milk, coffee, tea, drugs, preserved fruits, etc.

Dr. Alphonse Normandy, who had also written a work on adulteration—the result of ten years' investigation—said, in giving evidence as to the aluming of bread, that he had seen alum in bread in crystals of the size of a large pea. "In the bread of one baker I found alum actually in the state of large crystals; I went to him and showed him his bread, and he said, 'I cannot help it.'" In extreme instances he had found as much as from 250 grains to twice that quantity of alum in the 4lb. loaf, and in 1847 he had found magnesia carbonate in three samples of bread. In 1847 and 1848, years of great scarcity, he had discovered bean and pea meal in flour, but this he considered quite exceptional. With regard to beer, he thought that brewers often made use of *cocculus indicus*; and, finally, he gave evidence of the great adulteration of drugs.

Mr. Blackwell, of the firm of Crosse & Blackwell, gave some very interesting evidence as to the 'coppering of preserved vegetables,' practised before the food articles appeared in the *Lancet*. The process in use by his firm was to boil the pickles or vinegar several times in copper boilers. After each operation they became greener, and when the proper hue was attained, the process was finished; but since the outcry on coppered vegetables, this process had been abandoned.

Another witness, Mr. O. L. Simmonds, the author of a work upon 'Commercial Products,' in giving evidence on the adulteration of drugs, estimated that there was a loss to the revenue from this cause of no less than £3,000,000 per annum. As an instance of the manner in which the revenue suffered, he cited the substitution of cassia for cinnamon; cassia paid 1d. per lb. duty, cinnamon 2d. The dealers sold cassia under the name of cinnamon to such an extent as to affect seriously the cinnamon trade.

§ 24. *Adulteration Acts, 1860 and 1872.*—Upon the report of the Select Committee, the first general Adulteration Act was drafted, and became law in 1860. The first section enacted, "That every person who shall sell any article of food or drink with which, to the knowledge of such person, any ingredient or material injurious to the health of persons eating or drinking such article, has been mixed, and every person who shall sell as pure or unadulterated any article of food or drink which is adulterated or not pure, shall for every such offence, on summary conviction of the same, pay a penalty not exceeding £5, with costs." A second offence was punishable in addition by publishing the offender's name, place of abode and offence. The Act permitted, but did not compel, the appointment of analysts. The bodies which might appoint such analysts were—in the City of London, the Commissioners of Sewers; in the metropolis generally, Vestries and District Boards; in the counties, Courts of Quarter Sessions. Section 4 provided that any purchaser of any article of food in any of the districts in which analysts existed might have such article analysed on payment of a sum not less than 2s. 6d. and not more than 10s. 6d.; the purchaser, on the completion of the analysis, was entitled to receive a certificate of the result of the analysis.

These appointments were at first confirmed by the Secretary of State,

but afterwards the Local Government Act of 1871 transferred the regulation of the appointments to the Local Government Board. The Act existed, and was in partial operation for twelve years, when it was entirely recast and interspersed with various sanitary considerations.

In an Act passed in the year 1872 (35 and 36 Vict., c. 74), it was enacted that "Every person who shall wilfully admix, and every person who shall order any other person or persons to admix, any ingredient or material with any drug to adulterate the same for sale, shall be liable to a penalty for the first offence not exceeding £50, with costs." The second offence was punishable by a term of imprisonment not exceeding six months, with hard labour. By the second section, "Every person who shall sell any article of food or drink, with which to the knowledge of such person any ingredient or material injurious to the health of persons eating or drinking such article has been mixed, and every person who shall sell as unadulterated any article of food or drink or any drug which is adulterated, shall for every such offence, on a summary conviction of the same, pay a penalty not exceeding £20, with the costs of conviction." By the third section, "Any person who shall sell any article of food or drink, or any drug, knowing the same to have been mixed with any other substance with intent to fraudulently increase its weight or bulk, and who shall not declare such admixture to any purchaser thereof before delivering the same and no other, shall be deemed to have sold an adulterated article of food or drink, or drug, as the case may be, under this Act."

The Act, with doubtful advantage, also extended the right of appointing analysts to boroughs having separate police establishments. The appointment was optional, save on the direction of the Local Government Board. The sixth section provided that inspectors of nuisances or other local officers were to procure samples for analysis. Private purchasers might have articles analysed as before, the only difference being that, under this Act, they were to hand the substance, not to the analyst, but to the inspector. There were also provisions as to the sealing and division of samples. Since the Act of 1860 remained unrepealed, the two Acts were both in force simultaneously, and under their joint operation the following offences were punishable¹ :—

1. Selling any article of food, drink, or medicine, that contains any ingredient injurious to health, and knowing it to contain such ingredient.
2. Selling any adulterated food, drink, or drug.
3. Wilfully mixing with any article of food or drink any ingredient or poisonous ingredient to adulterate the same for sale.
4. Wilfully mixing any ingredient with any drug to adulterate the same for sale.
5. Selling any article of food, drink, or any drug, knowing the same to have been mixed with other substances with intent fraudulently to increase its weight or bulk, unless such admixture be declared at the time of sale.

§ 25. *The Select Committee, 1874.*—These Acts by no means worked well. Many of the analysts were inexperienced, and even those who had considerable chemical knowledge differed widely in the conclusions they drew from their analyses. The reason of this was evident, for the standards had scarcely been settled. There was, for example, no general agreement as to the amount of 'fat' and 'total solids' in milk; the question of

¹ *The Law of Adulteration*, by Sidney Woolf. London, 1874.

whether tea should be permitted to be faced, or not, was then (as, indeed, now) unsettled; there was no method in use which distinguished alum added to flour and alumina existing as sand. Analyst contradicted analyst. Magistrates were perplexed as to the meaning of the word 'adulteration,' and conflicting decisions on mere legal technicalities offered a still further obstacle to the healthy operation of the Act. The public generally were dissatisfied with an Act which inflicted real hardships on many retail dealers—*e.g.*, tea, paid for at the highest market price, and imported direct from China, would be examined by a local analyst and pronounced to be *faced* with Prussian blue, gypsum, etc.; while, from the peculiar nature of the statute, the seller, however innocent of the fraud himself, could not defend the charge on anything like equal terms. Petitions, moderate in tone, came in from most of the large towns, and the Government decided to appoint another Select Committee. A large number of witnesses—tea merchants, tea brokers, tea retailers, butter merchants, cocoa and coffee manufacturers, milk sellers, bakers, and analysts—were examined by this new Committee in 1874; and on their evidence a report was based, which stated that after having sat fourteen days, and examined fifty-seven witnesses, the Commissioners had arrived at the unanimous conclusion that, while the Act had done much good, it had, at the same time, inflicted considerable injury, and enforced heavy and undeserved penalties upon some respectable tradesmen. "This appears to have been mainly due to the want of a clear understanding as to what does, and as to what does not, constitute adulteration, and in some cases to the conflicting decisions and inexperience of the analysts. Your Committee are, however, of opinion that the Act itself is defective and needs amendment."

The report went on to say that the adoption of the Act had been by no means general, and in many cases where it had been applied its operation was of a very restricted character; for, even with competent analysts, if inspectors were not appointed at the same time the Act remained a dead letter. All the London vestries had made appointments, but in only twenty-six out of seventy-one boroughs, and thirty-four out of fifty-four counties, were there at that date official analysts. The examination of tea was recommended to be made on importation by the Customs. The Committee did not consider that the exact proportion of mixtures need be stated on a label, and they wished to record that mixed mustard and prepared cocoa had been long manufactured at Deptford for the supply of the Navy. They recommended that small districts should be consolidated, and that, as a rule, the boroughs in a county should be united with the county for the purpose of appointing one analyst for the entire district; and they pointed out that the only way to secure "the services of really efficient analysts is to offer them a fair remuneration, which can hardly be done without the union of several Local Authorities in one appointment." The Committee concluded their report by remarking that the public was 'cheated' rather than 'poisoned.'

§ 26. *Sale of Food and Drugs Act*, 1875, 1879, and 1899.—On this report was based the Act of 1875, which is at the present moment, with its amendments of 1879 and 1899, the existing law, and the full consideration of which will be reserved for another Section; an early defect in the Act, however, may be at once alluded to, for it had not been long in operation before its action was almost entirely stopped by legal ingenuity. The sixth section provides that "No person shall sell to the prejudice of the purchaser any article of food or any drug which is not of the nature,

substance, and quality of the article demanded by such purchaser"; and in a Justiciary Appeal case at Edinburgh, in which an inspector had purchased cream not for his own use but for analysis, the Scotch court discussed the 'prejudice' question—three out of seven judges adopting the view that a purchase made under these conditions was not to the 'prejudice' of the purchaser, and five out of the seven dismissing the summons on other grounds. The impression produced in this country, however, by the decision of the court, was that the sale, to be effectual, must be made in the ordinary way, and not merely for the purposes of analysis. The same question was raised in quite a different but equally ingenious way in a 'mustard case' argued before the Court of Queen's Bench. The purchase in this case was by an officer; the defence being that, as it was well known that mustard was mixed with flour and other things, such a purchase could not be to the prejudice of the purchaser. The point, however, was left undecided; the question again came before the Court of Queen's Bench, in the case of *Sandys v. Small*, and the 'prejudice' question was argued on both points. Whisky was alleged to have been mixed with water, and the defence set up—that it was known to be so mixed, and therefore not to the prejudice of the purchaser—was held by the court to be good, and the case having been decided upon this point, the other was not proceeded with. Finally the question was settled by the case of *Hoyle v. Hitchman*, March 27, 1879. The facts in this instance were of the simplest character: the appellant had purchased milk in the usual official way; the milk was found to be adulterated, and the defence was that, as he did not use the milk, therefore he did not buy the milk for his own use; he was not prejudiced. The magistrate who heard the case considered the defence good, and dismissed the summons.

Justice Mellor, in giving judgment, observed that the 'prejudice' view of the Act "would absolutely nullify its beneficial effect. For if the meaning of the enactment is that the offence cannot be complete without its being 'to the prejudice of the purchaser,' it is hardly possible that the offence should be brought home to any one. And this observation, in my view, goes far to show that this construction cannot be the right one. So far as authority is concerned, there is no direct decision in favour of such a view; and, indeed, in the English courts there is hardly any authority upon the point. For in the first of the two cases in this court referred to, the mustard case, my brother Lush distinctly said that, in his view, if the article were adulterated, it must be presumed that it was 'to the prejudice of the purchaser,' and I could not have dissented from that opinion, or I could not have concurred in sending the case down to be re-stated on the other point. And as to the other case, no doubt in the course of the argument the Lord Chief-Justice made some such remark, but not by way of a decided *dictum*, and rather by way of query or suggestion, and the decision went upon the other point, so that there is no authority in the English courts in favour of the view now presented. It cannot be said that the weight of judicial authority is against, and I rather think it is in favour of, the view which we have arrived at after the best consideration given to the question, as to the true construction of the enactment. It is quite general in its terms, and its terms are very large, nor is there anything to limit them,—'if any one shall sell, to the prejudice of the purchaser, any article of food not of the nature, substance, or quality of the article demanded by the purchaser.' There is nothing to limit the application of the enactment (as some of the Scotch judges seem to have

supposed) to articles deleterious in their nature. And in several of the sections (13 to 17) provisions are made for purchases by public officers for the purpose of analysis and prosecution, assuming that, if the article is found to be adulterated, the offence will have been committed. It would be strange indeed if all these provisions were to be made nugatory by a construction which would, in effect, come to this—that proceedings could only be taken by private individuals. Here the purchase was made by the inspector under those sections; but surely the case must be treated as though the purchase had been by a private individual. Now, in the case of a private individual, no one could dispute that in such a case as this the offence would have been completed, and the magistrate has so found, in fact. That being so, what difference can it make as to the nature of the offence, that the purchase was by an officer on behalf of the public, and furnished with public money for the purpose? If the purchaser asks for a certain article, and gets an article, which by reason of some admixture of a foreign article is not of the nature or quality of the article he asks for, he is necessarily 'prejudiced'; and how can the fact that the purchase is not with his own money at all affect the question of the commission of the offence? The offence intended to be prevented by the Act was the fraudulent sale of articles adulterated by the admixture of foreign substances, which would necessarily be 'to the prejudice of the purchaser'; and those words were inserted only to require that such an adulteration should be shown to have been made. Taking all these matters into consideration, I cannot bring my mind to the conclusion that in such a case as this the offence is less complete, merely because the money with which the purchase was made was not the money of the purchaser, which must be wholly immaterial to the seller, and cannot affect the offence he has committed. I come, therefore, to the conclusion that the magistrate was wrong in dismissing the case on that ground, and, therefore, that the case must be remitted to him to be determined on the evidence as to the offence alleged to have been committed."

Mr. Justice Lush, in expressing his entire concurrence, said that the differences of opinion which unfortunately prevailed as to the true construction of the sixth section of the Act had crippled the operation of a most beneficial Act.—Judgment for the appellant.

The Act of 1875 was amended by the 'Sale of Food and Drugs Act, 1879,' which became law on July 21st in that year. This Act settled the 'prejudice' question, authorised the obtaining of samples of milk for the purposes of analysis, and established standards for spirits. (See sections on the 'Existing Law relative to Adulteration.')

V.—THE HISTORY OF THE PRESENT SCIENTIFIC PROCESSES FOR THE DETECTION OF ADULTERATION.

§ 27. If an attempt were made to write the full history of the modern system of the practical assaying of foods, beverages, and drugs, the result would be neither more nor less than a history of the development of the chemical, physical, botanical, and medical sciences; for there has scarcely been a single advance in any one of those sciences which has not some bearing, immediate or remote, on our subject. Hence, the more useful

and less ambitious method to pursue will be merely to notice the chief writings and the more noteworthy discoveries of those who have explored this special field of investigation.

The very early and brief notices in the old writers have been already mentioned. The first general works on adulteration were devoted to drugs rather than to foods, and the herbals and the older works contain here and there, scattered through their prolix pages, casual mention of substitutions or falsifications. For example, Saladin of Ascala, a physician to the Grand Constable of Naples, who wrote in the fifteenth century a work on the aromatic principles of drugs, describes methods of preserving food, and in speaking of the adulteration of manna with sugar and starch, cites the case of an apothecary who was fined heavily and deprived of his civil rights.¹

§ 28. In the early part of the seventeenth century Bartoletus discovered by analysis milk-sugar (see chapter on 'Milk'), and to this epoch belong also some observations and experiments of another Italian, Francesco Redi² of Florence, published in 1660, on the amount of mineral substances in pepper, ginger, and black hellebore. He burnt 100 lbs. of each and weighed the ash: black pepper yielded 5 lbs. 2 oz. 4 drs. of ash, ginger 5 lbs. 3 ozs. 2 drs., while black hellebore burnt in the same quantity gave 4 lbs. of ash. These ash percentages, as we know, are accurate. He treated the ash with water, and noticed that all the salts lixiviated, and had a peculiar and definite figure, which they kept, although they were often dissolved and afterwards crystallised. "If in one liquid you dissolve together two or three sorts of salts of different figure, when they congeal they all resume their ancient and proper figure." He gives examples of this among mineral salts, and further states that if vitriol of cyprus (copper sulphate), rock alum, and nitre be dissolved, on evaporation and crystallisation the different salts can be readily detected.³

§ 29. The Honourable Robert Boyle, whose numerous writings and discoveries are well known to all scientific men, may be said in a way to have written the first treatise the sole object of which was to make known a method of detecting adulterations. The title of his work is *Medicina Hydrostatica; or Hydrostatics applied to Materia Medica, showing how by the weight that divers bodies used in physic have in water, one may discover whether they be genuine or adulterated*, 8vo, Lond. 1690. His method of determining specific gravity was similar to that now used. He determined the specific gravity⁴ of pure rock crystal, which he took as a standard, comparing the specific weight of various minerals with it. He showed that impure mercury sublimate, weighed in this manner, would be deficient, and that Roman vitriol mixed with alum might also be similarly detected. He observed that there were several forms of soluble salts in

¹ This work, *Compendium Aromatarium*, was published in Augsburg, 1481. There is no separate copy in the British Museum, but it will be found as the *Liber Saladinii* in the beautiful folio edition of the Arabic physician (Yūmannā ibn Massawāih), *Joannis mesuæ damasceni medico clarissimi opera, etc., de medicamentorum detectu, castigatione et usu, etc., etc., folio, Venice, 1623*. The work is in the old dialogue style, consisting for the most part of question and answer. The books preceding the *Liber Saladinii* also contain some observations on adulteration.

² Francesco Redi, 1626-1697; he was at once a poet, a chemist, and a physician.

³ *Phil. Trans.*, 1693, p. 281.

⁴ We believe that the oldest tables of specific gravity extant are those in Lord Francis Bacon's *Historia Densit et Rari*, fol., Lond., 1741. A cube of gold was taken as a standard, and cubes of other substances, of a size as exactly similar as possible, were made. He was conscious, however, of the want of complete accuracy.

plants, but always some that were cubical. Boyle also determined the percentage of ash in about forty different vegetables, and the amount of soluble ash. Boyle's was not a work of general scope, for the most part confining itself to the recommendation of a particular although widely applicable method.

An early general work on the adulteration of drugs was that of J. B. Vanden Sande,¹ who may be called the pioneer of applied quantitative chemistry, for he not only described the mere external characteristics of various articles of the *materia medica*, but also made alcoholic and ethereal extracts, and determined the weight of the extracts thus obtained. He also, after the manner of his time, distilled the substances and obtained various products.

§ 30. The invention of the microscope, revealing the most intimate structure of animal and vegetable tissues, and the regular and mathematical forms of salts and minerals, gave a great impetus to all the sciences. Antony Van Leeuwenhoek (b. 1632, d. 1723) was the first who in any philosophic manner occupied himself with this instrument. Gifted with rare powers of observation and manipulative dexterity, he made his own microscopes, and prepared all objects with his own hands. His microscopes were what we should now call lenses. Each object, permanently mounted, had a separate microscope, which merely consisted of a small double convex lens let into a socket between two plates riveted together, and pierced with a small hole. The object was placed, if liquid, on a fine little plate of talc, which was then glued to a needle, or, if solid, was attached to the needle itself. There was a mechanical arrangement by which the needle could be depressed or raised, or placed in any position desired. None of his lenses were very powerful—he rather preferred clear definition; nor were they all of the same magnifying power, but varied according to the nature of the object. He possessed an incredible number of these instruments, and at his death bequeathed many of them to the Royal Society. He investigated daily during his long life all kinds of objects in the three kingdoms of nature, and made perhaps a greater number of discoveries as to minute structures than any other man in his time; indeed, there was scarcely a competitor, for the method was peculiarly his own. These discoveries did not attract so much attention in his day as they deserved, the reason probably being that scarcely any one possessed the suitable means of corroborating his researches. Leeuwenhoek seems to have been the first to discover the active principle of tea and coffee, and to describe the structure of the coffee berry.² Speaking of coffee beans, he says: "I placed some of the beans in a proper chemical vessel over the fire, and observed that, in the roasting or burning them, a great quantity of oily substance and also of watery moisture was expelled. The roasted bean I broke into small pieces, and after infusing these in clear rain water I suffered the water to evaporate after pouring it from the grosser parts of the coffee, and then I discovered a great number of oblong saline particles

¹ *Les falsifications des médicaments dévoilées, ouvrage dans lequel on enseigne les moyens de découvrir les tromperies mises en usage pour falsifier les médicaments tant simples que composés, et où on établit des règles pour s'assurer de leur bonté, ouvrage non seulement utile aux médecins, chirurgiens, apothicaires, et droguistes, mais aussi aux malades*, by J. B. Vanden Sande, Maître Apothicaire de Bruxelles, à la Haye, 1784. A well-printed 8vo of 430 pages. The same author wrote a *Lettre sur la sophistication des vins*, Amsterdam, and one or two other works.

² The authors believe that this (as given in the first edition) is the first notice of the separation of caffeine by Leeuwenhoek.

of different sizes, but most of them exceedingly minute; all of them with sharp points at the ends and dark in the middle."¹ He figures these 'saline particles,' and from the description and the figure they can be scarcely other than crystals of 'caffeine' or 'theine.' He also cut thin slices of the berry, and one of his plates is a very good illustration of the cellular structure of coffee. He noted that "it was of an open and spongy texture . . . and some of the parts, which in the figure appear closed up, consisted of globules, and were filled with oil." Still more decisive are his observations on tea, in which it is absolutely certain that he obtained 'caffeine' or 'theine' by sublimation, for he distilled it and collected the 'volatile salt.' "All these saline particles were of the same shape, that is, very long and pointed at both ends. . . . I afterwards endeavoured, for my further satisfaction, to discover, if possible, how many saline particles could be produced from a single leaf of tea; but having reckoned up only a part of the volatile salts contained in one leaf, I forbore any further observations, because the number I had already reckoned up was so great that I dared not publish it, as I had proposed to do, and indeed many persons could not believe that the leaf itself could be divided into so many parts visible by the microscope, as I saw volatile saline particles produced from one single leaf." He also examined the ash of tea, and noticed its deliquescent character. He separated several distinct salts, of which one kind had small cubical crystals, and was probably common salt. He also turned his attention to pepper, and extracted from it a crystalline principle, probably 'piperine.' He powdered long pepper, and placed it in a glass vessel, covering it with rain water to about one-third of an inch. "After the water had stood about two hours, I poured it off, but it being evening I let the water stand all night. The next morning I saw in the place where it was most evaporated an incredible number of saline particles, many of which were almost twice as long as broad, but one side always longer than the other." He also distilled pepper, and extracted from it an oil. He considered the difference between white and black pepper to be that one was decorticated, the other not, and proved that he was right by direct experiment. In speaking of vinegar, he noticed that it was neutralised by chalk, and that it often contains minute eels—these 'eels' he figures and describes.

To Leeuwenhoek, then, may fairly be accorded the credit of having made several unnoticed discoveries in food analysis. Contemporaneously with Leeuwenhoek, Dr. Hy. Power published some microscopical observations, describing the appearances of sand, sugar, and salt, the eels in vinegar, and the mites in oatmeal. He also observed how easy it was to discover the particles or globules of mercury in compound powders. "In those chymical preparations of mercury which they call 'turbith mineral,' 'mercurum vitæ,' sublimate precipitate, and mercury cosmetical, you may most plainly and distinctly see the globular atoms of current and quick mercury besprinkled all among the powders, like so many little stars in the firmament."² He also notices the minute structure of several leaves, and may be considered, together with Dr. Hooke,³ as the English representative of microscopical science at that date.

¹ *The Select Works of Antony Van Lecurvenhoek*, 4to. Lond., 1798.

² *Experimental Philosophy, in Three Books, containing New Observation Experiments, Microscopical, Mercurial, Magnetical*. London, 1663.

³ Dr. Hooke published his "Micrographia Illustrata" in 1656, in the *Philosophical Transactions*, in which he made known his invention of glass globules applied to the

Microscopical observers rapidly multiplied as the instrument itself was perfected, and by about the year 1825, really good instruments, although not absolutely achromatic, could be purchased. In 1838, Ehrenberg brought out his great folio on *Infusorial Life*. The beauty of the illustrations in this have never been surpassed; they amply prove that very early in the nineteenth century, for those who could afford the expense, there were instruments of great power, precision, and definition.¹

In 1844, Donn ² published his beautiful plates containing, among other things, some accurate representations of the milk corpuscles (see article on 'Milk'). Dr. Ure, in an important case in which an attempt was made to evade the duty on cassava starch by calling it arrowroot, and importing it as such, detected the fraud by the microscopic appearances alone. An excellent collection of objects illustrating the minute anatomy of plants was to be found in 1845 in the Museum of the College of Surgeons, the catalogue of which was edited in an illustrated form by Professor Quekett.³ About the same time, Quekett also delivered several lectures on histology, in the course of which he pointed out the value of the microscope in the detection of fraud.⁴

§ 31. In the latter part of the eighteenth and the beginning of the nineteenth century, chemistry advanced with rapid strides: Neumann Caspar⁵ made various experiments on milk, wine, butter, tea, coffee, and other substances; the Boerhaave School⁶ analysed milk; Berzelius issued his chemical papers; Scheele instituted a variety of researches, and thus the foundation was being laid of those processes which were improved and perfected by the philosophical mind of Liebig, and applied in the analyses of various vegetable products⁷ by Mulder, many of whose methods are still quoted and taken to a certain extent as standard. This advance in chemical science was naturally accompanied by more elaborate and scientific works on food, and for the first time it became possible to study the subject in a philosophical manner, and to apply a variety of processes for the detection of fraud.

microscope, by which an immense magnifying power was obtained. He afterwards published a work entitled, *Micrographia; or, Some Physiological Descriptions of Minute Bodies, made by means of Magnifying Glasses, with Observations and Enquiries thereupon*, by R. Hooke, F.R.S. London, 1765. The work is a folio illustrated with well-executed copper plates. He describes and figures, like Leeuwenhoek, poppy seeds, vinegar eels, etc. He was a man of great ingenuity and celebrity. In a theoretical manner he anticipated the telephone, for in the preface he says: "'Tis not impossible to hear a whisper a furlong's distance, it having been already done, and perhaps the nature of the thing would not make it more impossible though that furlong should be many times multiplied. . . . I can assure the reader that I have, by the help of a distended wire, propagated the sound to a very considerable distance in an instant, or with as seemingly quick a motion as that of light, at least incomparably swifter than that which at the same time was propagated through the air, and this not only in a straight line, or direct, but in one bended in many angles."

¹ *Die Infusionsthierchen als vollkommene Organismen*, by D. Christians Gottfried Ehrenberg. Leipzig, 1838.

² A. Donn : *Cours de Microscope*.

³ *Descriptive and Illustrated Catalogue of the Histological Series in the Museum of the Royal College of Surgeons*, Vol. II., 1850.

⁴ Lectures on Histology, delivered at the Royal College of Surgeons, 1850-1.

⁵ Neumann Caspar, M.D., *Vom Thee, Caffee, Bier und Wein*. Leipsic, 1735. *The Chemical Works of Neumann Caspar, abridged and methodised*, by William Lewis. London, 1773.

⁶ See chapter on 'Milk.'

⁷ *The Chemistry of Animal and Vegetable Physiology, translated from the Dutch*, by P. F. H. Fromberg. Edinburgh, 1845.

§ 32. There was published, in 1820, a work on the adulteration of food, by Frederick Accum,¹ which is sometimes inaccurately referred to by writers of the present day as *Death in the Pot*. Accum, however, wrote no work bearing that title, which belongs properly to a little book by an anonymous writer, to be noticed presently. Accum's work, appearing just at a time when several brewers had been fined heavily for having in their possession illegal substances, and being reviewed most favourably by the press, exercised a very great influence on the public mind—the more so as it was written with considerable ability and knowledge of the subject.

After giving a general review of adulteration, and proving that it was a widespread evil, affecting more or less every industry—that woollen goods were adulterated with cotton, soap with clay, paper with plaster of Paris, provisions of all kinds with a number of worthless, or actually injurious, substances, that even hardware, such as cutlery and the like, did not escape—he is surprised at the great ingenuity applied to such bad purposes. "The eager and insatiable thirst for gain," he observes, "which seems to be a leading characteristic of the times, calls into action every human faculty, and gives an irresistible impulse to the power of invention, and where lucre becomes the reigning principle, the possible sacrifice of a fellow-creature's life is a secondary consideration." From generalities, Accum then proceeds to describe more or less minutely the fraudulent tricks of each particular trade: "The baker asserts that he does not put alum in his bread, but he is well aware that in purchasing a certain quantity of flour, he must take a sack of 'sharp whites,' a term given to flour contaminated with a quantity of alum, without which it would be impossible for him to produce light, white, and porous bread from a half-spoiled substance." He also states that the bakers used a powder technically called 'stuff,' consisting of one part of alum in minute crystals and three parts of salt.

In speaking of brewing frauds he supports his assertions by reference to actual convictions taken from the papers of the day, and he cites among others the *King v. Richard Bowman*, *Times*, May 18, 1818, in which the defendant, a brewer of Wapping, was convicted of having a substance called 'multum'² on his premises, and fined £200; and the *King v. Luke Lyons*, in which the defendant was convicted of having various deleterious drugs in his brewery, among which were capsicum and copperas, and fined £420.

From these and similar trials and cases, he gives a list of the adulterations in use by the brewers, among which figure multum (just mentioned), cocculus indicus, sold to tanners and dyers under the name of 'black extract,' and 'bittern,' composed of calcined sulphate of iron, extract of cocculus indicus, extract of quassia and Spanish liquorice—a compound not much dissimilar from some of the 'hop substitutes' of the present day. He also gives a list of the publicans convicted, between 1815 and 1818, of adulterating beer; these cases were nineteen in number; but the only adulterations proved seem to have been 'salt of steel,' which was doubtless sulphate of iron, common salt, molasses, and the mixing of table beer with strong beer. The fines ranged from £5 to £400.

In a notice of Mr. Accum's work in No. 156 of the *Literary Gazette*, there occurs the following passage, which may serve as an example of the

¹ *A Treatise on the Adulteration of Food and Culinary Poisons, exhibiting the Fraudulent Sophistication of Bread, Beer, Wine, Spirituous Liquors, Tea, Coffee, etc.*, by Frederick Accum. London, 1820.

² *Multum*, a mixture of extract of quassia and liquorice.

spirit of the press :—"Devoted to disease by baker, butcher, grocer, wine merchant, spirit dealer, cheese-monger, pastry cook, and confectioner, the physician is called to our assistance. But here again the pernicious system of fraud, as it has given the blow, steps in to defeat the remedy. The unprincipled dealer in drugs and medicines exerts the most potent and diabolical ingenuity in sophisticating the most potent and necessary drugs—Peruvian bark, rhubarb, ipecacuanha, magnesia, calomel, castor oil, spirits of hartshorn, and almost every other medical commodity in general demand, and chemical preparation used in pharmacy."

A few years after Accum's work a small duodecimo appeared, familiarly known under the name of *Death in the Pot*, but the full title of which was—*Deadly Adulteration, and Slow Poisoning, and Death in the Pot and the Bottle, in which the blood-empoisoning and life-destroying adulterations of wines, spirits, beer, bread, flour, tea, sugar, spices, cheese-mongery, pastry, confectionery, medicines, etc., are laid open to the public, with tests or methods for ascertaining and detecting the fraudulent and deleterious adulterations, and the good and bad qualities of those articles, with an exposé of medical empiricism and imposture, quacks and quackery, regular and irregular, legitimate and illegitimate, and the frauds and malpractices of pawnbrokers and madhouse keepers. By an enemy of fraud and villany* London.¹ This little brochure of 137 pages, written in a popular style, and rejoicing in a startling title, enjoyed a large circulation, and, despite its small intrinsic merit, had more influence on the popular mind than any similar work that had ever appeared. As may be expected from the title, the book is grossly sensational, and in speaking of water, the author says : "The filthy and unwholesome water supplied from the Thames, of which the delicate citizens of Westminster fill their tanks and stomachs at the very spot where one hundred thousand cloacinae, containing every species of filth and all unutterable things, and strongly impregnated with gas, the refuse and drainage of hospitals, slaughter-houses, colour, lead, and soap works, drug mills, manufactories, and dunghills, daily discharge their abominable contents, is so fearful that we see there is no wisdom in the well, and if we then fly to wine we find no truth in that liquid. Bread turns out to be a crutch to help us onward to the grave, instead of being the staff of life. In porter there is no support, in cordials no consolation, in almost everything poison, and in scarcely any medicine cure." In another place he ascribes the sudden deaths of people in the streets, etc., to the adulteration of their food. Among the sophistications of beer he enumerates, following Accum, cocculus indicus, St. Ignatius bean, nux vomica, tobacco, and extract of poppies.

¹ No date, but probably about 1825. A small Latin work, with a somewhat similar title, had been published about a century previous. *Mors in Olla, seu metallicum contagium in ciborum, potuum et medicamentorum*. Schulze: 1722. This is, however, of but little merit, and only points out the danger of metallic contamination by articles cooked or preserved in copper vessels. At the end is the following :—

"Herr Anhalt zeigt den Tod in Töpfen
Das ist erschrecklich ! doch dabey,
Lehrt er ! aus welchen Grund man schöpfen,
Soll Gegengift und Artzeney,
Wodurch er dann giebt deutlich zu erkennen
Dass man ihn bald mit Ruhm wird Doctor nennen."

There were besides other 'Deaths'; as for example—*Mors in vitro seu lethifera vini adusti damna ex sola ejusdem astringenti virtuti contrabeta*, N. B. Noel. 1709. A poor tract, full of words, and offering nothing new.

§ 33. About nine years after Accum's book had been published in England, A. Bussy and A. F. Boutron-Charlard published in France a work of considerable merit on the adulteration of drugs,¹ the arrangement of which is strictly alphabetical. In the preface, the authors assert that the great development in the art of adulteration had taken place particularly since the wars of the Republic and the establishment of the Continental system, and that it was due more especially to the action of the government, who encouraged the use of products of home growth, in consequence of which roots grown in France were being substituted for those of foreign origin. Chestnut bark, French rhubarb, and poppy were proposed to replace the quinine of Peru, the rhubarb of China, and the opium of the Levant.

Garnier and Harel, in 1844, published their treatise,² a well-written work, moderate in tone, and without exaggeration. A great number of the chemical reactions and tests mentioned by them are in actual use at the present time. Two years later, J. B. Friedrich, in Germany, published his observations on the same subject.³ The arrangement of Friedrich's work is alphabetical. He paid much attention to the composition of diseased milk; and although he made little, if any, use of the microscope, the chemical details of the work are superior to any that had hitherto appeared (see article on 'Milk').

§ 34. About the same time, after more than twenty years had elapsed since the publication of any English work—Accum's being the last—the subject of adulteration was revived here by John Mitchell,⁴ who published what must be considered a very useful volume, although many of the tests he gives would scarcely stand the ordeal of a court of justice at the present day. He states, *e.g.*, that if an infusion of tea, treated with sulphate of copper, and heated, throws down a copious chocolate precipitate, 'hawthorn' is present; if the infusion becomes of a bright green colour on adding caustic soda, sloe leaf⁵ is probable; but if, on the addition of acetic acid, the solution possesses a very bright colour, 'its presence is certain.' Mr. Mitchell's confidence in these reactions is amusing; but on the other hand, the greater number of his observations are still valid.

§ 35. In 1850 Chevallier issued his dictionary of adulteration,⁶ which, through successive editions, has from the time of its appearance been, *par excellence*, the standard French work on the subject. Many years before the publication of his great work, however, M. Chevallier had practically studied the question, as is proved by documentary evidence, and by his numerous representations to the Government on the necessity

¹ *Traité des moyens de reconnaître les falsifications des drogues simples et composées*, by A. Bussy et A. F. Boutron-Charlard. Paris, 1829.

² *Des falsifications des substances alimentaires et des moyens chimiques de les reconnaître*, by Jules Garnier et Ch. Harel. Paris, 1844. Small 8vo.

³ *Handbuch der Gesundheitspolizei, der Speisen, Getränke, und der zu ihrer Bereitung gebräuchlichsten Ingredientien*, edited by J. B. Friedrich, Ansbach, 1846. 8vo.

⁴ *Treatise on the Falsifications of Food, with the Chemical means employed to detect them*, by John Mitchell, F.C.S. London, 1848.

⁵ The belief in the adulteration of tea by the leaves of the sloe is almost contemporaneous with the introduction of tea itself into England, and there are numerous allusions to the practice scattered throughout the various fugitive contributions to literature. However, that tea has been actually adulterated with sloe leaves rests on no direct evidence worthy of consideration.

⁶ *Dictionnaire des altérations et des falsifications des substances alimentaires*, by M. A. Chevallier. Paris, 1st ed., 1850.

of amending the law. In a petition presented to the National Assembly in 1848, he says¹: "Since 1833 I have constantly addressed to the Chambers of Deputies petitions on the same subject, but these petitions have ever been abortive, and fraud has progressively augmented." The first edition of his dictionary, written in a clear style, contained an excellent *résumé* of what was already known with regard to falsifications, and, besides, was enriched with many new facts—the result of a long experience.

In the same year, 1850, Alphonse Normandy, who published the results of thirteen years' labour in a *Handbook of Commercial Analysis*, was one of the first who recommended the use of the microscope for the detection and discrimination of starches: "The admixture of potato flour or fecula with wheat flour may be very well detected by the microscope," p. 210. The scope of his work embraced not only the analysis of food, but also the examination of a variety of commercial substances,² such as ores, agricultural manures, soaps, etc. The arrangement is alphabetical; successive editions have brought the work to the present time.

§ 36. A year after the appearance of Normandy's English and Chevallier's French works appeared the papers of Dr. Hassall, in the pages of the *Lancet*, as already mentioned. The publication of these papers marked a new era in legal medicine and the investigation of foods, and the technical application of the microscope was fully developed in the English use. It was not so, however, among Continental chemists, for Hureauux, in his *Histoire des Falsifications*, published in 1855, scarcely mentions the microscope, although, so far as chemical tests go, his work leaves nothing to be desired. This is the more curious, since the author was aware of the evidence given before the select committee, as is obvious from more than one reference.

§ 37. In 1874 a movement took place in England, the effect of which has been to give an extraordinary stimulus to analytical chemistry—viz., the establishment of the Society of Public Analysts. The movement originated with a few of the leading analysts, who, after one or two private meetings, called a general gathering, which all those engaged in actual practice were invited to attend. This meeting took place at the Cannon Street Hotel in August 1874; and in a few months the society was fully organised, and a definition of adulteration within certain 'limits' had been laid down as follows for the guidance of members:—

DEFINITION OF AN ADULTERATED ARTICLE.

An article shall be deemed to be adulterated—

A. In the case of food or drink:—

1. If it contain any ingredient which may render such article injurious to the health of a consumer.
2. If it contain any substance that sensibly increases its weight, bulk, or strength, or gives it a fictitious value, unless the amount of such substance present be due to circumstances necessarily appertaining to its collection or manufacture, or be necessary for its preservation, or unless the presence thereof be acknowledged at the time of sale.
3. If any important constituent has been wholly or in part abstracted or omitted, unless acknowledgment of such abstraction or omission be made at the time of sale.
4. If it be an imitation of, or be sold under the name of, another article.

¹ *Pétition sur les falsifications, adressée à l'assemblée nationale*, by A. Chevallier. 1848.

² *A Handbook of Commercial Analysis*, by A. Normandy. London, 1850.

B. In the case of drugs:—

1. If when retailed for medicinal purposes under a name recognised in the British Pharmacopœia, it be not equal in strength and purity to the standard laid down in that work.
2. If when sold under a name not recognised in the British Pharmacopœia, it differ materially from the standard laid down in approved works on *Materia Medica*, or the professed standard under which it is sold.

LIMITS.

The following shall be deemed limits for the respective articles referred to:—

Milk shall contain not less than 9·0 per cent., by weight, of milk solids not fat, and not less than 2·5 per cent. of butter fat.

Skim Milk shall contain not less than 9·0 per cent., by weight, of milk solids not fat.

Butter shall contain not less than 80·0 per cent. of butter fat.

Tea shall not contain more than 8·0 per cent. of mineral matter, calculated on the tea dried at 100° C., of which at least 3·0 per cent. shall be soluble in water, and the tea as sold shall yield at least 30·0 per cent. of extract.

Cocoa shall contain at least 20 per cent. of cocoa-fat.

Vinegar shall contain not less than 3·0 per cent. of acetic acid.

§ 38. The 'Proceedings,' which appeared first in the *Chemical News*, were afterwards published in the special organ of the Society—the *Analyst*—throughout the pages of which will be found details of numerous processes, discoveries, and improvements in practical chemistry, which it is certain would, for the most part, not have been invented or known, had there been no such encouraging organisation. With this brief account of the establishment among us of the Society of Public Analysts, we may bring our sketch to a close.

VI.—THE PRESENT LAW IN ENGLAND RELATIVE TO ADULTERATION OF FOOD.

THE SALE OF FOOD AND DRUGS ACT, 38 AND 39 VICT. C. 63, 1875; SALE OF FOOD AND DRUGS ACT AMENDMENT, 1879, 42 AND 43 VICT. C. 30; THE MARGARINE ACT, 1887, 50 AND 51 VICT. C. 29; THE SALE OF FOOD AND DRUGS ACT AMENDMENT, 1889, 62 AND 63 VICT. C. 51.

§ 39. The preamble of the 'Sale of Food and Drugs Act' repeals the Acts in force relating to the adulteration of food.

Section 2 of the 1875 Act defined the term food to include every article used for food or drink by man, other than drugs or water, and the term 'drug' to include every medicine for external or internal use, and section 26 of the 1899 Act expressly extended the definition to flavouring matters and condiments.

Section 3 of the 1875 Act. No person shall mix, colour, stain, or powder, or order or permit any other person to mix, colour, stain or powder, any article of food, with any ingredient or material so as to render the article injurious to health, with intent that the same may be sold in that state; and no person shall sell any such article so mixed, coloured, stained or powdered, under a penalty in each case not exceeding fifty pounds for the first offence: Every offence after a conviction for the first offence shall be a misdemeanour, for which the person, on conviction, shall be imprisoned for a period not exceeding six months with hard labour.

Section 4 is very similar to this, and relates to drugs: "No person shall, except for the purpose of compounding as hereinafter described, mix, colour, stain, or powder, etc., etc., any drug with any ingredient or material so as to affect injuriously the quality or potency of such drug, with intent that the same may be sold in that state, and no person shall sell any such drug so mixed, coloured, stained, or powdered, under the same penalty in each case respectively as in the preceding section, for a first and subsequent offence."

The sections above quoted, formidable as they appear, possess in reality no deterrent powers, but are perfectly harmless, since no prosecution is likely to succeed under these sections, save when supported by very exceptional circumstances; for the next section expressly provides that no conviction is to take place if the person accused "did not know of the article of food or drug sold by him being so mixed, coloured, stained, or powdered," and were able to show that he "could not with reasonable diligence have obtained that knowledge."

§ 40. The real working sections of the Act are the following:—

Section 6. No person shall sell, *to the prejudice of the purchaser*, any article of food or any drug¹ which is not of the nature, substance, and quality of the article demanded by such purchaser, under a penalty not exceeding twenty pounds,² provided that an offence shall not be deemed to be committed under this section in the following cases, that is to say:—

1. Where any matter or any ingredient, not injurious to health, has been added to the food or drug, because the same is required for the production or preparation thereof as an article of commerce, in a state fit for carriage or consumption, and not fraudulently to increase the bulk, weight, or measure of the food or drug, or conceal the quality thereof.

2. Where the drug is a proprietary medicine, or is the subject of a patent in force, and is supplied in the state required by the specification of the patent.

3. Where the food or drug is compounded as in this Act mentioned.

4. Where the food or drug is unavoidably mixed with some extraneous matter in the process of collection or preparation.

In the amended Act, the second section, 42 and 43 Vict., c. 30, states that in any prosecution under the provisions of the principal Act for selling to the prejudice of the purchaser any article of food or any drug, which is not of the nature, etc., it shall be no defence to any such prosecution to allege that the purchaser, having bought only for analysis, was not prejudiced by such sale. Neither shall it be a good defence to prove that the article in question, though defective in nature or substance or quality, was not defective in all three respects.³

¹ The case of *Dickins v. Randersen*, King's Bench Division, *Times*, Jan. 30, 1901, establishes the principle that if a drug is in the Pharmacopœia and that drug is asked for, it is an offence, under section 6, to supply the same drug of a different strength or standard to that of the British Pharmacopœia. In the case of *Fowle v. Fowle*, 60 J. P. 758 Q. B. D., adulterated beeswax sold by a grocer was held under the special circumstances of the case not to be a drug, although mentioned in the P. B.

² In the amended Act of 1899, by section 17, a second offence is punishable by a fine of £50 or less; a third or subsequent offence, of £100 or less. The second or subsequent offences may under certain circumstances be punishable by imprisonment, with or without hard labour, for a period not exceeding three months.

³ Mr Justice Mellor, in the case of *Hoyle v. Hitchman*, L. R. 4 Q. B. D. 230, 43 J. P. 431, gave a judgment clearly expressing the meaning of prejudice to the purchaser. "The offence intended to be prevented by the Act was the fraudulent sale of articles adulterated by the admixture of foreign substances, which would necessarily be to the prejudice of the purchaser, and these words were inserted only to require that such adulteration should be shown to have been made; and further, if the purchaser asks for

The sixth section of the amended Act is to be read with the sixth section of the principal Act, for it states that "in determining whether an offence has been committed under section six of the said Act, by selling to the prejudice of the purchaser spirits not adulterated otherwise than by the admixture of water, it shall be a good defence to prove that such admixture has not reduced the spirit more than twenty-five degrees under proof for brandy, whisky, or rum, or thirty-five degrees under proof for gin."¹

Section 7 of the principal Act enacts that "No person shall sell any compound article of food, or compounded drug, which is not composed of ingredients in accordance with the demand of the purchaser; penalty not exceeding £20."

Section 8 provides "That no person shall be guilty of any such offence as aforesaid in respect of the sale of an article of food or a drug mixed with any matter or ingredient not injurious to health, and not intended fraudulently to increase its bulk, weight, or measure, or conceal its inferior quality, if at the time of delivering such article or drug he shall supply to the person receiving the same a notice by a label distinctly and legibly written or printed on or with the article or drug, to the effect that the same is mixed." This label, by the 12th section of the 1899 Act, is to be legibly written or printed, and not obscured by other matters on the label.

Section 9 enacts "That no person shall, with the intent that the same may be sold in its altered state without notice, abstract from an article of food any part of it so as to affect injuriously its quality, substance, or nature, and no person shall sell any article so altered without making disclosure of the alteration, under a penalty in each case not exceeding £20."

§ 41. One of the chief loopholes which offenders against the Act have diligently availed themselves of is the label section, Section 8. A label will often have a description of the article in large letters, such as COCOA, COFFEE, etc., and then in miserably small type a statement that the article is mixed. In the case of *Liddiard v. Reece* (44 J. P. 233), a grocer had sold half a pound of a mixture of coffee and chicory to an inspector; the mixture was contained in a canister, and was duly weighed, and the full price of coffee was paid for it. After the sale had been completed, the purchaser informed the appellant that he intended to have the article analysed. Thereupon, while the packet was still on the counter, the appellant called the purchaser's attention to the label, on which the purchaser noticed for the first time the words, "This is sold as a mixture of chicory and coffee," printed in distinct and legible characters. The label was affixed in a conspicuous position on the outside of the packet. The purchaser then said that he had asked for 'coffee,' and not for 'chicory and coffee.' The mixture was found by the analyst to consist of 60 parts coffee and 40 parts chicory. On the hearing of the case before the magistrates, they convicted the vendor on the following grounds:—"The fact that the purchaser asked for coffee and was supplied with an

a certain article, and gets an article which, by reason of some admixture of a foreign article, is not of the nature or quality of the article asked for, he is necessarily prejudiced. . . . The words 'to the prejudice of the purchaser' are necessary for the purpose of not interfering with the sale of an article of an inferior nature or quality to that demanded. The prejudice contemplated was not confined to a pecuniary prejudice, for it would very much diminish the probability of bringing home offences against the Act to those who were really guilty, and this was a sufficient argument against such a reading."

¹ By section 14 of the Licensing Act, 1874, it is provided, "That all convictions for selling adulterated drinks shall be entered in the proper register of licenses, and may be directed to be recorded in the license in the same manner as if the offence had been committed against this Act."

article consisting of only 60 per cent. coffee and 40 per cent. chicory, without having his attention called to the label; and without, in fact, seeing it until the purchase was completed, and also the fact that the price he paid for the said article was a usual and fair price for pure coffee, and much more than would have been given for coffee mixed with chicory to the above extent . . . and that, therefore, the appellant was not protected by the said eighth section."

On appeal the conviction was affirmed. In *Petchey v. Taylor*, 62 J. P. 360, Q. B. D., a conviction for selling condensed milk in a tin, the label of which stated the milk was skimmed, whereas the milk was separated, not skimmed, was affirmed on appeal.

There is also a similar decision in the case of *Holder v. Meddings*, 44 J. P. 234.¹

In *Jones v. Jones*, 58 J. P. 132, it was decided that it was not necessary to call the buyer's attention to the label, provided the label is distinct and that there is no fraudulent concealment as to the low quality of the article sold. The case was that the vendor sold a mixed cocoa, the packet being legibly labelled as a mixture, and it was held that the seller was protected by such label. Any verbal declaration is no protection unless it is uttered before the sale is completed. The sale, again, is evidently not completed until the goods are delivered into the purchaser's hand, and the vendor has received the money. Should a person buy any substance in a shop, and (after having tendered his money, and the same has been accepted) proceed to state that the article is required for analysis, and the vendor *then* attempt to return the money; if the purchaser does not accept the money, the sale is evidently complete. On the other hand—an inspector went into a druggist's shop and asked for quinine wine. The chemist served him with the wine, wrapped it up, and laid it on the counter. The inspector then produced his bottles, and declaring the nature of his errand, was about to divide the wine into three parts, when the druggist seized the bottle and refused to sell the wine, which, a moment before, by his actions he seemed ready to do. In this case, the sale was not complete. But now, let us suppose that the inspector had been a little quicker than the chemist, and seized the sample, and, notwithstanding the expressed refusal of the druggist to sell, the inspector had cast his money on the counter—Would the drug have been sold? This question is somewhat difficult to answer, but we think that it would

¹ This case probably overthrows the case reported in the *Times*, June 8, 1879, *Gibson v. Leaper*, a prosecution undertaken under the old Act, 35 and 36 Vic., c. 74, sections 2 and 3. On conviction the vendor appealed. The case was that of a Spalding grocer, who sold a packet of 'Epps's Cocoa' without making any verbal statement of its contents. The packet was labelled with the words "prepared cocoa—for ingredients, see the other side," and on the other side was a notification to the effect that it was necessary in order to make the oil in the cocoa soluble and easy of digestion, to combine with it arrowroot and sugar. The court quashed the conviction, holding that assuming the cocoa to be adulterated, it had not been sold as unadulterated. In the case of *Pope v. Turle* (43, *Law Journal*), May 28, 1874, the Justices of Bedford dismissed a summons for selling adulterated mustard, and the purchaser appealed. It was stated in the case that at the time the respondent delivered the mustard to the appellant he said: "I do not sell you this as pure mustard." The mustard was found to be the common mixture of flour and mustard. Lord Coleridge, Mr Justice Brett, and Mr Justice Grove were undivided in their opinion that the seller was entitled to their judgment on the ground of his having declared to the purchaser that the mustard was mixed with some other ingredient, and that, even had he not done so, he could not come within the section to incur the penalty, because if the admixture was such as to make it an adulterated article, still he had not sold it as an unadulterated article.

have been a sale, and, if adulteration had been proved, the vendor would probably not have escaped through adopting the defence that there had been 'no sale.'¹

§ 42. There is an important question as to how far a vendor can be protected by having a board in or over his shop or place of business, giving notice to the effect that all the goods are adulterated.

The English law is made for those who cannot read as well as for those who can: and presuming a purchaser to be uneducated, the notice gives him no information. Again, it is certain that a very large number of purchasers, even should the notice be in a conspicuous place, fail to observe it. In most cases in actual practice such notices are distinct evasions of the Act, being inconspicuous, and in dark corners.

A seller of milk had a van on which a notice was placed, "Country skimmed milk, sold as adulterated milk." The man with his can went on foot from door to door, the van being in the road. It is evident that, in such a case, very few of the customers could have seen the label. An inspector who bought a sample of the milk did not see it, and the magistrate convicted the defendant.² The important appeal case of *Sandys v. Small*,³ decided before the Court of Queen's Bench, June 25, 1878, bears upon this and lays down the law. A publican put up a notice in his house: "All spirits sold here are mixed." The inspector of weights sent a messenger to buy some whisky, which was given without anything more being said on either side; but the purchaser admitted that before he bought the whisky he saw the notice, "All spirits sold here are mixed, 38 and 39 Vict., c. 63, sec. 8 and 9," although at the very moment of buying the whisky he did not see it. It was proved that a similar notice was posted at the bar window in full view of persons purchasing. Chief-Justice Cockburn said:—"If the seller chooses to sell an article with a certain admixture, the onus lies on him to prove that the purchaser knew what he was purchasing. With respect to the alteration of the article, the Act has provided him with the means of protecting himself against such a presumption, and says that if he attaches to the article a notice of the adulteration which has been made in its quality, then he shall be protected against any charge of an offence against the Act. If he does not resort to this protection, then the presumption of law attaches, and is un rebutted. If he can show that he brought home by other ways to the knowledge of the customer that the quality of the article was altered by admixture, then he does not commit the offence, because both parties knew it, and the seller does not sell an article to the prejudice of the purchaser, and the parties are perfectly free to contract on that footing. In that view the seller, if he has stuck up a notice, would not commit an offence though he might not have affixed a label to the bottle, because he did not sell 'to the prejudice of the purchaser.' . . . It was sufficiently manifest that the man who was sent to buy the whisky knew of the notice stuck up, and hence it was clear that the defendant committed no offence."

From this judgment it is sufficiently evident that where the general label or notice has been clearly seen and understood before making the purchase, then no offence is committed. The decision of *Liddiard v. Reece* does not cover exactly the same ground as the case just quoted, but both appear to support the view here put forward—viz., that the defendant is

¹ In any case, the druggist might have been prosecuted under sect. 17 of the principal Act, for refusing to sell.

² *Analyst*, 1880, p. 225.

³ L. R. 32 Q. B. D. 449; 42 J. P. 550.

bound to prove that the purchaser had a clear knowledge of the quality of the goods before purchasing.

In *Palmer v. Tyler*, 61 J. P. 389 Q. B. D., a notice of general dilution of spirits was hung in a conspicuous place in a bar, a sample of rum 29° U.P., bought by an inspector who did not see the notice, formed the subject matter of a magisterial conviction. On appeal the conviction was quashed.

§ 43. Section 10 provides for the appointment of public analysts in England, Scotland, and Ireland, by various local bodies, such as, in England, the Commissioners of Sewers for the City of London, the District Councils of the Metropolis, the County Councils of Counties, and the Town Councils of Boroughs with a separate police establishment; in Scotland, the Commissioners of Supply, or the Commissioners of Boards of Police, or, where there are no such Commissioners, the Town Councils of Burghs; and in Ireland, the Grand Juries of the Counties and the Town Councils of the Boroughs.

These appointments must be confirmed by a central authority, which in England is the Local Government Board; in Scotland, one of Her Majesty's Secretaries of State; and in Ireland, the Irish Local Government Board. The appointment is now compulsory under section 3 of the Amendment Act 1899.

The qualifications of the analyst are, to a certain extent, defined by the Act of 1875, but the amended Act of 1899 compels proofs of competency according to regulations made by the Local Government Board, section 3 (5), and an order and circular issued by the Local Government Board, March 7, 1900, defines the qualifications as follows:—

“Every person appointed on or after the first day of January, one thousand nine hundred, to the office of Public Analyst, shall furnish such proof as we may deem sufficient of his competent skill in, and knowledge of, (a) analytical chemistry, (b) therapeutics, and (c) microscopy.

“Such proof shall in every case comprise documentary evidence that such person holds the requisite certificate, diploma, license, or document conferring the qualification, or attesting his possession of the skill or knowledge to which the same applies, and granted or issued by any person or body of persons for the time being recognised by us as competent to confer such qualification, or to test such skill or knowledge. Such proof shall also comprise such further evidence as we may in any particular case require.

“All such documentary evidence as is hereinbefore mentioned shall be furnished by such person to the local authority by whom he is appointed, and shall be transmitted to us by the local authority when applying for our approval of the appointment.

“Provided that nothing in this regulation contained shall, in the case of any person who was appointed to the office of Public Analyst with our approval, between the first day of January, one thousand eight hundred and ninety-one, and the date hereof, or of any person who is so appointed for the first time after such last-mentioned date, apply upon any subsequent appointment of such person to the said office.”

“As regards the reference in the Order to a person or body of persons whom the Board may from time to time recognise as competent to confer the requisite qualification, or to test the skill or knowledge of which proof is required by the Order, the Board may state that it would accord with their existing practice to accept as sufficient documentary evidence of the

requisite qualification under the Acts the Diploma of Fellowship or Associateship of the Institute of Chemistry of Great Britain and Ireland, together with the Certificate granted by the Institute after an examination, conducted by them on lines approved by the Board, in therapeutics, pharmacology, and microscopy."

"The possession of a diploma as a registered medical practitioner is accepted as sufficient proof of competency in microscopy and therapeutics, and it would only be necessary that a medical practitioner appointed as a public analyst should furnish evidence of competent skill in, and knowledge of, analytical chemistry."

"Evidence of skill or knowledge on the part of a candidate in respect of any of the qualifications referred to as requisite, which is tendered by an individual, must be from a person recognised as entitled to speak with authority as to proficiency in the particular qualification in question."

The eleventh section distinctly lays down the principle of combination, enacting that the town council of any borough may unite with that of any neighbouring borough in appointing an analyst jointly; or the analyst for the county in which the borough is situated may act upon arrangement as analyst for their borough. Those who are practically acquainted with the subject know that it is only in the largest and most populous places in England that any kind of living can be made out of a public analytical appointment. Hence it follows that an analyst for a small place must either have private means or that his chief occupation must be of a more remunerative nature; it is, therefore, highly desirable that the analysis of foods and drugs should be in a few hands only, and that an analyst should hold many appointments of the same nature. In this way, and in this way only, will it be possible to have properly-fitted laboratories, supplied with all the expensive appliances of modern research, and in this way only will it be possible to improve the processes of analysis. It is also a fact, from the very few cases in which an experienced analyst has to attend as witness, that there would be no inconvenience, were all the northern counties to have their samples analysed at Sheffield, Manchester, or York; the western and south-western counties at Bristol; and the rest of England at the London laboratories. Probably also the whole of the Scotch samples could be dealt with in Glasgow and Edinburgh, and the Irish, in like manner, in two of the chief cities.

§ 44. Section 12 of the principal Act provides for the purchase of samples by any purchaser for analysis by the public analyst for the district in which the purchase is made, on payment to such analyst of a sum not exceeding ten shillings and sixpence; or if there is no analyst appointed for the district, to the analyst of another place. In this latter case the fee appears to be a matter of private arrangement, for the words of the Act are—"such sum as may be agreed upon between such person and the analyst." In either case, the analyst must give a certificate of his results to the purchaser. Whether the purchaser is bound to purchase the article in the manner directed in section 14 is still an undecided point.

It is evident that, for legal purposes, the official analyst must be employed, and that under the Act no prosecution can be undertaken except on his certificate. Thus, at the Manchester Police Court, the Milk-Dealers' Protection Society attempted to prosecute on the certificate of a private analyst, but on this technical ground alone the magistrate dismissed the case.¹

¹ *Analyst*, 1879, vol. iv. p. 74.

There is no authority given by the Act for a Public Analyst himself to appoint a deputy. At Bristol the analyst suffering from illness deputed an analyst in a neighbouring district to do his work. The deputy certified to the adulteration of a sample of milk. The defendant took advantage of this omission in the Act and successfully disputed the power of an analyst to appoint, even under the circumstance of illness, a deputy to do his statutory duty.

Whether this case should have been otherwise decided on appeal is not clear, for the next section gives power to a purchaser to take a sample to 'the analyst of another place' should there be no analyst 'acting for such place'; and it might be reasonably argued that an analyst disabled by illness is for the time 'not acting for such place.'

§ 45. The thirteenth section of the old Act and the third section of the amended Act should be read together:—

"Any medical officer of health, inspector of nuisances, or inspector of weights and measures, or any inspector of a market, or any police-constable under the direction and at the cost of the local authority appointing such officer, inspector, or constable, or charged with the execution of this Act, may procure any sample of food or drugs, and if he suspect the same to have been sold to him contrary to any provision of this Act, shall submit the same to be analysed by the analyst of the district or place for which he acts; or if there be no such analyst then acting for such place, to the analyst of another place; and such analyst shall, upon receiving payment as is provided in the last section, with all convenient speed, analyse the same and give a certificate to such officer, wherein he shall specify the result of the analysis."

By section 3 of the 1879 Act, the same individuals "may procure at the place of delivery any sample of any milk in course of delivery to the purchaser or consignee, in pursuance of any contract for the sale to such purchaser or consignee of such milk," and by section 10 of the Act of 1899, a sample of milk "or of margarine or margarine cheese forwarded by a public conveyance, the person taking the sample shall forward by registered parcel, or otherwise, a portion of the sample marked, and sealed, or fastened up, to the consignor if his name and address appear on the can or package containing the article required."

Section 4 of the 1879 Act provides a penalty for refusal to submit samples of milk to be taken, of a sum not exceeding £10.

Section 17 of the principal Act also provides a penalty not exceeding £10, for refusal to sell to the persons appointed to carry out the Act any "article of food or any drug exposed for sale, or on sale by retail on any premises, or in any shop or stores. The purchaser shall tender the price for the quantity which he shall require for the purpose of analysis, not being more than shall be reasonably requisite."

Any street or place of public resort is held to come within the meaning of this section.

It is perfectly clear from the sections quoted, that if a sample be taken of milk in transit, that sample must be taken at the place of delivery. If, for example, a milkman is driving his cart through Oxford Street, it would not be legal for an inspector to stop the cart and require a sample of the milk. The sample must be taken at the place where the milk is delivered. This may be a house, or it may be a railway-station, or it may be a public booth where the milk is sold at so much a glass.

Previous to this amendment of the law, according to the case of Rouch

v. Hall,¹ it was not necessary to divide the sample at the place of delivery.

§ 46. Section 14 fully details the method to be pursued by any purchaser under the Sale of Food and Drugs Act.

The person purchasing any article with the intention of submitting the same to be analysed, shall, after the purchase shall have been completed, forthwith notify to the seller or his agent selling the article, his intention to have the same analysed by the public analyst, and shall offer to divide the article into three parts to be then and there separated, and each part to be marked and sealed up or fastened up, as its nature will permit, and shall, if required to do so, proceed accordingly, and shall deliver one of the parts to the seller or his agent. He shall afterwards retain one of the said parts for future comparison, and submit the third part, if he deem it right to have the article analysed, to the analyst.²

Section 15. If the seller or his agent do not accept the offer of the purchaser to divide the article purchased in his presence, the analyst receiving the same article for analysis shall divide the same into two parts, and shall seal or fasten up one of those parts, and shall cause it to be delivered, either upon receipt of the sample, or when he supplies his certificate to the purchaser, who shall retain the same for production, in case proceedings shall afterwards be taken in the matter.

In the case of *Horder v. Scott*,³ heard in the Queen's Bench Division before Justices Lush and Field, on the 4th of May 1880, it was made clear that an inspector could appoint a deputy. The case was an appeal from a decision of justices in the county of Stafford. An inspector under the Act had deputed his assistant to purchase a sample of coffee, which was duly divided in conformity with the Act, and the analyst certified to its adulteration with chicory. The magistrates, however, considered that as the proceedings were initiated by the inspector in his official capacity, he having laid the information, and having regard to sections 13, 14, and 17 of the Act, should personally have purchased the article, and the case was dismissed. This, Mr. Justice Field said, was entirely wrong—"It did not signify whether the inspector purchased by his own hand or by his agent. Then the magistrates had decided, secondly, that Samuel Toy, being the purchaser, should have submitted the article to the county analyst; there again he thought the magistrates were wrong. . . . If the thing were properly analysed, it does not signify through whose hands the article was bought."

On the purchase of an article it is evidently essential to say, not only that it is the purchaser's intention to have it analysed, but "analysed by the public analyst," care being taken to use the exact words of the Act. This objection has been several times raised with effect. When a deputy purchases samples, it would be a mistake for the inspector to appear and seal the samples. This had better be left to the purchaser, who can then immediately, or at any subsequent period, hand the samples to the inspector, by whom they should be delivered to the analyst. It is obvious that legal proof will be required as to the proper keeping and delivery of the samples.

It has been argued that the division of the sample into three parts

¹ L. R. 62 Q. B. D. 17; 50 L. J. 6; 45 J. P. 220.

² In a recent appeal case (*Mason v. Cowdary*) it was held that if, for example, six separate bottles be purchased, it is not sufficient to give two bottles to the vendor; each bottle must be divided into three parts.

³ L. R. 52 Q. B. D. 552; 49 L. J. 78; 44 J. P. 520, 795.

means three equal parts; but there is no direction in the Act as to an equal division. At the same time, should the purchaser leave with the seller, or keep himself an insufficient quantity for any further analysis, there would be an infringement of the spirit of the Act; for the purpose of the division evidently is to provide against any mistake or wrong interpretation of facts on the part of the analyst. Should another analysis be required, it would not be right that the seller should be put at a disadvantage by any marked or great inequality in the division of the parts; hence it will be prudent for purchasers to divide the substance into three parts as nearly equal as may be, but it is unnecessary to use for this purpose balances or measures.

On the seller or his agent not accepting the offer of the purchaser to divide the sample into three parts, it becomes the duty of the analyst to divide it into two parts. There is no direct stipulation as to when this is to be done, for the analyst is permitted to keep, if he chooses, the whole, until the termination of the analysis; but it is evidently the course most free from objection to divide it into two approximately equal parts immediately on receipt of the sample, to seal it in the presence of the purchaser, and deliver one of the parts to the purchaser.

§ 47. Section 16¹ permits articles to be sent by post after being duly registered, and the Postmaster-General has made the following regulations with regard to the transmission of samples:—"Each packet must be addressed according to the official designation of the analyst, as 'public analyst,' or otherwise; the nature of its contents must be stated on the front of the packet. Any postmaster, at whose office a packet for a public analyst shall be tendered for registration, may refuse to accept it for this purpose, unless it be packed in so secure a manner as to render it at least unlikely that its contents will escape, and injure the correspondence. Liquids for analysis shall be contained in stout bottles or bladders, which shall be enclosed in strong wooden boxes with rounded edges—the boxes being covered by stout wrappers of paper or cloth, and no such packet shall exceed eight inches in length, four in width, or three inches in depth. No packet whatever addressed to a public analyst shall exceed the dimensions of eighteen inches in length, nine inches in width, or six inches in depth. The postage and registration-fee on each packet must be prepaid."

§ 48. Section 18 states that the certificate shall be in the form set forth in the schedule, or to the like effect. These last few words are important, for the analyst thereby is not absolutely confined to the certificate in the schedule. Notwithstanding this, it is safer to adhere strictly to the exact form of certificate, and not to attempt to modify it in any way. It has been laid down by *Fortune v. Hemson*, 60 J.P. 88, that the grounds upon which the analyst bases his opinion must be given; it is not sufficient to say, *e.g.*, that a sample of milk is diluted by the addition of 20 per cent. of water; the reason why the analyst thinks that the 20 per cent. has been added must be stated. In certain foods—one of which is milk—the public analysts of this country have agreed upon forms of expression, the certificate for milk being as follows:—"I am of opinion that the said sample contains the parts as under:—

Milk,
Added water,

100·00

¹ See also sect. 15 of the Act of 1899, which verbally amends this section.

This opinion is based upon the facts that the sample contained only...per cent. of non-fatty solids, whereas normal milk contains at least 8·5 per cent. of non-fatty solids." Several prosecutions have resulted in convictions on the above form of certificate, and since it embodies the principle of the case, it is likely to stand.

In the case of any article liable to decomposition, the analyst must certify specially as to whether "*any change has taken place in the constitution of the article that would interfere with the analysis.*" Milk and butter are specifically mentioned, but the rule would evidently apply to all foods preserved in tins, provided the tin has been opened. It might also be argued that many other substances (such as wine or beer) are liable to decomposition; hence it will be better for the analyst to give this matter rather a wide interpretation, and insert in his certificate the necessary words, if called upon to certify in reference to any substance that, under any conditions, is liable to decompose. The exact words must be used, for in an appeal heard at the Middlesex Sessions, October 1880, *Peart v. Edwards*,¹ the analyst certified that the milk was fresh when delivered to him, but omitted to specify whether "any change had taken place in the constitution of the article, so as to interfere with the analysis"; and on this ground the assistant-judge quashed the conviction.

Section 19 provides for the regular quarterly reports of the analyst, copies of which are to be transmitted to the Local Government Board. If, as in many cases, no work at all has been done under the Act, it is evidently the duty of the analyst to send a 'nil' report.

Section 20 provides for the institution of proceedings. The Act says—"The person causing analysis may take proceedings." He, therefore, need not be the actual purchaser; and it is usual for an inspector to take the summons out on behalf of the public body for which he acts.

In all prosecutions under the Act, and notwithstanding the section just quoted, the summons must be served within a reasonable time, and in the case of a perishable article—*e.g.*, milk—not exceeding twenty-eight days from the time of the purchase, etc. The summons must state the particulars of the offence or offences, and also the name of the prosecutor; and it must not be made returnable in less than seven days from the day it is served upon the person summoned.

Section 21 of the principal Act provides that the certificate of the analyst shall serve as evidence; therefore unless specially required, he need not attend. Section 22 of the Act of 1899 in addition makes the certificate of a public analyst in like manner evidence. In the first case the defendant may, however, require the attendance of the analyst, in the second the prosecutor has the like privilege.

§ 49. Section 22 provides for a part of the sample, or samples, to be analysed at Somerset House, in case of any dispute as to the correctness of the analysis. The sample must be sent by order of the justices (or a stipendiary magistrate), and would not be received if sent by either party direct. The results of the official analysis should be considered as the written opinion of a referee not final but yet entitled to great weight.

Section 23 provides for an appeal to Quarter Sessions.

Section 25 gives the opportunity to the defendant to prove, by written warranty, "that he had no reason to believe at the time when he sold it that the article was otherwise than of the nature, quality, etc., demanded;

¹ 44 J. P. 699, 763.

that he sold it in the same state as when he purchased it." On proof of this, the defendant may be discharged from the prosecution, but he will have to pay costs, unless he has given notice to the prosecutor that he will adopt this line of defence.

The warranty or invoice is not available as a defence, unless the defendant has sent to the purchaser a copy of such warranty or invoice seven days after service of the summons, stating he intends to rely upon it, with the name and address of the warrantor, to whom he must send a similar notice.—Sale of Food and Drugs Act, 1899, section 20, where the defendant is a servant of the person who has purchased under warranty and may use such warranty for defence.—*Ib.*

In the case of *Rook v. Hopley*,¹ it was decided that an invoice containing a description of an article sold to a retail dealer is not such a written warranty as is required by section 25; and a retail dealer who sells an adulterated article in the same state as he purchased it will not, by virtue of such a document, be entitled to be discharged on being summoned before a magistrate.

Section 26 provides for the payment of penalties recovered to the authority, for the purpose of defraying the expenses of the Act.

Section 27 has stringent clauses relative to persons convicted of forging warranties, wilfully applying a certificate or warranty of an article of food or drug, to any other article of food or drug, the giving of a false warranty, and wilfully giving a label falsely describing the article sold. See also section 20 of the Act of 1899.

This latter clause of the section—viz., "Every person who shall wilfully give a label with any article sold by him, which shall falsely describe the article sold, shall be guilty of an offence under this Act," etc.—would apply to a great many cases of adulteration in which the article is wrongly described by label; but it is evident that guilty knowledge must be proved, for the word 'wilfully' presupposes guilty knowledge. In most cases, unless the actual manufacturer were summoned, ignorance would be pleaded.

Section 28 provides that nothing in the Act shall affect the power of proceeding by indictment, or take away any other remedy against any offender under the Act, or in any way interfere with contracts and bargains between individuals, and the rights and remedies belonging thereto, provided that in any action brought by any person for a breach of contract on the sale of any article of food or any drug, such person may recover alone, or in addition to any other damages recoverable by him, the amount of any penalty in which he may have been convicted under this Act, together with the costs paid by him upon such conviction, and those incurred by him in and about his defence thereto, if he prove that the article or drug, the subject of such conviction, was sold to him as and for an article or drug of the same nature, substance, and quality as that which was demanded of him, and that he purchased it not knowing it to be otherwise, and afterwards sold it in the same state in which he purchased it—the defendant, in such action, being nevertheless at liberty to prove that the conviction was wrongful, or that the amount of costs awarded or claimed was unreasonable.

The 30th section of the Act provides for the examination of tea on importation.

The effect of this examination has been so good that adulterated tea,

¹ L. R. 34 D, 209; 42 J. P. 551.

in comparison with the period before the Act, has decreased in a very marked degree.

§ 50. *Milk, Margarine, Margarine Cheese and Butter.*—The margarine Act of 1887, and the Sale of Food and Drugs Act of 1899, are specially directed against frauds with regard to the above articles, besides which the Act of 1899 creates a new Authority superintending the Sale of Food and Drugs Acts—viz., the Board of Agriculture.

The latter Board is empowered (Sale of Food and Drugs Act, 1899, sect. 4) to make regulations; in other words, to fix standards or limits, with regard to milk, cream, butter or cheese, and under this section the Board has made the following regulations:—

SALE OF MILK REGULATIONS, 1901.

The Board of Agriculture, in exercise of the powers conferred on them by section 4 of the Sale of Food and Drugs Act, have made the following Regulations:—

Milk.

1. Where a sample of milk (not being milk sold as skimmed, or separated, or condensed, milk) contains less than 3 per cent. of milk-fat, it shall be presumed for the purposes of the Sale of Food and Drugs Acts, 1875 to 1899, until the contrary is proved, that the milk is not genuine, by reason of the abstraction therefrom of milk-fat, or the addition thereto of water.

2. Where a sample of milk (not being milk sold as skimmed, or separated, or condensed, milk) contains less than 8·5 per cent. of milk-solids other than milk-fat, it shall be presumed for the purposes of the Sale of Food and Drugs Acts, 1875 to 1899, until the contrary is proved, that the milk is not genuine, by reason of the abstraction therefrom of milk-solids other than milk-fat, or the addition thereto of water.

Skimmed or Separated Milk.

3. Where a sample of skimmed or separated milk (not being condensed milk) contains less than 9 per cent. of milk-solids, it shall be presumed for the purposes of the Sale of Food and Drugs Acts, 1875 to 1899, until the contrary is proved, that the milk is not genuine, by reason of the abstraction therefrom of milk-solids other than milk-fat, or the addition thereto of water.

Extent.

4. These Regulations shall extend to Great Britain.

Commencement.

5. These Regulations shall come into operation on the First day of September, One thousand nine hundred and one.

Short Title.

6. These Regulations may be cited as the Sale of Milk Regulations, 1901.

SALE OF BUTTER REGULATIONS, 1902.

The Board of Agriculture, in exercise of the powers conferred on them by section 4 of the Sale of Food and Drugs Act, 1899, do hereby make the following Regulations :—

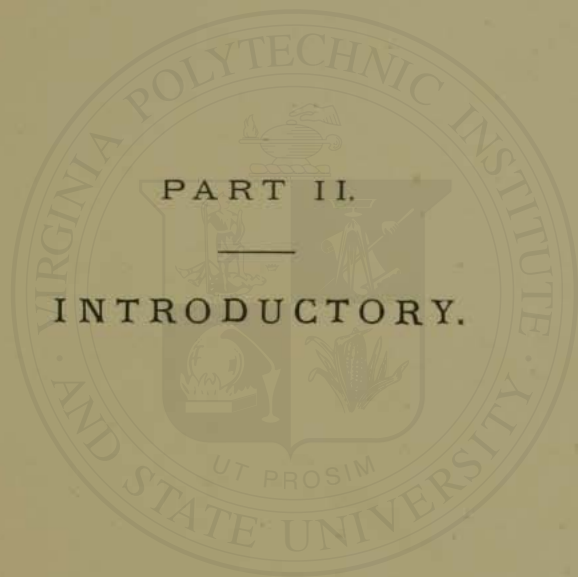
1. Where the proportion of water in a sample of butter exceeds 16 per cent. it shall be presumed for the purposes of the Sale of Food and Drugs Acts, 1875 to 1899, until the contrary is proved, that the butter is not genuine by reason of the excessive amount of water therein.
2. These Regulations extend to Great Britain.
3. These Regulations shall come into operation on the fifteenth day of May, One thousand nine hundred and two.
4. These Regulations may be cited as the Sale of Butter Regulations, 1902.

A standard has also been made by the same statute with regard to margarine (section 8), making it unlawful "to manufacture, sell, expose for sale, or import any margarine the fat of which contains more than 10 per cent. of butter fat."

Margarine is defined by the Margarine Act (section 3) to mean all substances, whether compounds or otherwise, made in imitation of butter, whether mixed with butter or not. It is unlawful to sell such a substance otherwise than under the name of margarine. Margarine cheese is defined by the 1899 Act to mean any "substance whether compound or otherwise, which is prepared in imitation of cheese, and which contains fat not derived from milk."

The Act of 1899 introduces into the statute some new terms—such, for example, as 'separated milk' as distinguished from 'skimmed milk'; there is also a distinction drawn between an adulterated and an 'impoverished' article. Both the Local Government and the Board of Agriculture are empowered to take samples and to have them analysed at the cost of the Local Authority, the Local Government Board in relation to matters affecting the *general interest of the consumer*, the Board of Agriculture to matters affecting the *general interests of Agriculture* (section 2). Both Boards may also, under similar conditions, empower an officer to purchase samples and submit them for analysis in cases in which the local authority fails to do its duty in enforcing the Sale of Food and Drugs Acts (sections 2 and 3).

For various other important details as to the registration of margarine-sellers, wholesale or retail, as to labelling packages or tins, as to warranty, as to protection from importation of certain adulterated or impoverished articles, the reader is referred to the Acts themselves, published in the Appendix to this work.



PART II.

INTRODUCTORY.

PART II.—INTRODUCTORY

A DESCRIPTION OF A FEW SPECIAL FORMS OF APPARATUS USEFUL IN FOOD-ANALYSIS

§ 51. As stated in the first edition of this work, it is no part of the author's plan to describe the elementary apparatus to be found in every text-book, and to be seen in every laboratory. Notwithstanding, it will be convenient here to give a brief notice of some special forms of apparatus useful in food-analysis.

APPARATUS FOR THE TREATMENT OF SUBSTANCES BY VARIOUS SOLVENTS.

It is a matter of some moment to economise alcoholic and ethereal solvents, and it is always advantageous to keep a laboratory as free as possible from vapours and odours. Where a solid has to be exhausted by ether or petroleum, one can scarcely imagine anything more convenient than the apparatus invented by Soxhlet, and proposed by him for the purpose of treating milk solids with ether, but in point of fact widely applicable. It consists of a glass tube (fig. 1), the size of which is perfectly under control, and may be made very large or very small, according to individual requirements. For the purpose of milk analysis a capacity of 100 c.c. is ample. The tube is quite closed at the bottom, A; the volatile vapours ascend through the tube D, and are condensed in an upright condenser attached to A; the liquid, therefore, falls drop by drop on to the substance at A. When the condensed liquid reaches X, the syphon BB acts, and the whole of the liquid runs into the flask. The apparatus works quite automatically, and scarcely any ether is lost, however long the operation may last.

Clausnizer has modified this apparatus for small quantities of substances (fig. 2). D is a tube drawn out pipette-like. J is an inner tube made so as to slip easily into the outer one, and pulled out in the blowpipe flame into a long, almost capillary stem, which is then bent up into a syphon, and terminates in the flask, being made a little longer than the drawn-out portion of D. The mode of action is precisely similar to that just described. The volatile vapour escapes between the two tubes, until it reaches the upright condenser; it is then condensed, and falls in drops on the substance in the inner tube; and when the bend of the syphon is reached, the little tube is at once emptied of the saturated solvent, and the process commences again.

Another method is as follows (see fig. 3):—Take a flask with a wide

neck, fit a small short test-tube, T, into a cork that will go tightly down into the neck of the flask; cut two or three notches in the cork, as shown in the figure. The ether or other solvent continually drops on to the substance in the tube, and when the tube is full, runs over into the flask, and thus the substance is at length exhausted. Similarly, any little apparatus may be suspended from the cork by means of a wire; or, lastly, a little tube may be supported from the bottom by means of a platinum wire-support fused into the flask.

Since the last edition of this work, there have been published a large number of modifications of the details of Soxhlet's and other apparatus for the extraction of dry solids, but those described answer well.

In the *extraction of liquids* by solvents there are three methods—(1) simply shaking of the liquid to be extracted and the solvent in a separating funnel; (2) allowing the solvent to bubble through the liquid, and if

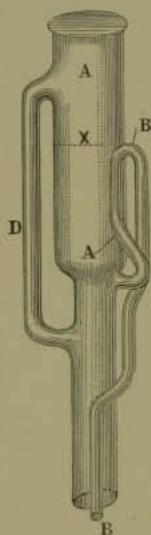


FIG. 1.



FIG. 2.



FIG. 3.

lighter than the liquid to flow from the top layer back to the flask; (3) the solvent flows over the liquid in a thin film.

The principle of the latter two methods is illustrated by the apparatus suggested by A. Wröblewski¹ and by Hans Malfatti² respectively. Wröblewski's apparatus (fig. 4) consists of a flask *d*, in which is placed the volatile solvent. On heating the flask the vapour ascends the tube *g*, and the ether bubbles up through the liquid to be extracted, is condensed by the cold tube *t*, which is surrounded by the water jacket *k*, and falls into the tube *b*, which is led to the bottom of the liquid. The ether forms an ever-increasing layer on the top of the liquid, and at length reaches the level of the mouth of the tube *c*, and then continuously flows back into the flask. Samples of the solvent can be drawn off from time to time by means of the stopcock at *h*, in order to ascertain whether the extraction is complete. If it is, a few drops evaporated on a watch-glass should leave no appreciable

¹ *Z. f. analyt. Chemie*, 1897, 671.

² *Ib.*, 1898, 374.

residue. Malfatti's apparatus (fig. 4A) consists of a flask A for the ether; the vapour from the boiling ether is conveyed by the rather wide right-angled tube *a* to a condenser B, consisting of a tube of lead or tin coiled round the glass tube. In the metal tube circulates cold water, the ether falls as it distils through a long narrow tube nearly to the bottom of the metre long extraction vessel C; near the top of the tube are two side tubes, the one bent *b*, so as to form a trap, and provided with a funnel, the other connected with the flask A. Lastly there is a vessel D, provided with a

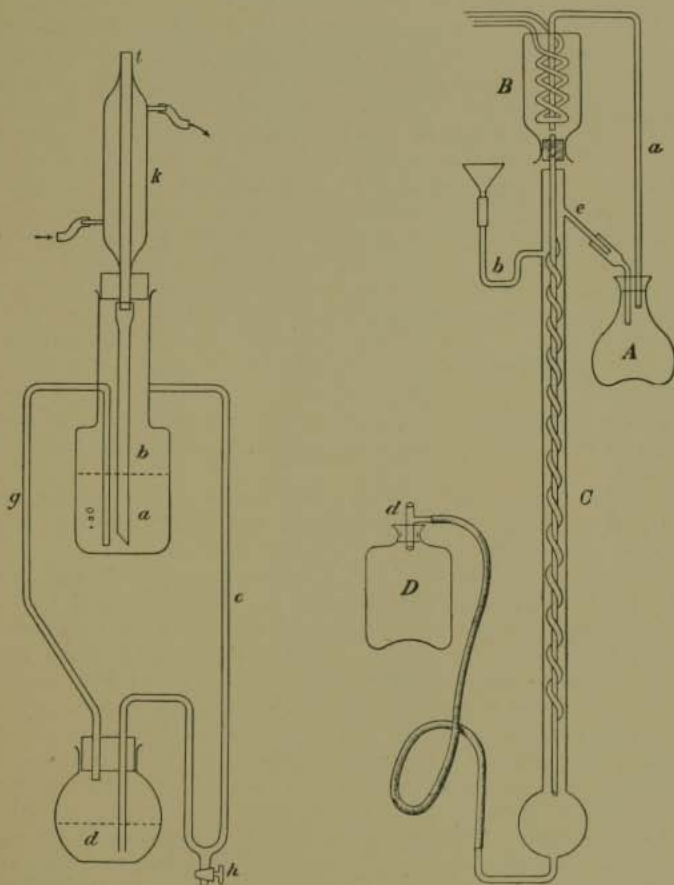


FIG. 4.

FIG. 4A.

T-piece *d* to receive the extracted fluid. The fluid to be extracted is allowed to flow drop by drop from a Mariott's flask into the funnel, and flows on to a spiral of fat-free wool curled round the inner tube. As this liquid flows down it comes in contact with the ether flowing up; and when the ether reaches the level of *e* it flows into the flask to be again distilled. Large quantities of liquid can be exhausted in this way by a volatile solvent.

There are many ingenious devices other than these just described for effecting the same purpose, but all are modifications of the two types.

When it is necessary to treat substances in open dishes with volatile solvents, the authors use the following apparatus (see fig. 5), which, since the first description of it in the *Chemical Society's Journal*,¹ is to be found in most laboratories.

The principle of the arrangement is simply this, that it converts an ordinary dish into a closed vessel, so that ether and volatile liquids may be boiled without loss; or, on the other hand, a volatile liquid may be distilled and recovered with as much ease as in operating with a retort.

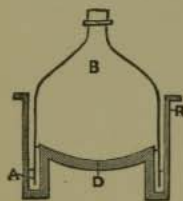


FIG. 5.

The essential part of the apparatus consists of a cast-iron body, R, externally drum-shaped, and having a deep groove, A, in which a little mercury or other 'sealing liquid' is placed. Into this groove fits a bell-jar, B, and the part marked D is hollowed out for the reception of a dish. The size of the dish is quite indifferent; any dish will do, so long as it is not too large for the bell-jar to cover. The neck of

the bell-jar is attached to a Liebig's condenser. Should a substance require exhaustion with the solvent, the Liebig is placed in an upright position; should an evaporation or distillation be required, the condenser is placed in the usual slanting position, and in this way all the liquid evaporated is saved. As a matter of convenience it is well to have a pair of these apparatuses in a laboratory, one with an upright, the other with a slant condenser.

§ 52. *The Spiral Balance.*—A spiral balance, proposed and used some years ago by Prof. Jolly, has been figured and described in a former edition of this work.

§ 53. *Vacuum Processes.*—There are a variety of analytical operations, especially those employed in toxicological and food chemistry, which for their proper performance require an efficient vacuum.

There are three chief varieties of mercury pumps in use, viz., the Geissler, the Sprengel, and the Toepler; the disadvantage of the Geissler is the numerous stopcocks, of the Sprengel, that it is not very suitable for evacuating large vessels, and of the Toepler the fragility of the numerous branches. The mercury pump the authors use is free from most of the above objections, and is known as the 'Physiological Mercury Pump'; it is to be found in the stock of most dealers in physical and chemical apparatus. The diagram is sufficiently explanatory; the reservoir is filled with mercury, and on being raised expels the air by flowing into the bulb B and then flows over through the small bored tube T, into a small vessel charged with mercury. When the reservoir is in the position shown in the diagram, the mercury is prevented flowing backwards and upwards into the drying tubes by a glass float valve. By first partially exhausting a large vessel such as a Winchester quart by means of a good water pump or a good air pump, the operation may be finished by the pump just described within half-an-hour, and smaller pieces of apparatus in proportionately less time.

¹ A new and simple Apparatus for the treatment of substances in open dishes by volatile solvents. By A. Wynter Blyth, *Journ. Chem. Soc.*, March 1880.

THE MICROSCOPE, THE SPECTROSCOPE, AND THE ART OF PHOTOGRAPHY AS APPLIED TO THE CHEMISTRY OF FOOD.

§ 54. There are so many special works describing the microscope, that it will be quite unnecessary to burden the pages of this book with informa-

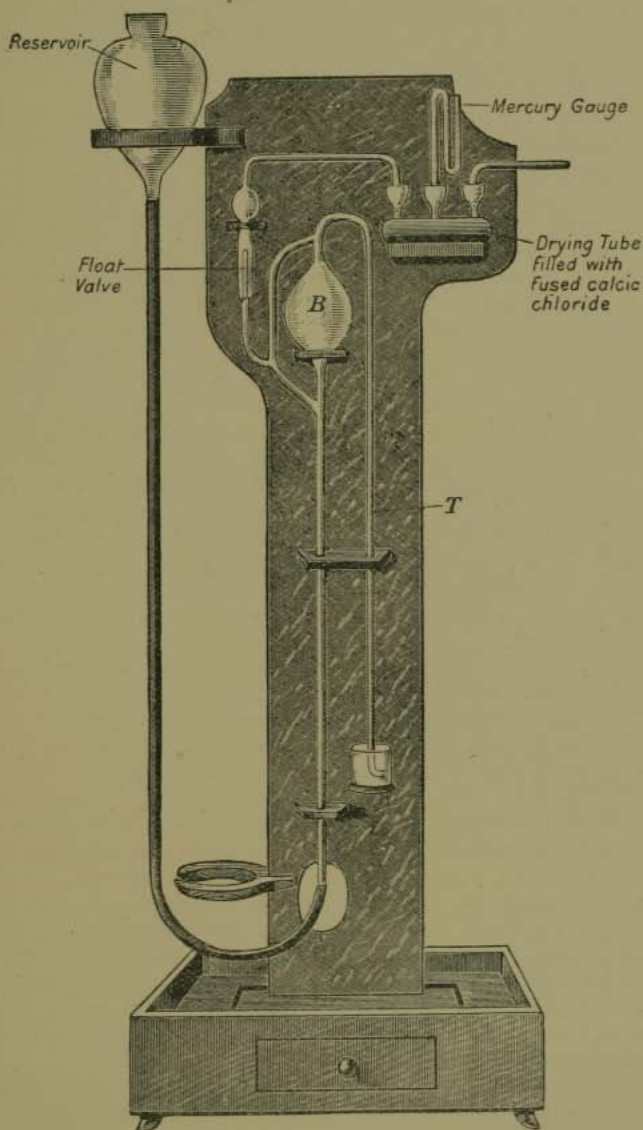


FIG. 6.

tion so readily accessible. The chemist, as a rule, will find a binocular

most suitable for his purpose, for it is only with a binocular that it is possible to have a really good view of crystals. Besides, the instrument is so readily converted into a monocular, that it possesses the advantages of the latter combined with its own. For certain branches of research, and more especially for observing reactions under the microscope, the inverted microscope of Dr. L. Smith, of Merton College (or those of similar pattern), by which the object glass is placed below the substance to be examined, has this advantage, that it is possible without injury to the instrument, and without being annoyed by acid fumes, to treat substances under observation with strong acids, even at a boiling temperature.

The analytical student will require to familiarise himself with the use of the micrometer and the polariscope. The most suitable micrometer for the measurement of starches and similar substances is what is called an eyepiece micrometer. A glass, ruled, either in squares, or as a simple scale, is placed between the eye and field piece, so that both the object magnified and the scale are seen clearly at one and the same time. In order to find the value of the divisions of the eye micrometer, it is necessary, in the first place, to determine them by noting how many divisions correspond with one or more of a slip of ruled glass placed on the stage, and containing divisions equalling the hundredths of an inch, or any other convenient measurement. Suppose, for example, that it is found that one-hundredth of an inch on the stage when measured by the eyepiece required 18 of the eyepiece divisions, then it is obvious that each one of the divisions is $\frac{1}{18}$ of $\frac{1}{100}$ or $\frac{1}{1800}$ of an inch; therefore, any object that measured, say four divisions, would be $4 \times \frac{1}{1800} = \frac{4}{1800}$, or would measure the one four hundred and fiftieth of an inch. There is another method of measurement which is extremely accurate and applicable to all cases; this is, to take a micro-photograph of the subject, and to photograph a glass with suitable ruled divisions, with the same arrangements and with the same powers; afterwards a measurement with ordinary compasses can, with great ease and convenience, be made.

Chemical reactions, under the microscope, are either observed in shallow cells ground in the glass slide itself, or simply on the ordinary flat slide, or, as is sometimes convenient, in almost capillary tubes with flattened sides, the microscope being in a horizontal position. Reactions, as a rule, should be observed with only a moderate magnifying power. It is quite possible to execute, on a very small amount of material, a complete qualitative analysis on the stage of the microscope, mixing with drops of the solution under observation droplets of the ordinary test solutions, such as sulphuretted hydrogen water, ammonium sulphide, ammonia, oxalic acid, sodic phosphate, etc.¹ Dr. Beale has recommended glycerine to be used instead of water for these reactions, and he states that although the reactions are slower, yet that they are more perfect.² The method of subliming alkaloids, and its important bearing in the determination of the nature of substances in tea or coffee, is described in the article on 'Tea,' together with the microscopic appearance of the ash of various leaves, and the method of obtaining 'skeleton ashes.'

In cutting sections of seeds, leaves, etc., no difficulty is experienced when they are in the entire state, nor are any special instruments required save a sharp razor, for with a little practice sections quite as fine as those it is possible to cut by a section-cutting machine can be made with a razor.

¹ See *Beiträge z. mikrochemischen Analyse*, by H. Behrens, 1902.

² *How to Work with the Microscope*. London, 1880.

It is, however, quite different with such matters as tea leaves which have been dried and crumpled, or seeds in the state of powder. Here considerable difficulty may be experienced, and it is often not possible to get a section at all satisfactory of any given dark microscopic particle. The authors have had tolerably fair results by sprinkling opaque powders on a piece of smooth wood, and embedding the powders in a tenacious glue.¹ When the cement has set, there is no difficulty in getting sections. Similarly, the known processes for embedding soft substances answer well with tea. A simple method is also to gum the leaf, or fragment of leaf, on to a solid substance, and then horizontal sections can be obtained. Sometimes scraping a leaf in the same manner as when a blot is being erased from paper, brings away very beautiful pieces of the epidermis and stomata. Sections of leaves are easily obtained by placing the leaf between two pieces of cork, pressing them well together, and then cutting the finest possible layers with a sharp razor. In all these cases the razor should be wet with some fluid, either water or (which is for the most part better)

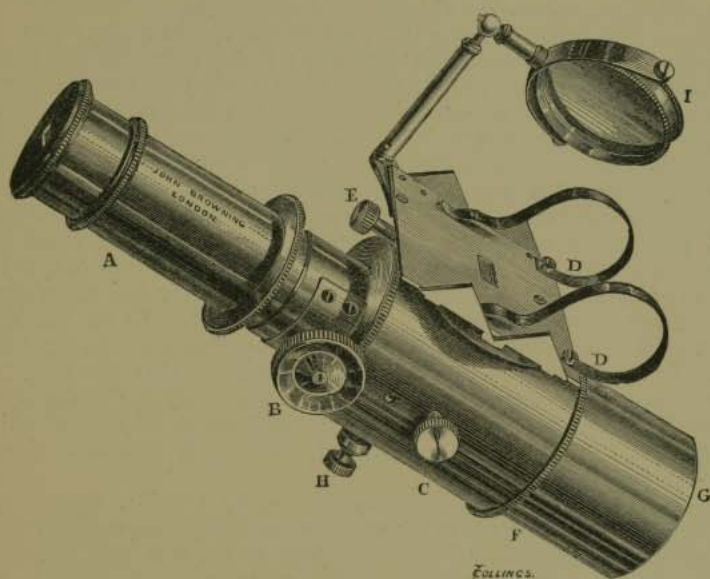


FIG. 7.

glycerine, a little diluted. The section floats on the water, and may be transferred to a dish of dilute glycerine. It is well to cut a great number of sections in this way, and select the most transparent from the dish for microscopic examination. The authors' method of observing and preparing leaves is described in the article on 'Tea.'

§ 55. *Micro-Spectroscope.* — The micro-spectroscope has become a leading instrument in food-analysis, more especially since the introduction of so many artificial colouring materials. Fig. 7 shows the various parts of the 'Sorby-Browning' micro-spectroscope. An eyepiece fits into the

¹ Sealing-wax does admirably: the particles of powder are placed on a thin layer of sealing-wax, and the wax softened by heat; on cooling, the particles are held with sufficient firmness to allow of sections.

microscope tube, having the upper lens made achromatic; at the focal point of this lens is fixed a narrow slit. A rectangular prism is fixed so as to extend over about one-half of the slit, and reflect the light coming through an aperture. In the stage attached to the side of the eyepiece, the other half of the slit transmits the light, passing up the main body of the microscope through the ordinary object glass.

When all is properly arranged and illuminated, in looking through the lens a narrow line of light is seen, one-half the length of which has passed through an object placed on the stage of the microscope, and the other half through any other placed on the side stage attached to the eyepiece of the prism; and if the prism has been properly adjusted, these two portions should appear perfectly continuous, without any break at their junction; but if not properly adjusted, the line appears broken, and would then give false results if the spectra were compared together. The analysing prism is compound, and fits over the eyepiece like a long cap. It consists of two rectangular prisms of crown glass, and two others with angles of 75° , a combination which gives direct vision.

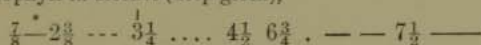
B is a milled head adjusting the focus of the eye lens (fig. 7); C is a milled head for adjusting the slit vertically; H for adjusting the breadth of the slit; D, D are springs for holding a small tube; E is for the purpose of regulating the slit of the second spectrum; F is the position of the field lens of the eyepiece; G is a tube which fits on the microscope. The prisms give that amount of dispersion which is admirably fitted for the purposes to which this instrument is applied, and is in itself sufficient to divide the absorption-bands seen in coloured solids and liquids, while it is not so great as to spread them over too wide a space, and make them obscure, as is the case when the dispersion is great. Since the light which passes through the opening does not extend over the same surface as that which passes through the object glass, it would be far too bright unless modified by means of a small shutter, opening and closing with a screw. In each case this can easily be adjusted so that the light from the two sources is equal, or may be made to vary for some special purpose; there is also a contrivance, so that when very small objects are examined no light shall pass except that which has come through them. (*Sorby.*)

Improvements have been made by Mr. Sorby and Mr. Browning, by which every line or band in the spectrum, when being measured, is brought into the centre of the field of view; the jaws of the slit open equally, so that, whatever their width may be, the zero remains unchanged. The micrometer is self-registering, and the whole turns of the micrometer screw, as well as fractional parts, can be read off at the same time by inspection. The instrument may also be used for opaque as well as transparent objects, and two spectra can be compared at the same time with one lamp. Moreover, the spectrum of the smallest object, or a particular part of any object, may be obtained without difficulty. Mr. Sorby's method of measurement is of the most accurate description. He uses an apparatus giving an interference spectrum, divided by black bands, all of equal optical value. The apparatus is composed of two Nicol's prisms, with an intervening plate of quartz, about $\cdot 043$ inch thick, cut parallel to the principal axis of the crystal, the thickness being so adjusted with the sodium line that the sodium line is exactly at 3.5, counting the bands from the red end towards the blue. He makes use of the following symbols to express the intensity of absorption:—

Not at all shaded,	Blank space.
Very slightly shaded, Dots wide apart.
Decidedly shaded, Dots closer.
More shaded, Dots very close.
Strongly shaded, but so that a trace of colour is seen,	- - - Three hyphens close.
Still darker,	— Single dash.
Nearly black,	— Double dash.

Definite narrow absorption-bands are indicated by * printed over their centre. It is assumed that there is a gradual shading off from one tint to the other, unless the contrary is expressed, which is done by means of a small vertical line, as in the following example:—

Normal chlorophyll in alcohol (deep green),



Nothing could be more accurate than Mr. Sorby's method of measurement, and for the actual worker his system of notation will also be found most convenient. For the purpose, however, of graphical illustration, 'Vogel's'¹ method is preferable, and it has been used in this work to delineate various coloured spectra. The system may be at once understood by reference to the diagram (fig. 16). The amount of absorption is shown by curves. Where the curve is highest, there the band is blackest; where it is lowest or absent, the least absorption is present. There is no doubt that the most permanently useful way to express spectra, whether absorption or spark, would be by wave lengths. Then, however the scales of different spectroscopes might differ (and scarcely two will give the same values), still the results would be the same for all spectroscopes. The following values of wave lengths are sufficient for absorption spectra:—

MILLIONTHS.	
A. 760·4	b ₄ 576·7
B. 686·7	F. 486·1
C. 656·2	G. 430·7
D. 589·2	h. 410·1
E. 526·9	H. 396·8
b ₁ 518·3	K. 393·3

By constructing a diagram similar to the following one, only many times larger, having the values of the scale marked at the top, and intersected by the lines giving wave lengths, and then determining the exact position of Fraunhofer's lines on the scale, and marking them by crosses on the chart as in the diagram, and lastly, joining the points in a uniform curve, it is possible to get very simply the wave lengths of every portion of the scale. The more uniform the curve, the greater the number of lines determined; and the larger the chart, the more accurate are the values. Supposing the centre of an absorption-band to be at 10 on the scale: on referring to the diagram the curve at 10 is exactly cut by the horizontal line 660, therefore the wave length would be 660, and so on. It will be necessary to measure in all cases the middle of the absorption-band, or the middle of the spark line.

A useful spectroscope for the examination of colouring-matters is the 'hema-spectroscope compareteur' of Maurice de Thierry.

It is a direct vision spectroscope (see fig. 9) furnished with a prism at

¹ Hermann Vogel, "*Praktische Spectral Analyse.*" Nordlingen, 1877.

P of considerable dispersive power. By casting the light through A, a scale is reflected, the divisions of the scale extending from 0 to 250, divided into tenths; by means of the screw at *b*, the reflected scale may be adjusted to the edge of the spectrum under observation, while the arm, A, admits of a slight angular movement, so that any division of the scale may be made to coincide with the spider thread carried by the ocular, O. Similarly the arm carrying O admits of angular movement by means of *a*, and the red, green, or blue may be brought in the centre of the field. The slit is at C; it is divided into two equal parts by a prism; by means of the

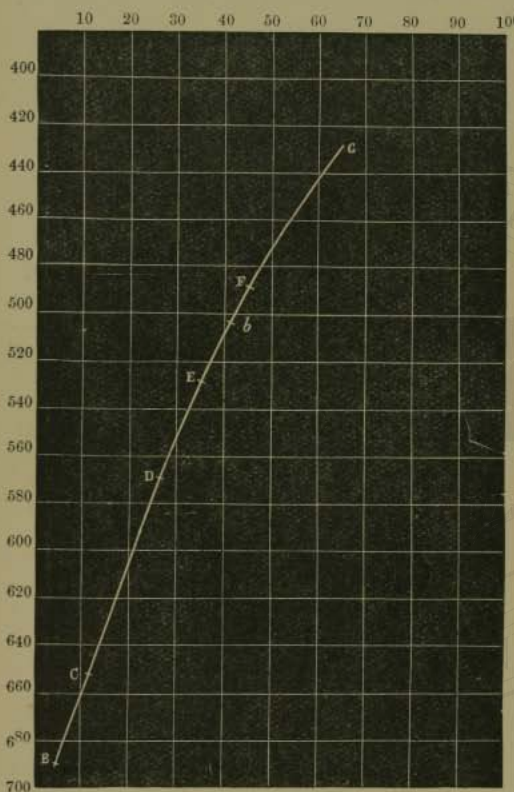


FIG. 8.

mirror, M', light may be cast through small glass vessels with parallel sides, and thus a second spectrum obtained for purposes of comparison. In the trough, G, rests a tube 50 centimetres long, quite similar to tubes for use in a saccharometer. Three of such tubes are furnished of the respective lengths of 10, 20, and 50 centimetres; a system of lenses at L condenses the light and makes the rays parallel thrown through the tube by aid of a mirror, M'', or from a direct source of light. The spectroscope can be detached altogether from the stand, fitted on to any ordinary stand, and be used for seeing line spectra emitted by coloured flames or from sparks.

Quite weak solutions of colouring matters, which give no spectra in thin layers, will give bands in one or other of the tubes. Oxyhaemoglobin thus reveals its presence in so weak a dilution as 1 to 85,000.

Measurements may be made with great facility by means of the thread in the eyepiece and the scale. Analysts generally will, however, prefer some form of spectrometer, with the circle divided into angular degrees; such an apparatus has the advantage of being available (by the use of hollow prisms) for determining the refractive index of liquids.

§ 56. The spectroscope, as applied to coloured flames, or to spark spectra, is not much used by food analysts. It will, however, probably be more used when the ash constituents of food have been thoroughly and scientifically worked out. A very careful search after the rarer metals and elements in the ash constituents of plants would, in all probability, be rewarded with the discovery of—if not *new* elements—yet of the wide

dispersion of the elements that are presumed not to be widely disseminated. The spectroscope in general laboratory use has only been applied to the diagnosis of potash, lithium, copper, barium, strontium, and a few other flame spectra easily obtained without the aid of electricity; but the interesting and convenient method of examination introduced by Lecocq

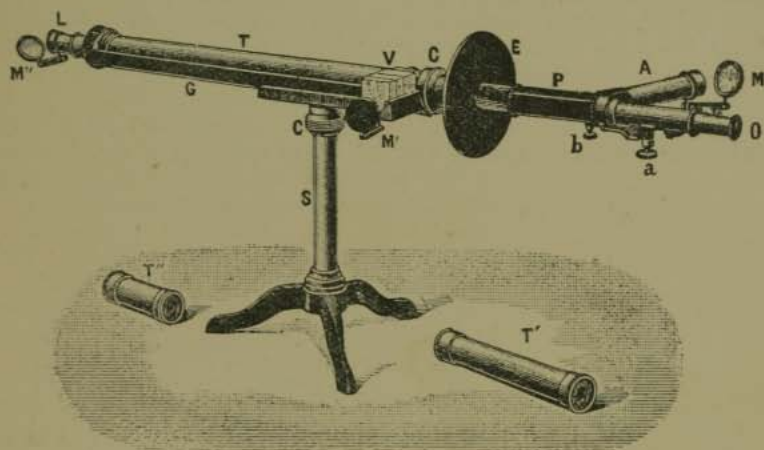


FIG. 9.

de Boisboudran has made spark spectra so very easy to be obtained by any one who has a battery, and a Ruhmkoff coil capable of giving a good spark, that there is no reason why an examination of the spark spectra of a body should not become the daily matter-of-course process in all laboratories, and not be restricted to pure scientific inquiry. Boisboudran's method is simply to pass the spark through a solution of the substance to be examined, and for this purpose the following apparatus can be constructed out of the ordinary apparatus of the laboratory (fig. 10).

A Woulfe's bottle is fitted with two good india-rubber corks. In the one a stout glass rod is placed, bent at right angles, serving as a support for a glass tube, through which the wire of the negative pole projects; the other neck carries a little test-tube with the wire, *g*, which comes up through the cork, and the test-tube supports a still smaller one, capable of holding a very small quantity of the liquid to be examined; the wire is fused into the bottom of this tube, and terminates a little below the mouth. Over the wire there is a minute tube, somewhat funnel-shaped at the end, which prevents the spark flying to the side of the test-tube; in the larger tube there is a little mercury to ensure contact. One effect of this arrangement is that the lower pole has always a thin film of the liquid over its surface, and on passing the current the spark volatilises the substances in solution, and their characteristic spectra are easily observed.

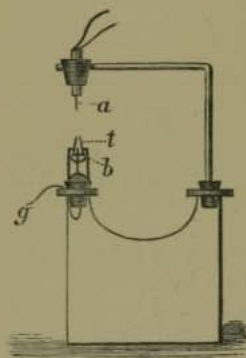


FIG. 10.

QUANTITATIVE ESTIMATION OF COLOURING MATTERS.

§ 56a. (1) *The Polarising Colorimeter.*—The simplest method is to imitate the solution of colouring matter, the strength of which is unknown, by diluting a stronger solution of known strength and placing the two solutions in two glass cylinders of equal bore; accurately match the colours by pouring some of the solution of the stronger shade out until the colours are equal, then the colouring matter is proportional to the depth or height of the liquids. The results with practice are in many cases fairly correct. A beautiful and more accurate method of colorimetry is made by the aid of a special apparatus—viz., the 'polarisation colorimeter'; this, in its most improved form, consists of two graduated cylinders, A and B, having

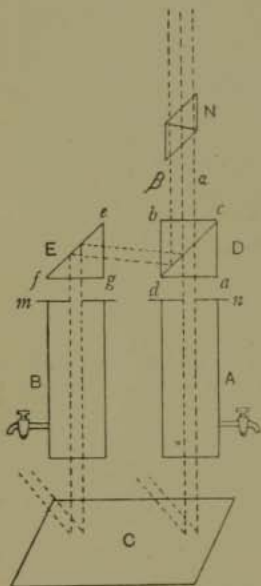


FIG. 11.—The Polarising Colorimeter.

at the bottom a white reflecting surface, C, which throws the light equally through the two cylinders (fig. 11). Above the cylinder, A, is a prism, D, of Iceland spar, the so-called 'Glans air prism.' The prism is cut in the direction *cd*, and both halves again joined together. The surfaces, *ad*, *cb*, and *bd*, are polished; *m* and *n* are diaphragms, admitting the rays of light thrown up from C. The ordinary ray passing from A is reflected at the surface, *cd*, while the extraordinary ray, *a*, passes on to the Nicol. The rays of light passing through B into the Iceland spar half prism, E, divide into extraordinary and ordinary rays; the extraordinary rays pass through the prism, the ordinary are reflected from the polished surface, *ef*, are again reflected by the surface, *cd*, and pass on to the Nicol, forming the bundle, β . The Nicol prism thus receives ordinary rays from B and extraordinary rays from A. The Nicol prism can be rotated on its axis, and the amount of rotation measured by a divided circle.

There will be two points, in one of which the extraordinary rays will vanish, and in the other, 90° from the former, the ordinary ray will vanish. Let these points be 0° and 90° .

If both rays come with equal clearness from D, then at 45° —that is, exactly between 0° and 90° —the two fields will be equally clear.

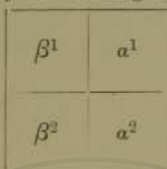
The clearness of the ray, *a*—that is, the extraordinary ray—is at $0^\circ = 0$, at $90^\circ = 1$; the other ray, the ordinary, β , is exactly the reverse—that is, at zero it is equal to 1, at 90° it is equal to 0. The angle, ϕ , measured by the rotation of the Nicol with regard to the ray, *a*, is proportional to the square of the sine, that of the ray, β , is proportional to the square of the cosine.

If both rays are equal in intensity, then for any angle, ϕ ; $a : \beta = \sin^2 \phi : \cos^2 \phi = \tan^2 \phi : 1$. The simplest method of using the polarisation colorimeter is always to fill the cylinders, so that the liquid of known concentration stands at the same height as the liquid of unknown concentration. The Nicol is turned until both fields are of precisely equal brightness, and the angle, ϕ , being read, the cylinders are now reversed—that is, the cylinder A is put in the place of B, and the cylinder B in place of

A, and the angle, ϕ' , again read; then the calculation is very easy, denoting the concentration of the one liquid as c , the other as c' , and the angle as ϕ .

$$c = c' \frac{\tan \phi}{\tan \phi'}$$

An improvement, even on this method, is the introduction of a quartz plate between the Nicol and D; the quartz plate has a thickness of 3.75 mm., and consists of one-half of right-handed, the other of left-handed quartz. The result of this is that the eye sees four fields, the ray β dividing into two and the ray α dividing into two.



The Nicol being at 0° , or at 90° , the fields, β^1 and β^2 , are of an equal colour as well as those of α^1 and α^2 , at a point between the two angles; α^1 and β^2 are of an equal yellow colour, while β^1 and α^2 are of a blue colour.

If in the cylinders, A and B, are placed solutions of a colouring material of different concentration, the one known, the other not, both fields lying to the right have a different degree of clearness to the fields lying to the left; and since two solutions of a substance of different concentration show a different spectrum, in this case the field α is differently coloured to the field β^2 , and β^1 differently coloured to α^2 .

By diminishing the height of the stronger liquid, by letting a little flow out of the tap, after a few trials not only the adjacent fields have the same brightness, but also the same colour.

If the Nicol was placed at 45° , and the height of one or the other fluid be decreased by means of opening the tap, then the concentration of the unknown liquid is given by the equation:—

$$c = c' \frac{h'}{h}$$

c = unknown concentration.

c' = known concentration.

h' = height of c' .

h = height of c .

Krüss¹ gives examples of the accuracy of the determinations of various substances by this method, as follows:—

	Actual Contents.	Amount found by the Colorimetric Method.
Copper sulphate solution— (1 c.c. = 0.025 gm.), .	0.05000 gm.	0.05009 gm.
Nickel sulphate— (1 c.c. = 0.025 gm.), .	0.05000 „	0.05013 „
Potassium monochromate— (1 c.c. = 0.375 gm.), .	0.07500 „	0.07535 „
Ammonia solution, to which Nessler's solution has been added,	0.000652 mgrm.	0.000650 mgrm.

¹ *Kolorimetrie u. Quantitative Spektralanalyse*, by Dr. G. Krüss and Dr. H. Krüss. Hamburg and Leipzig, 1890.

The small error appears generally to be in excess, and, no doubt, a correction factor could be got out for any solution.

An improved form of colorimeter has also been invented by Hugo Krüss,¹ called "colorimeter with the Lumner-Brodhuns prism." The essential part of the instrument is the Lumner-Brodhuns prism (see A, fig. 12); the one part of the prism is an ordinary reflecting prism, but the other part has a polished plane surface, gr ; but from e to g and from r to s the section is that of a portion of a circle.

The other prisms have the shape shown in fig. 12, and the path of the rays of light is indicated by the dotted lines. The effect of the arrangement is that the eye, looking through the lens on to the surface $ergs$, sees an elliptic spot, which, if the light coming from the two cylinders is unequal, is surrounded by a field of a different tone with a sharply defined line; but if the light is equal in intensity, then the line of demarcation vanishes, and the spot is of the same shade and hue as that of the surrounding field. This colorimeter is stated to be of great delicacy and accuracy.

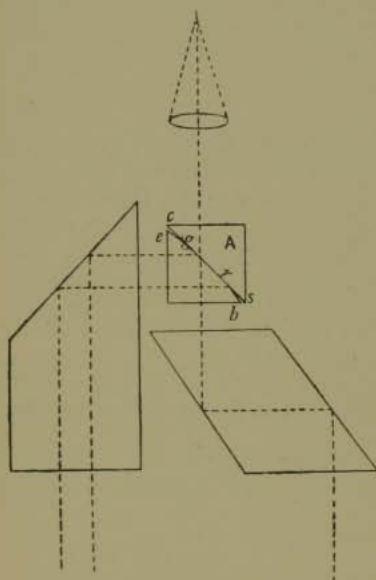


FIG. 12.—Colorimeter with the Lumner-Brodhuns Prism.

Colorimetry is applicable to the estimation of ammonia, nitrites, nitrates, copper, permanganate, and solutions generally, either coloured in themselves or striking colours with reagents.

§ 56b. (2) *Quantitative Spectroscopy*.—Quantitative estimations by the spectroscope may be made by an appliance, the invention of Karl Vierordt,² known as the 'double slit'; instead of a single slit, the slit is divided into two halves, as shown in fig. 13; each half of the slit can be opened at a different degree, and the

width to which they are respectively opened, accurately estimated by means of the divisions on the milled heads of the respective micrometer screws, i and i . Both of these slits correspond to two spectra, as seen through the observation tube. If the light is properly placed and both slits equal in breadth, the illumination of both spectra will be equal; on the other hand, if one slit is open of a greater width than the other more light will pass through. If in front of one half of the slit is placed a liquid which absorbs the light more or less, in order to make the illumination, say, of the brightest part of the spectrum equal to the other spectrum illuminated directly by the source of light, the two slits will have to be of a different width, the one having to be narrowed or the other widened, and by reading from the milled heads the width of the slit, a basis

¹ Kolorimeter mit Lumner Brodhunschen Prismen-paare v. Hugo Krüss. *Zeit. f. anorgan. Chemie*, v.

² *Die Anwendung des Spectral-apparates zur Photometrie der Absorptions-spectren u. zur quantitativen chemischen Analyse*. Tübingen, 1873.

for calculation of the concentration of the solution is obtained by determining what is called the 'extinction coefficient.'

The following is the method of obtaining the extinction coefficient of a solution:—A small glass trough, with parallel sides, is taken, the walls of the trough being exactly 11 mm. from each other; the lower half of this glass trough is occupied by a Schulz's glass body, a solid bit of glass, which occupies 10 mm.; hence, it is obvious that when the trough is charged with a liquid, the light will pass through 1 mm. in the lower half and 11 mm. in the upper half of the trough. In other words, light passing through the upper part will be weakened in the proportion to the lower as 10 is to 1; it will pass through 1 mm. of the lower half and 11 mm. of the upper half. This trough is so adjusted before the divided slit of the spectroscope that the light passing through the one half slit is wholly derived from the upper half of the trough, and that passing through the other half slit is wholly derived from the lower half.

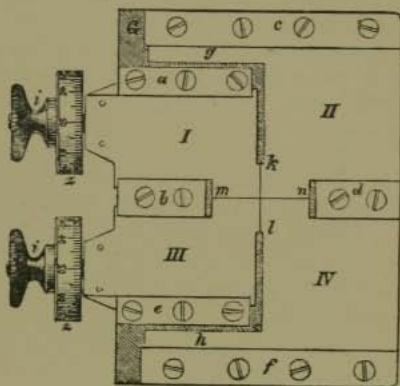


FIG. 13.—Vierordt's Double Slit.

Before the trough is placed in position both slits are opened an equal width, and the light adjusted until an exactly equal illumination is obtained. On now adjusting the trough the illumination will, of course, be unequal.

A complete revolution of the micrometer screw is equal to 100 divisions, and supposing that to make the light equal, the one micrometer screw has to be turned down to 30, then, as the intensity of the light falling through the 1 mm. is to be considered as equal to 1, the unabsorbed light is in the proportion of 30 to 100, or as 0.3 to 1. The light passing through the 1 mm. being equal to 1, and the light passing through the solution to 0.3, then the extinction coefficient is equal to the reciprocal or arithmetical complement of the logarithm of 0.3, or $e = -\log 0.3$.

If a table of logarithms be consulted, the logarithm of 0.3 will be found to be $\bar{1}.47712$; if the last figure in the decimal portion be subtracted from 10, and the remainder from 9, and the index or characteristic be diminished by 1, this gives the arithmetical complement—that is, 0.52288, which is the extinction coefficient.

The extinction coefficient being known, the absorption proportion must be obtained, and this is known if the concentration is known, for

If c = the concentration expressed as grammes per cubic centimetre.

e = the coefficient extinction.

A = the absorption proportion.

Then,

$$A = \frac{c}{e}.$$

If the absorption proportion is known, the concentration unknown, all that the experimenter has to do is, for one or more definite regions of the spectrum, to work out the extinction coefficient, multiply this by the

absorption proportion for that particular region of the spectrum, and the result equals the grms. per c.c.—that is,

$$c = e A.$$

In other words, once the absorption proportion for various regions of the spectrum is known, it is easy to ascertain the percentage composition. The more regions of the spectrum investigated, of course, the more accurate is likely to be the determination.

As an example, let us take the absorption spectrum of permanganate. For wave lengths, 680.7 to 650, Krüss found that a solution containing 0.001 gm. per c.c. gave a coefficient extinction of 0.47238; and for a concentration of 0.00025 per c.c. a coefficient extinction of 0.11351.

Therefore dividing c by e in each case, for the one we get as the absorption proportion 0.002116, for the other 0.002202, the mean of which is 0.002159.

In the region embraced between the wave lengths 596.4 and 582.8, for solutions containing respectively 0.00025 gm. and 0.000125 gm. per c.c., Krüss found the coefficient extinction to be 0.40561 and 0.16242; the mean of the results of

$$c \text{ divided by } e \text{ is } 0.006845;$$

and so on for various portions of the spectrum. For example, if it were desired to ascertain the strength of a solution of potassic permanganate, the unknown solution would be diluted until, from its colour, it was judged to be somewhere near the strength of the solution whose absorption proportion was known and several extinction coefficients obtained. Thus, in the present instance, supposing for the wave lengths 680.7 to 650.1 an extinction coefficient of 0.47238, and for the wave lengths 613.2 to 596.4 a coefficient extinction of 1.08093 was obtained, the absorption proportion for the respective wave lengths being known to be 0.00215 and 0.0009186, we should have—

$$[e \times A = c.]$$

$$(1) 0.47238 \times 0.002159 = .00102$$

$$(2) 1.08093 \times 0.0009186 = .00124$$

giving a mean value of 0.0011, the real value in this case being in a c.c. .001; a nearer approximation could be made to the true value by more determinations.

For every coloured substance there are special regions of the spectrum most suitable for quantitative estimation, and it is necessary in ascertaining the 'absorption proportion' to measure carefully the proportions of the spectrum observed; for example, Krüss finds that the most suitable regions for the quantitative estimation of potassic permanganate solution are as shown in the following table, which also gives the absorption for those regions:—

Wave Lengths.	Absorption Proportion.
λ 494.7 to λ 486.5	0.0001909 ¹
λ 486.5 ,, λ 480.9	0.0002251
λ 480.9 ,, λ 474.8	0.0003277

¹ If oxygen is to be determined by permanganate, then the absorption, in terms of oxygen, is for the wave lengths given—0.00004833, 0.00005699, and 0.00008296.

Three solutions of permanganate gave the following strengths, as estimated by the spectral method, using alone the three regions mentioned: 0.00189 grm., 0.00218 grm., and 0.00228 grm. per c.c., while titration with sodic thiosulphate gave 0.00191, 0.00217, and 0.00229, thus proving the great accuracy and convenience of the method.

As before mentioned, any spectroscope with an accurate scale may be used, but Krüss has invented an instrument which has some special advantages; it is called the 'Universal Spectral Apparatus,' and is represented in fig. 14.

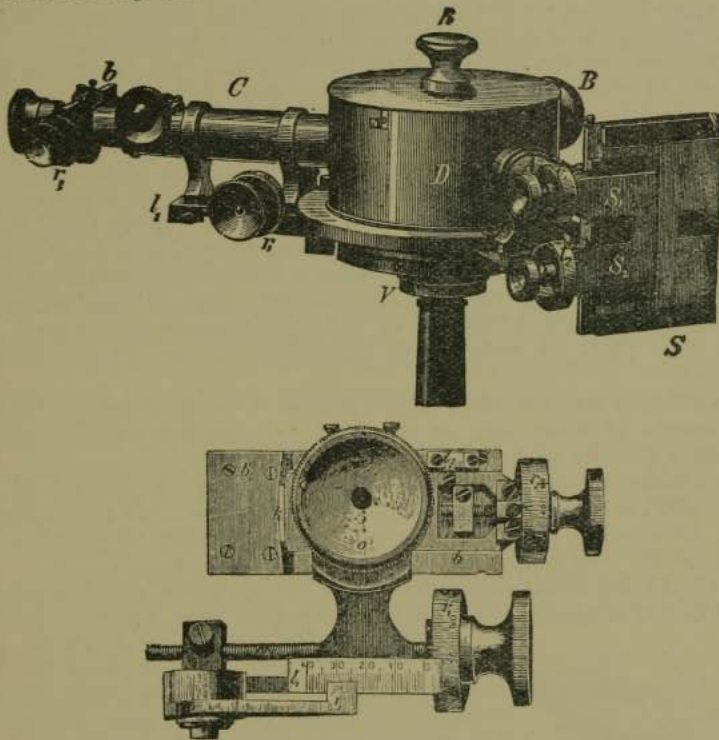


FIG. 14.—Krüss's 'Universal Spectral Apparatus.'

S is the double slit before described, and is carried by the collimator, A; the edges of the slits are of platinum, and both edges open equally; B is a tube carrying the ordinary photographic scale, the image of which can be thrown on the prism; the 100 on the scale is made to coincide with the sodium line, D. The prism is contained in D; two prisms are supplied with the apparatus, one a flint-glass prism with a refracting angle of 60° and a dispersion extending from A to $H_2 = 4^\circ 30'$, the other a Rutherford prism with a dispersion from A to H_2 of 8° to 11° . The first is used for qualitative work, and in cases where good illumination is preferred; the second when a long spectrum is necessary. C is the telescope, and the eyepiece contains cross threads; this piece carries the special measuring apparatus, which is represented in detail in the figure. This measuring apparatus is accessory to the scale, already mentioned, carried by B, which,

for most purposes, is all that is necessary ; but for fine measurements the micrometer arrangements, r_1 , l_1 , and r_2 , l_2 are useful.

The lower milled head micrometer screw has its circumference, r_1 , divided into 100 divisions ; it moves the telescope round the axis, V, and the cross threads of the telescope being made to coincide with any line or band, by means of the scale, l_1 , i_1 and the divided circle, r_1 , a measurement expressed by four figures can be made. Similarly the upper milled head works a micrometer screw, and is supplied with a similar scale, l_2 , r_2 . This upper screw moves the cross threads, and by its aid very fine measurements may be made ; k is a vertical slit in a shutter, and its use is to shut out all other parts of the spectrum save that under observation.

Whoever uses the double slit for the first time will experience some difficulty in educating his eye to appreciate small differences in the upper and lower half of the strip of colour. The main errors of observation are due to placing the eye in such a position that more of the one slit is seen than the other. It is essential to success for the two portions observed to be nearly equal. The double slit can also be used as a colorimeter ; that is to say, by the use of a reflecting prism and two fairly equal lights, the illumination by a little management is made equal and then the light from the one solution, whose value is known, is caused to pass through the one slit, and the other rays from the second source of illumination are made to pass through the solution of unknown concentration through the other slit. Then by widening or narrowing the one slit the light is equalised.

In a similar way the colour of two samples of water can be appreciated ; or a sample of water can be compared with distilled water. For this purpose a stratum of from 10 to 20 c.c. is necessary.

In making comparative colorimetric observations by the aid of the double slit, the solutions by suitable dilution should be made approximately equal, for equality produced by a moderate narrowing or widening the slit gives the best results.

§ 57. *Photography.*—The introduction of dry plates and the general simplification of photography will, in a very little time, make its practice general in all the larger laboratories for purposes of registration. In important analyses, likely to entail evidence in the higher courts of justice, it might be useful (and will always be possible) to photograph certain analytical results.

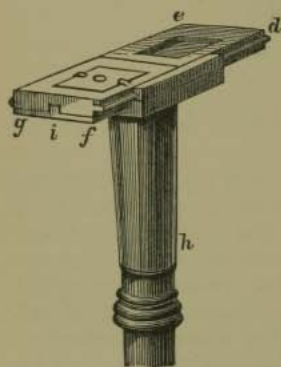


FIG. 15.

In the quantitative determination of mixtures of starches, a micro-photograph of the mixed starch and the 'imitation' mixtures renders the counting of the number of starches in the field a very easy operation. Similarly, if a measurement of any object be required, a micro-photograph having been taken, and next a photograph of the stage micrometer with the same powers, the object may be measured more easily than in the ordinary way. It is also most useful for the analyst to have by him, in

this way, a series of 'picture records' for reference. Stein's photographic microscope,¹ when the object is not to make pretty pictures, nor to use

¹ *Das Licht im Dienste wissenschaftlicher Forschung*, by Dr. Th. Stein. Leipzig, 1877.

high powers, is excellent. It merely consists (see fig. 15) of a conical tube, turned out of boxwood, and fastened on to the microscope tube by means of a screw, *h*. Combined with this funnel tube is a wooden tray, in which the frame, *e, d, f, g*, easily slides backwards and forwards. Thin panes of glass are let in the cassette into this frame. Whilst the image is being adjusted, the thin glass, *e, d*, stands over the tube, and the prepared plate is put under the little cover at *g, f*. If the picture is well defined, the frame, *e, d, g, f*, is pushed into the tray, so that the part, *g, f*, can stand over the microscope tube, and by a simple arrangement the photographic plate can be exposed. Direct sunshine will, in some cases, be necessary, and the rays should be transmitted through a cell containing the ammonia sulphate of copper.

Quite as good results may, however, be obtained without special apparatus.

Place the microscope without the eyepiece in a dark room. Support the ground glass in a frame by means of a clamp in such a position that it is exactly in focus, lift it off without shaking the instrument, and substitute a sensitive plate.

An ordinary electric lamp is enclosed in a box of such size that it can be placed underneath the stage of the microscope, the light from the lamp passing through a circular hole. Should an electric light not be available, then a wooden box with a loosely-fitting lid should be mounted on stout legs. The edges of the lid should be lined with velvet. Underneath the box a hole is cut, which is adapted to the wide end of the bellows of a quarter-plate camera.

The narrow end of the box is adapted to the top of the microscope tube; a couple of projections in the interior of the box are placed to support the sensitive plate or ground glass. The source of light may be any ordinary lamp.

Provided the microscope has a short barrel, and that all the interior is well blackened, excellent photographs can be taken in this way.

To photograph with high powers it is, however, best to purchase the more elaborate apparatus sold by opticians.

§ 58. *Colour*.—It will often be necessary to ascertain the exact colouring matter used to make articles of food attractive, more especially confectionery, jellies, pickles, etc. The question will generally resolve itself into deciding as to whether the colour is harmless or poisonous, and if the latter, whether the poison is in sufficient quantity to injure the consumer's health. The poisonous colouring matters are those containing lead, copper, arsenic, chromium, and zinc, all of mineral origin; together with a few injurious organic colouring substances, such as gamboge and picric acid. The non-poisonous colouring matters are some of the aniline colours, *so long as they are pure, and contain no arsenic*—saffron, turmeric, annatto, chlorophyll, and generally (with some exceptions) all organic colours obtained from the vegetable and animal kingdoms.

The first thing for the analyst to ascertain is whether the colouring material is insoluble or soluble in water, for, as a rule, with the exception of gamboge, the harmless colours are soluble, while the mineral are insoluble in water. The organic colours are also bleached by a solution of hypochlorite of soda. The aniline colours are soluble in alcohol.

The search for poisonous matters more properly belongs to, and is treated of in, the work on 'Poisons' by one of the authors. With the exception of salts of lead and copper in small quantities, they are rarely met with in

food, and even in the matter of confectionery, of late years, there has been a great improvement. As a rule, sweetmeats in England are not coloured with injurious matters.¹

The analyst having settled that the colouring matter is one of organic origin, by its being bleached by sodic-hypochlorite, and by its solubility in water or alcohol, will next proceed to study its spectroscopic characters, either by using a pocket spectroscope or the micro-spectroscope already described.

Mr. Sorby makes use for his instrument of little cells, cut from barometer tubing. They are half-an-inch long, and with an external diameter of somewhat under half-an-inch; they are ground flat at each end, and cemented with Canada balsam near one edge of a glass plate, so that they may be examined sideways or endways. In examining an unknown colouring matter, he adopts the following divisions:—

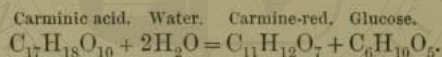
1. Soluble in water, and not precipitated by alcohol.
2. Soluble in water, but precipitated by alcohol.
3. Insoluble in water, but soluble in alcohol.
4. Insoluble in water and alcohol.

He next subdivides his main divisions according to the action of bisulphite of soda.

The organic colouring matters most likely to be found may be treated of in the order of the spectrum, beginning with the red.

§ 59. REDS.—The common reds are—cochineal, aniline reds, alkanet, and the madder colours alizarine and purpurine.

Cochineal.—Cochineal is a red complex colouring matter, secreted by certain species of a peculiar family of insects feeding on the *Cactus coccinellifera*, *C. opuntia*, *C. tuna*, *C. pereskia*. The chief colouring matter of cochineal is 'carminic acid,' the formula of which appears to be $C_{17}H_{18}O_{10}$. By the action of dilute acids carminic acid splits up into sugar, and a beautiful colour known as *carmine-red* thus—



Cochineal imparts its colouring matters both to alcohol and water, and is precipitated by acetate of lead, carminate of lead being one of the constituent parts of the precipitate. The solutions of cochineal are purplish-red to crimson, turning a more or less rich violet-purple with alkalis, and becoming of a yellow colour on the addition of acids. The colour is well-known to chemists, as it is much used as an indicator for acids, being especially useful in titrating an alkaline liquid containing carbonates, since carminic acid is not affected by carbon dioxide like so many other colouring matters.

Cochineal in neutral solutions gives absorption bands, but not very

¹ This is the more necessary to state clearly, since, on the Continent, very erroneous ideas prevail. Thus, in the "*Dictionnaire des Alterations et Falsifications*," par M. A. Chevallier et M. Fr. Baudrimont, Paris, 1878, the adulterations of half a century ago are enumerated; and the reader is informed that the English confectioners not only falsify their sweetmeats with plaster, lime, starch, baryta, but frequently employ bronze powder, the leaf foil of copper, tin, and carbonate and arsenite of copper, verdigris, chromate of lead, red lead, and vermilion; and, further, that nearly all the ginger lozenges contain lead. Similarly, in Dr. Hermann Klencke's "*Lexicon der Verfälschungen*," in the article, "Conditiorwaaren," it is stated that almost all the English confectionery contains lead salts, often to the extent of one and a half per cent.!! All this is nonsense. Such adulterations have been found, it is true; but instead of being common, they are rare and exceptional.

definite when examined by the spectroscope; if, however, it be made ammoniacal, then there are bands which differ in position only slightly from the absorption bands of blood.

No. 18 (fig. 16) is a graphical illustration of the spectrum of cochineal in water; No. 19, in alcohol; and No. 20, on the addition of nitric acid (a) or NH_3 (b). If alum is added to cochineal it loses its power of turning yellow with acids, and the purpurine band becomes so broad that the two bands almost run into each other. On addition of acetic acid they are separated, and appear as tolerably sharply defined bands between D and E, and there is another at D.

On dissolving cochineal with alum solution, a lake is obtained; on dissolving this in tartaric acid or dilute nitric acid, the solution gives a band at b and E, and another close on D. The nitric acid solution gives a spectrum very similar to blood.

An aqueous solution of cochineal may be distinguished from the red solutions of brazil-wood, sapan-wood, peach-wood, and a few others, by the fact that the calcium salt of their colouring matters is violet, and readily soluble in water, while the calcium salt of cochineal-red is dark-purple or almost black, and insoluble in water.

The following are the absorption factors for carminic acid, as obtained by using a solution containing 0.0001 grm. per c.c. with a drop of NH_3 ¹ :—

At Temp. 20.0°.	Absorption Factor.
λ 627.5 to λ 609.5	0.00113
λ 583.6 ,, λ 576.6	0.00015
λ 552.6 ,, λ 540.6	0.00010
λ 522.7 ,, λ 513.2	0.00008
λ 501.6 ,, λ 493.6	0.0001
λ 482.8 ,, λ 475.0	0.00014

Aniline Reds.—The aniline reds are numerous; the chief are fuchsine, safranine, and coralline. These three may be roughly distinguished from each other by adding a dilute mineral acid: fuchsine becomes yellow, safranine violet-blue, and coralline gives a yellow precipitate.

Fuchsine, or *Rosaniline*, also called magenta, aniline red, and other names. It is a mixture of hydrochloride or acetate of para-rosaniline (triamido-triphenyl-carbinol) and rosaniline (triamido-diphenyl-tolyl-carbinol). It is distinguished from coralline, which gives a very similar spectrum (see No. 28) by the yellow colour with acids already mentioned.

The absorption spectrum of fuchsine has been studied by many observers, among others by Vierordt² and by Hartley.³

In weak solutions (0.024 mgrm. per c.c.) it shows an absorption band in the visible spectrum when viewed through a stratum 4 mm. thick, extending from λ 546.7 to λ 535.0; in stronger solution, 0.12 mgrm. per c.c., and viewed in a layer 4 mm. in thickness, the band in the visible spectrum

¹ Ueber den Einfluss der Temp. gefärbter Lösungen u. die Absorption Spectren derselben zur quantitativen Spektralanalyse, by G. Krüss u. H. Krüss. *Zeit. f. anorgan. Chemie*, i.

² *Op. cit.*

³ "The molecular structures of carbon compounds and their absorption spectra." *Journ. Chem. Soc.*, li., 1887.

occupies the region from $\lambda 462$ to $\lambda 580$; there are also two other bands in the ultra-violet—viz., one from $\lambda 300$ to $\lambda 283$, and another from $\lambda 247$ to $\lambda 231$, both, of course, invisible save with a Soret's ocular or other similar arrangement. A solution of rosaniline hydrochloride in alcohol (0.155 mgrm. per c.c.) in a layer of 5 mm. gives an absorption in the visible spectrum from $\lambda 591$ to $\lambda 456$, and in the ultra-violet from $\lambda 310$ to $\lambda 274$.

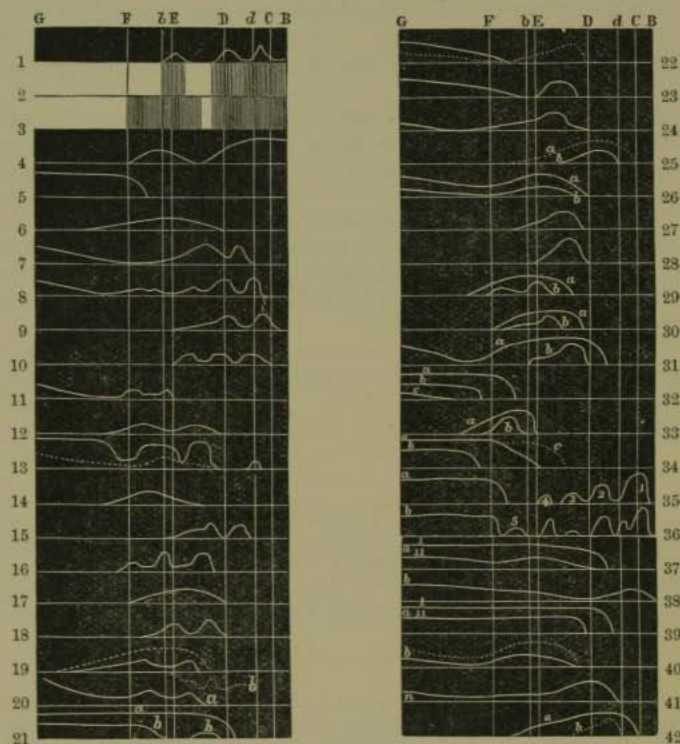


FIG. 16.

The absorption factor for the spectrum, from *a* to *H*, has been worked out by Vierordt, and, reducing his notation to wave lengths, the following are the results from *B* to *F* :—

- | | |
|-----|---|
| No. | 1. A diagrammatic representation of the bands of No. 2. |
| " | 2. Absorption spectra of clear blue cobalt glass. |
| " | 3. Absorption spectra of dark blue cobalt glass. |
| " | 4. Diagrammatic representation of No. 3. |
| " | 5. Alcoholic solution of alizarine. |
| " | 6. Alcoholic solution of alizarine made alkaline with ammonia. |
| " | 7. Aqueous ammoniacal solution of alizarine. |
| " | 8. Alcoholic solution of alizarine made alkaline by potash. |
| " | 9. Sulphoxanthraquinone in alcoholic solution alkalisied by potash. |
| " | 10. Aqueous solution of sulphoxanthraquinone made alkaline by potash. |
| " | 11. Alcoholic solution of purpurine. |
| " | 12. The same alkalisied by ammonia. |
| " | 13. The same alkalisied by potash. |
| " | 14. A neutral solution of alizarinamid. |
| " | 15. An ammoniacal solution of alizarinamid. |
| " | 16. A neutral solution of purpurinamid. |

- No. 17. The same alkalisied by baryta.
 ,, 18. An aqueous solution of cochineal.
 ,, 19. A dilute alcoholic solution of cochineal.
 ,, 20. Cochineal in concentrated watery solution with (a) nitric acid, (b) ammonia.
 ,, 21. Logwood, (a) concentrated watery solution, (b) dilute.
 ,, 22. The same with the addition of nitric acid.
 . . . The same alkalisied by ammonia.
 ,, 23. A decoction of brazil-wood.
 ,, 24. The same alkalisied by ammonia.
 ,, 25. Litmus, (a) concentrated, (b) dilute.
 ,, 26. The same made acid, (a) concentrated, (b) dilute.
 ,, 27. Dilute solution of fuchsine.
 ,, 28. Alcoholic solution of coralline.
 ,, 29. Alcoholic solution of eosin, (a) concentrated, (b) dilute.
 ,, 30. Safranine, (a) concentrated, (b) dilute.
 ,, 31. Naphthaline red, (a) concentrated, (b) dilute.
 ,, 32. Curcuma, (a) concentrated, (b) dilute, (c) strongly diluted.
 ,, 33. Fluoresceine, (a) somewhat concentrated, (b) dilute.
 ,, 34. Fustic extract.
 ,, 35. Fresh chlorophyll in alcoholic solution.
 ,, 36. Old chlorophyll solution.
 ,, 37. Wine colouring-matter, (I.) pure, (II.) diluted.
 ,, 38. Wine colouring-matter + NH₃.
 ,, 39. (I.) Mallow colouring-matter concentrated, (II.) Elderberry concentrated.
 ,, 40. Acid cherry :—(b.) Acid cherry, with addition of tannin.
 ,, 41. Mallow colouring-matter, with the addition of alum.
 ,, 42. Indigo solution.

BRIGHTER PORTION OF THE SPECTRUM EXTENDING FROM B NEARLY TO D.

Wave Lengths.	Absorption Factor.
679.9 to 663.2,01447
663.2 ,, 646.2,00449
646.2 ,, 629.4,00207
629.4 ,, 612.6,00117
612.6 ,, 595.9,000517

DARKER PORTION OF THE SPECTRUM FROM ABOUT D TO F.

595.9 ,, 582.3,000127
582.3 ,, 569.9,0000592
569.9 ,, 557.0,00002238
557.0 ,, 546.8,00001248
546.8 ,, 535.0,000007819
535.0 ,, 523.6,000009669
523.6 ,, 516.3,00001222
516.3 ,, 508.5,00001376
508.5 ,, 501.1,00001394
501.1 ,, 494.2,0000153
494.2 ,, 486.0,0000176

Safranine dissolves in alcohol with a fine rose-red colour, with a weak red fluorescence. Its spectrum is shown in No. 30 (a, concentrated solution, b, dilute); it is like that of eosin (No. 29), but the spectrum of eosin is changed by nitric acid, that of safranine is unchanged. From a solution containing safranine, safranine may be extracted by shaking it up with its own volume of amyl-alcohol in any ordinary separating tube.

The spectrum of naphthaline-red is figured No. 31, after Vogel, but it is not quite accurate, for it gives a delicate shaded band from $\lambda 536$ to $\lambda 558$, and a band extending from E to $\lambda 472$.

Coralline and *Aurine* (rosolic acid) give a very similar spectrum. The spectrum of aurine, according to Noel Hartley, in 0.00058 grm. per c.c., viewed in a layer of 3 mm., consists of a band, $\lambda 516$ to $\lambda 361$, in the visible, and also a band in the ultra violet spectrum, $\lambda 286$ to $\lambda 256$; in weaker solution (0.000116 grm.) there is a single band, $\lambda 484$ to $\lambda 467$.

Ponceau 2 R ($C_{18}H_{14}N_2O_7S_2Na_2$), the sodium salt of xylene-azo- β -naphthol-disulphonic acid ;

Ponceau 3 R, the similar salt of cumene ;

Congo-red, $C_{32}H_{22}N_6O_6S_2Na_2$, the sodium salt of diphenyl-diazo binaphthionic acid, all give a similar spectrum, a broad band in the green, from about $\lambda 448$ to $\lambda 488$.

Erythrosine (phloxine), $C_{20}H_6O_5I_4Na_2$, the sodium (or potassium) salt of tetra-iodo-fluoresceine, gives two bands in dilute solution, the one extending from $\lambda 488$ to $\lambda 530$, then a narrower band between D and E, $\lambda 550$ to $\lambda 558$.

Fast Red, $C_{20}H_{13}N_2O_4SNa$, the sodium salt of *p*-sulpho-naphthalene-azo- β -naphthol, gives in solutions of 0.04 mgrm. per 100 c.c. viewed through 1 mm., a band between F and G, $\lambda 441$ to $\lambda 439$.

Biebrich Scarlet, $C_{22}H_{14}N_4O_7S_2Na_2$, the sodium salt of sulpho-benzene-azo-sulphobenzene-azo- β -naphthol, gives in a solution, 3 mm. thick, strength 0.56 mgrm. per 100 c.c., a band extending from $\lambda 518$ to $\lambda 438$ (*Hartley*).

Croceine Scarlet, $C_{24}H_{18}N_4O_7S_2Na_2$, the sodium salt of sulpho-toluene-azo-toluene-azo- β -naphthol- β -sulphonic acid, gives in a solution, 2 mm. thick, strength 0.45 mgrm. per 100 c.c., a band from $\lambda 520$ to $\lambda 479$.

Alkanet, the root of *Anchusa tinctoria*, contains a red colouring-matter, insoluble in water, but soluble in alkalis, alcohol, ether, and fatty oils. The colouring-matter appears to be anchusic acid, $C_{35}H_{30}O_8$. In dilute solutions the spectrum shows three absorption-bands; on the addition of a trace of a magnesium salt, a fourth absorption-band appears, hence alkanet-red is a test for magnesium salts, and conversely a magnesium salt is a test for alkanet-red.¹ It may not unfrequently be found as the colouring-matters of tooth tinctures, hair oil, etc.

Madder.—The root of the madder, *Rubia tinctorium*, contains two colouring-matters—alizarine and purpurine—with others less studied.

Alizarine, $C_{14}H_8O_4$, crystallises from an alcoholic solution in yellowish-red crystals, and may be sublimed as brilliant red needles at temperatures a little above 100° . The needles are sparingly soluble in water, but dissolve freely in alcohol and ether. Alizarine is now made artificially on a large scale. The alcoholic solution of artificial alizarine shows no bands, but there is extinction of the violet up to the green (see No. 5). On the addition of ammonia the solution changes to a beautiful red, with weak bands in the green (No. 6); the aqueous ammoniacal solution gives two bands (No. 7); the alcoholic solution made alkaline with potash gives evidence of a third feeble band (No. 8). Natural alizarine is not now much used, but it may be at once distinguished from the artificial by its giving absorption-bands in an alcoholic solution.

Alizarine may be also distinguished by chemical tests: copper acetate added to a solution in alcohol gives a purple precipitate, aluminium acetate gives a red precipitate in an alkaline solution, and ferrous acetate and other iron salts give a dark blue violet precipitate. Blue precipitates are also formed on the addition of either barium chloride or calcium chloride to alkaline solutions. These precipitates have respectively the composition $C_{14}H_6O_4Ba$ and $C_{14}H_6O_4Ca$.

Purpurine, $C_{14}H_8O_5$, crystallising from an alcoholic solution in yellow

¹ Magnesium salts also alter more or less characteristically the spectrum of juice of elder berries, the colouring-matters of the beet, dahlia, dragon's mouth, horse chestnut, hyacinth, violet, rhododendron, purple aster, and *Primula farinosa*. *Berichte der deutsch. chem. Gesellschaft*, xiii. 766-768.

needles, and subliming like alizarine in red needles, dissolves in alkalis with a dark red colour; it gives not blue but purple precipitates with the chlorides of calcium or barium. The alcoholic solution gives (see No. 11) weak absorption-bands at F and at *bE*. The spectrum on the addition of ammonia or potash becomes very characteristic (see Nos. 12 and 13).

Safflower.—The safflower, *Carthamus tinctorius*, contains in its petals several colouring-matters, chief among which is carthamine or carthamic acid, $C_{14}H_{16}O_7$. Carthamine turns red in alkaline solutions, and may be precipitated red by an acid. It is met with usually as a delicate pink or red dye, and forms the usual colouring-matter of rouge.

Logwood contains a colouring-matter named *hæmatoxylin*, $C_{16}H_{14}O_6 + 3H_2O$, crystallising from water in yellow prisms; this changes by the action of the air and ammonia into a red or purple substance named *hæmatein* ($C_{16}H_{13}O_6$)₃N. It may also be produced by adding nitric acid to an ethereal solution of hæmatoxylin. Hæmatein always exists in the free state in the wood, and an alcoholic tincture gives the reaction of hæmatein. The spectrum of the logwood colouring-matters is delineated in Nos. 21 and 22. Alkalies turn the tincture first red, and then violet. The best test, however, is the addition of an aluminous salt to an ammoniacal tincture of logwood, the result being the formation of an abundant bluish-violet precipitate. To test for alum, chips of logwood are boiled with water and the solution diluted until the spectrum shows the edge of the band close to the D line, as represented in fig. 16, curve 22; on now adding a drop of a solution of alum, 1 per cent., the absorption in the blue disappears, and a single band makes its appearance, having its centre about the line D, and extending from E to between C and D, or to about $\lambda 628$ —the curve then having the position shown in fig. 17: 1 mgrm. of either alum or Al_2Cl_6 may be in this way recognised. Iron interferes with the test, and should it be present, Vogel recommends its conversion into the ferrous condition, to add ammonium sulphocyanide, and to extract the sulphocyanide with ether, iron sulphocyanide being soluble in ether; in this way he has been able to detect alum in a mixture of 1 of alum and 40 of an iron salt (*op. cit.*). Coloured liquids, such as wine, are decolorised by a little *Eau de Javelle* and hydrochloric acid, boiled with alcohol, neutralised, and filtered.



FIG. 17.

Brazil-wood, sapan-wood, lima-wood, peach wood, and some others, yield a glucoside, which splits up into sugar and *brasiline*, $C_{22}H_{20}O_7$. Alkalies turn brasiline a crimson colour, and the crimson solution gives blackish-violet precipitates with aluminium and stannic salts. It is oxidised slowly in the air, or rapidly by the action of nitric acid into a red crystalline body, *brasilein* ($C_{22}H_{10}O_7$)₃N. The spectrum of brazil-wood extract in solution is delineated in Nos. 23 and 24.

There is also a red colouring-matter yielded by the santal-wood named '*santalín*,' and occurring in microscopic red crystals insoluble in water, but dissolving in alcohol, with a red colour, turning to violet on the addition of alkalis.

The Red Juices of Fruits and Berries.—Red currants when fresh show a band between D and F, the centre of the band being nearly at E. In

dilute solutions there are indications that this is really a double band ; when acidified, the colour can be shaken out with amyl alcohol.

The red colouring-matter of the strawberry is also similar, if not identical. It gives a double band in the green, and the colour is dissolved from the acid solution by amyl alcohol.

The raspberry has also a similar, if not identical, colouring-matter.

The spectrum of these vegetable juices appears to be analogous to the red in the chromatophore of the lobster (*Thudichum*).

The red berries of the common asparagus contain a yellow colour soluble in water, and a red colouring-matter soluble in alcohol. The latter colouring-matter gives two well-defined bands between F and G.

The red berries of *Crataegus* also give a two-banded spectrum, the one between E and F, the other midway between F and G.

All these colouring-matters, however, require further investigation, especially with regard to the ultra-violet and infra-red portions of the spectrum, both of which are invisible. The infra-red spectrum will specially repay study. A method of examination of this portion has been discovered of great accuracy. A fine wire, through which a feeble current of electrical current passes, is slowly, by means of a delicate micrometer arrangement, moved along the invisible spectrum, and where there are absorption bands or lines the electrical current is affected as measured and shown by a galvanometer. The ultra-violet part, if a quartz train of prisms and lenses be used, can be easily photographed or may be viewed by a Soret's ocular.

The red colouring-matters of the grape and various reds used to colour wines are dealt with specially in the article on *Wine*.

As with other colours, so with red tints generally ; if they are once identified, imitation mixtures can be made and estimated colorimetrically ; or, if the absorption factor is known, they may be estimated by a spectroscope with a double slit and a suitable shutter connected with the ocular.

§ 60. ORANGE AND YELLOW.—The most common oranges and yellows are the annatto colours : curcuma, picric acid, fustic, chrysophanic acid, gamboge, and aniline oranges and yellows.

The *Annatto Colours*.—The colouring-matters of the annatto are two substances, one a yellow, *orellin* ; another a cinnabar red substance, *bixin*. The latter is described in the article on 'Annatto.' The annatto colouring-matters are not soluble in water, but are easily dissolved by alcohol. The alcoholic solution is orange red, and non-fluorescent. On the addition of nitric acid it becomes turbid. On dilution with water, there is a strong fluorescence, and it becomes yellow-green. It then absorbs, like ferric chloride, the whole left side of the spectrum E, and half to D.

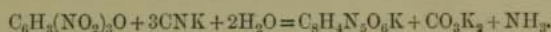
Turmeric is the root of *Curcuma longa*. The colouring-matter is *curcumin*, $C_6H_{10}O_3$, insoluble in cold water, and sparingly soluble in boiling water. It is very soluble in alcohol, and forms brilliant yellow crystals. Turmeric moistened with boric acid and dried assumes an orange colour, changed by alkalis into a blue ; this is due to the formation of a compound soluble in alcohol, forming a red solution, and crystallising in lustrous green crystals, to which the name of *rosocyanin* has been given. No. 32 shows the spectroscopic appearances of curcumin.

Picric Acid ($C_6H_3(NO_2)_3O$), also called *carbazotic acid* and *trinitrophenol*, is formed commercially by dissolving phenol in sulphuric acid, and then treating the solution with nitric acid. It crystallises from hot water

in yellow plates, having a very bitter taste. The salts are explosive. It is taken up from acid watery solutions by petroleum-ether, ether, or benzene, and hence can be readily obtained pure enough for examination.

Picric acid is not precipitated by acetate of lead. The chief chemical test is the production of isopurpurate of potash, which is the result of adding cyanide of potassium, and gently warming.

The reaction is represented as follows:—



Isopurpurate of potash is of a blood-red colour.

Fustic is the general name for yellow colours found in the wood of the *Morus tinctoria*.

The wood contains two distinct colouring-matters; the one, *moritannic acid*, $C_{13}H_{16}O_6 + H_2O$, soluble in hot water, and forming yellow crystals; the other, *morine*, $C_{12}H_8O_5$, is but sparingly soluble in water; crystallised from alcohol the substance forms yellow needles. Both give yellow precipitates with acetate of lead. The spectroscopic appearances of fustic are shown in No. 34 (a.) in concentrated, (b.) in dilute solution.

Chrysophanic Acid, $C_{14}H_5(CH_3)O_2(OH)_2$, melting point 178° , appears to be dioxymethylanthraquinone. It is contained in the rhubarb and wall lichen (*Parmelia parietina*). In commerce it is in the form of six-sided tabular crystals, of a pale to an orange-yellow colour. It is very readily extracted from rhubarb which has been previously macerated in water, pressed, and dried, by digesting the rhubarb in benzene. The crystals are soluble in ether, oil of turpentine, coal-naphtha, benzene, and other hydrocarbons.

The spectrum of chrysophanic acid is very similar to that of natural alizarine. Solutions of chrysophanic acid give, with alkalis, a rich purple colour. An ammoniacal solution of chrysophanic acid yields, with alum, a beautiful rose-coloured precipitate. An alcoholic solution of subacetate of lead gives, in the alcoholic solution, a red-white precipitate. Chrysophanic acid, on being oxidised with nitric acid, yields methylanthraquinone, $C_{15}H_{10}O_2$.

Gamboge.—This is a colouring-matter derived from the *Garcinia morella*, var. *pedicellata*. It is a gum resin, to which the formula of $C_{30}H_{35}O_6$ has been ascribed. It is very insoluble in water, but soluble in alcohol.

In order to detect it in sweetmeats, to which it has occasionally been added, the yellow colouring-matter is dissolved in alcohol, and precipitated by water, a reaction at once showing that it is of a resinous nature. The precipitate, if gamboge, will give a red colour with an alkali. It is without doubt a poisonous colour, but the question of quantity must be considered. Sixty grains have caused death, and five grains is a medicinal dose; and since some persons, especially children, are peculiarly susceptible to the action of medicines, even quantities so small as a third of a grain in a couple of ounces of sweets, should be considered as having possibly an injurious action.

Aniline Oranges and Yellows.—*Eosine*, $C_{20}H_6O_5Br_4Na_2$, the alkali salt of tetra-bromo-fluoresceine, is a reddish-brown powder soluble in water, the solution being of a reddish-orange with green fluorescence; it gives a yellow-red colour, or, if in strong solution, a red precipitate with hydrochloric acid. All the derivatives of fluoresceine have a similar

spectrum, of which eosine may be taken as the type. Moderately diluted, there is an intense dark band in the green; this, farther diluted, divides into two well-marked bands, the respective centres of which (in an alcoholic solution) are $\lambda 525$ and $\lambda 491$; the same bands, slightly altered in position, are seen in ethyl-eosine, rose bengal and phloxin. Eosine as a dye may often be recognised by the spectrum of the fabric strongly lit by the reflected rays of the sun, or by the lime or electric light. The fibre may also be digested in slightly acidified alcohol; the alcoholic extract is then taken up by amyl alcohol, a drop of ammonia added, and the spectroscopic appearances noted. The absorption factors of eosine from a solution of 0.01 mgrm. per c.c. have been ascertained by Krüss for the following wave lengths:—

Temp. 20° Wave Length.	Absorption Factor.
678.9 to 656.2	0.00025
627.5 ,, 609.5	0.00020
583.6 ,, 576.6	0.00012
552.6 ,, 540.6	0.000055
522.7 ,, 513.2	0.0000096
501.6 ,, 493.6	0.0000137
482.8 ,, 475.0	0.0000221
469.0 ,, 461.5	0.0000344

Croceine Orange (Ponceau 4), $C_{16}H_{11}N_2O_4SNa$, is the sodium salt of benzene-azo- β -naphthol- β -sulphonic acid. This dye is soluble in water with an orange colour. Hydrochloric acid gives a brownish-yellow precipitate. It gives a spectrum in which a single broad band occupies half of the green and some of the blue; the band in dilute solution extends from about $\lambda 526$ to $\lambda 438$.

Orange T (Mandarin G R), $C_{17}H_{13}N_2O_4SNa$, is the sodium salt of sulpho-*o*-toluene-azo- β -naphthol, it is soluble in water, and precipitated by hydrochloric acid in brown flocks. It gives a similar spectrum to the above.

Orange I (Tropæoline OOO), $C_{16}H_{11}N_2O_4SNa$, is the sodium salt of *p*-sulpho-benzene-azo- α -naphthol, while *Orange II* is the similar compound of β -naphthol; the former gives a brownish-yellow colour to hydrochloric acid, and precipitates in brown flocks. It gives a spectrum similar to croceine orange; 0.2 mgrm. in 1 c.c., viewed in a stratum of 4 mm., gives a band extending from $\lambda 530$ to $\lambda 399$.

Resorcin Yellow (Tropæoline O), $C_{12}H_9N_2O_3SNa$, is the sodium salt of *p*-sulpho-benzene-azo-resorcinol; it is soluble in water, and not changed by hydrochloric acid; dissolved in the proportion of 0.2 mgrm. in a c.c. of water, and viewed in a stratum of 4 mm., it gives a single band from $\lambda 475$ to $\lambda 361$ (Hartley).

Diphenylamine Orange (Orange IV—Tropæoline OO—Fast yellow), $C_{18}H_{14}N_2O_3SNa$, is the sodium salt of *p*-sulpho-benzene-azo-diphenylamine. The salt dissolves in water readily; it gives a violet precipitate with hydrochloric acid. It gives no bands, but cuts off the violet and blue rays.

Chrysoidine, $C_{12}H_{13}N_4Cl_7$, is the hydrochloride of diamido-azo-benzene. In commerce it is more often mixed with other colours; thus, mixed with magenta, it is known as 'cardinal'; with safranine, it is known as 'scarlet for cotton'; it is soluble in water, and strikes with hydrochloric acid an orange-brown colour. It gives a band in the visible spectrum, when

in the concentration of 0·8 mgrm. per c.c., and viewed through 2 mm., extending from $\lambda 494$ to $\lambda 439$; in a stratum of 3 mm. there is a band in the ultra violet from $\lambda 247$ to $\lambda 232$ (*Hartley*).

Methyl Orange (Orange III—Helianthine), $C_{14}H_{14}N_3SO_3Na$, is the sodium salt of *p*-sulpho-benzene-azo-dimethylaniline; it is a common laboratory reagent as an indicator for alkalis and acids. It is readily soluble in water to an orange solution, and strikes a magenta-red with hydrochloric acid and other acids. Methyl orange in a concentration equal to 0·3 mgrm. per c.c., and viewed through a stratum of 3 mm., gives a band extending from $\lambda 509$ to $\lambda 361$; it, therefore, shuts off all the violet and blue rays, as well as some of the green. It also, under the same conditions, gives a spectrum in the ultra violet, extending from $\lambda 270$ to $\lambda 256$. In a more dilute solution (0·06 mgrm. per c.c.) it gives a band from $\lambda 458$ to $\lambda 407$ (*Hartley*).

Metanil Yellow, $C_{15}H_{14}N_3O_3SNa$, is the sodium salt of *m*-sulpho-benzene-azo-diphenylamine; it is soluble in water, and gives a crimson colour with hydrochloric acid. It gives no band in the visible spectrum, but shuts off all the violet and blue rays.

Phosphine (Chrysaniline), $C_{16}H_{16}N_4O_3$, is the nitrate of diamido-phenyl-acridine and its homologues. This colour is soluble in water, and does not change colour with hydrochloric acid. It gives a shadowy band from $\lambda 486$ to $\lambda 508$ in the visible spectrum.

§ 61. The GREENS—are chlorophyll and the aniline greens.

Chlorophyll, the green colouring-matter of plants. It would appear that it is now possible to separate chlorophyll in a crystalline state. The merit of this discovery belongs to A. Gautier, who crystallised chlorophyll in 1877,¹ and two years afterwards Hoppe-Seyler,² without knowing of Gautier's paper, described the same substance under the name of chlorophyllone. Gautier's method is as follows:—The leaves are pounded in a mortar, and sodic carbonate added in quantity nearly sufficient to neutralise the acid juices, and the product strongly pressed. The marc is then exhausted at 55°, and again strongly pressed; to the exhausted substance alcohol is added, and the whole digested at 83°; chlorophyll, wax, pigments, fat, and other matters dissolve. The liquid is filtered and digested with coarse animal charcoal, which has been previously washed. At the end of four or five days it has lost its green colour, and becomes yellowish- or brownish-green. The charcoal retains the chlorophyll, and is now washed with alcohol at 65°; the latter removes a yellow crystallisable substance generally accompanying chlorophyll, and intimately allied to it in composition.

From the animal black thus freed from the yellow substance, chlorophyll may be extracted by anhydrous ether, or very light petroleum ether. A slow evaporation in the dark will yield it in crystals. Thus obtained, chlorophyll forms flat, often radiating crystals, which may be more than a centimetre in length, soft in consistence, and of an intense green colour when recent, but slowly changing to yellowish-green, or greenish-brown. If the crystallisation is too rapid, these long crystals are not obtained, but green masses composed of microscopical crystals, belonging to the rhombohedral system.

Diffused light changes chlorophyll to yellowish-green, and ultimately

¹ *Bulletin de la Société Chem.* T. xxviii., 1877, p. 147. *Comptes rendus*, lxxxix., p. 861.

² *Berichte der deutschen chem. Gesellschaft*, 1879.

decolourises it. The crystals dissolve in ether, chloroform, petroleum, carbon disulphide, and benzine. Digested with hydrochloric acid, chlorophyll splits up into two new substances, one giving a beautiful blue solution, the other remaining insoluble, but dissolving with a brown colour in hot ether and alcohol, from which it appears inclined to crystallise. Fremy, who was the first to notice this splitting-up of chlorophyll, called the first substance phyllocyanine, and the last phylloxanthine.

The ultimate analyses of chlorophyll by Hoppe-Seyler and Gautier agree fairly well, especially as Hoppe-Seyler's chlorophyll being derived from monocotyledonous and Gautier's from dicotyledonous plants, it is possible there may be some slight difference in their composition. Chlorophyll has not yet been obtained free from ash.

	Hoppe-Seyler.	Gautier.
Carbon,	73.4	73.77
Hydrogen,	9.70	9.80
Nitrogen,	5.62	4.15
Phosphorus,	1.37	
Phosphates, Ash,		1.75
Magnesia,34	
Oxygen,	9.57	10.33

Mr. Sorby has studied the colouring-matters of leaves, and has divided them into five groups:—1, Chlorophyll; 2, xanthophyll; 3, erythrophyll; 4, chrysophyll; and 5, phaiophyll.

The *Chlorophyll* group are all green in colour; the members are insoluble in water, but soluble in alcohol or carbon disulphide. The absorption-band is in the red, but the green is more or less completely transmitted, so that the prevailing tint is a more or less modified green. There appear to be several members of the group, one kind (which is probably the crystalline chlorophyll just described) occurs nearly pure in small aquatic plants, allied to the *Oscillatoria*; the green leaves of plants contain this, along with one which gives special absorption-bands. A third kind is the result of the action of acids on this, found more especially in autumnal leaves which have become brown. A fourth is in faded *Confervæ*, and turns blue when acted upon by hydrochloric acid (Fremy's phyllocyanine?).

Xanthophyll.—This is a group of orange colours. They are insoluble in water, but soluble in alcohol or carbon disulphide. The general tint of the spectrum is orange, there is absorption at the blue end, often more or less marked narrow bands. These bands are best seen in carbon disulphide solution. Examples of two species of xanthophyll are the inner and outer layers of the common carrot.

Erythrophyll.—A group of reds. They are soluble in water and weak alcohol, but not in carbon disulphide. There are many varieties. There is a strong absorption in the green parts of the spectrum.

Chrysophyll is a golden-yellow colour, soluble in water and weak alcohol, like the last, and insoluble in carbon disulphide.

The *Phaiophyll* group consists of various browns, due to the oxidation of chlorophyll. They give no definite absorption-bands.

The spectrum of chlorophyll is given in Nos. 35 and 36 (fig. 16). No. 35 is an alcoholic solution of fresh chlorophyll. There are two well-defined absorption-bands in the red, and two others, somewhat weaker, between E and D. The blue end of the spectrum, up to F, is dark. Old solutions (No. 36) alter the bands somewhat, and a fifth band appears between F and C.

Green, if not due to chlorophyll, nor to colouring-matters of copper or metallic origin, may be one of the aniline greens, such as aldehyde or iodine green.

Aldehyde Green, $C_{22}H_{27}N_3S_2O$, is made by the action of aldehyde on rosaniline and treatment of the product with sodium hyposulphite. It is only manufactured in small quantities, its place having been taken by other greens. It is insoluble in water, but soluble in alcohol. The spectrum of aldehyde green (1), methylrosaniline picrate (2), iodine green (3), and malachite green (4) are all similar, but by the aid of tests and comparative solutions may be distinguished from each other. The following diagram (fig. 18), after Vogel, sufficiently indicates the differences between the spectra.

Mr. Hartley has investigated the spectrum of iodine green in a concentration of 1.25 mgrm. per c.c., viewed in a stratum of 5 mm.; it gives the following bands:—In the visible part two bands, one from $\lambda 694$ to $\lambda 545$, the other from $\lambda 434$ to $\lambda 410$; in the ultra violet a single band from $\lambda 324$ to $\lambda 286$; but in a layer of 4 mm. two bands are seen in the ultra violet, one from $\lambda 317$ to $\lambda 286$, the other from $\lambda 256$ to $\lambda 235$. Probably most, if not all, of the aniline greens give bands in the ultra violet.

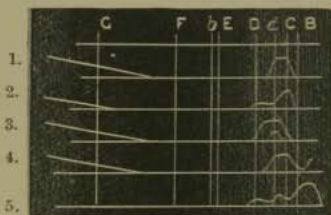
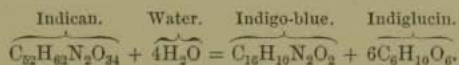


FIG. 18.

§ 62. INDIGOS AND VIOLETS.—The chief blues to be found, not of mineral origin, are indigo, litmus, and the aniline colours.

Indigo.—Indigo is the produce of a great number of plants, most of which belong to different species of *Indogifera*. It would seem that the indigo exists in these plants in the form of a syrupy glucoside, which has been named *indican*, and splits up into *indiglucin* and *indigo-blue*, according to the equation,



In the treatment of the crude substance by acids, other red and yellow colouring-matters are thrown down with the indigo. Pure indigotin has to be freed from these impurities, and may be crystallised in minute crystals. It is also capable of sublimation.

Crystalline indigotin is of a deep purple colour, and is insoluble in water, dilute acids, and alkalies. It is converted into orange crystals of *isatin*,

$C_6H_4 \left\langle \begin{array}{c} \text{CO} \\ \text{NH} \end{array} \right\rangle \text{CO}$, by treatment with oxidising agents. Indigotin is dissolved in concentrated sulphuric acid, and two compounds are formed—the

one, *indigo monosulphonic acid*, or *indigo-purpuric acid*, $C_{16}H_9N_2O_2(SO_3H)$, which is first formed; and the other indigo disulphonic acid, $C_{16}H_8N_2O_2(SO_3H)_2$, which is formed later. The first is easily separated from the second, for it is precipitated by the addition of water. A sodium salt of indigo-sulphuric acid is used much in laboratories under the name of indigo-carmine as a test for nitrates, as well as for their estimation, oxidising agents converting it into *isatin-sulphonic acid*, $C_{16}H_8N_2O_4(SO_3H)_2$, which has an orange colour; hence, when a liquid containing a nitrate is acidified strongly with sulphuric acid, and a solution of indigo-carmine run

into the hot solution, the colour changes at first to yellow until an excess of indigo has been added. The spectrum of indigo, in concentrated solution, is shown in No. 42. There are no bands, but absorption of the red, orange, and yellow end of the spectrum. Indigo can be determined quantitatively by the spectroscope; the absorption factor for $\lambda 412.7$ to $\lambda 595.9$ is 0.0000142.

Litmus is a blue colour obtained from lichens, and very familiar to chemists as an indicator of acids. As a colouring agent of either foods or stuffs, it is of little importance, not being a 'fast colour.' Its spectrum is shown at Nos. 25 and 26.

Aniline Blues.—The aniline blues and violets are numerous, and give spectra in which may be seen one or more bands in the red.

Methylene Blue, $C_{16}H_{18}N_3SCl$ or $(C_{16}H_{18}N_3SCl)_2 + ZnCl_2 + H_2O$, is the hydrochloride, or may be found as the zinc double chloride of tetra-methylthionine. It is easily soluble in water, but less soluble in alcohol. Reducing agents reduce it to leucomethylene blue, the air oxidising it back again to methylene blue. Concentrated solutions shut off all red and yellow up to $\lambda 547$; more dilute ones show a band from D to C ($\lambda 589$ to $\lambda 622$).

The absorption factor has been worked out for certain wave lengths.¹

Temp. 20° Wave Lengths.	Absorption Factor.
$\lambda 678.9$ to $\lambda 656.2$	0.0001996
$\lambda 627.5$ to $\lambda 609.5$	0.000265
$\lambda 583.6$ to $\lambda 576.6$	0.000725
$\lambda 552.6$ to $\lambda 540.6$	0.00125
$\lambda 522.7$ to $\lambda 513.2$	0.00308
$\lambda 501.6$ to $\lambda 493.6$	0.00326
$\lambda 482.8$ to $\lambda 475.0$	0.005
$\lambda 462.0$ to $\lambda 461.5$	0.0176

Mauve (Perkins' Violet), the colouring-matter of postage stamps, is soluble in water, and gives a band from $\lambda 614$ to $\lambda 558$.

Aniline Blue (soluble in spirit) is the hydrochloride, sulphate or acetate of triphenyl-rosaniline and triphenyl-para-rosaniline. These blues are insoluble in water; properly diluted they give a band extending from *d* to *D*.

Methyl Violet gives a band the centre of which occupies the line *D*.

Alkali Blue 6 B gives no band, but extinguishes the red and yellow; on acidifying with HCl a band is produced between *E* and *D*, and there is absorption of the violet and blue.

§ 63. BROWN COLOURS.—*Bismarck Brown* (Vesuvine), $C_{18}H_{20}N_8Cl_2$, is the hydrochloride of benzene-diazo-phenylenediamine. It is soluble in water, and does not change by acidifying with hydrochloric acid. Hartley has investigated its spectrum. In a stratum 5 mm. thick, with a concentration 0.09 mgrm. per c.c., it gives a band extending from $\lambda 467$ to $\lambda 407$.

Acid Brown, $C_{18}H_{15}N_6O_9SNa$, is the sodium salt of benzene-azo-phenylene-diamine-azo-benzene-*p*-sulphonic acid; it is soluble in water, and does not change by the addition of hydrochloric acid. A solution of 4 mgrms. per c.c., viewed in a stratum 8 mm. thick, gives an absorption band from $\lambda 451$ to $\lambda 399$.

Caramel.—All shades of brown, from a rather delicate fawn colour up to black or nearly so, can be given by caramel. Caramel is not a single

¹ G. and H. Krüss, *Zeit. f. anorgan. Chemie*, I.

simple substance, but a mixture of various colouring-matters derived from the dehydration of sugar. Caramel is soluble in water, and is precipitated by alcohol.

Caramel, as it usually occurs, is almost all soluble in water, and gives precipitates with alcohol, ammoniacal lead acetate, and baryta water. It gives a spectrum without definite absorption-bands, but extinguishes the blue side of the spectrum, like ferric chloride. Caramel may be suspected in a brown liquid not coloured by a mineral substance, which is not decolorised when sufficient tannin is added, for the colouring-matter of berries is precipitated by tannin. Among the members of caramel, there are three principal substances—viz., caramelan, $C_{12}H_{28}O_9$, carameline, and caramelin.

Caramelan makes up the chief bulk of ordinary caramel. It is brown, hard, and brittle, bitter to the taste, without odour, deliquescent, and very soluble in water, sparingly soluble in weak alcohol, insoluble in ether. It does not precipitate metallic salts in aqueous solutions, but reduces an alkaline solution of oxide of copper, and also the salts of gold and silver.

Carameline may be separated from caramelan by first extracting the latter by alcohol of 84 per cent., exhausting with cold water, and precipitating with absolute alcohol. It is a brittle solid, of great tinctorial power; it is easily soluble in water, sparingly in alcohol, insoluble in ether.

Caramelin is a mixture of three substances of different solubilities; it is black, shining, and infusible. Its solutions precipitate metallic salts, and reduce gold and silver solutions.

§ 64.—SCHEME FOR THE DETECTION OF THE ANILINE AND OTHER COLOURING MATTERS¹ BY CHEMICAL REAGENTS.

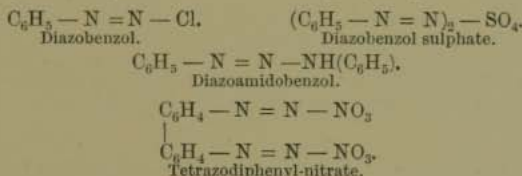
The aniline colours admit of a chemical classification as follows:—

I. *Nitro-Colours*.—The examples of which are picric acid, the dinitrocresols, bromonitro-phenol-*p*-sulphonic acid.

II. *Azoxy-Colours*, such as the sodium salt of azoxystilbene disulphonic acid, known in commerce as "sun yellow."

III. *Hydrazine Colours*, such as "tartrazines."

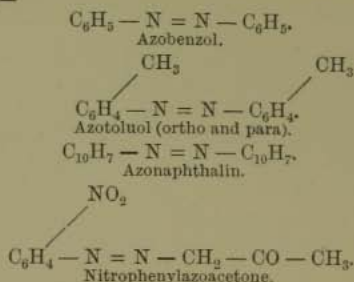
IV. *Azo-Colours*.—These are very numerous. The bivalent azo-group ($-N=N-$) is found in two classes of organic bodies—in the diazo and in the azo compounds. In the diazo compounds the one valency is satisfied by a benzol residue, and the other by a halogen, hydroxyl, an inorganic acid, or an amido-group; e.g.—



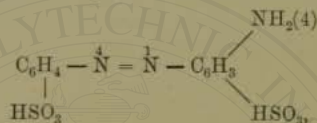
These diazo bodies possess no colouring properties, but they are the more important sources of the azo bodies.

¹ The above classification is borrowed from *The Systematic Survey of the Organic Colouring-Matters*, by Dr. G. Schultz and P. Julius, translated, with additions, by Arthur G. Green, F.I.C., F.C.S., London, 1894. On the interesting chromophore theory of Witt, see *Ber. der deutschen chem. Ges.*, ix., p. 522, and *Chemiker-Zeitung*, iv., 422. On the poisonous properties of the tar colouring-matters, see *Die Theerfarben, mit besonderer Rücksicht auf Schädlichkeit u. Gesetzgebung, hygienisch u. forensisch-chemisch untersucht*, by Dr. Th. Weyl. Berlin, 1889.

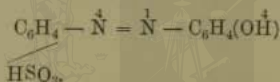
The azo bodies contain two benzol residues, each of which is bound to a valency of the nitrogen atoms, thus—



The azo bodies possess colour, but are not dyes until an acid or basic group is introduced into the molecule; azo-benzol, for example, is a yellow substance not capable of dyeing, but azo-benzol mono- or di-sulphonic acid possesses weak-dyeing properties. Similarly, if an hydroxyl or an amide group enters into the molecule, the dyeing properties are much enhanced; for example, amido-azo-benzol disulphonic acid—



in the form of a sodium salt, is the chief constituent of 'acid yellow'; tropæolin Y is the sodium salt of oxy-azo-benzol sulphonic acid—



About half the number of tar colours known belong to the azo-group.

Examples of azo colours are;—

YELLOWS.—Acid yellow, Sudans—croceine—orange—butter yellow.

REDS.—Various Ponceaux, Bordeaux, Roccelline.

BLACK.—Azonigrine—naphthylamine black D.

PURPLES AND BLUES.—Hessian purple, diamine violet, sulphone-azurine.

V. *Nitroso Colours*.—All the colours in commerce belonging to this group are green; examples are dinitroso-resorein, naphthol green.

VI. *Oxyketone Colours*.—Examples are alizarines—purpurine.

VII. *Diphenyl-methane Colours*.—Examples, auramine, acridine red, pyronines.

VIII. *Triphenyl-methane Colours*.—Examples, malachite green, victoria blue, magenta, acid magenta, methyl violet, eosine, erythrosine, rose Bengal, phloxines, rhodamines.

IX. *Indophenols*.

X. *Oxazines and Thiazines*.—Examples are Meldola's blue (chloride of dimethyl-phenylammonium- β -naphthoxazine), methylene blue B and B G H (hydrochloride of tetramethyl thionine), thionine blue (hydrochloride of trimethyl-ethyl-thionine).

XI. *Azines*.—(a) Eurhodines.—Example, neutral violet (hydrochloride of dimethyl-diamido-phenazine). (b) Safranines and indulines.—Examples, safranine, naphthalene red, mauve, induline.

XII. *Artificial Indigo*.

XIII. *Quinoline Colours*.—Examples, quinoline red and quinoline yellow, aldehyde green.

XIV. *Acridine Colours*.—Examples, acridine yellow, acridine orange, phosphine.

XV. *Thiobenzoyl Colours*.—Examples, thioflavine, primuline.

To these must be added a number of colours like canarine and murexide, which either do not fall within the above divisions, or their constitution is insufficiently known.

The group reagents used by Schultz and Julius are—(1) a solution containing 10 per cent. tannin and 10 per cent. sodic acetate; (2) zinc dust and hydrochloric acid, or zinc dust and ammonia; (3) 1 per cent. solution of chromic acid; (4) 1 per cent. solution of chromic acid and 5 per cent. sulphuric acid.¹

¹ *Systematic Survey of the Organic Colouring-Matters*, by Drs. G. Schultz and P. Julius. Translated by Arthur G. Green, F.I.C., F.C.S. London, 1894.

GROUP I.—COLOURS SOLUBLE IN WATER.

A. PRECIPITATED BY TANNIN SOLUTION: Basic colours. Reduce aqueous solution by zinc dust and hydrochloric acid, and put a drop or two of the decolorised solution on filter paper. If the colour does not quickly return on exposure to air, the spot is touched with a drop of 1 per cent. chromic acid solution.

(1.) The original colour quickly reappears on exposure to air; azine, oxazine, thiazine, and acridine colours.

RED.—TOLUYLENE RED, SAFRANINE, PYRONINE, ACRIDINE RED.

ORANGE AND YELLOW.—PHOSPHINE, BENZOFLUORINE, ACRIDINE YELLOW, ACRIDINE ORANGE.

GREEN.—AZINE GREEN.

BLUE.—METHYLENE BLUE, THIONINE BLUE, TOLUIDINE BLUE, MELDOLA'S BLUE, MUSCARINE, NEUTRAL BLUE, BASLE BLUE R and B B, NEW METHYLENE BLUE G G, NILE BLUE, CAPRI BLUE, FAST BLACK, INDAZINE M, METAPHENYLENE BLUE B, PARAPHENYLENE BLUE, INDAMINES.

VIOLET.—MAUVE, AMETHYST, NEUTRAL VIOLET, FAST NEUTRAL VIOLET, PRIME, PARAPHENYLENE VIOLET, INDAMINES.

(2.) The colour reappears very slowly, or not at all, on exposure to air, but returns on spotting with 1 per cent. chromic acid solution; triphenylmethane colours and basic phthaleins.

RED.—MAGENTA, ISORUBINE, RHODAMINE, ANISOLINE.

GREEN.—MALACHITE GREEN, BRILLIANT GREEN, METHYL GREEN, IODINE GREEN.

BLUE.—VICTORIA BLUE B, VICTORIA BLUE 4 R, NIGHT BLUE.

VIOLET.—METHYL VIOLET, CRYSTAL VIOLET, HOFMANN'S VIOLET, BENZYL VIOLET, ETHYL PURPLE, REGINA PURPLE.

(3.) Original colour does not return at all.

YELLOW AND BROWN.—AURAMINE (this colour, when a solution is reduced, and a drop placed on filter paper and warmed over a flame until dry, gives a beautiful violet); THIOFLAVINE T (this colour is reduced with difficulty) CHRYSOLINE, BISMARCK BROWN.

B. Not precipitated by Tannin Solution: Acid colours. The aqueous solution is reduced with zinc dust and ammonia, or zinc dust and hydrochloric acid, and a drop of the decolorised solution put on filter paper. If the colour does not quickly return on exposure to air, the spot is touched with a drop of chromic acid solution (1 per cent. $\text{CrO}_3 + 5$ per cent. H_2SO_4), warmed over a flame, and then held in the vapour of ammonia.

(a.) The solution is decolorised.

(a.) The colour quickly reappears on exposure to air; SULPHONATED AZINES, OXAZINES, THIAZINES, etc., SOLUBLE INDULINES, SOLUBLE NIGROSINES, RESORCIN BLUE, AZURINE, THIOCARMINE, BASLE BLUE R S and B B S, GALLAMINE BLUE, GALLOCYANINE, GALLANILIC INDIGO P S, INDIGOCARMINE, SAFROSINE, AZOCARMINE, MIKADO ORANGE.

(b.) The original colour does not reappear on exposure to air, or only very slowly, but returns with chromic acid and exposure to ammonia vapour. The aqueous solution of the dyestuff is acidified and shaken with ether.

(a.) Ether extracts more or less completely the phthaleins and aurine, such as URANINE, CHRYSOLINE, EOSINE, ERYTHRINE, PHLOXINE, ERYTHROSINE, ROSE BENGAL, CYCLAMINE, AURINE, CORALLINE. (Of these, eosine, erythrine, and phloxine contain bromine; erythrosine, rose bengal, and cyclamine contain iodine; and the bromine or iodine may be set free by the action of strong sulphuric acid.)

(b.) Ether does not extract the sulfonated triphenyl-methane colours—that is the following:—ACID MAGENTA, ACID VIOLETS, FORMYL VIOLETS,

ALKALI BLUES, SOLUBLE BLUES, ALPINE BLUE, PATENT BLUE, FAST GREEN-BLUIISH, ACID GREENS, GUINEA GREENS, CHROME VIOLET.

- (κ.) The original colour does not appear at all ; azo, nitro, nitroso, and hydrazine colours. Heated on platinum foil, nitro colours deflagrate with the production of coloured vapours ; such nitro colours are—PICRIC ACID, VICTORIA YELLOW, AURANTIA, MARTIUS YELLOW, NAPHTHOL YELLOW S, BRILLIANT YELLOW, AUROTINE.
- On the other hand, azo, nitroso, and hydrazine colours burn quietly or deflagrate slightly, giving off at the same time (like the nitro colours) coloured vapours. Small pieces of unmordanted cotton are soaked in the solution, and the resulting coloured cotton submitted to the action of warm soap. The substantive azo colours, such as ORIEL YELLOW, CLAYTON CLOTH RED, ALKALI BROWN, ATLAS RED, VIOLET BLACK, COTTON SCARLET, NAPHTHYLENE RED, HESSIAN PURPLE, CONGO-YELLOW, CONGO-RED, BENZO-PURPURINE, resist the action of warm soap, but the ordinary azo colours are stripped by them.
- (λ.) Colour not decolorised by zinc and ammonia, but changed to brownish-red. Original colour returns quickly on exposure to air—ALIZARINE S, ALIZARINE BLUE S, CÆRULINE S.
- (μ) Colour by the action of zinc and ammonia slowly and incompletely disappears—CLAYTON YELLOW, THIAZOL YELLOW, TURMERINE, MIMOSA.
- (ν.) Colour not altered by zinc and ammonia ; very slowly or not at all by zinc and hydrochloric acid—QUINOLINE YELLOW, PRIMULINE, THIOFLAVINE, OXYPHENINE.

GROUP II.—DYESTUFFS INSOLUBLE IN WATER.

The colouring-matter is treated with water, and a few drops of 5 per cent. caustic soda solution.

- (1.) The colour dissolves. The alkaline solution is heated with zinc dust and ammonia, and a drop is put on filter paper. The colour fades or is changed to light brown.
- (a.) Original colour reappears quickly on exposure to air—CÆRULINE, GALLEINE, GALLOCYANINE, GALLANILIC VIOLET B S, GALLANILIC BLUE P, GALLOFLAVINE, ALIZARINE, BLUE, ALIZARINE BLACK, ALIZARINE, CYANINE, ALIZARINE-CYANINE BLACK, RUFIGALLOL.
- (b.) Original colour does not reappear on exposure to air—ALIZARINE, ANTHRA-PURPURINE, FLAVOPURPURINE, ALIZARINE ORANGE, ALIZARINE BROWN, ALIZARINE BORDEAUX, ALIZARINE YELLOW G & R, CHRYSANINE, SUDAN BROWN, PATENT FUSTIN, MYRTLE or RUSSIAN GREEN, GAMBINE R and Y DIOXINE.
- (2.) The colour remains insoluble.

(A) Soluble in 70 per cent. alcohol.

- (a.) Solution not fluorescent. On adding caustic soda (33 per cent.) to the alcoholic solution—INDULINE OPAL, NIGROSINE OPAL, ROSANILINE BLUE OPAL, and DIPHENYLAMINE BLUE OPAL change to reddish-brown ; while INDOPHENOL, SUDAN II and III, and CARMINAPHTH are not altered.
- (β.) Solution fluorescent. On adding caustic soda as above, Magdala red fluorescence is destroyed ; while the fluorescence of the spirit eosine and cyanosine is unaltered.

(B) Insoluble in 70 per cent. alcohol.

INDIGO, ANILINE BLACK, PRIMULINE BASE.

§ 64a. Otto Witt and E. Weingärtner classify aniline colours as follows :—

I. Soluble in water.

- (a.) Basic colours.
(b.) Acid colours.

II. Insoluble in water.

To isolate the basic soluble colours, the aqueous solution is alkalisied by baryta, soda, or potash, and the colouring-matter extracted by shaking out with acetic ether. The acid soluble colours are tested for, as follows :—To the aqueous solution is added an excess of calcined magnesia, and then a solution of mercuric acetate (20 per cent. strength), the whole boiled and filtered; the filtrate is either coloured or colourless, should the filtrate be colourless, if an acid aniline colour be present, acidification with acetic acid will reproduce the colour.

The aniline dyes insoluble in water are dissolved either in soda solution or in alcohol. The chief basic colours in presence of baryta water dissolve in acetic ether; they are equally soluble in ammoniacal amylic alcohol, which they for the most part colour; but if not, the colour is reproduced by adding to the amylic solution acetic acid. They are also precipitable by tannin.

These remarks apply to the following :—

RED COLOURS.—FUCHSINE, TOLUYLENE RED, SAFRANINE.

YELLOW AND ORANGE COLOURS.—PHOSPHINE, FLAVANILINE, AURAMINE, CHRYSOIDINE, VESUVINE.

GREENS.—VICTORIA GREEN, MALACHITE GREEN, METHYL GREEN, METHYLENE GREEN.

BLUES.—METHYLENE BLUE, VICTORIA BLUE.

VIOLETS.—METHYL VIOLET, HOFMANN'S VIOLET, MAUVEINE.

The acid aniline colours are not precipitated by tannin; they are insoluble or but slightly soluble in acetic ether in the presence of baryta water, and in any case the addition of acetic acid does not increase the tint. They are also insoluble or but little soluble in amylic alcohol. The chief of these colours are :—

REDS.—Eosine, eosine scarlet, phloxine, Bengal red, Biebrich's scarlet, croceine, Congo red, coccinine, ponceau from xylydine, ponceau R, 2R, 3R, roccelline (R solid), Bordeaux B, acid fuchsine, coralline, aurine, benzo-purpurine (B) 4 B, delta-purpurine 5 B, Congo-cornith, Congo-cornith B, erythrosine, ponceau S, purple N, heliotrope, rosazurine G and B, induline, nigrosine.

YELLOW AND ORANGE.—Fluoresceine, benzil fluoresceine, picric acid, Martius yellow, naphthol S yellow, aurantia, chrysamine G and R (this colour is not very soluble in cold water), chrysophenine, tropæoline O, tropæoline O O, methyl orange (tropæoline D), orange I, orange II, orange III (ethyl orange), Poirier's yellow, metaniline yellow, luteoline, citronine, orange (D R P 3,229), chrysoine, mandarine, tartrazine, alizarine S.

VIOLETS.—Acid violet, nitro-violet, heliotrope, induline, and nigrosine.

BLUE.—Alkaline blue, soluble blue, benzo-azurine G and R, nitro-blue, alizarine blue S, indigo-carmine, soluble indulines.

GREENS.—Helvetia green.

§ 64b. Stein's method of procedure to detect colours generally is as follows :—

RED COLOURS.

A. HEAT WITH AMMONIUM SULPHIDE.—A greenish or bluish colour, which, by the action of baryta water, is changed into green—ALOE'S PURPLE. If the liquid becomes purple, ARCHIL is also present.

B. BOIL WITH A SOLUTION OF ALUMINIUM SULPHATE.

a. The liquid is coloured red, with a beetle-green reflection—MADDER. (Confirm by spectroscopy.)

b. The liquid becomes red, but there is no reflection. Add an equal volume of sodium sulphite.

(1.) It is bleached.

Presence of BRAZIL-WOOD, SANTAL, MAGENTA, CORALLINE, SAFRANIN.

C. BOIL WITH ALCOHOL OF 80 PER CENT.

- a. The liquid colours distinctly. If bluish-red, **MAGENTA**; if yellowish-red, **SANTALIN**.
- b. Liquid colours very little, or not at all—**BRAZIL-WOOD, CORALLINE, SAFFLOWER**.
- (1.) Heat with lime water. No colour—**SAFFLOWER**. Red colour—**BRAZIL-WOOD, CORALLINE**.
- (2.) Heat with dilute sulphuric acid. Orange colour—**BRAZIL-WOOD**. Yellow turning to grey on addition of copper chloride—**CORALLINE**. (Confirm by spectroscope.)

(2.) *It is not bleached.*

COCHINEAL LAC-DYE, LAC-DYE, KERMES, ARCHIL.

- (a.) Boil with alcohol; liquid becomes red—**ARCHIL**. If only faintly colours, or at least if the colour is not decided, it may be **COCHINEAL, LAC-DYE, KERMES**.
- (b.) Heat with baryta water; no change—**LAC-DYE**; the liquid becomes red—**COCHINEAL, KERMES**.
- (c.) Heat with lime water: a red colour—**KERMES**; a violet colour—**COCHINEAL**.

YELLOWS.

Heat with a dilute solution of neutral ferric chloride.

N.B.—This test must be applied when the colouring-matter is separated in a fairly pure state.

- A. Colour but little altered—**ANNATTO, TURMERIC, ANILINE YELLOW, PICRIC ACID, NAPHTHALINE YELLOW**. Test with a drop of concentrated sulphuric acid: a blue or green colour is produced—**ANNATTO**. If the spot becomes at once, or after a little time, more or less brown or red, then add alcohol, with a few drops of hydrochloric acid and some boric acid.
- (a.) Liquid becomes of an intense pink colour, and on diluting with water there is a reddish-yellow colour—**TURMERIC**.
- (b.) Pale pink colour, on dilution with water, crimson—**ANILINE YELLOW**.
- (c.) There is no change of colour on the addition of hydrochloric acid, etc. Heat with ammoniacal copper solution, *bluish-green*; confirm the presence of picric acid by the cyanide of potassium test, a blood-red colour—**PICRIC ACID**; the colour becomes an olive green—**NAPHTHALINE YELLOW**.
- B. Various shades of colour from green to almost black—**MADDER YELLOW, FUSTIC, FUSTET, QUERCITRON, FLAVIN, BERRIES, WELD**. Boil with aluminium sulphate, with the addition of an equal volume of water; liquid becomes yellow, with a red reflection—**MADDER YELLOW**, with tin. Yellow with a bluish-green reflection—**FUSTIC**. Liquid yellow without reflection. Heat with baryta water, a red colour—**FUSTET**. The colour is only darkened. Boil with glacial acetic acid. On cooling, if the liquid is yellow, or greenish-yellow—**ENGLISH FLAVIN**. If the solution is not at all, or only faintly, coloured, boil with basic lead acetate. Thus, with regard to fabrics, if a tissue is *dyed* with **WELD**, the tissue will not change colour; if with **QUERCITRON** or **BERRIES**, the tissue will change to orange-brown; but articles of food will be scarcely coloured with these substances.

GREEN COLOURS.

If the green colouring-matter is not soluble in water, it is probably chlorophyll unless indeed it is a mineral colouring substance. Chlorophyll is best recognised by the spectroscopic characters of its alcoholic solution (see p. 78). If not chlorophyll nor a mineral substance, then

- A. Boil with a moderately concentrated solution of potassic cyanide.
- (a.) Colour changes into brown or yellow—**ANILINE GREEN, GREEN**, containing **INDIGO-SULPHURIC ACID** (carmine green).
- (b.) Does not change, or changes into a brownish- or yellowish-green—**GREEN**, containing **INDIGO** with or without **CARMINE GREEN**.

B. In any of the foregoing cases add an equal volume of water, and then a solution of aluminium sulphate until an abundant precipitate is formed; filter and wash. (Excess of the precipitant must be avoided.)

(a.) Filtrate yellow or reddish—**ANILINE GREEN**.

(b.) Filtrate blue—(1) precipitate colourless—**CARMINE GREEN** with **PICRIC ACID**; (2) precipitate yellow—**CARMINE GREEN** with a vegetable yellow. Dissolve the yellow precipitate in water, add sulphuric acid, and filter: a green fluorescence—**FUSTIC**; no fluorescence—**WELD, TURMERIC**. Test for turmeric in the original substance by boiling with alcohol, and adding boric and hydrochloric acids.

(a.) Filtrate colourless, precipitate yellow—**INDIGO**.

(b.) Filtrate blue; if the precipitate is colourless, **PICRIC ACID** may be present, and should be tested for; if coloured, there is probably a vegetable colour present.

BLUE COLOURS.

Dissolve out the colouring-matter with alcohol of 80 per cent., or treat the substance itself with alcohol, allowing it to remain in the liquid, and add a few drops of hydrochloric acid: a red colour—**LOGWOOD**. Confirm by adding to an alcoholic solution ammonia and then alum; a blue or violet precipitate should result. If the liquid does not dissolve any of the colour, it is most probably Prussian Blue, but it may also be Indigo. If, on the other hand, although some of the colour is extracted, yet the substance still remains blue, it is probably **ANILINE BLUE** or **INDIGO-SULPHURIC ACID**. Add strong sulphuric acid, with **INDIGO-SULPHURIC ACID**, no change. The colour changes into a yellow or reddish-brown in presence of **ANILINE BLUE**. On heating with sodium carbonate again, **INDIGO** is not changed, but **PRUSSIAN BLUE** is changed into yellow or brown.

VIOLET AND PURPLE COLOURS.

Heat with ammonium sulphide.

A. The tissue is bleached; soluble **ANILINE VIOLET, MAGENTA**, with **INDIGO CARMINE**. These two may be distinguished by the action of boiling alcohol: **SOLUBLE VIOLET** remains violet, **MAGENTA** becomes red, the substance becomes brownish-red.

Presence of **MAUVE**, or **HOFMAN'S VIOLET**. These may be distinguished by the addition of hydrochloric acid, which colours **HOFMANN'S VIOLET** yellow, but **MAUVE** becomes purple.

B. Turns olive brown—**ALKANET**. (Confirm by spectrum.)

D. Hardly any change. Presence of **ARCHIL, ARCHIL** with **INDIGO, LOGWOOD, or MÄDDER**.

Boil with alcohol—

a. The solution is pink, and changes to violet on the addition of ammonia **ARCHIL**; if the **ARCHIL** is accompanied with **INDIGO**, hot chloroform will colour blue.

b. Solution in alcohol remains colourless. Heat with dilute hydrochloric acid: **LOGWOOD** is coloured red, and may be further identified by test already given; if **INDIGO** is associated with it, hot chloroform will colour blue. With hydrochloric acid, madder, if it is coloured at all, becomes yellow.

J. Formanek,¹ has recently published an elaborate study of colouring-matters; he divides the colouring-matters into green, blue, red and yellow, and subdivides these into groups, based on their solubility in water, alcohol, amyl alcohol, and their chemical reactions when treated with ammonia nitric acid, and alcoholic potash, and the absorption spectra in the different menstrua. The visible spectrum is alone studied, and the appearances are represented graphically by a fairly complete series of curves.

The accompanying Tables will also be found useful in the identification of organic colours.

¹ *Spektralanalytischer Nachweis künstlicher organischer Farbstoffe*. Berlin, 1900.

TABLE I.—EFFECT OF VOLATILE

NAME.	The Acid Watery Fluid is coloured	The Colouring Matter dissolved	
		Petroleum Ether.	Benzine.
Aniline red.	Reddish.	Nothing.	Only impurities.
Aniline violet.	But little coloured, for the greater part separates.	Nothing.	Only impurities.
Aniline new violet.	Violet from the solution of a portion.	Nothing.	Only traces.
Aniline blue (insoluble).	All separated; water colourless.	Nothing.	Traces which, on exposure to air, are coloured, and give a blue residue.
Aniline blue (soluble).	Blue.	Nothing.	As with the insoluble blue, but residue greenish-blue.
Aniline yellow.	Bright yellow.	Is coloured yellow; deposits yellow crystals spontaneously and on evaporation.	As with petroleum ether.
Picric acid.	Bright yellow.	Is not coloured, but leaves on evaporation a yellow residue.	As with petroleum ether.
Styphnic acid.	Bright yellow.	As with picric acid, but a smaller portion is dissolved.	As with petroleum ether.
Chrysammic acid.	Yellow neutral, solution red.	Nothing.	Is coloured yellow; the separated benzine is red when shaken with caustic potash.
Aniline orange.	Bright yellow, with greenish flocks.	Only impurities.	Is coloured yellow, and gives a brownish-yellow residue.
Havanna brown.	Dark brown.	Only impurities.	Is coloured blue-yellow; residue amorphous brown.
Vesuvium.	Brown.	Only impurities.	Is coloured yellow; residue brown amorphous.
Coralline.	Yellow, but little dissolved.	Only impurities.	Only impurities.

SOLVENTS ACTING ON ACID SOLUTIONS.

or suspended in Water acidified with Sulphuric Acid is extracted by

Ether.	Chloroform.	Amyl alcohol.
Traces which, when dissolved, are colourless, but, on evaporation, red.	As ether.	Is coloured red; extracts easily, gives a residue.
Is coloured lilac, and leaves behind traces of a violet residue.	But little coloured; gives but small residue.	Extracts both dissolved and suspended, is coloured violet, and gives a residue.
As benzine.	As benzine.	As with aniline violet.
Extracts copiously; is coloured strongly pale gold.	As ether.	As ether, but a larger quantity dissolved.
Only traces.	Only traces.	As with the insoluble.
As with petroleum ether, only in greater quantity.		
Is coloured yellow, and gives a yellow residue.	As with petroleum ether.	As with ether, only in greater quantity.
?	As with petroleum ether.	Is dissolved easier than picric acid, the solution yellow.
?	As with benzine.	As with benzine, only easier soluble.
As benzine, but easier soluble.	As benzine.	Dissolves easily; solution greenish - yellow, residue brownish.
Extracts very little.	Extracts very little.	Dissolves easily; solution deep red-brown, residue brown amorphous.
Takes less up than benzine.	Takes less up than benzine.	Dissolves easily; solution deep red-brown, residue amorphous.
Dissolves much; residue orange.	Solution yellow to deep brown.	As ether and chloroform.

TABLE II.—EFFECT OF VOLATILE SOLVENTS

NAME.	The Ammoniacal Solution is coloured	The Colouring	
		Petroleum Ether.	Benzine.
Aniline red.	Almost colourless.	Becomes fluorescent, but only dissolves a trace, probably impurities.	Is coloured yellow, strongly fluorescent; residue red.
Aniline violet.	Almost colourless.	As with aniline red.	As with aniline red, only the residue is violet.
Aniline new violet.	Almost colourless.	Nothing.	Nothing.
Aniline blue (insoluble).	Discoloured.	Is coloured red-brown; residue bluish.	As with petroleum ether.
Aniline blue (soluble).	Reddish.	Nothing.	Only a little extracted.
Aniline yellow.	Dark brown.	At first yellow, but again separating, and becoming colourless.	Extracts traces.
Aniline orange.	Brown.	Nothing.	Extracts traces.
Havanna brown.	Clear brown and discoloured.	Is fluorescent in the green; residue brownish.	As with petroleum ether.
Vesuvia.	Clear brown.	Is coloured yellow; residue brown.	Is coloured orange; residue brown.
Coralline.	Splendid purple colour.	Only impurities.	Only impurities.

ACTING ON AN ALKALINE SOLUTION.

Matter dissolved in Ammoniacal Water, and extracted by

Ether.	Chloroform.	Amyl alcohol.
Is coloured bluish ; the residue as with benzine.		Is coloured deep red ; the residue blue-red.
Is coloured bluish ; the residue violet.	Is coloured a violet-blue ; residue likewise violet.	Is coloured a deep violet-red ; residue violet.
Only traces.		Less than from acid ; the residue the same as from acid.
As with petroleum ether.		More is dissolved, but otherwise as petroleum ether.
Only a little extracted.		Is coloured yellow, and gives a blue residue.
Is coloured yellow, but little is extracted.	As with benzine.	Takes up less than out of acid ; is coloured yellow, residue yellow.
Is coloured yellow, and leaves a small yellow residue.	As with benzine.	Extracts less than from an acid solution.
Extracts less than with benzine.		Becomes deep brown, with a green fluorescence ; residue brown.
Is coloured yellow ; residue less, but as with benzine.		Is coloured deep brown ; residue brown.
Is coloured pale yellow ; residue red-brown, is coloured purple by NH_3 .	Extracts less than out of an acid fluid.	Is coloured raspberry-red ; residue as from ether.

TABLE III.—VARIOUS COLOUR REACTIONS.

NAME.	Concentrated sulphuric acid.	Nitric acid.	Solution of ammonia.	Dissolved in sulphuric acid fluid.			Tannic acid.	In hydrochloric acid solution.	
				Iodine.	Iodide of potassium and bismuth.	Iodide of mercury and potassium.		Platin chloride.	Gold chloride.
Aniline red.	Dissolves yellow.	Dissolves green, then brown, on evaporation again red.	Dissolves red-violet, is quickly discoloured.	Greenish coloration.	No precipitate, or but a slight one.	Violet-red precipitate.	No precipitate.	Greenish precipitate.	
Aniline violet.	Dissolves dark yellow brown.	Dissolves brown, then deep green, on evaporation red-brown.	Dissolves violet, later almost colourless.	Greenish coloration.	Blackish precipitate.	Blue precipitate.	Blue precipitate.	Black precipitate.	
Aniline new violet.	Dissolves blood-red.	Dissolves deep blue.	Dissolves violet.	Greenish coloration.	No precipitate.	No precipitate.	No turbidity.		
Aniline blue (insoluble).	Is coloured blood-red to brown.	It is dissolved blue.	Is dissolved without coloration.	Greenish coloration.	No precipitate.	No precipitate.			
Aniline blue (soluble).	Is coloured blood-red to brown.	Dissolved blue.	Is dissolved without coloration.	Brown coloration.	No precipitate.	No precipitate.			
Aniline yellow.	Dissolves yellow.	Dissolves yellowish.	Dissolves orange.	No precipitate.	No precipitate.	Slight turbidity.	Slight turbidity.	No turbidity.	
Picric acid.	Dissolves brownish red.	Dissolves yellow.	Dissolves yellow.	No precipitate.	No precipitate.	No precipitate.		No turbidity.	
Styptic acid.	Dissolves almost colourless.	Dissolves colourless.	Dissolves yellow.	No precipitate.	No precipitate.	No precipitate.	No turbidity.		
Chrysammic acid.	Dissolves with separation of a violet powder.	Dissolves greenish-yellow.	Dissolves red.	No precipitate.	No precipitate.	No precipitate.	No turbidity.		
Aniline orange.	Dissolves brownish.	Dissolves pale yellow.	Dissolves reddish.	No precipitate.	No precipitate.	Slight turbidity.	Slight turbidity.		
Havana brown.	Is not made darker.	Dissolves brown.	Dissolves with difficulty brownish.	No precipitate but a red colour.	Brown colour when shaken with ammoniacal solution.	Red-brown precipitate.	Brown precipitate.	Brown precipitate.	
Veauvin.	Dissolves brown.	Dissolves blood-red, later red-brown.	Dissolves easily — colour, brown.	No precipitate but a red colour.	Brown colour when shaken with ammoniacal solution.	Brown precipitate.	Brown precipitate.	Brown precipitate.	
Coralline.	Dissolves yellow.	Dissolves yellow.	Dissolves purple.	No precipitate.	No precipitate.	No turbidity.	Slight turbidity.	No turbidity.	

THE MINERAL MATTERS OR 'ASH' OF FOOD.

ANALYSIS OF THE ASH OF ORGANIC SUBSTANCES.

§ 65. As a general rule, testing the ash for abnormal metals and alkaline earths is necessary, and more especially if the ash present any unusual character, whether in weight, colour, or solubility. Leaving for the present the *special* tests, the number and nature of the constituents which require to be determined for the purpose of the food-analyst vary according to the particular substance under examination, *e.g.*—

In all substances, *the percentage.*

In such fluids as milk, *the alkaline phosphates and the chlorides.*

In seeds, such as wheat, cocoa, etc., *the total phosphoric acid.*

In beer-ash, *the amount of common salt.*

In bread-ash, *the presence or absence of alumina, magnesia, and proportion of silica to alumina.*

In tea-ash, *the alkalinity, the iron, the silica, and proportion of soluble to insoluble ash.*

In coffee-ash, likewise *the proportion of soluble to insoluble ash, but the presence or absence of silica becomes also of importance.*

From these illustrations (which might be multiplied) it follows that, for the purposes of the food-analyst, the general constitution of the ash will be sufficiently known when the following determinations have been made:—

- (1.) The total percentage of ash.
- (2.) The total percentage of ash soluble in water.
- (3.) The total percentage of ash soluble in acid
- (4.) The alkalinity of the ash.
- (5.) The percentage of chlorine.
- (6.) The percentage of phosphoric acid.

(1.) *The Total Percentage of Ash.*—Of the various methods of estimating an ash, the simplest and most practical appears to be—to place a sufficient quantity of the substance to be burnt in a capacious platinum or porcelain dish, and to consume at the lowest possible temperature in a muffle or by heating with a ring burner.¹ The quantity to be taken is regulated by the amount of ash in the substances. For example, flour, containing only 7 per cent. of ash, would give with 50 grms. 35 ash, which is about as small a quantity as it is possible to work with conveniently, whilst in the case of coffee, tea, and cocoa, from 5 to 20 grms. is for most purposes ample.

(2.) *The Soluble Ash.*—The ash is boiled up two or three times with water in the same platinum dish; filtered, and the filtrate evaporated to dryness, heated to dull redness, and weighed.

(3.) *The Ash Soluble in Acid.*—The portion of ash insoluble in water is boiled up with HCl, and filtered from the sand; the latter is washed, dried, and weighed.

¹ If the sulphuric acid in the ash is not to be determined, a wide glass tube (such as the chimney of a common paraffin lamp) adjusted over the dish, by its powerful up-draught greatly expedites the operation; but if the sulphuric acid is to be determined, the impure gas of commerce renders the results too high. It is, however, of course open to the analyst to make the gas pass through a proper absorption-apparatus, or to use as a fuel, alcohol.

(4.) *The Alkalinity of the Ash.*—The solution in water from (2) is coloured with cochineal, and titrated with d. n. acid: the result may be usually expressed as potash.

(5.) *The Percentage of Chlorine.*—The determination of chlorine in the ash usually gives results too low, especially if the substance burnt is one, like bread, of difficult combustion, or containing substances which decompose chlorides at a red heat. Notwithstanding this defect, in a series of ashes burnt under similar circumstances, the amount of chlorine found gives fair comparative results. Should there be any special necessity for an accurate determination of chlorine, no volatilisation will occur in the combustion of most articles of food, if they are simply well carbonised and not burnt to a complete ash, and if the charcoal be finely powdered and extracted with plenty of boiling water. The chlorine may be determined gravimetrically by nitrate of silver, or more conveniently by a standard solution of nitrate of silver, using as an indicator neutral chromate of potash. Should alkaline phosphates be present, they must be first removed by baryta water.¹

(6.) *The Phosphoric Acid.*—The usual method of determining phosphoric acid is to dissolve the ash in hydrochloric acid, evaporate to dryness, remove the silica, mix the acid filtrate with ammonia in excess, redissolve the precipitated earthy phosphates by acetic acid, filter off and estimate the insoluble phosphate of iron (and alumina, if present), precipitate the lime with oxalate of ammonia, and then in the fluid (free from lime and iron) precipitate the phosphoric acid, by the addition of ammonia and magnesia mixture.

§ 66. *General Method of Determining all the Constituents of an Ash.*—The best method of determining all the constituents of an ordinary ash is perhaps as follows:—A sufficient quantity of the ash (from 5 to 10 grms.) is placed in a flask, about 25 c.c. of water added, and saturated with CO₂; the liquid is now evaporated to dryness, heated with a small quantity of water to dissolve the alkaline salts—the solution is filtered through a small weighed filter, the filtrate evaporated to dryness, the saline residue treated with a small quantity of water, and the calcium sulphate which separates out filtered through a weighed filter, and estimated; the filtrate from this is put in a tared flask, made up to any convenient weight, and divided into five portions *by weight*, viz.:—

(1.) For CO₂.—This is most accurately determined by the use of the following little apparatus:—The solution is placed in a flask A, and sufficient acid is put into the short test-tube T, to more than neutralise the carbonate. A stout glass rod is passed through the doubly perforated caoutchouc cork, and supports the little tube in position. The carbonate solution is now boiled until steam hisses out of the tube G (which it is convenient to furnish with a Bunsen's valve²). G is then placed under the mouth of a graduated measuring tube filled with mercury, and it is at once seen whether all the air is expelled. The flame is withdrawn for a second, and the glass rod, which moves quite air-tight, is pulled a little up, so as to allow the acid tube to fall down and empty its contents into the alkaline

¹ Chlorine may also be estimated conveniently in the acid solution of an ash by Volhard's method, described in the article on "Water Analysis" (*post*).

² Bunsen's valve is made as follows: Take a piece of rather thick-walled india-rubber tubing, say, three inches in length; work it, by the aid of a little spirit, on to any wooden rod which is of sufficient size to stretch it well; then with a sharp chisel, by a single blow, cut a longitudinal slit; if well made, it allows air to go one way with the greatest ease, but effectually prevents a return.

fluid. The flame is again placed under the flask, and the CO_2 boiled out into the measuring apparatus, and measured in the ordinary way. Those who are not provided with gas apparatus will find it convenient to jacket their eudiometer with a tube open at both ends. The lower end is closed by the mercury in the bath; the upper is placed under a water-tap, and a syphon is adjusted so as to prevent overflow. In this way the gas is rapidly cooled, and the whole determination, from first to last, need not take more than a quarter of an hour.¹ Instead of boiling the solution in this way, those who possess the mercury pump described and figured at page 53 (fig. 6), will find it more convenient to make the flask vacuous, then upset the acid, and collect the gases expelled.



FIG. 19.

(2.) For the sulphuric acid, determined by chloride of barium.

(3.) For the phosphoric acid, determined as magnesian pyrophosphate.

(4.) For the chlorine, by precipitation as silver chloride.

(5.) For the alkalis, by boiling in a platinum dish with slight excess of baryta water, filtering, getting rid of the excess of baryta by ammonia and ammonium carbonate, evaporating the filtrate to dryness, converting the alkalis into chlorides, and determining their relative proportion from their total weight and their content in chlorine. This completes the analysis of the *soluble* portion of the ash.

The *insoluble* will contain lime, magnesia, ferric oxide, alumina if present, silica, phosphoric, sulphuric, and carbonic acids.

The main portion of the insoluble ash is dissolved in nitric acid, freed from silica in the usual way, evaporated again to dryness in a porcelain basin, dilute nitric acid added until the bases are completely dissolved, and strong fuming nitric acid added, until the solution begins to be turbid from the separation of calcic nitrate. The turbidity is now destroyed by a few drops of dilute nitric acid, the solution warmed, and tinfoil added in small portions at a time, in weight about equal to the amount of ash taken. When the tin is fully oxidised, the solution is evaporated *nearly* to dryness, water is added, and the solution filtered; the phosphoric acid is retained in the precipitate—the bases are all in the filtrate. The precipitate is dissolved in strong potash solution, acidified with sulphuric acid, and freed from tin by hydric sulphide, concentrated to a small bulk, filtered if any further sulphide of tin separates, and the phosphoric acid determined by magnesia mixture and ammonia.²

The filtrate from the tin phosphate must be freed from lead (if the

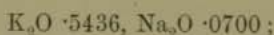
¹ Much ingenuity has been expended on the estimation of carbon dioxide, and the old method of estimating in light glass apparatus, by the loss of weight, is quite forsaken. The method given in the former edition of this work was to absorb the CO_2 in a clear solution of ammoniacal calcium chloride, collect the precipitated calcium carbonate, and titrate it with d. n. acid, using as an indicator cochineal. Another method which has been proposed, is to make a combustion of the substance with potassic bichromate, and absorb the CO_2 in the usual way in potash bulbs. A third, is to let the acid drop from a separating funnel on to the carbonates, and absorb the CO_2 as in the last, an aspirator and drying tubes being used. (*Annalen der Chemie*, clxxvi., 136-144.)

² Thorpe's "Quantitative Analysis." Lond., 1877.

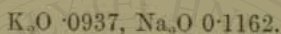
tin originally contained lead) by hydric sulphide, concentrated, the iron and alumina separated and determined by ammonia, the manganese separated as binoxide by bromine-water, the lime by oxalate of ammonia as oxalate, and the magnesia determined in the usual way as pyrophosphate.

A weighed portion of the insoluble ash must also be taken for the carbon dioxide, sulphuric acid, and sand. The carbon dioxide is determined in the manner already described.

The process just given is not quite accurate with regard to the estimation of the alkalis; for Bunge¹ has shown that since the alkalis form insoluble compounds with the alkaline earths, a watery extract of the ash gives low results. For example, Bunge incinerated 300 c.c. of cow's milk; from a watery extract of the ash he obtained



while from a subsequent nitric acid extract of the same ash,



If chlorides of the alkalis be heated with tribasic phosphate of lime, the soda is specially likely to combine with the lime in insoluble combination—in far less proportion the potash.

Bunge recommends the following method:—The watery extract is decomposed with baryta water until a film forms on the surface of the solution, the mixture is warmed and filtered hot. The excess of baryta is got rid of by CO_2 , subsequent warming, and filtration; the filtrate is evaporated in a platinum dish, the residue gently ignited, dissolved in a little water, filtered through a small filter, and evaporated with HCl in a small platinum dish. The chlorides are then ignited, weighed, and separated by platinum chloride.

The hydrochloric or nitric solution of the insoluble portion of the ash is evaporated to dryness in a platinum dish, the residue again dissolved in a little of the acid and water, treated like the former with baryta water, and filtered hot. Ammonia and carbonate of ammonia are now added, the liquid filtered, and the filtrate evaporated in a platinum dish, and ignited at the lowest possible temperature. The residue still containing a trace of alkaline earth, is extracted with water, evaporated with oxalic acid, ignited again, taken up with water, filtered, evaporated in a small platinum dish, ignited again, dissolved in a little water, and lastly, evaporated with HCl, and the alkaline salts separated by bichloride of platinum.

Since a determination of the ash only gives those mineral substances which are fixed in the fire, and destroys nitrates, and changes oxalates, citrates, and tartrates into carbonates, while other constituents, under the influence of heat, undergo a new arrangement, it becomes a question whether the ingenious method recommended by E. Laugier² in the analysis of sugar, would not be applicable in several cases.

M. Laugier takes two portions of sugar, one for the ash, the other for the organic acids, the latter being exactly double the quantity of the former. To the larger quantity of the sample, dilute sulphuric acid is added drop by drop to set free the organic acids; the acidified sugar is mixed with pumice stone and exhausted with ether; half of the ethereal

¹ Liebig's *Annalen der Chemie u. Pharmacie*, April 15, 1874.

² *Compt. rend.*, lxxxvii., 1088-1090.

solution is added to the ash obtained from the smaller portion, and evaporated down upon it and weighed. By this means M. Laugier thinks that he reconstructs the original salts in the sugar. This, however, cannot be entirely true. The other half of the ether solution is titrated with an alkali.

METHODS OF ESTIMATING NITROGEN AND NITROGENOUS SUBSTANCES IN FOODS.

§ 66a. A complete analysis of foods, especially with the view of ascertaining their dietetic value, necessitates the following determinations:—

1. Total nitrogen.
2. Nitrogen as albumen.
3. Nitrogen as acid-amines.
4. Nitrogen as amido acids.
5. Nitrogen as nitrates and nitrites.

Total Nitrogen.—The method of burning organic substances with copper oxide and copper, so as to obtain all the nitrogen as a gas, and also the other well-known method, by which the substance is mixed with soda lime and burned, so as to decompose the nitrogenous substances into ammonia, are both too well known to be farther described. At the present time, nitrogen is most frequently determined by Kjeldahl's process, as modified by Gunning, Arnold, and others. From 0.5 gm. to 2 grms. (according to richness in nitrogen), of the substance is introduced into a flask of hard Jena glass, and treated with 20 c.c. of strong sulphuric acid, at the same time adding a small globule of mercury; the flask is closed by a balloon stopper, and heated over a gas flame very gradually; after frothing has ceased, the heat is increased to brisk boiling, and after a short time 10 grms. of potassium bisulphate are added. On again boiling, the liquid gradually becomes clear and colourless. The contents of the flask are cooled, washed into a spacious distilling flask, provided with a doubly perforated cork, through the one hole of which passes a thistle funnel provided with stopcock, while the other carries a tube connected with a condensing apparatus; through the funnel is poured a sufficient quantity of a solution of sodium hydrate (containing a little sodium sulphide), to more than neutralise the acid. To prevent bumping, a little zinc foil should be added. The ammonia is now distilled over into a known quantity of d. n. sulphuric acid, the loss of acidity being finally determined in the usual way and expressed as nitrogen. To avoid the error from the soda solution being carried over, Dr. Bernard Dyer uses, as a communication between the distilling and the receiving flask, a block tin tube bent in the form of an arch; this rises to a height of 18 inches before turning over; the other end of the tube is united by a cork to a pear-shaped adaptor having a large expansion, which dips into an Erlenmeyer flask containing the acid. If nitrates are present, the sulphuric acid used for oxidation is charged with about 2 per cent. of salicylic acid. While the contents of the flask are still cold, some zinc dust (1–2 grms.), as well as the globule of mercury before mentioned, is added, and the zinc is allowed to dissolve before heating.¹

¹ Dyer, *Journ. Chem. Soc. Trans.*, 1895, 811.

Besides the above, there are other modifications, *e.g.*, Arnold and Wedemeyer¹ oxidise 0.5 gm. of the substance by means of 30–50 grms. sulphuric acid, 15–30 grms. potassium sulphate, 1 gm. mercuric oxide, and 1 gm. copper sulphate. With small quantities of ammonia the iodometric titration described in the chapter on 'Water,' *post*, has some advantages.

Estimation of the Nitrogen from Albumen.—The method of Stutzer is probably the best. From 1 to 2 grms. of the finely divided substance are boiled for a few minutes with 100 c.c. of water, or, if the substance contains much starch, it is simply digested on the water bath for from ten to fifteen minutes, then from 0.3 to 0.4 gm. of moist hydrate of copper is added, and, after cooling, the liquid is filtered through Swedish filter paper, the filter washed with water, and the residue with the filter submitted to Kjeldahl's process. The ammonia thus obtained is considered to be derived from albumen only, and the difference between the total nitrogen and that from Stutzer's process gives non-albuminoid nitrogen, which is sufficient for most purposes. Should the substance contain difficultly soluble alkaloids, it is first treated with absolute alcohol, acidified with acetic acid and heated to boiling, the alcohol decanted off as far as possible, and the residue washed with a little warm alcohol; then the substance is treated as before with water and copper hydrate.

Estimation of Non-Albuminoid Nitrogen.—10 grms. of the substance are boiled with about 300 c.c. of weak alcohol (40 per cent.), acidified with a few drops of acetic acid, the flask being connected with an upright condenser. The boiling should continue for about one and a half to two hours; the liquid is then cooled, filtered, and made up to a definite bulk, of which aliquot parts are taken for the following determinations:—

1. *Estimation of Ammonia.*—One-third of the fluid extract is evaporated down to 50 c.c., cooled, some recently calcined magnesia added, placed in a small flask attached to a tube loosely packed with glass wool, moistened with 10 c.c. of normal hydrochloric acid, and the air evacuated by a mercury or a good water pump, and after a vacuum has been obtained, a clip is put on the india-rubber tube connecting the apparatus with the pump, and the apparatus put on one side for three days. By the end of that time it is to be presumed that any ammonia in the apparatus will have evaporated and have been condensed in the glass wool moistened with hydrochloric acid; the glass wool tube is now disconnected, and the wool washed with a little distilled water, the acid water is evaporated to dryness on the water bath, and the ammonium chloride determined by the well-known method of precipitation with platinum chloride.

2. *Estimation of Amido-acid Amide Nitrogen.*—Another third of the extract is boiled with dilute hydrochloric acid (7 to 8 c.c. of strong acid added to every 100 c.c. of liquid extract) for from one and a half to two hours. At the end of that time the liquid is cooled and treated with hypobromide of soda in a nitrometer, and the resulting gas measured.

The difference between the nitrogen found by the first process and that by the second process equals the nitrogen as amido-acid amide.

3. *Estimation of Amido-acid Nitrogen.*—Another portion of the liquid extract is also boiled with hydrochloric acid, and then cooled and subjected in a suitable apparatus to the action of sodic nitrite. The resulting gas is submitted in a Hempel's burette to strong alkaline permanganate, which

¹ Arnold and Wedemeyer, *Zeit. anal. Chem.*, xxxi., 525. See also Henri E. Causse, *J. Pharm.*, 1895 [6], I., 543.

absorbs both NO and CO₂, and leaves, if the operation has been properly conducted, only nitrogen; the gas is measured in the usual way.

Since nitrite evolves nitrogen from both amido-acid amides and acid amides, the nitrogen of 2 must be subtracted from the nitrogen evolved in 3.

By the processes described, the following are estimated:—

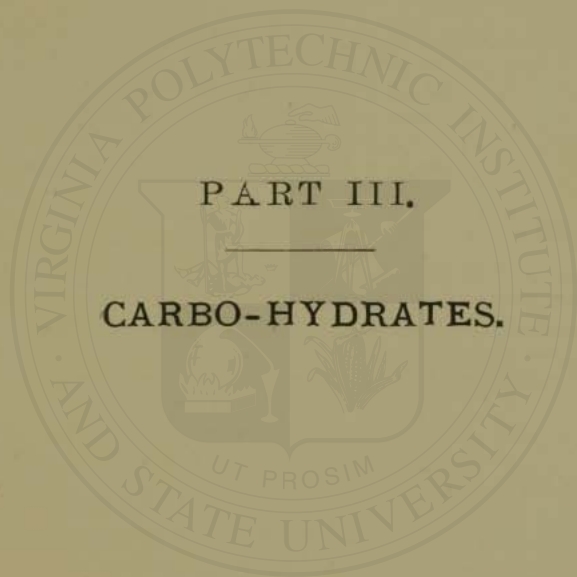
1. Ammonia.
2. Nitrogen of amido-acid amides from difference between Nos. 1 and 2.
3. The nitrogen from amido-acids from the difference between Nos. 2 and 3.



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PART III.

CARBO-HYDRATES.

PART III.—CARBO-HYDRATES.

STARCHY AND SACCHARINE SUBSTANCES.

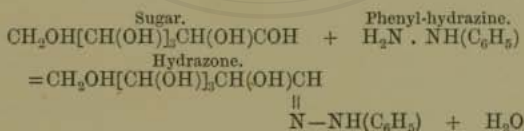
§ 67. The carbo-hydrates are divided into three great groups, viz., that of grape sugar, that of the saccharoses, and that of cellulose. They mostly contain 6, or a multiple of 6, atoms of carbon; and hydrogen and oxygen in the proportion of 2 : 1—that is, in the same proportion as the constituents of water. Recently, however, sugars have been synthetically formed containing 3, 4, 5, 7, 8, and 9 carbon atoms.

I. The Grape Sugar Group.

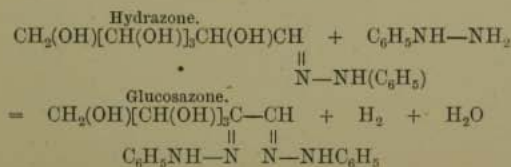
To this group belong sugars containing 3, 4, 5, 6, 7, 8, and 9 carbon atoms, known under the names of trioses, tetroses, pentoses, hexoses, octoses, and nonoses. These possess the following general properties:—

1. They are all easily oxidisable, and reduce Fehling's solution.
2. Most of the glucoses under the action of dilute mineral acids form levulinic acid and humus substances.
3. Warming with acetic acid and phenyl-hydrazine they all form more or less difficultly soluble and crystalline 'osazones.'

The formation of osazones takes place in two stages; first, one molecule of the sugar unites by means of its aldehyde or ketone group with one molecule of phenyl-hydrazine to form the easily soluble hydrazone, thus:—



with separation of one molecule of water. Then, in presence of an excess of phenyl-hydrazine, another molecule of phenyl-hydrazine unites with the hydrazone with separation of a molecule of water and a molecule of hydrogen, thus:—



The osazones are distinguished from each other by their differences of solubility and their melting-point; glucosazone melts at 145° C., lactosazone at 200° C., galactosazone at 193° C., maltosazone at 206° C., arabinosazone at 160° C., and sorbinazone at 164° C.

4. The glucoses all form additive compounds with hydric cyanide, the cyanide thus formed being converted, by the action of hydrolysing agents, into acids, which acids on reduction yield aldehydes; these are true sugars.

5. Several, not all, of the glucoses are capable of fermentation.

An example of a triose is glycerose, $C_3H_6O_3$; of a tetrose is erythrose, $C_4H_8O_4$; of a pentose is arabinose, $C_5H_{10}O_5$; the hexoses are glucose, mannose, fructose, and galactose.

II. The Cane Sugar Group.

To this group belong sugars of the formula $C_{12}H_{22}O_{11}$ —*e.g.*, cane sugar, milk sugar, maltose, and sugars of the formula $C_{18}H_{32}O_{16}$ (raffinose).

III. The Carbo-hydrates.

To this group belong the so-called polysaccharides, cellulose, starch, glycogen, gummy matters, and dextrin.

Cane Sugar, $C_{12}H_{22}O_{11}$, occurs in a very large number of plants, but is only manufactured from beet-root, the sugar-cane, sorghum, and the sugar-maple. Its specific rotatory power is + 66.5 [a]_D. It crystallises from its solutions in water or dilute alcohol in anhydrous crystals, the specific gravity of which is 1.606. It is soluble in one-third of its weight of cold water, and is very soluble in hot; in absolute alcohol it is insoluble, the solubility rising in proportion to the weakness of the alcohol. Thus, according to Scheibler, the numbers in Table IV. are the percentages of sugar dissolved, and the specific gravity of the solution at the common temperature of 14° C.

TABLE IV.—SOLUBILITY OF SUGAR IN ALCOHOL OF DIFFERENT STRENGTHS.

Per cent. alcohol.	100 c.c. of the solution contain	Specific gravity of the saturated solution.
0	85.8	1.3248
5	82.4
10	79.4	1.2991
15	76.5
20	73.4	1.236
25	69.8
30	66.0	1.2293
35	61.6
40	56.7	1.1823
45	51.6
50	45.7	1.1294
55	39.6
60	32.9	1.050
65	25.6
70	17.8	0.9721
75	11.2
80	6.4	0.8981
85	2.7
90	0.7	0.8369
95	0.2
97.4	0.08
100	000

When solutions of sugar are boiled with dilute mineral acids, the sugar is split up into two glucoses: the one rotating to the right, hence named dextrose, the other rotating the plane of polarised light, to the left—levulose.¹ Long boiling with water has, to a slight degree, the same effect, and it is also shown in the action of ferments, when exposed to light. An uncorked solution of sugar (or one imperfectly sealed) will in a few days, according to the temperature, show some degree of inversion. But a boiled solution of sugar, which, while actually boiling, has been hermetically sealed, will keep unchanged for years. Citric acid in a 2 per cent. solution readily inverts cane sugar; it does not act on milk sugar, a fact utilised in the detection and determination of mixtures of these sugars.²

Carbon dioxide, especially under pressure, inverts sugar.³ Pure cane sugar, if free from glucose, undergoes no change of colour when boiled with the alkalies; if, however, glucose be present, there is a very decided change.

Sugar forms a few well established compounds with bases, and many with salts. The most definite of the sugar compounds combined with bases are those which it forms with baryta and lime. If a solution of sugar be boiled down with sulphide of barium or baryta, a sandy precipitate forms, having the composition $C_{12}H_{22}O_{11}BaO$; and on decomposition of this with CO_2 , pure sugar is obtained. A commercial process based upon this reaction is in use in order to recover the sugar from molasses, and it may be employed in certain cases in the laboratory with advantage.

There is a monobasic lime sucrate ($C_{12}H_{22}O_{11}CaO$) corresponding to the barium compound, and a tribasic sucrate of lime ($C_{12}H_{22}O_{11}3CaO$). Crystalline compounds are also easily obtained with certain sodium salts; thus, there is a chloride of sodium compound $C_{12}H_{22}O_{11}NaCl.2H_2O$; and another having the formula $2C_{12}H_{22}O_{11}3NaCl.4H_2O$. An iodide of sodium compound may be obtained in large crystals having the following composition: $2C_{12}H_{22}O_{11}3NaI.H_2O$.

Sugar heated to 160° melts to a colourless liquid; on cooling, the melted mass is at first clear and transparent, but in a little time it becomes crystalline and opaque. At about 170° to 180° it loses water, and is said to be transformed into dextrose and levulose; as the heat is increased, water is continually being lost, and more or less brown products are formed (see 'Caramel,' p. 80). If sugar is fused with zinc chloride, a liquid is obtained which yields, on distillation, aldehyde, acetone, metacetone, formic acid, acetic acid, furfural, and apparently mesityl oxide; carbon dioxide, carbon oxide, and hydrocarbons are also formed; and there is also a sublimation of crystals of hexamethylbenzene, $C_6(CH_3)_6$. (Lippmann.)

Bromine, according to E. Reichardt, transforms one-third of cane sugar into gluconic acid, one-third into glucose, and the remainder into gum.

§ 68. *Adulterations of Sugar.*—Loaf sugar is, as a rule, chemically pure. It is probably, indeed, the purest food-substance in commerce, and a large quantity may be burnt up without obtaining a trace of nitrogen, and without leaving any residue. The only sugars that may be impure are the 'raw' sugars.

¹ To this mixture of dextrose and levulose, the term 'invert sugar' is applied, because the polarisation is the opposite of that of cane sugar; for, although glucose rotates to the right and levulose to the left, yet the latter is so much more active that the solution polarises to the left.

² Stokes and Bodmer, *Analyst*, x., 62-65.

³ V. Lippmann, *Ding. Poly. J.*, cxxxvij., 146-163.

The adulterations of sugar usually enumerated are: Glucose or starch sugar, sugar of milk, dextrin, chalk, plaster, sand, and various species of flour; few of these have been found of late years. A new adulteration is the colouring of sugars with yellow and yellow-brown aniline dyes; this is somewhat common. The detection is on the principles detailed in the chapter on Colouring-matters. One of these dyed sugars is at once detected by moistening the sugar with hydrochloric acid; a red colour is immediately produced. Phosphine is now often used (see p. 83).

To detect glucose (dextrose) in the presence of other sugars, B. Bottger¹ mixes the solution with an equal quantity of carbonate of soda solution [1 of the salt to 3 of water], and then adds a little basic bismuth nitrate, boils, noticing whether there is any blackening, which is taken as an indication of dextrose.

E. Brucke² has modified this method, so as to eliminate any blackening (which might occur from the sulphur in albuminous matters), by using potassium bismuth nitrate, which precipitates albumen. The reagent is made by dissolving basic bismuth nitrate in a hot solution of potassic iodide with the addition of hydrochloric acid. The albumen precipitated by the reagent is of course filtered off, and the filtrate is boiled.

The simplest and best method, however, of detecting starch sugar when mixed mechanically with cane sugar is undoubtedly that recommended by P. Casamajor.³ The suspected sugar is thoroughly dried, and is then treated with methyl-alcohol which has been saturated with starch sugar. 100 c.c. of methylic alcohol of 50° strength dissolves about 57 grms. of starch sugar, the 100 c.c. becoming in volume 133 c.c. Such a saturated solution dissolves cane sugar readily enough, but leaves starch sugar undissolved. After stirring the sugar in the methylic alcohol for about two minutes, the residue is allowed to settle, and the clear solution decanted. The residue is now washed with the same solution, and after stirring and allowing the residue to settle again, if starch sugar were present there will remain a certain quantity of chalky white specks, accompanied by a fine deposit of starch sugar. By collecting this on a filter, and washing rapidly with nearly absolute methyl, approximate quantitative results may be obtained.

The best method of detecting dextrin when mixed with sugar has been specially studied by Scheibler.⁴ He took a sugar which gave the following analysis:—

	Per cent.
Water,	3.35
Ash,	1.73
Organic matter,	2.32
Sugar,	92.60

and mixed this sugar with various proportions of dextrin—from 1 to 3 per cent.—and examined the behaviour of the sugar, both optically and chemically. The polarisation indicated the following amounts of cane sugar:—

0.0	0.5	1.0	2.0	3.0 per cent. dextrin
92.6	93.4	94.0	95.6	96.3
				„ sugar.

Thus, a sugar adulterated with 3 per cent., if examined optically, would

¹ *Journ. für prakt. Chem.*, lxx., 432.

² *Wien Akad. Ber.*, 1875, 52.

³ *Chemical News*, 1880.

⁴ "The Sugar Cane," 1871, p. 469.

indicate 96·3 per cent. of cane sugar instead of 92·6. On inverting the sugar, there were great discrepancies, quite enough to make even an inexperienced observer suspect something wrong. Thus with the same amounts of dextrin, the pure sugar showing, before inversion, 96·3 per cent. of sugar, and after inversion, 67·0 :—

	0·5	1·0	2·0	3·0 per cent. dextrine.
Direct,	93·4	94·0	95·6	96·3 ,, sugar.
After inversion,	80·4	78·0	73·1	67·0 ,, ,,

It was no use to experiment beyond 3 per cent., because it was then impossible to clarify the solution by lead acetate sufficiently for the purposes of optical analysis. Scheibler summarised his results as follows: Dextrin may be detected by its thus raising the degree of rotation, by the great difference of the results before and after inversion, by the blue colour it gives with iodine (although there are dextrins which, when added to sugar, may show this test imperfectly), by the impossibility of clarifying, by lead acetate, the liquid should any amount be present, and lastly, by partial separation of the dextrin by animal charcoal.

Insoluble mineral matters, such as sand, present in low-class sugars as an impurity, may be readily detected by simple solution of the sugar and filtration. Gummy matters may also be separated by precipitation by alcohol in the way to be described in the article on 'Tea'; mineral matters, generally, may be detected in the ash. Beet sugars, and to a less degree cane sugars, will contain a large amount of potassic and sodic carbonates, arising from the decomposition of the citrates, malates, oxalates, etc. Beet sugars may also contain nitrates. Cane sugar leaves an ash containing but little soda, with much more lime, magnesia, iron, and alumina. Thus, the following is the ash of raw cane and beet sugars, obtained in the following manner: All the mineral matters in the sulphuric acid residue of a large sugar factory were kept for a whole year, and analysed at the end of the year. Of course the carbonates, nitrates, and chlorides have all been decomposed, and the analysis is true only with regard to the bases.

	Cane Sugar Ash. Per cent.	Beet Sugar Ash. Per cent.
Potash,	28·79	34·19
Soda,	·87	11·12
Lime,	8·83	3·60
Magnesia,	2·73	·16
Oxides of iron and aluminium,	6·90	·28
Sulphuric acid [anhyd.],	43·65	48·85 ¹

Dividing each factor of the ash of the beet sugar by that of the cane, we get the following proportions for the bases :—

	Cane.	Beet.
Potash,	1	1·19
Soda,	1	12·78
Lime,	1	·41
Magnesia,	1	·06
Oxides of iron and aluminium,	1	·04

In other words, the potash is almost equal in the two ashes, but there is nearly thirteen times more soda in beet ash than in cane sugar ash; lime,

¹ J. Wallace, *Chem. News*, xxxvij., 75.

magnesia, and oxides of iron and aluminium are in very small quantities in beet sugar ash.

An analysis of the ash of a Demerara cane sugar growing near the sea-coast, by Dr. Wallace, is as follows :—

	Per cent.
Potash,	29.10
Soda,	1.94
Lime,	15.10
Magnesia,	3.76
Sulphuric anhydride,	23.75
Phosphoric acid,	5.59
Chlorine,	4.15
Carbon dioxide,	4.06
Iron peroxide,55
Alumina,65
Silica,	12.38
	<hr/>
	101.03
Deduct oxygen = chlorine,93
	<hr/>
	100.10

If sugar be ever adulterated by any of the starches, so clumsy a fraud is readily detected by a microscopical examination, and the use of iodine to the residue obtained by dissolving the sugar in cold water, and then filtering.

§ 69. *Full Analysis of Sugar.*—The full analysis of a raw sugar consists in :—

1. Determination of the water driven off at a heat not exceeding 55° to 60° C.
2. An optical estimation before and after inversion.
3. Titration with Fehling's solution before and after inversion.
4. Estimation of the organic acids by treating with sulphuric acid, and shaking up this acid extract in a separating funnel with ether, until it has dissolved out all the organic acids.
5. Titration of the organic acids with d. n. soda or potash.
6. Estimation of any insoluble matter, whether organic or inorganic.
7. Estimation of the ash and its constituents.

It may also be necessary to estimate the matters precipitated by basic lead acetate; it would be, however, quite sufficient for commercial, and, indeed, for most purposes, to merely estimate the percentage of cane sugar, fruit sugar (if present), water, and organic matter, or water and ash, as in the following analyses (Tables V. and VI.).

The difference between the totals and 100 would be returned as "unestimated matters and loss."

The methods of estimating the different kinds of sugar are fully considered in the next section, and it only remains to detail the best methods of taking the ash of a sugar.

There are two methods of taking the ash of sugar.

The one is simply to burn the ash in the ordinary way in a platinum dish heated to redness in a current of air. In the case of all substances like sugar or starch, this method is very tedious, and without doubt there is some loss by volatilisation. Landolt¹ determined the amount of this volatilisation by a series of careful experiments, and gives the following

¹ Landolt, H., *Journ. für praktische Chem.*, ciii. Also, "Sugar Cane," 1873.

TABLE V.—COMPOSITION OF RAW BEETROOT SUGARS OF GOOD QUALITY.

	Cane Sugar. Per cent.	Organic Matter. Per cent.	Water. Per cent.
1. Clear light mixed product, . . .	85.0	1.6	2.0
2. Light mixed product, . . .	93.0	2.0	2.3
3. Half white, 1st product, . . .	94.3	2.3	2.5
4. Light mixed product, . . .	93.5	2.4	3.3
5. Light mixed product, . . .	94.4	3.1	2.2
6. Half white, 1st product, . . .	96.1	2.3	3.4
7. Clear white, 1st product, . . .	92.0	3.7	2.8
8. Light mixed, 1st product, . . .	93.5	3.0	2.8
9. Clear light mixed, 1st product, . . .	92.0	2.3	2.6
10. Clear light, 1st product, . . .	93.1	3.3	3.0
11. Clear light, 1st product, . . .	94.0	3.0	2.9
12. Clear yellow mixed product, . . .	92.0	3.0	2.3
13. Yellow mixed product, . . .	92.0	2.4	3.5
14. Clear yellow mixed product, . . .	92.0	3.2	3.1
15. Clear yellow, 1st product, . . .	93.7	2.6	2.8
16. Yellowish mixed product, . . .	91.8	2.9	3.6
17. Yellow mixed product, . . .	93.0	3.5	2.7
18. Yellow mixed product, . . .	93.4	2.9	2.3
19. Yellow mixed product, . . .	93.0	2.3	2.9
20. Yellowish product, . . .	93.0	3.7	3.1
21. Yellow mixed product, . . .	93.0	3.3	2.7
22. Light mixed product, . . .	91.6	3.0	3.0
23. Yellow mixed product, . . .	90.7	4.6	4.3
24. Yellow mixed product, . . .	93.8	2.1	2.8
25. Yellow mixed product, . . .	92.5	2.4	3.5
26. Yellowish mixed product, . . .	90.0	4.3	3.7
27. Yellowish brown, 2nd product, . . .	91.3	5.1	2.5
28. Yellow, 1st product, . . .	92.4	3.5	2.6
29. Yellow mixed, 1st product, . . .	89.5	4.0	3.7
30. Light yellow, 1st product, . . .	92.0	2.8	3.4
31. Yellow mixed product, . . .	89.6	4.3	3.8
32. Brownish, 2nd product, . . .	89.0	6.3	3.0
33. Second product, . . .	87.0	6.2	4.1

TABLE VI.—SOME ANALYSES BY MR. HALSE OF CONCRETES.

	Cane Sugar. Per cent.	Uncrystallised Sugar. Per cent.	Water. Per cent.	Ash. Per cent.
1.	87.20	4.00	4.50	1.38
2.	89.60	1.90	5.50	.85
3.	90.20	1.95	4.03	.99
4.	91.70	3.30	2.15	.88
5.	87.00	5.00	4.72	1.24
6.	95.10	1.40	1.56	.86
7.	94.30	1.70	2.21	1.16
8.	92.50	1.92	2.70	1.18

Table (VII.), which may be used as a guide to the correction of the final weight of the ash.

TABLE VII.

Weight of Residue.	Loss by Heating after			
	Half an Hour.	One Hour.	One and a Half Hours.	Two Hours.
·01 grm.	·002 grm.	·004 grm.	·006 grm.	·008 grm.
·02 "	·002 "	·004 "	·007 "	·009 "
·03 "	·002 "	·005 "	·008 "	·010 "
·04 "	·003 "	·006 "	·009 "	·012 "
·05 "	·004 "	·007 "	·011 "	·015 "
·06 "	·004 "	·008 "	·013 "	·017 "
·07 "	·005 "	·010 "	·015 "	·019 "
·08 "	·005 "	·010 "	·016 "	·020 "

A method recommended and practised by Scheibler was to moisten the ash with sulphuric acid, whereby the combustion is much hastened, and the bases, being obtained as sulphates, approximate more nearly in weight to that of the organic salts naturally in the sugar, which in the other method are obtained as carbonates. It has also been proposed to precipitate the sugar with acetate of lead, and thus obtain the lead salts of the organic acids. The lead compounds are decomposed in the usual way, and the acids set free titrated by potash. The potash combination approximates somewhat more closely to the actual salts of the sugar.

But the best of those methods which attempt to reconstruct from the ash the original salts, is probably that of Laugier, already described at p. 96. Laugier extracts the organic acids by ether, and then adds them to the ash, and evaporates them down with it. As to raw beet sugar ash, the experiments of Landolt appear to show that simply multiplying the potassic carbonate found by 2, gives the amount of organic salt from which it was derived.¹ His experiment was as follows:—Two pounds of syrup were fully precipitated by lead acetate, then decomposed by SH_2 , and exactly neutralised by potash. The solution was next partly evaporated, passed through animal charcoal, and dried. It gave the reactions of chlorine, and of oxalic, malic, and tartaric acids, with a trace of sulphuric acid. Three separate portions were now carbonised, and the proportion for every one part of organic salt of carbonate of potash was—in experiment 1, 2·04; in experiment 2, 2·05; in experiment 3, 1·98: the mean of the three being 2·02.

§ 70. *Glucose, Dextrose, Dextro-Glucose, Grape Sugar*, $\text{C}_6\text{H}_{12}\text{O}_6\text{H}_2\text{O}$.—The specific rotatory power of glucose is 53.² It is soluble in 100 parts of

¹ Or if Scheibler's process be followed, the sulphates of the alkalis may be multiplied by 1·54.

² $[\alpha]_D = +52.8$ up to 15 per cent. concentration, the rotation usually accepted for the 3·86 divisor is +51·7.

cold water, and very soluble in boiling water; it is soluble in glycerine, in about two parts of rectified spirit, and two of amylic alcohol; but it is insoluble in ether and in chloroform. Dextrose is widely spread in the vegetable kingdom, but is never found unaccompanied by levulose. Dextrose is artificially obtained by heating carbo-hydrates, such as starch or cane sugar, with acids; in the former case, it is accompanied by dextrin, from which it is difficult to purify it. The rotation increases with the concentration—this may be calculated for anhydrous glucose by the following formula, p being the per cent. of sugar in solution $[\alpha]_D^{20} = 52.50 + 0.0188p + 0.000517p^2$.

The best way to obtain dextrose from cane sugar in a pure state is, according to Soxhlet, the following:—3 litres of 90 per cent. alcohol and 120 c.c. of concentrated hydrochloric acid are made to act at 45° C. for two hours on 1 kilo. of cane sugar. After ten days, crystals of dextrose form, when the liquid may be concentrated by distillation, and the crystals which have formed removed. In a few days, the whole of the dextrose will have been deposited as a white powder. The crystals are washed with 90 per cent. alcohol and with absolute alcohol, and crystallised out of the purest methyl-alcohol. Crystallised grape sugar is in the form of little masses of six-sided tables, which melt at 86° C., and lose at 100° C. their water of crystallisation.

§ 71. *Levulose* (or *Levoglucose*) is isomeric with dextrose, but distinguished from it by its action on a ray of polarised light which is turned to the left, instead of to the right: for a 10 per cent. solution specific rotation at 17° C. is $[\alpha]_D = 91.55$, but the rotation usually taken for the 3.86 divisor is -93.7 . It is obtained in company with dextrose when sugar is 'inverted' by the action of a dilute acid. To isolate levulose the acid must be got rid of; for example, if hydrochloric acid has been used, it is precipitated by silver solution; if sulphuric, by baryta water, etc. The solution of invert sugar must be about 10 per cent. strength. To every 100 c.c. 6 grms. of freshly burnt lime must be added, and the whole shaken. By artificially cooling the solution with ice, a crystalline magma is obtained, and by filtration the more soluble dextrose lime-compound can be obtained from the less soluble levulose lime-compound. The sugar thus obtained can be freed from lime by carbon dioxide.

Levulose is uncrystallisable, but it has not been found possible to separate it entirely from the crystalline glucose, by crystallising the latter out of it. It presents when pure simply the characters of a colourless syrup.

§ 72. *Estimation of Carbo-hydrates*.—Carbo-hydrates in solution are estimated by English chemists by a combination of physical and chemical processes, that is to say, by determinations of the reducing power on certain solutions of copper salts, by the action of invertase, of diastase, by a determination of the specific gravity and by observations by means of a polarimeter.

(1.) *Determination of the Cupric Reducing Power.*

This is capable of great accuracy if the conditions of strength of solutions, temperature, and time are always maintained as nearly uniform as practicable.

The standard solution of copper (Fehling) is made as follows:—34.5 grms. of copper sulphate are dissolved in 500 c.c. of water, and the solution made up to 500 c.c. 173 grms. of Rochelle salt (sodic-potassic tartrate) and 63

grms. of anhydrous soda are dissolved in water, and this solution also is made up to 500 c.c.; the two solutions are kept separate, but for use equal bulks are mixed together. It is important never to vary these quantities, and in particular to have the exact quantity of soda.

A slight reduction takes place on heating this solution, which, in important determinations, must be estimated; this reduction under the usual conditions is from 2-3 mgrms. CuO for every 50 c.c. of the Fehling solution used.

The reduction of a reducing sugar must be so arranged that the resulting copper oxide lies between the values of 150 and 350 mgrms.; that is to say, equal to from 60 to 120 mgrms. of dextrose.

This knowledge can usually be obtained from the specific gravity of the solution (see p. 113), or by a rough volumetric preliminary test.

The process of reduction is performed as follows:—50 c.c. of Fehling's solution are placed in a wide-mouthed flask of 250 c.c. capacity, and immersed in boiling water. When the temperature of the copper solution continues stationary, an accurately weighed or measured solution of the sugar is added, and the whole made up to 100 c.c. with boiling distilled water. The heating is continued exactly twelve minutes. The precipitated copper oxide is now filtered off while hot, and the resulting cuprous oxide is converted into CuO at a red heat, or the copper oxide is reduced by hydrogen, and the copper weighed as such, or the copper oxide is dissolved in nitric acid and the copper deposited electrolytically.

Most chemists filter the cuprous oxide through a small plug of asbestos pulp in a Soxhlet tube; others use a double filter paper. In the one case, however well washed the asbestos may be, it loses weight; in the other case, the copper in solution adheres in small quantity to the filter paper, and cannot be entirely washed out; if, however, blank experiments are made, this last source of error can be allowed for, and on the whole an ashless filter paper is the more convenient. The filter is well washed with hot water, and then dried with its contents in a porcelain or platinum dish; when dry, the filter is slowly charred, then brought for some time to a dull red heat, and finally heated to bright redness in a muffle or over a good Bunsen flame. If, however, the copper is to be determined electrolytically, the filter may be destroyed rapidly and a solution made of the copper.

The use of any kind of filter may, however, be avoided by transferring the liquid to a special tube, and whirling in a centrifugal machine. The special tube is in two parts; the one an ordinary glass tube, the other a cone; this latter may be of glass, of porcelain, or of platinum; the two are connected by a sound caoutchouc joint. On whirling, the precipitate collects in the cone. The supernatant fluid is syphoned off, hot water added, and the operation repeated until the washing water gives no copper reaction.

In order to estimate copper oxide by electrolysis, so that the copper is deposited as a coherent film on a platinum electrode or dish, the following conditions are necessary:—

The copper oxide is dissolved by a known quantity of nitric acid, and the liquid diluted, so that the strength of nitric acid in solution does not exceed 3 per cent. The solution is then electrolysed at from 20° to 30° C.; the current density must not be more than from 0.5 to 1.0 ampère, and the tension from 2.2-2.5 volts. The time taken for the deposition is from four to five hours.

§ 73. *The K and R Values.*—O'Sullivan took dextrose as the type of reducing bodies, and used it as the standard to which all other reducing carbo-hydrates or mixtures of reducing with non-reducing ones were compared. He designated by the letter K the cupric oxide reduced by 100 parts of a substance calculated as dextrose; hence K 50 means that a substance has half the reducing power of dextrose.

Just as K is the specific reducing power of a substance referred to dextrose as standard 100, so also the symbol R is used to denote the specific cupric reducing power of a substance referred to maltose as a standard (100).

Since, moreover, the strength of solutions is mostly determined by using as a divisor the specific gravity minus 1000, and the ordinary divisor for a 10 per cent. of cane sugar is 3.86, this divisor is used for most solutions of unknown value; when this is done, then the factor is suffixed to K—thus $K_{3.86}$. If, on the other hand, the true divisor is used, as, for example, for particular solution of sugar or starch conversions as obtained from the curves in fig. 19A, on page 114, then this is the absolute value, and is designated as such or simply as K.

To convert absolute values into 3.86 values is easy. For example, 1.371 grm. of CuO is reduced by 1 grm. of absolute maltose, that is maltose divided by the gravity as determined by the true divisor 3.92. Hence for 1 grm. of 3.86 maltose we have $1.371 \times \frac{3.86}{3.92} = 1.350$ CuO.

TABLE VIII.—THE CUPRIC REDUCING POWERS OF DEXTROSE, LEVULOSE, AND INVERT SUGAR. Brown, Morris and Millar, *Journ. Chem. Soc. (Trans.)*, lxxi., 281.

Mgms. of Sugar.	Dextrose.			Levulose.			Invert Sugar.		
	Cu grams.	CuO grams.	CuO corresponding to 1 gram.	Cu grams.	CuO grams.	CuO corresponding to 1 gram.	Cu grams.	CuO grams.	CuO corresponding to 1 gram.
50	0.1030	0.1239	2.578	0.0923	0.1155	2.310	0.0675	0.1231	2.442
55	0.1134	0.1422	2.585	0.1027	0.1257	2.341	0.1076	0.1349	2.453
60	0.1228	0.1552	2.587	0.1122	0.1407	2.345	0.1176	0.1474	2.457
65	0.1342	0.1682	2.589	0.1216	0.1524	2.346	0.1275	0.1598	2.459
70	0.1443	0.1809	2.585	0.1312	0.1645	2.350	0.1373	0.1721	2.459
75	0.1543	0.1935	2.580	0.1405	0.1761	2.349	0.1468	0.1840	2.454
80	0.1644	0.2061	2.577	0.1500	0.1881	2.351	0.1566	0.1963	2.454
85	0.1740	0.2187	2.572	0.1593	0.1993	2.345	0.1662	0.2084	2.451
90	0.1834	0.2299	2.555	0.1686	0.2114	2.340	0.1755	0.2200	2.445
95	0.1930	0.2420	2.547	0.1774	0.2224	2.341	0.1848	0.2317	2.439
100	0.2027	0.2538	2.538	0.1862	0.2331	2.331	0.1941	0.2430	2.430
105	0.2123	0.2662	2.535	0.1952	0.2447	2.331	0.2034	0.2550	2.429
110	0.2218	0.2781	2.528	0.2040	0.2558	2.335	0.2128	0.2668	2.425
115	0.2313	0.2900	2.522	0.2129	0.2669	2.331	0.2220	0.2783	2.420
120	0.2404	0.3014	2.512	0.2215	0.2777	2.314	0.2311	0.2898	2.415
125	0.2496	0.3130	2.504	0.2303	0.2887	2.310	0.2400	0.3009	2.407
130	0.2585	0.3241	2.493	0.2390	0.2997	2.305	0.2489	0.3121	2.400
135	0.2675	0.3354	2.484	0.2477	0.3106	2.300	0.2578	0.3232	2.394
140	0.2762	0.3463	2.473	0.2559	0.3209	2.292	0.2663	0.3339	2.385
145	0.2850	0.3573	2.464	0.2641	0.3311	2.284	0.2750	0.3448	2.378
150	0.2934	0.3673	2.448	0.2723	0.3409	2.273	0.2832	0.3546	2.364
155	0.3020	0.3787	2.443	0.2805	0.3517	2.269	0.2915	0.3655	2.358
160	0.3103	0.3891	2.432	0.2889	0.3622	2.264	0.3002	0.3764	2.352
165	0.3187	0.3996	2.422	0.2972	0.3726	2.258	0.3086	0.3869	2.345
170	0.3268	0.4098	2.410	0.3053	0.3828	2.252	0.3167	0.3971	2.336
175	0.3350	0.4200	2.400	0.3134	0.3930	2.245	0.3251	0.4076	2.329
180	0.3431	0.4302	2.390	0.3216	0.4032	2.240	0.3331	0.4177	2.320
185	0.3508	0.4399	2.377	0.3297	0.4134	2.234	0.3410	0.4276	2.311
190	0.3590	0.4501	2.369	0.3377	0.4234	2.228	0.3490	0.4376	2.303
195	0.3668	0.4599	2.358	0.3457	0.4335	2.223	0.3570	0.4476	2.295
200	0.3745	0.4689	2.344	0.3539	0.4431	2.216	0.3656	0.4570	2.285
205	0.3822	0.4792	2.338	0.3616	0.4534	2.211	0.3726	0.4672	2.279

The *Cyanide Copper Process*¹ gives a fairly accurate estimation of reducing sugars, and is performed as follows:—To 20 c.c. of Fehling (or, as it is better to keep the solutions separate, to 10 c.c. of copper sulphate solution of Fehling's strength, to which 10 c.c. of the alkaline tartrate are added), add 40 c.c. of water in a porcelain dish, and heat to boiling; while boiling, drop in from a burette a 5 per cent. solution of potassic cyanide until there is only the faintest blue colour observable—now add a second 20 c.c. of Fehling, and heat to boiling, and, while boiling, drop in the solution of sugar from a burette until complete decolorisation. The liquid should be standardised by solutions of 0.5 per cent., 1.0 per cent., and 2 per cent. of the various reducing sugars.

Mr. W. A. Rogers has made a number of determinations of lactose in milk by the copper cyanide process in our laboratory, and has observed that, when the end of the reaction is nearly reached, it is necessary to add the sugar solution very carefully, since the addition of any excess of the

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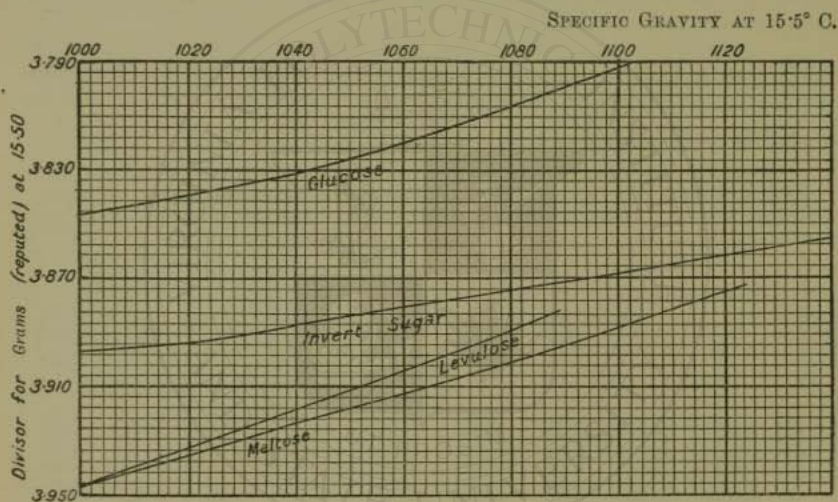


FIG. 19A.

latter, more than necessary to just decolorise the solution, produces a green colour which may rather easily be mistaken for the faint blue which precedes the end of the reaction. In making a number of determinations, it is also advisable to allow the same length of time (say a minute) to elapse between the addition of the portions of the sugar solution; otherwise strictly comparative results may not be obtained.

(2.) *Dr. Pavy's Process.*

An excellent method of determining sugar has been invented by Dr. Pavy.² The principle of the process depends on the decolorisation of an ammoniacal copper solution by glucose in the absence of air.

The copper solution is made by dissolving 20.4 grms. of potassic sodic tartrate and 20.4 grms. of caustic potash in 200 c.c. of water; in another 200 c.c., 4.158 grms. of cupric sulphate are dissolved by the aid of heat,

¹ Gerrard, *Pharm. Journal*, 3d series, xxv., 912.

² *Lancet*, 1884.

and the two solutions are mixed together; when cold, 300 c.c. of strong ammonia (sp. gr. .880) are added, and the whole made up to 1 litre with water. 10 c.c. of this liquid diluted to 20 c.c. with water equals 5 mgrms. of glucose.

To make the estimation, 10 c.c. of the ammonia copper solution are placed in a flask with a rather wide mouth, an equal bulk of water is added, and a good caoutchouc stopper having two perforations is fitted to the neck of the flask; through one of the holes is adapted air-tight the conical end of a burette, having either a glass stopcock or a clip; while the other hole carries a bent tube for the exit of the vapour. The sugar solution is placed in the burette. Heat is now applied to the flask, and when the liquid is boiling violently the sugar solution is run in slowly, after each addition boiling up; the blue colour fades gradually, and the end reaction is the complete absence of blue colour.

TABLE IX.—SHOWING THE AMOUNT OF SUGAR EXPRESSED IN PARTS BY WEIGHT PER 1000 BY VOLUME CORRESPONDING WITH C.C. IN 10THS REQUIRED TO DECOLORISE 10 C.C. OF THE AMMONIATED COPPER TEST.

C.c. to Decolorise.	Parts per 1000 of Sugar.	C.c. to Decolorise.	Parts per 1000 of Sugar.	C.c. to Decolorise.	Parts per 1000 of Sugar.
6.0	.833	7.4	.673	8.8	.568
6.1	.819	7.5	.661	8.9	.561
6.2	.806	7.6	.653	9.0	.555
6.3	.793	7.7	.646	9.1	.549
6.4	.781	7.8	.638	9.2	.543
6.5	.769	7.9	.631	9.3	.537
6.6	.757	8.0	.625	9.4	.531
6.7	.746	8.1	.617	9.5	.526
6.8	.735	8.2	.609	9.6	.520
6.9	.724	8.3	.602	9.7	.515
7.0	.714	8.4	.595	9.8	.510
7.1	.703	8.5	.588	9.9	.505
7.2	.691	8.6	.581	10.0	.500
7.3	.681	8.7	.574	10.1	.495

The authors use a modification of the process invented by Mr. Stillingfleet Johnson. The caoutchouc stopper is perforated by three holes instead of two; the third is for a tube which dips beneath the surface of the copper solution, and is closed or opened at pleasure by means of a short bit of india-rubber tubing adapted to the air-end of the tube and furnished with a clip; the second tube, for the outrushing ammonia vapour and steam, is also furnished with an india-rubber tube, the end of which dips under the surface of a considerable bulk of acidulated water, and is furnished with a Bunsen valve to prevent any back-rush. The use of the latter tube is, of course, to condense the ammonia-vapour, so that the operator is not inconvenienced. The use of the extra tube is to more accurately hit the end reaction—to do this, directly the decolorisation of the copper solution is complete, the flame is removed and the clip opened; as the flask cools, air passes in a stream of bubbles through the liquid; if the point has been exactly reached, the blue colour reappears after a very few seconds, but if, on the contrary, too much sugar solution should be run in, a longer time elapses.¹ By standardising the copper solution by

¹ It has been proposed to exclude the air by a layer of paraffin of high boiling point.

pure glucose, and working so that after complete decolorisation a certain number of seconds elapse, before the blue colour reappears, and taking with unknown solutions the same number of seconds, a high degree of accuracy is attainable. It is, of course, obvious that the sugar solution to be tested must be very dilute—viz., from .4 to .8 per 1000. A preliminary experiment must first be made, and then the solution so diluted that from 6 to 10 c.c. are required to decolorise 10 of ammoniacal copper.

The foregoing table, taken from Dr. Pavy's original paper, may be useful.

(3.) *Physical Processes for the Determination of Sugar.*

The saccharimeters in use are numerous; for the food analyst, the most useful are the larger instruments, which admit of the use of tubes up to 500 mm., for in this way, and in this way only, can solutions containing 0.5 per mil. of sugar be physically estimated. *Mitscherlich's polariscope* (see fig. 20) consists of a stationary Nicol's prism in *a*, a plano-convex lens in *b*, and a rotating Nicol's prism *c*. The first prism polarises the light, and the use of the second is to indicate the plane of the polarised ray coming from the first. The second prism is therefore set in a graduated circle, *d d*, and is provided with an index, *f*, and there is a handle, *e*, which turns both prism and index. If the index be either at 0° or 180°, and an observer look through the tubes towards the source of light, the flame is seen divided by a vertical line into two equal parts; if now the tube, supplied with the instrument, be filled with the liquid to be examined, and interposed between the lens and the

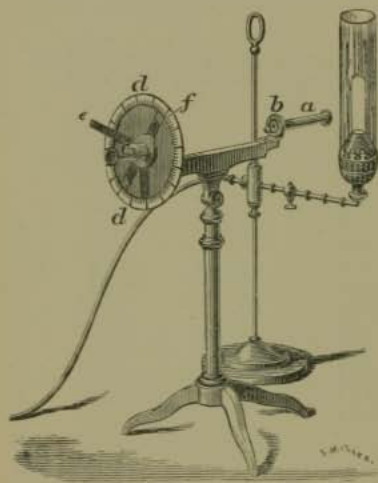


FIG. 20.

second prism, should it contain sugar or other polarising substance, the black stripe is no longer in the middle of the field, and the handle moving the index and prism must be turned until the black stripe is seen; or, if the stripe is broad and undefined, the prism is turned until the exact point is reached in which blue changes into red—the index at this point marking the amount of the polarisation by the scale and the direction; for if the index has to be turned to the right, the polarisation is +, or right-handed; if to the left -, or left-handed.

In order to make this quantitative, and to estimate the specific rotation of a sugar (*i.e.*, the number of degrees of rotation observed when 1 gm. of the sugar is dissolved in 1 c.c. of fluid and observed by yellow light through a tube 1 decimetre long), it is necessary to dissolve a known weight of the pure sugar in water; then if the length of the tube be known, and the temperature of the solution and the rotation be observed, all the necessary data are obtained. For example, let the rotation = α , the length of the

tube in decimetres = 1, the weight of substance in 1 c.c. of fluid = p , then the specific rotation for yellow light—

$$= \pm \frac{a}{p \cdot l} \quad \text{or } (a)_s = \pm \frac{a}{p \cdot l}$$

Provided there be only one sugar in the fluid under investigation, the specific rotation of which is known, the weight of the sugar in 1 c.c. of the fluid is estimated by the following formula: $P = \frac{a}{(a)1}$, where a equals the observed, and (a) the specific rotation. It is convenient to take the reciprocal of $(a) = r$, then the formula for 1 decimetre tube is simply $P = r \times a$. For example, taking the sp. rotatory power of cane sugar as 66.5, then the reciprocal = $\frac{100}{66.5} = 1.504$, and an angular deviation of 2° would equal 3 per cent. of sugar, for $2 \times 1.504 = 3.008$, or, if the 200 mm. tube is taken, the reciprocal is .75, the polarisation would, in such a case, be 4°, and $4 \times .75 = 3.0$.

TABLE X.—SHOWING THE ROTATORY POWER OF THE CARBO-HYDRATES.

Substance.	Divisor to get grms. per 100 c.c.	Specific Rotatory Power (absolute).		Specific Rotatory Power reduced to the Common Divisor, 3.86.
		$[\alpha]_D$.	Reciprocal.	
Dextrin,	3.95	+198.9°	.502	+194.4°
Sucrose,	3.85	+ 66.5	1.504	+ 66.8
Maltose,	3.92	+138.0	.724	+135.9
Lactose (anhyd.),	3.99	+ 55.4	1.805	+ 53.6
Lactose (cryst.),	3.99	+ 52.6	1.901	+ 50.9
Dextrose,	3.83	+ 51.3	1.950	+ 51.7
Levulose,	3.93	- 95.4	1.048	- 93.7
Invert sugar,	3.88	- 22.0	4.545	- 21.9

The readings may be converted into $(a)_s$ by simply multiplying by the factor 1.111; thus the reading for $(a)_D$ dextrose $\times 1.111$ equals 56.9, or practically 57.¹ If the specific rotatory power given is to be reduced to the common divisor 3.86 this is effected by dividing the given number in Table X. by the factor in column headed D and multiplying by 3.86. Thus, if 51.3 be taken as the absolute optical activity of dextrose, then $\frac{+51.3^\circ \times 3.86}{3.83} = +51.7^\circ$.

Soleil's Saccharimeter (see *Plate*) consists of three essential parts, two of which are fixed (fig. 1), AB and CD, the other movable, which is inserted

¹ The factor 1.111 is not absolutely correct for all sugars and concentrations. Brown, Morris, and Millar (*Journ. Chem. Soc. (Trans.)*, 1897, 93) give the following factors for converting $[\alpha]_D$ Biot into $[\alpha]_s$:—

	Per cent.	Factor.		Per cent.	Factor.
Cane sugar,	10	1.107	Dextrose,	5	1.111
Maltose,	10	1.113	Starch products,	10	1.111
"	5	1.111	"	5	1.111
Dextrose,	10	1.115			

(2.) *The Raw Sugar of Commerce.*—16·26 grms. of the sugar are powdered and dissolved in water, and the whole made up to 100 c.c.; if the solution is too dark, it may be clarified by sugar of lead. The tube BC is filled with the solution thus prepared and adjusted.

(3.) The next operation is to invert the sugar. 5 c.c. of fuming HCl are added to 50 c.c. of the sugar solution, and heated in the water-bath up to 68° C.; when that temperature is reached, the solution is put in the tube B'C, its rotating power (which is now inverse) observed, and at the same time the temperature at the moment of the observation.

Inversion of Cane Sugar by Invertase.—It is recommended in all cases in which other sugars, inulins, starches and glucosides are present to invert with invertase, because inversion by acids also acts upon the bodies mentioned. The only sugars inverted by invertase, besides cane sugar, are raffinose and gentianose, both of which bodies can easily be detected by their optical properties.

Invertase may be prepared¹ by allowing brewers' yeast to liquefy; this it does if kept at ordinary temperatures for a month or longer; the liquid filtered through a cloth has high hydrolytic power. Invertase may be precipitated by alcohol with some loss of activity. E. Bourquelot² shakes a top fermentation yeast with 95 per cent. alcohol, allows the mixture to stand for thirty minutes, drains off the liquid and dries the residue at 30°. One gm. of this product, triturated with 100 c.c. of water, saturated with thymol and filtered, yields an active solution preserving its properties for a week.

To invert with invertase a small quantity is added to the liquid, which must be slightly acid and contain no alcohol. The liquid is then kept at 50° C. for three to five hours. The liquid is then heated to 100° C. to destroy the invertase, and its polarimetric and reducing properties investigated. Instead of invertase, a small quantity of yeast may be added direct, and the liquid kept, as before, at 50° for some hours. From the polarimetric readings before and after inversion, the amount of sugar may be readily found by tables, such as those of M. Clerget, or by the formula as below.

Supposing the number given by the first observation is 75, by the second (inverted) 21, at a temperature of 12° C., the sum of the two numbers (75 + 21) makes 96. Now, on referring to M. Clerget's table, under 12°, or in the third column corresponding to the temperature of 12°, the nearest number to 96 is in this instance 95·6; the horizontal line in which 95·6 is placed is followed, and there is found, first, in the column A, the figures 70 per cent. of pure crystalline sugar; secondly, in the column B, the figures 114·45, placed by the side of 70, which indicates that the saccharine solution examined contains per litre 114·45 grms. of pure sugar. If, however, as sometimes happens, the solution contains a polarising substance not modified by acids, in such a case the difference of the two numbers, and not the sum, is to be taken and dealt with as before. It is scarcely necessary to remark, that if the substance is known to contain only crystallisable sugar, and the tube BC be filled, one observation alone suffices.

If tables are not at hand, the following formula can be used:—Let T be the temperature, S the sum or difference of the two determinations, P the rotatory power, R the quantity of sugar contained in 1 litre of the solution:—

$$P = \frac{200S}{288 - T} \quad R = \frac{P \times 16 \cdot 350}{10} = P \times 1 \cdot 635 \text{ grms.}$$

¹ O'Sullivan and Thompson, *Journ. Chem. Soc. (Trans.)*, 1890.

² *Journ. Pharm. Chim.*, 1901, xiv., 481-487.

Professor Jellet's instrument is a little more elaborate than *Soleil's*, and of great accuracy. The eyepiece or analyser of the apparatus consists of a suitably mounted prism, made from a rhombic prism of Iceland spar. The rhombic prism is cut by two planes perpendicular to the longitudinal edges, so as to form a right prism. The prism is next divided by a plane parallel to the edge just produced, and making a small angle with the longer diagonal of the base. One of the two parts into which the prism is thus divided is then reversed, so as to place the base uppermost, and the two parts are connected together.

Another distinctive feature of the instrument is, that the mechanical rotation of the analyser for the finding of any particular plane is dispensed with, this function being transferred to a fluid which has the power of turning the plane of polarisation opposite to that of the solution to be examined. The analysing tube slips into, and moves up and down in, the compensating fluid, so that different thicknesses of the latter fluid can be readily interposed and measured by a scale fixed to the instrument.

The *Saccharimètre à Penombyés*, of which the principle was enunciated by M. Jellet, as constructed by M. Duboscq, has some very great advantages. It requires the employment of a simple light, and the field does not present to the eye for comparison two different colours, but two intensities, sensibly diverse, of one and the same colour, so that the least variation can be appreciated. The simple light is best obtained by the insertion of a bead of some salt of soda on a platinum wire in the flame of a Bunsen burner. Landolt's with Lippich's polariser and Laurent's half-shadow are all excellent instruments, and suitable for the purposes of the analyst.

The polarising instruments in use are in France, Laurent's half-shadow, and in Germany Landolt's as constructed in numerous varieties by Schmidt and Haensch; in this country, probably, the latter class of instrument is the most common. Lippich has invented an instrument in which the visual field is divided into three parts; this is effected by placing behind a large Nicol prism two half prisms symmetrically disposed; one of the half prisms is fixed, the other can be rotated (*Zeit. f. Instrumentkunde*, 1894, xiv. 326). The saccharimeter scales are not identical. In the German instruments, the normal weight of sugar is 26.048 grms. in 100 c.c. of water at 17.5—in the modern French instruments 16.35; some instruments, e.g., Laurent, have angular degrees and sugar divisions marked on the same circular disc. The analyst, who has to assay sugar, finds sugar divisions convenient, but the public analyst has more frequently to estimate the percentage of sugar in solution than to assay the sugar.

As a rule, the most frequent determination is the percentage of cane or other sugars in a liquid; it is useful, therefore, to give the following relations:—

VALUES OF POLARISING INSTRUMENTS.

	Sugar Scale 1 Division =	Angular Degrees D.	Angular Degrees '	Cane Sugar corresponding to 1 Division.
German Instruments: Schmidt and Haensch, Ventze-Scheiber, etc., .	1	3468	3840	26048
Soleil Duboscq,	1	2175	2408	1635
Wild,	1	1331	1473	10
Laurent,	1	2167	2399	1627

Polarising instruments may be standardised by plates of quartz, accurately ground, of known thickness. The amount of right- or left-handed rotation of 1 mm. quartz for $[\alpha]_D$ is 21.67° , for $[\alpha]_J$ is 24° .

CONFECTIONERY.—SWEETMEATS.

§ 74. It would take many pages to describe the composition of the various kinds of sweetmeats in commerce: the basis of all is either cane or grape sugar, or honey, flavoured with appropriate essences, and coloured with various colouring-matters. A great many common sweetmeats have a most definite composition, and it is evident that a deviation from the ordinary process of manufacture, if it should take the form of substituting inferior articles for, or the addition of matters giving weight to, that which is ordinarily sold, would be an adulteration. As an example, 'peppermint lozenges,' or 'peppermint drops,' are composed of albumen, cane sugar, and oil of peppermint. None of these ingredients have any amount of mineral matter, and peppermint lozenges, when burnt, do not leave as much as .2 per cent. of ash. Since they are sold by weight it is easy to adulterate them by mineral substances; but such an addition would be most decidedly fraudulent, and the analyst may justly certify accordingly.

A large proportion of the common sweets contain nothing else besides sugar, for the manufacturer, by careful heating, is able to impart a quite surprising scale of colours, from the purest white to fawn colour, straw colour, reddish-brown, brown to almost a jet black, by this process alone.

SUGAR-CANDY is simply crystals of sugar obtained in a particular way, and is of all colours—from the white candy, largely used for the manufacture of artificial champagne, to all shades of yellow and red. As usually manufactured, the purified sugar solution is concentrated to a specific gravity of 1.420 to 1.450, and then run into copper cones, through which are passed a number of threads; these cones are heated with warm air, and the crystallisation occupies as much as from eight to fourteen days. The composition of white candy, made from pure loaf-sugar, is as follows:—

	Per cent.
Crystallisable sugar,	80.00
Uncrystallisable sugar,	Traces.
Ash,	0.0
Water,	20.00

The coloured candies may contain some mineral matter, and a good deal of uncrystallisable sugar; copper may, as an impurity, be present.

Composition of Sweetmeats Generally.

TOFFY.—Toffy is made by melting sugar with butter. The latter substance is readily extracted by ether and other fat solvents.

The ice-coating of cakes is composed of white sugar and albumen.

Mazzipan is made with sugar and almond paste.

A great many sweets are acidulated with citric acid, and a few have cavities within them, supposed to contain alcohol, but more often a little syrup. Gum tragacanth, citric acid, fruit sugar, chocolate, gelatin, albumen,

fatty and flavouring matters, with the following colouring-matters, make up the ingredients of the confectioner's shop:—

Red.—Cochineal, the juice of beet and of red berries, such as cherries, currants, and others. Rhodamine (French cream pink). Concentrated pink, which is a mixture of rosaniline hydrochlorate, with milk sugar, or starch. Fuchsine. Rhodites, which is the hydrochlorate of the phthalein of di-ethyl-meta-amido-phenol. Cherry red, which is the sodium salt of tetraiod-fluorescein. Ponceau red (Biebrich or crocein scarlet).

Yellow.—Saffron, safflower, turmeric, marigold, Persian berries. Saffron yellow, which is the sodium salt of amido-azo-benzene sulphonic acid. Primrose yellow (auramine). Imperial yellow (ammonium or sodium salt of hexa-nitro-di-phenylamine). Citron orange (sodium salt of xylenesulphonic acid-*azo-β*-naphthol).

Blue and Violet.—Indigo, litmus, heliotrope (Hofmann's violet); also mixtures of Hofmann's violet are sold under the name of 'lavender' and 'damson blue.'

Green.—Spinach juice and mixtures of yellow colours with blue.

Black.—Chinese ink, aniline blacks and browns.¹

Analysis of Sweetmeats.

§ 75. The analyst will naturally first turn his attention to the percentage of sugar, and estimate the total amount in the usual way; and, if necessary, investigate by optical and chemical means, whether there is more than one kind of sugar present. The essential oils may be dissolved out by petroleum ether, and identified by their odour; but the colouring-matter will, for the most part, be the chief substance necessary to examine. If the colouring is only on the external surface, it is better to detach it by scraping or rasping than to powder the whole substance up, for if the colour is carefully detached as pure as possible, tests may sometimes be directly applied without any further trouble. The colour by treatment with alcohol, with water, and with bleaching powder, is quickly referred either to the organic or to the inorganic division of chemical substances. With regard to organic colours generally, the reader may consult the sections treating of 'Colour,' where full directions are given for their identification. If, however, the colour is apparently inorganic, then the following substances may be particularly tested for:—

Among RED colours—*iron*;

„ YELLOWS—*chromate of barium*, and *lead compounds*, *arsenic* and *antimony*;

„ GREEN—*arsenic*, *copper*;

„ BLUE—*Prussian blue*;

„ WHITE—*sulphate of barium*, *salts of zinc*.

A weighed portion of the scraped-off colouring-matter is burned to an ash, which is dissolved in hydrochloric acid, and tested with hydric sulphide, after adding just sufficient soda to so neutralise the acid as to leave only a slight excess. Under these circumstances, lead, copper, or zinc, if present, will be precipitated; while, if it is strongly acid, zinc would remain almost entirely in solution. Ammonium hydrosulphide is next added to the solution, which has been boiled and filtered from any

¹ Some of the colours referred to are taken from Mr. Boseley's list of "Colouring Matters used in Confectionery."—Appendix to Report of Departmental Committee on the use of Preservatives and Colouring-Matters, 1901.

precipitate; this reagent will throw down iron, manganese, etc. To test for chromium, it is best to boil the colouring-matter with a solution of carbonate of potassium, when potassic chromate will be formed, which gives, in neutral solutions, a purplish precipitate with nitrate of silver. Barium is easily detected by fusing the ash with carbonate of soda, powdering the mass in a mortar, lixiviating with hot water, and treating the insoluble portion left with hydrochloric acid, and to the acid solution adding a little hydric sulphate; a heavy characteristic precipitate of barium sulphate is thrown down. If barium is present, it may exist with evidences of chromium, in which case, in all probability, the colouring-matter was chromate of barium, or if the sweetmeat is not coloured by barium chromate, baryta sulphate may have been added simply to give weight. Arsenic is tested for as more fully described *post*, but a preliminary treatment by Reimsch's test will give evidence of presence or absence of arsenic or antimony. Copper is also best detected by electrolysis, the substance being placed in a platinum dish, acidified, and then a rod of zinc inserted; or, the neater plan of connecting the dish itself with a battery may, where appliances are at hand, be preferred.

HONEY.

§ 76. Commercial honey is the saccharine matter collected and stored by one particular species of bee (*Apis mellifica*); but the production of honey is by no means limited to the bee, for there is a honey-ant¹ in Mexico, which stores a nearly pure syrup of uncrystallised sugar. This is slightly acid in reaction, and reduces salts of silver like formic acid.²

From determinations of the amount of saccharine matter in different flowers, it has been calculated that to make 1 kilogramme of honey, the bees must visit from 200,000 to 500,000 flowers.

A wasp of tropical America is said to yield a honey in which are found crystals of cane sugar, but the evidence as to this latter point is not decisive.³ A curious sample of honey has been analysed by A. Villiers.⁴ It was derived from Ethiopia, and is the produce of an insect resembling a large mosquito, which, like our wasp, makes its nest in cavities in the ground. It secretes no wax. The natives call the honey 'tazma,' and ascribe to it medicinal virtues, especially using it as a cure for sore throat. Its composition is as follows:—

	Per cent.
Water,	25.5
Fermentable sugar (levulose with a sixth of glucose in excess),	32.0
Mannite,	3.0
Dextrin,	27.9
Ash,	2.5
Loss and unestimated,	9.1

The honey contained a non-nitrogenous bitter principle.

The essential constituent of honey is a mixture of dextrose and levulose; it also contains mannite, wax, formic and other organic acids, pollen, not unfrequently alkaloidal and bitter principles from the plants,

¹ The *Myrmecocystus Mexicanus*. There are two kinds of workers—one the active form, the other sedentary—which produce the honey. The latter is the larger, and has a tumid abdomen; it never quits the nest. The honey is discharged into proper receptacles, and from it the Mexicans make a pleasant drink.

² H. Marsten, *Pogg. Ann.*, c., 550.

³ G. M. Wetherell, *Chem. Gaz.*, 1853, 72.

⁴ *Compt. Rend.*, lxxxviii., 292, 293.

possibly derived from the pollen, small quantities of cane sugar, of mineral matter, and invariably minute quantities of alcohol.

The properties of dextrose and levulose have been already described. The other saccharine constituent of honey—mannite, $C_6H_{14}O_6$ —crystallises in four-sided prisms, is soluble in 80 parts of alcohol of specific gravity 0.898, and in 1400 parts of absolute alcohol; in boiling alcohol it is more soluble, but in ether it is quite insoluble, and may be precipitated from alcoholic solution by ether. It has no action on polarised light. Its melting point is from 160° to 165° C.; at 200° C. it boils, and may be distilled, a portion being decomposed; at higher temperatures it carbonises. It does not reduce cuprous oxide. All these properties readily distinguish it from the other sugars. Chemically speaking, mannite is a hexatomic alcohol. Mannite may be separated from honey by boiling a weighed quantity of the honey with alcohol, evaporating down the alcoholic extract to dryness, and boiling this extract with absolute alcohol, concentrating the alcohol solution, and precipitating with ether.

Dr. Brown has published some analyses of honey, in which the different sugars have been identified; the general results of six of his samples may be thus stated:—

	Per cent.
Water (expelled at 100°),	18.07
Water expelled at a much higher temperature and loss,	7.99
Levulose,	36.22
Dextrose,	37.58
Ash,14
	<hr/> 100.00

The chief results of twenty-five analyses made by Mr. Hehner¹ of honey believed to be genuine, are as follows:—

	Mean of the twenty-five samples. Per cent.	Maximum. Per cent.	Minimum. Per cent.	
Moisture,	18.8	23.04	15.09	
Glucose,	67.85	75.34	61.42	
Difference,	13.35	19.17	8.48	
Mean of seven samples only.	Glucose after fermentation,	1.24	2.36	nil.
	Total solids after fermentation,	6.04	7.67	4.30
In five of the twenty-five samples, polarisation	= + 1°			
In one sample, polarisation	= - 1°			
In one sample very crystalline,	= - 11°			
In the remaining eighteen,	= 0			

Five samples which were considered adulterated yielded the following values:—

	1.	2.	3.	4.	5.
Moisture,	17.54	18.68	21.23	18.90	21.25
Glucose,	48.45	49.66	58.32
Difference,	34.01	31.66	20.45
Glucose after inversion,	43.33	48.77
10 per cent. solution polarises	+ 50°	+ 35°	+ 15°	+ 35°	+ 33°
Glucose after fermentation,	9.02	7.59	3.69	5.98	5.15
Total solids after fermentation,	31.45	25.33	53.29	23.36	18.38
Difference,	22.43	17.74	49.60	17.38	13.23
10 per cent. solution polarises after fermentation,	+ 30°	+ 28°	+ 7°	+ 16°	+ 10°

¹ *Analyst*, April, 1884.

Dr. E. Sieben¹ has published analyses of sixty samples of honey which he believed to be perfectly genuine. The general results are as follows:—

	Mean.	Maximum.	Minimum.
Moisture,	19·98	24·95	16·28
Grape sugar,	34·71	44·71	22·23
Levulose,	39·24	49·25	32·15
Invert sugar,	70·30	79·57	69·95
Cane sugar, by boiling with acid,	1·08 ²	8·22 ²	0·00
Total sugar,	75·03	81·74	70·20
Dry substance,	80·03	83·72	75·05
Substances other than sugar,	5·02	8·02	1·29

The chief adulteration of honey is the addition of starch sugar in the form of syrup. Cane sugar may be also found,² and mineral adulterations are possible.

Ernst Beckmann (*Zeit. f. analytische Chemie*, 1896, 263) has shown that if methyl alcohol be added to pure honey solution, there is no separation of dextrin bodies; and it is probable that the dextrin which may be in natural honey is a different dextrin to that which is the result of the hydrolysis of starch by acids.

He has applied this fact to the testing of honey for the addition of glucose as follows:—

To 5 c.c. of a 20 per cent. solution of honey are added 3 c.c. of a 2 per cent. hydrate baryta solution and then 17 c.c. of methyl alcohol. On shaking the mixture, pure honey remains clear or only slightly turbid. On the other hand, should the honey be adulterated with either glucose syrup or glucose there is a marked precipitate.

The same author has attempted to find a quantitative process on the weight of the compound of dextrin and baryta separated, but this latter process abounds with too many errors to be useful to the food analyst. It may happen that from the presence of phosphates and sulphates baryta gives a precipitate alone.

In this case these must first be separated.

Mr. Hohner determines the moisture at 100°C. The glucose is estimated by Fehling's solution—both before and after inversion; the inversion is produced by heating with 10 per cent. of hydrochloric acid to about 70°. The rotatory power of a 10 per cent. solution is determined both before and after fermentation.³ The fermentation is produced in a 10 per cent. solution by the addition of a little yeast, the vessel being kept in an incubator at 30°C. for from five to six days; this operation might be made more speedy by fermenting in a vacuum with plenty of yeast, as suggested by Boussingault. After fermentation, the solid matter is determined and subtracted from the percentage of glucose left unfermented. The proportion of unfermentable matter should be no larger than would be yielded by a pure glucose solution after fermentation, viz., about 5 per cent.

A pure honey has the following characters:—The moisture does not

¹ "Ueber die Zusammensetzung des Stärke-zuckersyrups, des Honigs, u. über die Verfälschungen des letzteren." *Zeitschrift der Rubenzucker Ind.*, 1884, 837.

² It is doubtful whether cane sugar occurs in pure honey, for, although Sieben gives 8·22 per cent. of cane sugar as a possibility in pure honey, he yet returns a honey containing 7 per cent. of cane sugar as adulterated, two statements that cannot be reconciled.

³ The honey derived from flowers turns a ray of light to the left, that from conifers to the right; it is obvious that honey derived from both species of plants may either have no influence on light, or deviate right or left, according to circumstances.

TABLE XI.—SOME ADULTERATED SAMPLES OF HONEY ANALYSED BY E. SIIBEN.

By titration with Fehling and Sacchse's solution.	Grape sugar.		Levulose.		3.	4.	5.	6.	7.	8.	9.	10.	11.	12.	
	P.c.	P.c.	P.c.	P.c.											
	25.63	31.80	25.42	31.80	56.39	19.45	76.84	20.85	79.15	2.31	33.05	0.0	0.0	...	Adulterated with cane sugar.
	37.20	19.60	61.97	18.54	69.90	18.54	61.97	36.48	81.46	12.46	44.40	7.20	4.2	4.4	" " and water.
	21.75	28.89	51.05	18.65	41.35	18.65	58.50	36.48	82.19	40.00	43.60	21.85	12.4	25.0	" about 15% starch sugar-syrup.
	34.61	23.51	51.05	17.81	58.83	17.81	58.50	36.48	82.19	23.69	51.43	16.82	7.6	13.0	" " 65%
	25.47	12.83	51.05	19.94	49.04	19.94	56.04	36.48	80.06	24.02	52.90	22.71	8.2	17.4	" " 40%
	21.92	22.30	51.05	18.12	85.00	18.12	34.75	36.48	81.88	57.13	56.02	34.10	15.2	34.0	" " 40% and also cane sugar.
	30.00	22.30	51.05	19.73	52.56	19.73	52.30	36.48	80.27	27.97	55.70	20.70	9.4	25.2	" " 80%
															" " 50%

exceed 23 per cent. The percentage of glucose before and after inversion is about the same. The unfermentable matter should not exceed 8 per cent. The polarising power of a 10 per cent. solution, both before and after fermentation, should be very small or *nil*.

There should be only a slight precipitate with either alcohol or baric chloride. The ash always contains from .01 to .035 per cent. of P_2O_5 . The ash from honey made from glucose larger quantities. The ash of pure honey is always alkaline, that made from glucose neutral.

There is an artificial honey in the market, sold as such, that consists of dextrose and levulose, and, according to Hehner, can only be distinguished from genuine honey by the entire absence of phosphoric acid from the ash.¹

A curiosity of food is a commercial American artificial honey, which is entirely composed of glucose syrup, while the comb is also artificial, and made of paraffin. The appearance of both comb and syrup is said to be superior to that of natural honey. It is not probable that this artificial honey will be met with; if there should be any suspicion that the comb is artificial, the presence or absence of paraffin is easily ascertained. Pure bees'-wax melts at 62° to 65° C. Its specific gravity is .962; it contains cerotic acid, myricine, as well as ceroleine; and, like other fatty matters, it is attacked and blackened by warm sulphuric acid. Paraffin, on the contrary, remains unacted upon, so that this test alone will suffice either to detect paraffin when pure, or to separate it from other matters, such as waxes and fats, which are carbonised by sulphuric acid.

§ 77. *Treacle, Golden Syrup, Molasses.*—The above-named syrups are by-products of the sugar industry, and all should consist essentially of sucrose and invert sugar, the common adulterant being glucose syrup. Considerable attention has been devoted of late years to this form of adulteration on account mainly of the possibility of arsenical contamination by arsenical glucose.

The composition of a genuine sample of ordinary treacle is as follows² :—

Cane sugar,	per cent.
Levulose,	44.80
Dextrose,	5.80
Ash,	7.12
Water,	8.21
Matters unaccounted for,	28.20
	5.87

Genuine syrups of this class on inversion turn the plane of polarisation to the left, and when treated by Beckman's test (see p. 125) give no indications of dextrin.

On the other hand, samples adulterated with glucose syrup give a decided dextrin reaction, and on inversion the polarisation is still to the right, for glucose syrup is not sensibly affected by methods of inversion. For example, the brands known as Crossfield's and Lyle's golden syrup, when pure, give the following polarimetric readings for $(a)_D$:—

	Before Inversion.	After Inversion.
Crossfield's at 17.5°,	+ 24.21	- 11.07
Lyle's at 17.5°,	+ 14.53	- 13.83

A simple method of analysis is given by E. W. T. Jones.³

¹ *Analyst*, Dec., 1885.

² C. J. Matthews and A. Hyde Parker, *Analyst*, xxv., 90.

³ *Analyst*, xxv., 88.

Mr. Jones makes a 10 per cent. solution at 15.5°, and takes the sp. gravity; on dividing the gravity, minus 1000 by 4, an approximation to the amount of water is obtained; thus a sp. gravity of 1032 would show 80 per cent. of water.

The polarimetric value of the liquid is obtained before and after inversion. The following formula is then used for calculating the amount of glucose syrup, provided the syrup shows dextro-rotation after inversion:—

$$\frac{([\alpha]_D \text{ after inversion} + 11) 100}{120} = \text{per cent. glucose syrup.}$$

Another simple method of calculating is given by Bernard Dyer.¹ He assumes the specific rotatory power of glucose syrup to be $(\alpha)_D$ 113.0° and the K value 42.

He polarises at 20°C. both before and after inversion, and calculates the percentage of glucose syrup by the following formula, in which R stands for the specific rotatory power at 20° C. of the uninverted sample.

S stands for the + rotation of sucrose present and K for the copper reducing power:—

$$\text{Per cent. of glucose syrup} = \frac{0.206K + (R - S)}{1.217}$$

The inversion of a 10 per cent. solution of syrup is best performed by invertase, or, if no invertase is at hand, a gramme of pressed yeast is added to 50 c.c. of the solution, and kept at 52° C. for five hours, then boiled to destroy bi-rotation; on cooling, it is made up to the original volume and examined polarimetrically. A more elaborate method of analysis, in which practically all substances are determined quantitatively, is as follows:—

Total Solids.—Make up a 10 per cent. solution at 15.5°C., and take the sp. gravity.

Determine the weight of mineral matter in, say, five grms. of the original syrup, shake up the ash with 100 c.c. of water, and take the sp. gravity; subtract the sp. gravity thus found for an equivalent amount yielded by a 10 per cent. solution from the original sp. gravity, and then make use of the usual divisor 3.86, which gives carbo-hydrates. For example, a 10 per cent. solution had a sp. gravity of 1032; five grms. of the original substance yielded 0.37 per cent. ash, which, when shaken up with 100 c.c. of water, gave a solution of 1003 sp. gravity. We have, therefore,

Sp. gravity of 10 per cent. solution = 1032.

Sp. gravity of a solution of ash from 10 grms. = 1006, which on subtraction leaves 26; this multiplied by 10 and divided by 3.86 equals 67.3 per cent. carbo-hydrates. The ash is 7.4 per cent., which gives total solids 74.7 per cent. and water 25.3 per cent. The cane sugar is obtained from the polarimetric readings before and after inversion.

Cupric Reducing Power.—The 10 per cent. solution is diluted to double its volume, and the cupric reducing power of 2 c.c., which is equivalent to 100 milligrams of the original syrup, is ascertained by Fehling's solution (see method at page 111).

The weight of CuO, multiplied by 1000 and divided by the mean cupric reducing power of 1 gm. of invert sugar (2.36), equals reducing sugars.

Estimation of Matters Disappearing during Fermentation.—A few grms. of pressed yeast are added to 100 c.c. of the ten per cent. solution and kept

¹ *Analyst*, xxv., 95.

at 18° for from three to five days, until all fermentation has ceased. After evaporating off the alcohol, the alcohol-free extract is made up to the original bulk and the specific gravity determined and also a polarimetric reading taken.

Pure samples show no optical activity after fermentation; samples in which dextrin is present show an optical activity in proportion to the undestroyed dextrin, fermentation always hydrolysing a portion of the dextrin.

From the original specific gravity, the extract specific gravity is subtracted; this figure multiplied by 10 and divided by 3.86 gives the per cent. of sugar which has disappeared during fermentation.

The cane sugar found by optical activity before and after inversion added to the reducing sugars found by the cupric estimation should about equal the sugar which has disappeared during fermentation, but on making a copper estimation after fermentation there is generally found some reduction due to unfermented dextrose and levulose. The CuO found \times by 1000 and divided by 2.36 (1 grm. of invert sugar = 2.36 grms. of CuO) equals the percentage of unfermented reducing sugars. The percentage of levulose and dextrose is estimated in the following way:—

$$\begin{aligned} \text{Let } x &= \text{per cent. of dextrose; } y = \text{per cent. of levulose,} \\ \text{then } x + y &= \text{total sugars,} \\ \text{and } 52.8x - 95.65y &= 0, \end{aligned}$$

the difference between the two, of course, equalling the levulose.

The total reducing sugars are found by subtracting the optical activity due to cane sugar from the optical activity of the original solution and taking into account the cupric reducing power.

Estimation of Maltose and Dextrin.—It has been already stated that after complete fermentation there should be no rotation in a sample which has not been adulterated with glucose syrup; if there is a dextro-rotation, it will be due to dextrin or to dextrin in combination with maltose. In order to ascertain this, the liquid which has been fermented and deprived of alcohol by evaporation is submitted to the action of malt extract or diastase. The malt extract is conveniently prepared as follows:—

100 grms. of pale malt are digested with 250 c.c. of water and allowed to stand in the cold for twelve hours; the liquid is then filtered and the filtrate used.

The specific gravity of this filtrate is taken; it is also examined optically and its K value ascertained by a copper estimation. An aliquot part (2–3 c.c.) is then added to the alcohol-free extract as above, and the whole kept at 50° for five hours. Any reducing sugars set free by the diastase will now be shown by an increased cupric reducing power. Therefore, a copper estimation is made on an aliquot portion, the result being, of course, corrected by subtracting any reduction due to the malt extract added.¹ To another portion some yeast is added, and the liquid kept at a fermentation temperature for some days. If fermentation takes place and the fermented liquid after evaporating off the alcohol shows no reducing power when treated with Fehling, it may be with confidence presumed that the reduction before fermentation was due to maltose alone; and from the reduction or optical activity or both the percentage of maltose and dextrin may be calculated.

¹ If more than 2–3 c.c. of the malt extract is added, the specific gravity, the opticity, and the K value of this extract must be obtained after heating to 50° under the same conditions as the mixture.

To show the mode of calculation, an example may be taken from Brown and Heron's paper ("History of Starch, and its Transformations," *Journ. Chem. Soc.*, 1879).

A starch conversion by cold malt extract gave the following values— $(\alpha)_{3.86} + 163.3^\circ$ and $K_{3.86} 48.8$.

(1.) Determination of the proportion of maltose and dextrin simply by the optical properties.

We have the optical activity of dextrin + 216, of maltose + 150°, difference 66.

$$\text{Then} \quad \frac{(163.3 - 150) 100}{66} = 20.1$$

therefore the mixture consisted of

Maltose,	79.9
Dextrin,	20.1
	100.0

(2.) Determination from the reduction.

The reduction is simply due to maltose, and taking $K_{3.86}$ as 61

$$\frac{48.8 \times 100}{61} = 80.0$$

therefore the mixture consisted of

Maltose,	80.0
Dextrin,	20.0
	100.0

This should give the following opticity:—

Maltose,	$\frac{150 \times 80}{100} = 120$
Dextrin,	$\frac{216 \times 20}{100} = 43.2$
	163.2

which is practically identical with the observed opticity, viz., $(\alpha)_j 163.3^\circ$.

Some analyses of adulterated treacle have been published by Mr. Chapman as follows:—

TABLE XII.

	No. 1.	No. 2.	No. 3.	No. 4.
Cane sugar,	20.63	26.10	27.40	4.10
Invert sugar,	10.80	12.00	23.60	...
Dextrose,	6.10	22.00	11.40	31.70
Maltose,	25.40	.40	...	7.50
Dextrin,	12.95	8.98	2.50	34.30
Ash,	1.82	5.20	7.24	1.10
Water,	19.08	20.50	22.69	21.30
Undetermined,	3.22	4.82	5.17	...
	100	100	100	100
Optical activity $[\alpha]_D$, . . .	+77.3°	+45.2°	+24.6°	+99.01°

It often happens in the examination of treacle, jams, and similar products that, after treatment with malt extract, the sugar produced is only partly or not at all destroyed by fermentation. This points to the setting free of a polysaccharide, which is unfermentable. Some chemists, however, ascribe this circumstance to 'gallisin,' a dextrin-like unfermentable body, which has not been obtained pure. The probable opticity of gallisin is $(\alpha)_D + 84^\circ$ and K absolute 41.

Treacle, golden syrup, and molasses should, as a matter of routine, be examined for arsenical contamination by the method described *post* (see Index).

JAM.

§78. Jam consists of various species of fruit preserved by boiling in strong syrup. Most jams are very readily adulterated, since any tasteless vegetable tissue, such as vegetable marrow, turnips, etc., when mixed in jam, cannot be readily detected by the palate. The chemical composition of the various jams is simply the chemical composition of the fruit juice and fruit itself, with the loss of a few volatile constituents and the addition of cane sugar. The latter may be in part inverted by the action of the organic acids or ferments so constantly found in fruit. A large proportion of jams and marmalade are preserved by antiseptics, more especially by salicylic acid (a common proportion is 3 grains to the lb.); to not a few glucose syrup is added. The carbo-hydrates in jams and marmalade are determined on precisely the same lines as the carbo-hydrates in treacle. Arsenic will also have to be tested for, as described *post* (see Index). The detection of other adulterations in jam are mainly microscopic; but, at the same time, in many cases a careful observation of the absorption-spectrum will assist the diagnosis. In order to carry out this successfully, in addition to the precautions before described, it will be safest in all cases to use comparison liquids; and those who devote themselves to this study should have at hand a variety of genuine jams of different ages. The mean composition of the more common kinds of fruits is detailed in the following table [*König*]:—

TABLE XIII.—100 PARTS OF THE SEED FRUIT.

	Water.	Nitrogenous substances.	Free acid.	Sugar.	Other non-nitrogenous matters.	Woody fibre, etc.	Ash.
Apple, . . .	83.58	0.39	0.84	7.73	5.17	1.98	0.31
Pear, . . .	83.03	0.36	0.20	8.26	3.54	4.30	0.31
Plum, . . .	81.18	0.78	0.85	6.15	4.92	5.41	0.71
Prune, . . .	84.86	0.40	1.50	3.56	4.68	4.34	0.66
Peaches, . .	80.03	0.65	0.92	4.48	7.17	6.06	0.69
Apricots, . .	81.22	0.49	1.16	4.69	6.35	5.27	0.82
Cherries, . .	80.26	0.62	0.91	10.24	1.17	6.07	0.73
Grapes, . . .	78.17	0.59	0.79	24.36	1.96	3.60	0.53
Strawberry, .	87.66	1.07	0.93	6.28	0.48	2.32	0.81
Raspberry, . .	86.21	0.53	1.38	3.95	1.54	5.90	0.49
Bilberry, . .	78.36	0.78	1.66	5.02	0.87	12.29	1.02
Blackberry, .	86.41	0.51	1.19	4.44	1.76	5.21	0.48
Mulberry, . .	84.71	0.36	1.86	9.19	2.31	0.91	0.66
Gooseberry, .	85.74	0.47	1.42	7.03	1.40	3.52	0.42
Currant, . . .	84.77	0.51	2.15	6.38	0.90	4.57	0.72

Brief Notes of the Microscopical Structure of Certain Fruits.

§ 79. *Apples and Pears.*—Both apples and pears contain numerous dotted ducts and spiral vessels. There is no very distinctive peculiarity about these ducts, but in the core will be found a strong horny membrane with spiculated cells, crossing one another at right angles, forming altogether a very singular tissue, and one which, once seen, can always be recognised.

Damson.—The skin of the damson is composed of at least two distinct species of cells underlying the transparent epidermis. One kind is a double row of reddish-purple oblong or oval cells, having, when seen in section, an average length of $\cdot 00232$ inch, and an average breadth of about $\cdot 000928$ inch; seen from above (as in tearing off a shred of the tissue) they form a beautiful five- and six-sided mosaic pattern, the size of the cells being from about $\cdot 000928$ to 00116 inch. The blue cells are very similar in shape and size to the reddish-purple; below the blue there are some loose cells containing chlorophyll. Hence the beautiful colour of the damson is the combined effect of the blue, the red, and the green shining

through the transparent epidermis. The pulp contains the usual large colourless globes or cells, of $\cdot 0116$ inch average diameter (*b*, fig. 21). Spiral vessels are numerous, stomata are occasionally to be seen on the surface of the dark-coloured epidermis. The breadth or thickness of the skin is $\cdot 00814$ inch. By the use of bleaching powder, a small portion of the skin may be deprived of its colour, either partially or wholly, according to the judgment of the operator, and then will be seen a mapping out of the whole

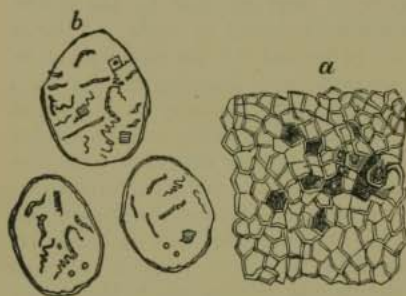


FIG. 21.—*a*, Epidermis of damson; *b*, pulp cells, $\times 115$.

surface into lobes by cells so placed that they form a network.

Plum.—There are at least three distinct structures to be seen in the boiled and preserved plum:—1. The epidermis, consisting for the most part of a pavement-like layer of little square or irregularly oblong cells, filled with a granular matter (*c*, fig. 22), the size of the cells averaging from about $\cdot 000696$ to $\cdot 00116$ inch; the general distribution of these cells is somewhat circular. Scattered tolerably uniformly are patches of a deeper colour, with larger cells, the patches being irregularly circular, and the centre of the patch an empty space, which possibly is a much deformed stoma. The pulp consists of the very common large globular cells (*a*, fig. 22), of about $\cdot 12$ to $\cdot 14$ inch diameter, almost perfectly transparent, with a shrivelled mass within. Lastly, there are some beautiful masses of compound cells, varying in size from $\cdot 016$ to $\cdot 48$ inch (*b*, *b*, fig. 22), the length usually being from one and a half to three times the breadth. These compounds are either prismatic in shape or oval, while a few resemble long tubes. The number of cells thus bound together is very variable, since from seven up to twenty-seven may be counted on one side. The little cellular members of the composite are five-sided cells of an average length of $\cdot 06$ inch.

Oranges—Lemons; Marmalade.—Marmalade is made by preserving

sliced up oranges or lemons, or both combined, in a strong syrup. Orange marmalade is properly made from Seville oranges only, lemon marmalade from lemons only. It is not practicable to distinguish by the microscopic structure alone whether the substance is orange or lemon, or to what species the orange or the lemon belongs. It is, however, most easy in marmalades to recognise substances foreign to marmalade, because the structure of the vegetable tissues used is very distinctive. Good marmalade is wholly composed of fine sections of the fruit: on selecting the thinnest of these sections, or (what amounts to the same thing) cutting and preparing a section, there will be three structures to notice. It will be observed that the colour layer is very thin, and composed of layers of yellow cells (*a*, fig. 23), many filled with oil, and here and there large cavities, covered with a thin transparent epidermal layer. The size of the cells is small, about $\cdot 000147$ inch in diameter. Beneath the yellow layer there is a deep layer of colourless cells, traversed by a network of bundles of vessels, each bundle consisting of a dozen or more spiral vessels, of small diameter, in the midst of the ordinary elongated fibre-like cells. In this layer are large

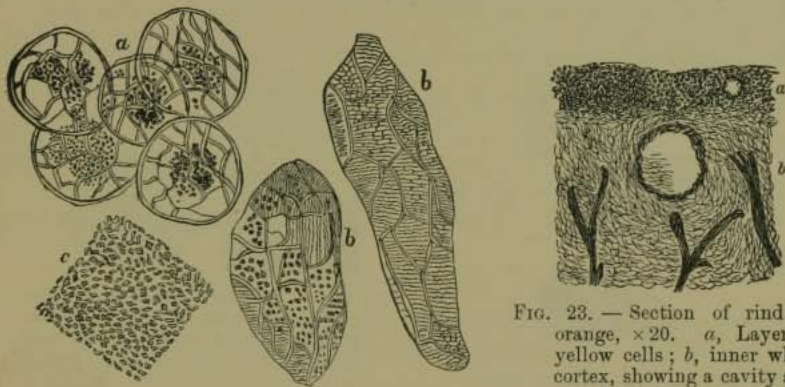


FIG. 22.—Structures found in the plum, $\times 115$.
a, Pulp cells; *b*, *b*, compound cells; *c*, a portion of epidermis.

cavities, $\cdot 0415$ inch diameter or more, and around these cavities the cells are applied in concentric layers. The pulp of the orange presents a number of soft and thin-walled cells without any very distinctive peculiarity. Large spirals, large oval cells and structures, dissimilar to the above, will be suspicious signs, and will denote adulteration.

The Strawberry may be readily distinguished under the microscope by the great number of very small seeds which are scattered on the exterior of the fruit. These seeds are pyriform and very regular in size, being about $\cdot 038$ inch wide at the broadest end, and $\cdot 07$ inch in length. The coat of the seed is almost smooth; under a high magnifying power, it may, however, be seen to be slightly tuberculated. Each seed is attached to the central part of the fruit by means of a vascular bundle formed of delicate fibres and spiral vessels; and the consequence of this structure is, that the strawberry is full of spiral vessels, all of minute size and very transparent. The cells seen when jam is examined are, for the most part, collapsed and shrivelled; those that are not so are large oval or pyriform cells, often containing shrivelled cell contents. Common measurements of these cells



FIG. 23.—Section of rind of orange, $\times 20$. *a*, Layer of yellow cells; *b*, inner white cortex, showing a cavity and vascular twigs.

are .008 inch small diameter, .0176 inch long diameter for the oval cells, and for the pyriform .0136 inch broad end, .0184 long diameter.

A general analysis of strawberries has been published by J. M. H. Munro. (*Chem. News*, 1, 227.)

	Per cent.
Water,	89.30
Organic matter,	10.27
Ash,43
The ash contained—Calcium phosphate,	22.91
Potassium carbonate,	60.77
Magnesia,	2.93
Soda,	1.29
Sulphuric anhydride,	3.88
Sand and insoluble matter,	6.61
Undetermined,61

The Raspberry has its seeds reticulated, and in most jams the form of the fruit is preserved quite sufficiently for recognition.

The Gooseberry has an epidermis in which can be seen a mosaic pavement of cells, and the fruit also possesses clavate hairs.

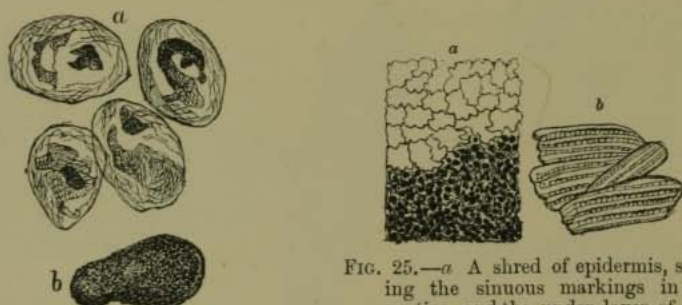


FIG. 24.—*a*, Pulp cells of strawberry, $\times 115$; *b*, strawberry seed, $\times 20$.

FIG. 25.—*a* A shred of epidermis, showing the sinuous markings in one portion, and the under layer of cells in another; *b*, the compound bodies, $\times 115$.

The Blackberry.—The seeds are reticulated, and the cuticle covered with stellate hairs.

Currants.—Both the black and the red currant are similar in structure; the epidermis is covered with an excessively thin membrane, showing sinuous wavy divisions, and set with simple hairs. Beneath the outer membrane are the colour layers, consisting of little square masses with rounded angles about .00029 to .00039 inch diameter (*a*, fig. 25). The pulp is made up of thin-walled cells, and, lastly, here and there may be found peculiar compound bodies, *b*, attached to the inner layer of the epidermis. These are about .0058 inch in length and .0015 inch in breadth, and are formed of a number of oblong cells. So far as known, these bodies are found only in the currant.

SACCHARIN.

This is the popular name given to a crystalline substance discovered by Fahlberg and Remsen (*Deut. Chem. Ges. Ber.*, xii., 469-573) in 1879. It was first obtained by the oxidation of orthotoluene sulphonamide by permanganate; its formula is $C_7H_5O_3SN$, and it has been named anhydro-ortho-sulphamine-benzoic acid $[C_6H_4 < \begin{smallmatrix} CO \\ SO_2 \end{smallmatrix} > NH]$ —or,

shorter, Benzoic sulphimide—it is in the form of white crystals, soluble in hot water, alcohol, and ether. It melts at 220°, and can be sublimed without undergoing decomposition; it forms crystalline compounds, with the alkalis and alkaline earths; on evaporating an aqueous solution strongly acidified with HCl, almost to dryness, it is transformed into ortho-sulpho-benzoic acid. It is so intensely sweet that 1 part in 10,000 of water is very perceptible; the sweetness is like that of sugar, but with a peculiar flavour. The substance is used in commerce, and the analyst will look for it in all sweet manufactured liquids, such as lemonades, temperance drinks, and liqueurs; it is said also to be added to sugar itself, to increase its sweetening power. It is not poisonous, but, on the other hand, it has no nutritive powers; it is said to pass through the kidneys unchanged. A general method of detecting saccharin is to shake liquids, after feebly acidifying, with ether; to separate the ether and evaporate to dryness. The ethereal extract obtained in this way, if saccharin be present, will taste extremely sweet, and if the residue is gently fused in a platinum dish with six times its weight of pure sodic carbonate and potassic nitrate, the sulphur will be oxidised into sulphate, and the fused mass will, when dissolved and the solution acidified with HCl, give a precipitate with boric chloride.

Saccharin fused gently with potash is converted into salicylic acid, and, therefore, with ferric chloride strikes a violet colour.

Solids, such as sugar, are also treated with ether, and the ethereal extract examined as before; but if the solid has an alkaline reaction, it is best first to extract with hot water, acidify the aqueous solution, and shake out with ether.

Fatty substances or liquids must first be freed from fat, by treatment with light petroleum. A quantitative estimation is most accurately made by oxidation of the sulphur by fusion and precipitating the sulphate with barium chloride—1 part of barium sulphate equals $\frac{7}{85}$ of saccharin.

Saccharin treated with NH_3 in the cold and the excess of ammonia got rid off by evaporation forms the compound $\text{C}_6\text{H}_4\text{CO}, \text{SO}_2\text{N}_2\text{H}_4$; on treating this with alkaline bromide, nitrogen is evolved. The number of c.c. of N divided by 0.89 = saccharin in centigrams. (H. Defournel, *J. Pharm.*, 1901 [vi.], xiii., 512).

Dulcine or Sucrol (Phenetol carbamide) $\text{CO} < \begin{smallmatrix} \text{NH} \\ \text{NH}_2 \end{smallmatrix} > \text{C}_6\text{H}_4, \text{OC}_2\text{H}_5$ is also used for sweetening purposes; it may be extracted from liquids by ethylacetate.

STARCH, $\text{C}_6\text{H}_{10}\text{O}_5$.¹

§ 80. It is convenient to consider the starches together, more especially as, however varied in form, the chemical composition of all starch is very similar, if not identical.

Every starch corpuscle is composed of at least two probably isomeric bodies, the one '*granulose*,' soluble in saliva, and coloured blue by iodine; the other coloured by iodine pale yellow, and only becoming blue after the addition of sulphuric acid; it is fully soluble in ammoniacal oxide of copper, and appears to agree very closely with the characters of *cellulose*.

These two substances may be most readily separated by diluted chromic acid, which dissolves granulose very easily, whilst cellulose remains unaltered. All starch is very hygroscopic: wheat starch, dried in a vacuum, still contains 11 per cent. of water, and air-dried from 16 to 28 per cent. of water. Starch is insoluble in cold water or spirit. Some chemists, indeed, assert that if finely powdered in agate mortars, or with quartz sand, a small portion dissolves; others contend that this is no true solution, but the starchy matter in a state of most minute division. If warmed with water, the starch granules swell, and when heated up to 100° most starches form a semi-solution in water. True compounds of starch with bases are scarcely established. Lime and baryta appear to form weak

¹ Brown & Millar (*Journ. Chem. Soc.*, 1899, 331), consider that soluble starch has a molecular weight of 32,400, and is formed by the combination of maltan and dextrin groups.

Maltan group.	Dextrin group.
80 $\text{C}_{12}\text{H}_{22}\text{O}_{10}$	40 $\text{C}_6\text{H}_{10}\text{O}_5$

unions, and the intense colour produced by iodine, as well as bromine, seems to point to the formation of haloïd combinations. Fritsche, indeed, states that he has isolated the iodide and the bromide of starch, the former containing ten equivalents of starch and one of iodine.

Starch heated in closed tubes up to 100° C. changes gradually into soluble starch. If the temperature is raised up to 160° or 200° C., it forms a transparent mass consisting wholly of dextrin. At 220° to 280° C. still further change is produced, and the result is *pyrodextrin*, a substance easily soluble in water (but insoluble in absolute alcohol and ether), and with the composition of $C_{48}H_{26}O_{36}HO$. At still higher temperatures there is carbonisation, and the formation of products similar to those caused by the decomposition of sugar.

Starch is easily changed into sugar by the action of dilute mineral acids, as well as by oxalic acid, aqueous chloride of zinc, and by certain ferments—diastase, saliva, yeast, etc.

The estimation of starch in organic bodies is always based on hydrolysing the starch and converting it into either a reducing sugar, and dextrin, or the hydrolysis is carried farther, and the dextrin also converted into sugar. This conversion is done in various ways. 1 to 1.3 grms. in 40–50 c.c. of .2 per cent. sulphuric acid for eight hours may be heated in closed tubes or flasks in a glycerin bath to 108° or 110° C.; or the substance, in some instances, may be heated in ordinary flasks in the water-bath, with a 2 per cent. solution of hydrochloric acid, for many hours until it ceases to give a starch reaction with iodine.

An excellent general method has been proposed by Dragendorff:—2 to 3 grms. of the powdered and dried substance are heated, with 25 to 30 c.c. of a 5 per cent. solution of potash in absolute alcohol, for from eighteen to twenty-four hours in the water-bath, filtered hot, through a weighed ash-free filter; the residue on the filter is washed first with hot absolute and then with cold ordinary alcohol, and lastly with water—the residue is now dried at 110° C. and weighed; the loss of weight corresponds to the albuminoid matters, the fat, the sugar, and the soluble salts, which have been removed by the alcoholic potash, the alcohol, and the water. The filter and its contents are now divided finely by scissors, and boiled with 5 per cent. hydrochloric acid until a blue colour is no longer struck with iodine, the liquid is then filtered through a weighed filter, and the residue washed, dried, and weighed—the difference between the weights of Nos. 1 and 2 gives very nearly the starch. This weight may, of course, be controlled by estimating the glucose by Fehling solution.

A general method for the estimation of starch in flours has been worked out by C. O'Sullivan (*Journ. Chem. Soc.*, Jan. 1, 1884, No. ccliv., 2–10); its principle is the freeing of the finely divided substance from fat, albuminoids, and amylans by suitable solvents, and then transforming the starch by the action of diastase into maltose and dextrin, the proportions of which are estimated by the Fehling solution and by the polariscope.

1. Preparation of the Diastase.

2 to 3 kilos. of finely-ground pale barley malt are steeped in water just sufficient to cover the whole. After standing several hours it is filtered by means of a filter press; and, if not clear, by passing it also through an ordinary filter. The diastase is now precipitated by alcohol sp. gr. .83, the alcohol being added so long as the precipitate is flocculent, but

discontinued when a milky or opalescent appearance commences. The diastase is washed with alcohol (.86-.88), dehydrated with absolute alcohol, and dried in vacuo over sulphuric acid. Diastase thus prepared is a white, dry, friable, soluble powder retaining its activity for a long time.¹

Freeing the Flour from Fatty Matters.—5 grms. of the flour are first saturated with alcohol sp. gr. .82, and from 20 to 28 c.c. of ether added. The flask containing the mixture is set aside for a few hours, and the whole is then filtered; the residue being washed with ether.

2. Removal of Sugars, Albuminoids other than Casein, and Matters soluble in Weak Alcohol.

To the flour now fat free, 80 or 90 c.c. of alcohol sp. gr. .90 are added, and the mixture kept at 35° to 38° C., with occasional shaking, for a few hours. The alcoholic solution is then passed through the same filter which has been used for the 'ether' operation, and the residue washed by decantation with the same strength alcohol, and at the same temperature.

3. Solution of the Amylans.

The flour which has been treated with alcohol and ether is now submitted to the action of water; the flour is digested with half a litre of water, and decanted through a filter at the end of twenty-four hours; it is then repeatedly washed with water at 35° to 38° C.

This part of the operation is tedious, for the filtration is sometimes very slow.

4. Conversion of the Starch.

The residue is finally transferred to a beaker, and boiled for a few minutes in 40 or 45 c.c. of water with constant stirring—it is then cooled to 62° or 63° C., and .021 to .038 grm. diastase, dissolved in a few cubic centimetres of water or a few cubic centimetres of normal malt extract added. In a very short time the solution ceases to give a starch reaction with iodine, but it is best to maintain the digestion for an hour, because filtration is then easier. At the end of that time the contents of the beaker are boiled for eight or ten minutes, thrown on to a filter, and the filtrate received into a 100 c.c. measuring flask. The residue is carefully washed with small quantities of boiling water at a time. When the flask is nearly full, its contents are cooled down to 15°·5 C., and made up to 100 c.c. with water at that temperature. Should the filtrate exceed 100 c.c., it is to be concentrated to the proper quantity. The specific gravity of the solution is now taken; its optical activity is determined, and its reducing power on copper solution estimated by boiling with Fehling.

The optical activity of maltose, according to Brown and Heron, is $[\alpha]_D^{20}$ 150° and K_{200} 61·0. That of dextrin $[\alpha]_D^{20}$ 216°. The principles of calculating dextrin and maltose from the optical and copper values of a mixture have been already detailed. An example from Mr. O'Sullivan's paper will make the above clear.

5 grms. of barley flour, treated as above, gave as the ultimate result

¹ Instead of diastase, what is called normal malt extract, made by steeping 100 grms. of finely divided pale malt in 250 c.c. of water for twelve hours and filtering, may be used.

100 c.c. solution having a sp. gr. = 1.01003 = 2.539 grms. solid matter.¹
 9.178 grms. of this solution reduced .241 cupric oxide, and a layer of it
 200 mm. in length, gave a deviation with the Soleil-Ventzke-Scheibler
 saccharimeter = 21.1 div.

From these data we have :—

$$0.241 \times 0.7256 \text{ (K of maltose } 62.5) = .1748 \text{ gm.}$$

maltose in 9.178 grms. solution.

The weight of 100 c.c. solution is from the sp. gravity 101.003 grms. ;
 hence, percentage of maltose 9.178 : 101.003 :: .1748 = 1.923. Then—

$$1.923 \times 8.02 = 15.422 \text{ optical activity of the maltose,}$$

$$21.1 - 15.422 = 5.678 \text{ optical activity of the dextrin,}$$

and

$$5.678 \div 11.56 = .491 \text{ the dextrin in 100 c.c. solution.}$$

We have then in the 100 c.c.—

Maltose,	1.923	grms.	
Dextrin,491	,,	
Diastase,030	,,	
	2.444		
Against,	2.539	,,	as represented by the sp. gr.
	.095	,,	matter unaccounted for.

This .095 gm. of unestimated matter was partly referred to .062 gm.,
 α amylan, leaving a total error of only .012 gm.

Mr. O'Sullivan's determinations by this method of the starch in flour,
 etc., are as follows :—

	Per cent.
Barley flour,	46.3
,, malt,	39.9
Wheat flour,	55.4
,, malt,	43.4
Rye,	44-46.0
Rice,	75.77
Maize,	54.58
Oats,	35.38

A method of starch estimation devised by Märcker and Morgen,² much
 used on the Continent, is as follows:—3 grms. of the finely-powdered
 substance is digested in a small metal vessel with 50 c.c. of water at a
 heat of about 100° C. for twenty minutes, then cooled to 70° C., and 5 c.c.
 of freshly-prepared malt extract (100 grms. malt to 500 water) added ;
 the mixture is now kept at 70° C. for twenty minutes, in order to liquefy the
 starch paste. Then 5 c.c. of a one per cent. solution of tartaric acid is
 added, the vessel covered with a metal cover and submitted in a Soxhlet's
 digester to heat at a pressure of 3 atmospheres for half an hour. The
 digester with its contents is now allowed to cool, the lid unscrewed, the
 metal vessel removed, a second 5 c.c. of tartaric acid added, and the whole
 heated once more to 70° C. for twenty minutes. The solution is transferred
 into a flask of 250 c.c. capacity, filtered, diluted to 200 c.c., and inverted by

¹ Mr. O'Sullivan has taken 3.95 for the divisor of the sp. gr.—1000. He has also
 taken the optical activity of maltose to be [α]_D 154, and K 62.5, the optical activity of
 dextrin as [α]_D 222, all numbers differing from those of Brown and Heron.

² *Handbuch der Spiritus-fabrikation*, 1886.

boiling with 15 c.c. of HCl of 1.125 sp. gravity. After three hours' boiling under an inverted condenser, the liquid is cooled, almost neutralised by soda, made up to 500 cubic centimetres, and the sugar estimated by Fehling's method.

A. Leclerc¹ has proposed to treat 2 grms. of the moistened starchy substance with 180 c.c. of concentrated neutral solution of zinc chloride, and to heat the mixture in a salt bath up to 108° C. for two hours or more, until a solution is effected. On cooling, the solution is made up to a definite bulk, filtered, and 25 c.c. of the filtrate precipitated by 2 c.c. of hydrochloric acid and 75 c.c. of 90 per cent. alcohol; the precipitate is stated to consist only of starch and dextrin. The precipitate is washed with acid holding alcohol, then with 90 per cent. alcohol, dried, and weighed. It is finally burned, and any ash subtracted.

M. Honig² heats the starch-holding substance in glycerin to 210°, and pours the solution into strong alcohol, and when quite cool adds a fifth of its volume of ether. The precipitate contains all the starch, which may be converted into sugar in the usual way.

Salicylic Acid Method.—Both salicylic acid and benzoic acids dissolve starch; and on heating the solution, the starch is converted into sugar. This is a good method in all cases in which the solution thus obtained can be sufficiently clarified for observation by a saccharimeter. 5.376 grms. of the starch are heated for half an hour with 100 c.c. of water and 0.5 gm. of salicylic acid. The solution is clarified by adding a few drops of ammonia or sodic hydrate solution; it is made up to 200 c.c., filtered, cooled, and examined by the polariscope.

Inversion by Oxalic and Nitric Acid.—3 grms. of the material are heated with 100 c.c. of a saturated solution of oxalic acid for one hour. The liquid is cooled and made up to 200 c.c. with 10 per cent. nitric acid, filtered, and the filtrate heated one hour in the water bath, the flask being connected with an upright condenser. The solution is then polarised.

W. E. Stone,³ in an elaborate study of methods for the determination of starch, gives preference to that in which the starch is converted by means of malt extract or diastase, for it appears that although the various processes by which starch (by means of a mineral or organic acid) is converted into dextrose are accurate with pure starches, such processes are often most inaccurate in the presence of the xylans (gum, etc.), substances which, without any starch at all, give sugar indications from 4 to 60 per cent., wheat flour giving results so widely divergent as 59.8 per cent. by precipitations with barium hydrate, and 77.7 per cent. by inversion with hydrochloric acid.

An easy and simple method of determining starch in flour has been suggested by Edwin Douzard.⁴ A gm. of flour is mixed with a small quantity of cold water, then with 35 c.c. of water at 100° C., and the whole kept at 100° C. for 30 seconds; it is then cooled to 48° C., treated with 20 c.c. of a 10 per cent. malt extract solution (or an equivalent quantity of diastase) maintained at 48° C. for 20 minutes, heated to boiling and filtered; the filtrate is made up to 100 c.c., some resolin added, the filtrate polarised, any rotation due to malt extract being, of course, subtracted. The specific rotation is calculated into dextrin and maltose, as at page 138.

¹ *Journ. Pharm. Chim.*, 1890.

² *Chem. Ztg.*, 1890, p. 902.

³ *Journ. Am. Chem. Soc.*, xvi., No. 12.; *Chem. News*, lxx. 308.

⁴ *Chem. News*, 1898, lxxvii., 107, 108.

Microscopical Identification of Starches.

The successful microscopical examination of starches requires practical study, and those who desire to identify them must use all drawings and descriptions as guides merely. It is not easy to preserve starches mounted as microscopical objects,¹ and the analyst is therefore recommended to fit up a little case, in small, wide specimen-tubes, so that he can have at hand a sample of every kind of starch possible to be obtained. These samples should be arranged in the five classes described (pp. 141-144), partly based on Dr. Muter's² classification.

A high magnifying power is not required, save for the very minute starches, such as rice and pepper. For ordinary work a magnifying power of 250 diameters is ample. Dr. Muter's classification of starches was founded on observations with a B micrometer eyepiece and a $\frac{4}{10}$ -inch power.

It is also useful to observe the various samples of starch, and make tables of their dimensions. The proper way to do this is to put the smallest possible quantity of the well-mixed starch on a glass slide, add a droplet of distilled water, cover with a thin glass, take the exact size of all the starches in the field, enumerate them, and work them out into percentages for future reference.

The illumination of starches is to be particularly attended to. The light must strike obliquely through the granules, in order to observe the rings, which are by no means so easily seen as diagrams would indicate.

Polarised light is also useful, especially in the diagnosis of certain starches. Thus, the polarised starch of wheat, when examined in water, exhibits a dull cross; that of jalap, in shape and size like wheat, polarises brightly. Polarised light, in conjunction with a selenite plate, will also be found of great service. Red and green selenites are best, and give a beautiful play of colours with the arrow-roots and potato starch; while the starches of wheat, barley, rice, and oats scarcely show any colour. The whole of the starches of the Leguminosæ are, so far as they have been hitherto examined, likewise destitute of this power of brilliant coloration. A $\frac{1}{2}$ -inch object-glass, with an A eyepiece, will be found better adapted for this method of research than higher powers.

If adulteration in any case has been made out, approximate quantitative results may be obtained by making a standard mixture of the genuine starch with the adulterant found, and then counting the individual grains in the microscopical field. Thus, for example, supposing oatmeal to be found adulterated with barley-starch, and from a preliminary examination the mixture is thought to be 40 per cent., we proceed as follows:—

Pure barley-meal and oatmeal are carefully dried at 100° and mixed so that the mixture is exactly 40 per cent. A few grains of this powder are now rubbed up with glycerine and alcohol into a smooth paste, which is then further diluted to a certain bulk, a drop taken out with a glass rod, and covered with a glass, which is gently pressed down. The number of grains of barley and oat starch are now counted, and their relative proportion noted, and an exactly similar process is applied to the oatmeal in question.³ If proper care be taken to repeat the experiments, the

¹ According to Muter, a mounting medium of 1 part of glycerin to 2 of water preserves the characters of starch longest.

² *Organic Materia Medica*. London, 1878.

³ See a paper by E. L. Cleaver, F.C.S., *Analyst*, January 31, 1877.

result is a near approximation to the truth. If photographs are taken of these mixtures they are always at hand for reference, and much time is saved.

DIVISION I.—STARCHES SHOWING A PLAY OF COLOURS WITH POLARISED LIGHT AND A SELENITE PLATE.

CLASS I.—*The hilum and concentric rings clearly visible, all the starches oval or ovate. The group includes tous les mois, potato, arrow-root, calumba, orris-root, ginger, galangal, and turmeric.*

Tous les mois, or Canna arrow-root, is furnished by the *Canna edulis*, nat. order *Marantaceæ*. The granules vary in diameter from $\cdot 0469$ to $\cdot 132$ mm. [$\cdot 0018$ to $\cdot 0052$ inch]. They present themselves under several forms, the smaller being granular or ovoid, the larger pyriform, whilst the largest granules are flat, oval, and pointed at their extremities. The hilum is annular, eccentric; the rings are incomplete, extremely fine, narrow, and regular. The starch dissolves easily in boiling water; solution of potash causes the granules to swell rapidly, and gives to the hilum and lines remarkable clearness.

Tous les mois can only be confused with the potato; the size is the chief distinction. The granules burst in water at 72° C., and they give a more regular cross when examined by polarised light than those of the potato.

Curcuma arrow-root, which is also called East Indian (though the arrow-root ordinarily sold as East Indian is a *Maranta*), is furnished by the *Curcuma angustifolia*. The granules are elongated triangular, or irregularly oval, flattened, and almost transparent. The normal measurement varies from $\cdot 0304$ to $\cdot 0609$ mm. [$\cdot 0012$ to $\cdot 00238$ inch]. The hilum is eccentric, not very distinct; the concentric rings are clearly visible, and form segments of a circle. The application of heat or a solution of potash deforms the grains in a very irregular manner; they begin to swell about 72° C.

Maranta arrow-root, syn. *Jamaica*, *St. Vincent*, is derived from *Maranta arundinacea*. The granules are somewhat ovoid, flattened, and tending to a triangular shape in the larger, but the smaller may be circular. The concentric layers are always visible and numerous, but not very marked. Nucleus is central, or about $\frac{1}{5}$ eccentric—in some circular, in others linear; from the nucleus a little slit, filled with air, often goes to the edge. Length of granule $0\cdot 010$ to $0\cdot 070$ mm., average $0\cdot 036$ mm. [= $\cdot 00138$ inch]. Tumefaction in water begins at 76° C. The specific gravity of the starch taken in petroleum or benzole is $1\cdot 504$; if dried at 100° , $1\cdot 565$.

Natal arrow-root is probably the produce of *Maranta arundinacea*, the same plant from which *Maranta* itself is derived, but growing in a different climate. The majority of the granules are broadly ovate, but some are occasionally circular. The dimensions are from $\cdot 0327$ to $\cdot 0375$ mm. [$\cdot 00129$ to $\cdot 00148$ inch]. The eccentricity of the hilum ranges between $\frac{1}{3}$ and $\frac{1}{2}$. The laminae appear under water with special clearness, and on this account granules of *Natal arrow-root* have been frequently mistaken for those of the potato.

Potato starch, syn. *Potato arrow-root*.—The starch derived from the potato (*Solanum tuberosum*). The granules vary greatly in shape and size, some being small and circular, others large, ovate, and oyster-shaped. The hilum is annular, and the concentric rings incomplete. In the larger

granules the rings are numerous and distinct. The normal dimensions are $\cdot 06$ to $\cdot 10$ mm. [$\cdot 0024$ to $\cdot 0039$ inch]. The eccentricity averages $\frac{1}{3}$. The granules float on chloroform.

Potato starch is frequently used as an adulterant of the arrow-roots. The most reliable method of examination is careful microscopic observation, but there is also a different behaviour with regard to reagents, viz. :—

(1.) Maranta arrow-root, mixed with twice its weight of hydrochloric acid, produces a white opaque paste, whereas potato starch treated similarly produces a paste transparent and jelly-like.

(2.) Potato starch evolves a disagreeable and peculiar odour when boiled with dilute sulphuric acid, which is not the case with arrow-root.

(3.) An acrid oil may be extracted from the starch of the potato, but not from that of the Maranta.

Ginger.—The granules are variable in shape, but characteristic. The usual form may be described as shortly conical with rounded angles; the hilum and rings are very faint. Measurement about $\cdot 0376$ mm. [= $\cdot 00148$ inch].

The remaining starches belonging to this group are distinguished as follows :—

Galangal granules, skittle-shaped, with faint incomplete rings, an elongated hilum, with a normal measurement of $\cdot 0342$ mm. [$\cdot 00135$ inch].

Calumba.—The starch granules of Calumba are variable in form, most of them are pear-shaped. They have a semilunar hilum, and faint complete rings. The measurement is about $\cdot 0469$ mm. [$\cdot 00185$ inch].

Orris-root.—The starch granules are of a characteristic, elongated, oblong shape, with a faint hilum. Measurement $\cdot 028$ mm. [$\cdot 0011$ inch].

Turmeric has oval, oblong, conical granules, with the rings well marked and incomplete. Normal measurement $\cdot 0376$ mm. [$\cdot 00148$ inch].

DIVISION II.—STARCHES SHOWING NO IRIDESCENCE, OR SCARCELY ANY, WHEN EXAMINED BY POLARISED LIGHT AND SELENITES.

CLASS II.—*The concentric rings all but invisible; hilum stellate. To this group belong the starches of the bean, pea, maize, lentil, dari, and nutmeg.*

The nucleus of the Leguminosæ is seen usually as a long, more or less stellate, air-filled black hollow. The concentric layers are recognisable if the starch is treated with chromic acid.

The starch from the *bean*, *pea*, and *lentil* are in shape oval, oblong, and almost identical; but both the bean and pea have a stellate hilum, whilst that of the lentil is a long depression. The granules of the bean are of two kinds, large and small; the large are fairly uniform in size, averaging $\cdot 0343$ mm. [$\cdot 00135$ inch] and are bean-shaped, the small are nearly round; those of the pea are variable in size, ranging from $\cdot 0282$ to $\cdot 0177$ mm. [$\cdot 00111$ to $\cdot 0007$ inch], the smaller size predominating. The lentil granules average $\cdot 0282$ mm. [$\cdot 00111$ inch]. The granules of the *nutmeg* are of small size and of characteristic shape. Measurement not exceeding $\cdot 012$ mm. [$\cdot 00047$ inch]. The starch from the *dari* is in small elongated hexagons; average size $\cdot 0188$ mm. [$\cdot 00074$ inch]. The starch from *maize* varies in shape from round to polyhedral; the granules are the same size as those of the *dari*; the distinguishing mark is the rounded angles of the polygonal granules.

CLASS III.—*Starches having both the concentric rings and hilum invisible in the majority of granules. This important class includes wheat, barley, rye, chestnut, acorn, and a variety of starches derived from medicinal plants, such as jalap, rhubarb, senega, etc., etc.*

Wheat starch is extremely variable in size, being from $\cdot 0022$ to $\cdot 052$ mm. [$\cdot 00009$ to $\cdot 0019$ inch]. The granules are circular, or nearly so, and flattened. Polarised light shows a cross, but in water the effect is not great.

Barley.—The granules of barley are of fairly uniform size—viz., $\cdot 0185$ mm. [$\cdot 00073$ inch], but a few measure $\cdot 07$ mm. The shape of the starch is that of slightly angular circles.

Rye.—Rye starch is similar in shape to barley starch. The measurements are from $\cdot 0022$ to $\cdot 0375$ mm. [$\cdot 00009$ to $\cdot 00148$ inch]. The small granules are perfectly round, and here and there cracked.

Chestnut.—The starch grains vary much in form; they are round or elliptical, or three- or four-angled, with the angles rounded. In the place of a nucleus there is almost always a central hollow filled with air. The size is small and regular, being from $\cdot 0022$ to $\cdot 022$ mm. [$\cdot 00009$ to $\cdot 0009$ inch], and this regularity of size is the chief means of distinction.

Acorn.—The starch granules of the acorn are almost round, or round-oval. A nucleus may be made out after treatment with chromic acid; eccentricity $\frac{1}{4}$. Normal measurement $\cdot 0188$ mm. [$\cdot 00074$ inch].

CLASS IV.—*All the granules truncated at one end. This class includes sago, tapioca, and arum, besides several drugs—viz., the starches from belladonna, colchicum, scammony, podophyllum, canella, aconite, cassia, and cinnamon.*

Sago.—A starch obtained from the pith of certain species of palms, especially *Sagus levis* and *S. Rumphii*. It exists in commerce as *raw* and as *prepared* sago; both have oval-ovate granules, the normal measurements of which are from $\cdot 0282$ to $\cdot 0660$ mm. [$\cdot 00111$ to $\cdot 0026$ inch]. There is a circular hilum at the convex end of the raw sago granules, and rings are faintly visible; but starch granules from prepared sago have a large oval or circular depression, covering nearly one-third of each granule.

Tapioca is a starch furnished by the *Manihot utilissima*, which is more or less altered by heat, having been dried on hot plates. This causes some of the granules to swell, and thus renders indistinct in some cases the original structure. The starch is in groups of two to eight, or in isolated granules. When resting on its flat surface, the granule shows a little circle, and round this is a broad flat zone; but if resting on its curved surface, the granule shows contours varying from a kettle-drum to a sugar-loaf shape; and it can then be recognised that the nucleus does not lie in the centre, but in the axis of the granule, and always nearer to the curved than to the flat surface. A conical hollow exists under the nucleus, filled with a substance slightly refracting light. The normal measurement is from $\cdot 0140$ to $\cdot 01879$ mm. [$\cdot 00055$ to $\cdot 00074$ inch].

Arum starch, sometimes called *arum arrow-root*, has somewhat smaller grains than tapioca; they are truncated by two facets; the hilum is eccentric. The normal measurement is about $\cdot 014$ mm. [$\cdot 00056$ inch].

CLASS V.—*In this class all the granules are angular in form; it includes oats, tacca, rice, and pepper, as well as ipecacuanha starch.*

Oat Starch or Meal.—The starch of the oat is mostly polyhedral, being irregularly from three- to six-sided—.0044 to .03 mm. [.00017 to .00118 inch]. The principal starch with which it has been found adulterated is barley; but great caution must be used, for oatmeal contains little round masses extremely similar to barley.

Tacca arrow-root, also called *Tahiti arrow-root*, is extracted from the *Tacca Oceanica* and *pinnatifida*. The granules, when viewed sideways, are muller-shaped, with truncate or dihedral bases; when seen endways they appear circular, occasionally angular or polyhedral; sometimes a sort of contraction gives them a subpyriform appearance. The hilum is well developed, often starred. The normal measurement is from .0094 to .0190 mm. [.00037 to .00075 inch]. It may be confused with maize starch, but tacca has sharp angles; maize, rounded.

Rice Starch.—Each individual grain is polygonal, mostly five- or six-sided, here and there three-sided. If a high magnifying power, such as $\frac{1}{2}$ or $\frac{1}{12}$, be used, a starred hilum may be seen. The normal measurement is from .0050 to .0076 mm. [.0002 to .0003 inch].

Pepper.—The starch of pepper is in small polygonal granules, each of which, with a high magnifying power, is seen to possess a hilum. The normal measurement is from .0050 to .0005 mm. [.0002 to .00002 inch].

§ 81. Vogel has given the following table to assist in the diagnosis of different starches:—

A. Granules, Single throughout, bounded by Rounded Surfaces.

I. Nucleus central, layers concentric.

(a.) For the most part round, at the side lens-shaped.

Nucleus round or a radiating slit.

(1.) Large granules, 0.0396 to 0.0528 mm. [.0015 to .002 inch]—**RYE STARCH.**

(2.) Large granules, .0352 to .0396 mm. [.0013 to .0015 inch]—**WHEAT STARCH.**

(3.) Large granules, .0264 mm. [.001 inch]—**BARLEY STARCH.**

(b.) Egg-shaped, kidney-shaped, mostly a long, often a ragged slit; diameter of starch, .032 to .079 mm. [.0012 to .003 inch]—**LEGUMINOUS STARCHES.**

II. Nucleus eccentric, rings markedly eccentric or meniscus-shaped.

(a.) Granule not flattened, or only slightly.

(1.) Nucleus mostly at the smaller end, .06 to .10 mm. [.0023 to .0039 inch]—**POTATO STARCH.**

(2.) Nucleus mostly at the broad end or towards the middle, .022 to .060 mm. [.0008 to .0023 inch]—**MARANTA STARCH** (W. India arrow-root).

(b.) Granule more or less markedly flattened.

(1.) Many of the granules drawn out more or less at one end into a short point near the nucleus; at the most, .060 long [.0023 inch]—**CURCUMA**; at the most .132 mm. [.0041 inch]—**CANNA.**

(2.) Many lengthened into a disc, bean, or club-shaped form; nucleus near the broader end, .044 to .075 mm. long [.0017 to .0029]—**BANANA STARCH.**

(3.) Many markedly kidney-shaped; nucleus near the edge—**SOUTH AMERICAN ARROW-ROOT** (*Sisyrinchium galaxoides*).

(4.) Egg-shaped, one end thinning into a wedge form, placed one against the other, nucleus at the smaller end, .05 to .07 mm. [.0019 to .0027 inch]—**YAM STARCH.**

B. Starch Granules, Single or Compound. Single Starches with Relation to the Little Granules they are made up of. Bounded by even, many-angled Surfaces, or partly by Rounded Surfaces.

I. Granules throughout many-angled.

(1.) With an evident nucleus, largest $\cdot 0066$ mm. [$\cdot 00025$ inch]—RICE.(2.) Without a nucleus, the largest $\cdot 0088$ mm. [$\cdot 00034$ inch]—MILLET STARCH.

II. Among many angular forms also some rounded.

(A.) No drum-shaped starches present, angular forms predominating.

(1.) Without a nucleus, very small, $\cdot 0044$ mm. [$\cdot 00016$ inch]—OAT STARCH.(2.) With a nucleus, $\cdot 0132$ to $\cdot 0220$ mm. [$\cdot 0005$ to $\cdot 0008$ inch].

(a.) Evident round nucleus, here and there the smaller combined granules in variously shaped groups—BUCKWHEAT.

(b.) Mostly a radiating or star-shaped fissure, none of the granules united—MAIZE.

(B.) More or less numerous drum-shaped to sugar-hat shaped granules.

(1.) Numerous eccentric layers. Largest granules, $\cdot 0220$ to $\cdot 0352$ mm. [$\cdot 0008$ to $\cdot 0014$ inch]—BATATA STARCH.(2.) Without concentric circles, $\cdot 008$ to $\cdot 022$ mm. [$\cdot 0003$ to $\cdot 0008$ inch].(a.) The slit of the drum-shaped particles enlarged towards the flattened side, $\cdot 008$ to $\cdot 022$ mm. [$\cdot 0003$ to $\cdot 0008$ inch]—CASSAVA STARCH.

(b.) Slit wanting or not large.

(aa.) Nucleus small, eccentric, $\cdot 008$ to $\cdot 016$ mm. [$\cdot 0003$ to $\cdot 0006$ inch]—PACHYRHIZUS ANGULATUS.

(bb.) Nucleus small, central, or wanting.

(aaa.) Irregular many-angled forms, $\cdot 009$, $\cdot 008$ to $\cdot 0176$ mm. [$\cdot 0003$ to $\cdot 0007$ inch]—SECHIU M STARCH (Cucurbitaceæ).(bbb.) Only a few angular forms, some with a radiated fissure, $\cdot 008$ to $\cdot 0176$ mm. [$\cdot 0003$ to $\cdot 0007$ inch]—CHESTNUT STARCH.

C. Granules Single and Compound, the Majority Egg-shaped and Rounded with Eccentric Nucleus and Numerous Concentric Layers, the Compound Masses composed of a Large Granule and one or a few very Small Flattened Drum-Shaped Bodies.

 $\cdot 025$ to $\cdot 066$ mm. [$\cdot 00097$ to $\cdot 0025$ inch]—SAGO STARCH.

Karmarsch has determined the maximum size of some of the starches. Karmarsch's values, as well as Wiesner's, are arranged in the following Table (XIV.). They differ in a few instances from the dimensions the author has given at pp. 143, 144.

TABLE XIV.

	Karmarsch.		Wiesner.	
	mm.	inch.	mm.	inch.
General size of potato, . . .	$\cdot 100$	$\cdot 0039$	$\cdot 06$ to $\cdot 10$	$\cdot 0024$ to $\cdot 0039$
Maximum,	$\cdot 185$	$\cdot 0079$
Maranta starch,	$\cdot 140$	0055	$\cdot 01$ to $\cdot 07$	$\cdot 0004$ to $\cdot 0028$
Hogbean, <i>Vicia faba</i> ,	$\cdot 075$	$\cdot 0029$	$\cdot 065$	$\cdot 0025$
Sago,	$\cdot 070$	$\cdot 0027$	$\cdot 033$	$\cdot 0012$
Linseed,	$\cdot 067$	$\cdot 0026$	033 to $\cdot 039$	$\cdot 0012$
Common bean,	$\cdot 063$	$\cdot 0024$	$\cdot 057$	$\cdot 0021$
Pea,	$\cdot 050$	$\cdot 0019$	$\cdot 0283$	$\cdot 0011$
Wheat starch,	$\cdot 050$	$\cdot 0019$
Batata,	$\cdot 045$	$\cdot 0018$	$\cdot 0369$	$\cdot 0014$
Rye,	$\cdot 031$	$\cdot 0012$
Maize,	$\cdot 030$	$\cdot 0012$	$\cdot 020$	$\cdot 0007$
Tapioca,	$\cdot 028$	$\cdot 0010$
Rice,	$\cdot 022$	$\cdot 0008$
Barley,	$\cdot 025$	$\cdot 0009$	$\cdot 0203$	$\cdot 0007$
Millet,	$\cdot 010$	$\cdot 0004$	$\cdot 009$	$\cdot 0003$

WHEAT—WHEATEN FLOUR.

§ 82. The wheat cultivated in this country is the *Triticum vulgare*, of which there are two varieties—the *T. aestivum*, or summer wheat, and the *T. hybernum*, or winter wheat. The mean composition of wheat from 250 analyses is as follows [König]:—

	Per cent.
Water,	13·56
Nitrogenous substances,	12·42
Fat,	1·70
Sugar,	1·44
Gum and dextrin,	2·38
Starch,	64·07
Fibre,	2·62
Ash,	1·79

These analyses do not include Russian wheat. In the central parts of South Russia a wheat is grown which has an amount of nitrogenous sub-

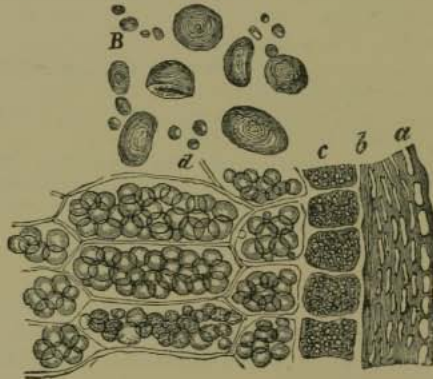


FIG. 26 is a representation of the microscopical structure of wheat when a fine section of the seed is made, $\times 199$. *a* is the cells of the bran; *b* the cells of the thin cuticle; *c* the gluten cells; *d* the starch cells; B, wheat starch, $\times 350$.

stances quite uncommon—the mean of twenty-four analyses giving a percentage of 3·45 nitrogen and 21·56 nitrogenous substances. The mean composition of the ash of entire wheat is as follows:—

	Winter wheat.	Summer wheat.
Potash,	31·16	29·99
Soda,	2·25	1·93
Lime,	3·34	2·93
Magnesia,	11·97	12·09
Ferric oxide,	1·31	·51
Phosphoric acid,	46·98	48·63
Sulphuric acid,	·37	1·52
Silica,	2·11	1·64
Chlorine,	·22	·48

According to the researches of M. Duvivier, of Chartres, the external part of the envelope of the grain is covered with fatty, odorous, and nitrogenous matters, which are in a particular combination, and form on the grain a very tenacious coating—a sort of preservative, and communicating an odour quite *sui generis*. These substances, which are named

cerealin, tritisealine, etc., are isolated by treating the entire grain with cold ether, and then allowing the ethereal extract to evaporate spontaneously. However, neither the nature nor even the presence of these matters can be considered satisfactorily established, and further investigation seems necessary.

§ 83. *Constituents of Flour.*—*Flour*, in the common acceptance of the term, is ground wheat freed from bran.

The *physical characters* which flour should possess are as follows:—It should be an almost perfectly white, fine powder, with only the slightest tinge of yellow, and it should be free from acidity; the odour should be sweet. It should exhibit no trace of bran when pressed smooth by means of a polished surface; and it should have a certain amount of cohesiveness sufficient to retain for some time any shape impressed upon it by squeezing. The microscopical characters of good flour are: the absence of foreign starches, of fungi, etc., and the presence alone of the elements of ground wheat. (See pp. 143, 144, and 145, for measurements of the starches.)

The chemical composition of ground wheat is represented in the following table, giving the mean of fourteen analyses by Peligot:—¹

	Mean of 14 analyses.	Extremes.	
		Max.	Min.
Water,	14.0	15.2	13.2
Fat,	1.2	1.9	1.0
Nitrogenous matter insoluble in water,	12.8	19.8	8.1
Soluble nitrogenous matter—albumen,	1.8	2.4	1.4
Soluble non-nitrogenous matter—dextrin,	7.2	10.5	5.4
Starch,	59.7	66.7	55.1
Cellulose,	1.7	2.3	1.4
Ash,	1.6	1.9	1.4

The percentage of ash from the whole wheat is much higher than that of ordinary flour, the bran containing much ash. Thus, 100 parts of bran contain—

	Per cent.
Water,	13.1
Albumen, coagulated,	19.3
Fat,	4.7
Husk and a little starch,	55.6
Ash,	7.3

The analyses of Millon² and Kekulé give the following:—

	Wheat bran.	
	Millon.	Kekulé.
Starch,	52.0	} 67.3
Gluten,	14.9	
Sugar,	1.0	..
Fat,	3.6	4.1
Wood fibre,	9.7	9.2
Salts,	5.0	5.6
Water,	13.8	13.8

whilst 100 parts of flour, according to Wanklyn, contain—

	Per cent.
Water,	16.5
Fat,	1.2
Gluten, etc.,	12.0
Starch, etc.,	69.6
Ash,7

¹ According to Church, Indian wheat contains from 10.3 to 16.7 per cent. of albuminoids, the average being 13.5 (*Food Grains of India*).

² *Comptes rendus*, xxviii., p. 40.

C. O'Sullivan [*Journal Chem. Soc.*, Trans., xli., 24] has described two varieties of starch found in barley, wheat, and rye, under the names of α and β amylan.

The principles of separation are briefly as follows:—The finely ground grain is freed from sugars, certain albuminoids, and other substances, by alcohol (specific gravity .90), the residue is then treated with water at from 35° to 38° C. for several hours. The solution is filtered and evaporated until it becomes fairly thick. Alcohol of specific gravity .83 to .84 is now added to complete precipitation; the precipitate is purified by repeated treatment with alcohol, and a dry, friable substance obtained which is an impure mixture of α and β amylan.

The substance is purified by (1) treatment with cold water which dissolves out the β amylan, (2) dilute hydrochloric acid which dissolves out traces of mineral matter, and (3) solution in boiling water, filtration, and precipitation by alcohol. By repetition of these processes a snow-white body was obtained, which only gave .3 per cent. of ash, and had the percentages of C, H, and O of a starch.

α amylan is nearly insoluble in cold water, it gelatinises in water, and in 2 per cent. solution forms a clear, jelly-like fluid. A solution containing 1 per cent. of dry substance has a specific gravity of 1.00396, and in a 200 mm.-tube an optical activity at 15.5° C., equal to $[\alpha]_D - 24^\circ$.

It does not reduce alkaline copper solution, and is converted into dextrose without yielding any of the intermediate products furnished by starch. β amylan obtained from the cold water solution of the impure mixture of the amylans, by precipitating by alcohol, and purified on pretty well the same lines as before detailed, is a snow-white body soluble in cold water, and having an optical activity three times as great as α amylan, and yielding by treatment with lime a bi-rotatory body.

Barley contains about 2 per cent. of α amylan, and 0.3 per cent. of β amylan. Wheat and rye contain from 2 to 2.5 per cent. of β amylan, and from .05 to .1 per cent. of α amylan. Malt does not contain α amylan, but a body like β amylan, but more soluble.

Determination of Nitrogenous Matter.

The nitrogenous matter of the cereals has been usually determined by a combustion and subsequent estimation of the nitrogen. Mr. Church has, however, shown that this method of estimation is not perfectly reliable as a measure of the flesh-formers, or albuminous matters properly so called. In a valuable series of researches on this point, with regard to wheat, barley, and oats, Wigner was one of the first to show that these nitrogenous flesh-forming constituents had been over-estimated, since nitrogen combined in other forms than proteid was present in larger quantities than had been hitherto supposed. This is more especially true as regards the husk and bran, very little non-coagulable nitrogenous matter being contained in the flour properly so called. These analyses were carried out as follows:—

50 grains of the sample were ground in a warm porcelain mortar with enough carbolic acid to form a paste. Two or three drops of dilute acid were added, and the paste then diluted with hot carbolic acid and allowed to cool, filtered, and the filter washed with carbolic acid of the same strength. By this treatment all the true albuminoids were coagulated and remained in the filter, while any nitrogenous matter, either as nitrates,

nitrites, alkaloids, or gluten, passed through the filter. The residue in the filter was washed down into the point as far as possible, and the filter dried; the residue detached, and the filter itself finely shredded with scissors and ground to powder, and then burnt in a combustion-tube in the usual way.

The samples were collected so as to give examples of every class of wheat, barley, and oats, and ground by the operator. Treated in this manner, Mr. Wigner found that 17·7 per cent. of the total nitrogen in wheat, 17·6 per cent. of that in oats, and 14·7 per cent. in that of barley, as an average, deduced from the examination of fifteen samples of each, was present in such a form as not to be capable of coagulation by carbolic acid. The extremes in various samples differed widely, and it would appear as though we may have by this process a method of distinguishing the nutritive value of one sample of grain from another.¹ In a research by the senior author, the nitrogen as nitrates in the bran was estimated as ·0071 gm. per cent.; peptone was precipitated by phospho-molybdic acid, and found to exist in the proportion of about ·75 per cent. In whole meal the nitrogen as nitrates was ·043 per cent., the peptone 1·00 per cent.

Weinwurm² has made a similar research by a different process; 10 grms. of the substance were treated with 200 c.c. of water and 0·5 c.c. of acetic acid, the whole warmed twenty minutes on the water bath, the solution cooled, made up to 500 c.c., and in 50 c.c. the soluble nitrogen estimated. Weinwurm erroneously calls this 'amido-nitrogen'; but it is obvious that the solution will contain other forms of nitrogen than amido. The results differ from Wigner's because of the difference in the process.

The vast improvement in modern mills worked with steel rollers separates wheat into nine varieties of flour and three varieties of bran of different degrees of fineness. Weinwurm ascertained the soluble nitrogen in all these qualities, as well as other principles; his results are contained in Table XV. The fineness decreases from Nos. 0 to 9; and it is clear that the finer the flour there is less nitrogen, more starch and less ash. It has also been ascertained that coarse flour is more digestible than fine.

TABLE XV.

	Yield.	Water.	Insoluble N.	Soluble N.	Fat.	Starch, etc.	Fibre.	Ash.
Original wheat,	13·37	10·69	2·93	1·98	80·41	1·90	2·09
Flour, No. 0, . . .	6·0	12·56	8·38	3·06	0·83	87·26	Trace	0·47
" " 1, . . .	14·0	12·54	8·32	3·06	0·92	87·20	"	0·50
" " 2, . . .	6·0	12·48	8·87	2·95	0·97	86·69	"	0·52
" " 3, . . .	4·0	12·50	8·94	2·89	1·05	86·57	"	0·55
" " 4, . . .	5·0	12·50	8·75	3·17	1·10	86·45	"	0·53
" " 5, . . .	6·0	12·48	8·94	3·00	1·15	86·36	"	0·55
" " 6, . . .	4·0	12·39	9·38	3·00	1·17	85·87	0·02	0·56
" " 7, . . .	12·0	12·35	9·82	3·06	1·28	85·01	0·09	0·74
" " 8, . . .	6·0	12·41	10·06	3·22	1·30	84·55	0·06	0·81
" " 8, . . .	5·0	12·40	12·56	2·72	1·91	81·52	0·08	1·21
" " 8, . . .	5·0	11·72	14·34	3·00	3·51	75·90	1·02	2·23
" " 9, . . .	3·0	10·64	15·02	2·55	4·02	74·20	1·55	2·66
Fine bran, . . .	16·0	11·35	13·50	3·06	4·54	63·64	8·71	6·55
Medium fine bran,	2·0	11·55	13·38	2·72	3·96	63·97	9·08	6·89
Coarse bran, . . .	2·0	12·37	13·44	3·17	3·46	62·13	9·79	8·01

¹ See *Analyst*, July, August, 1878.² *Oesterr.-Ungar. Ztschr. f. Zuckerindustrie u. Landw.*, 1890, Heft ii.

A similar more recent research has been made by Professor Snyder¹ of the University of Minnesota, on experiments on bread which necessitated careful analyses of the different forms of flour.

In order to avoid the error inseparable from analysing the varied mill products of different varieties of wheat, various flours were extracted by the process of milling from the same wheat.

Hence the analyses show, with greater accuracy than heretofore, the composition of the several kinds of flour which may be produced by the most modern and improved system of 'milling.'

The explanation of the various terms is as follows:—

First Patent Flour.—This is the highest grade of flour. The gluten having considerable power of expansion, it produces a very white and large loaf.

Second Patent Flour.—Similar to the first, but not quite so white.

Standard Patent Flour.—Ordinary flour as found in the American market, and formed of a mixture of the first and second patent.

First Clear Grade Flour.—This contains slightly more protein than the flours described above, but apparently does not contain the right proportions of gliadin and glutenin to produce so good a quality of bread as the patent flours.

Second Clear Grade or Low Grade Flour.—This, in the process of milling, is only separated in small quantity, about 0.5 per cent. It contains a high percentage of protein, but it does not make good bread, the gluten being of poor quality.

Red Dog Flour.—This consists largely of the germ or embryo and adjacent portion of the wheat, and contains a relatively high proportion of protein. It is distinctly coloured, and produces a small dark coloured loaf.

Middlings or Shorts.—This mainly consists of fine bran.

Bran.—The epispem or outer covering of the wheat.

Entire Wheat Flour.—This must not be considered as denoting literally what its name indicates. A portion of the bran is removed, and then the rest is ground.

Graham Flour.—The entire wheat kernel, bran included, is ground into meal. Hence this might truly have been called 'entire-wheat flour.' The presence of the bran prevents the meal being finely ground; hence it contains many coarse products.

Wheat Ground in Laboratory.—This is the wheat from which the above products were derived. The wheat was a hard Scotch Fife spring wheat, plump and of good quality, weighing 60 pounds to the bushel.

Gluten Flour.—A flour containing as high a percentage of protein as it is possible to obtain by the ordinary roller-process rolling.

The acidity in the Table XVI. is calculated as lactic acid. The value for proteids, it will be observed, has been calculated by using the factor 5.7 instead of the usual one of 6.25. The ash increases from the first patent up to the 'bran,' so that with regard to flours derived from the same sample of wheat the different grades of flour can be determined by a simple determination of the ash.

Chemically the lower grades have a higher nutritive value, but physiologically the experiments of Snyder and others show that the protein of the lower grades is in such an insoluble form that it passes away undigested, and that the higher grade flours are more nutritious.

The nitrogenous constituents of flour comprised under the name of

¹ Studies on Bread and Bread-making at the University of Minnesota in 1899-1900. By Harry Snyder, B.S., Washington, 1901.

gluten are probably, from the researches of T. Weyl and Bischoff,¹ not readily formed in the flour, but the result of the action of a ferment like myosin, which, however, has not been isolated. This theory would account for the fact that (as observed by Mitscherlich and Krocher) wheat in which no sugar could be found before being sent to the mill, on being moistened and ground yielded as much as 4 per cent. of sugar.

TABLE XVI.—COMPOSITION, ACIDITY, AND HEATS OF COMBUSTION OF FLOURS AND OTHER MILLING PRODUCTS OF WHEAT.

Milling product.	Water.	Protein N × 57.	Fat.	Carbo-hydrates.	Ash.	Phosphoric acid.	Acidity.	Heats of combustion.	
								Calculated.	Determined.
First patent flour, . . .	10·55	11·08	1·15	76·85	·37	·15	·08	3·989	4·032
Second patent flour, . . .	10·49	11·14	1·20	76·75	·42	·17	·08	3·992	4·006
Straight or standard patent flour, . . .	10·54	11·99	1·61	75·36	·50	·20	·09	4·022	4·050
First clear grade, . . .	10·13	13·74	2·20	73·13	·80	·34	·12	4·087	4·097
Second clear grade, . . .	10·08	15·03	3·77	69·37	1·75	·56	·27	4·153	4·267
'Red Dog' flour, . . .	9·17	18·98	7·00	61·37	3·48	...	·59	4·349	4·485
Shorts, . . .	8·73	14·87	6·37	65·47	4·56	...	·14	4·219	4·414
Bran, . . .	9·99	14·02	4·39	65·54	6·06	2·20	·23	3·988	4·198
Entire wheat flour, . . .	10·81	12·26	2·24	73·67	1·02	·54	·32	4·026	4·032
Graham flour, . . .	8·61	12·65	2·44	74·58	1·72	·71	·18	4·123	4·148
Wheat ground in laboratory, . . .	8·50	12·65	2·36	74·69	1·80	·75	·18	4·114	4·140
Gluten flour, . . .	8·57	16·36	3·15	70·63	1·29	...	·14

Gluten is composed of at least four bodies—*gluten-casein*, *gluten-fibrin*, *mucedin*, and *gliadin*.

Gluten may be obtained by merely kneading the flour into a paste, and then washing all the starch out of the paste in a thin stream of water. For this purpose a rose with very fine holes may be fixed to a water tap, the flour made into a paste with water, the paste spread out on a rather fine hair sieve, and the streams of water made to play upon the paste, which is gently kneaded. When all the starch is washed out, the effluent runs almost clear. As thus obtained, it is, in the moist state, a yellowish-grey, very elastic, adhesive mass; and when dry, somewhat horny. It dissolves for the most part in alkaline liquids and in acetic acid. From the gluten the four bodies mentioned may be separated as follows:—

1. *Gluten-casein*.—The well-washed gluten is digested a few days with potash solution (for every 100 grms. of gluten about 3 to 4 grms. KHO). The clear solution is decanted from the insoluble residue, and precipitated by acetic acid in the least excess. The precipitate is exhausted successively with 60 per cent. and with 80 per cent. alcohol, then with absolute alcohol, and lastly with ether. The insoluble portion now consists of *gluten-casein*, which may be purified by solution of weak potash lye, precipitated by acetic acid, washed with water and alcohol, and dried in a vacuum. It forms a whitish-grey, voluminous, earthy mass, soluble in dilute alkaline solutions, but insoluble in water, whether hot or cold. At 100°

¹ *Ber. der deutsch. chem. Gesellschaft*, 1880, p. 1064.

it soon changes into a modification insoluble in alkaline fluids. Its solution in very dilute alkaline fluids becomes turbid on exposure to the air, and is precipitated in a flocculent condition by the heavy metals. The elementary analysis of gluten-casein gives the following percentages:—Carbon 52.9, hydrogen 7.0, nitrogen 17.1, oxygen 22.0, and sulphur 1.0. On heating gluten-casein with sulphuric acid tyrosin, leucin, glutamine acid, and asparagic acid with ammonia, are among the products.

The gluten-casein of rye seems to be similar to that of wheat [Ritthausen].

2. *Gluten-fibrin*.—The substances remaining in solution from 1 are gluten-fibrin, mucedin, and gliadin. The first is separated by distilling the united alcoholic extracts to one-half, when it separates as a brownish-yellow mass. It may be purified by repeatedly dissolving in a little 60 to 70 per cent. alcohol, from which it separates on cooling. This property is, indeed, characteristic of gluten-fibrin. It forms a tenacious brownish-yellow mass, becoming horny on drying. It is insoluble in cold water; boiling water partly decomposes and changes it into a modification insoluble in alcohol, acetic acid, and potash. The elementary composition of gluten-fibrin, according to Ritthausen, is carbon 54.3, hydrogen 7.2, nitrogen 16.9, oxygen 20.6, and sulphur 1.0.

3. *Mucedin*.—The alcoholic extracts from 2. contain mucedin and gliadin, and are united and evaporated to dryness. The varnish-like residue is treated with ether to remove fat, dissolved in warm 60 to 70 per cent. alcohol, allowed to cool, and filtered from any gluten-fibrin still remaining. The *mucedin* is now precipitated in a flocculent state by strong alcohol, and by a repetition several times of this operation is obtained pure. When fresh, it is a whitish-yellow, slimy mass; when dry, a horny, crumbling mass. It dissolves easily in 60 to 70 per cent. alcohol, but is precipitated by 90 per cent. alcohol. The acetic acid solution is coloured a beautiful violet on the addition of sulphate of copper and potash, and slightly warming.

The mucedin from rye and barley is similar to that of wheat [Ritthausen]. The elementary composition of mucedin is as follows:—

	Wheat.	Rye.	Barley.
Carbon,	54.1	53.6	54.0
Hydrogen,	6.9	6.8	7.0
Nitrogen,	16.6	16.8	17.0
Oxygen,	21.5	22.3	21.3
Sulphur,9	.5	.7

4. *Gliadin*.—Gliadin is obtained by evaporating the alcoholic solution left from 3. It then remains behind as a clear yellow varnish, which is so tenacious that it may be drawn into threads. By treating it with absolute alcohol and ether, it is changed into a friable, lustreless mass. It is easily dissolved by 40 to 80 per cent. alcohol, and this solution is made milky by absolute alcohol and water, turbid by ether. By boiling it is changed into an insoluble modification. If digested with cold water it dissolves, and the solution is opalescent and frothy, giving a precipitate with tannic acid and soda. Gliadin dissolves in dilute alkaline and alcoholic solutions and in acetic acid; on neutralisation, or on addition of salts of the heavy metals, precipitates are formed.

A simpler composition than that just ascribed to the proteids is given by Osborne,¹ who considers the wheat flour proteids to consist of (1)

¹ *Amer. Chem. Journ.*, xv. (1893), pp. 468-471.

albumen, soluble in water and coagulable by heat; (2) a globulin, soluble in dilute salt solution and coagulable by heat; (3) a proteose body, soluble in water and not coagulable by heat; (4) gliadin, soluble in dilute alcohol; and (5) glutenin, insoluble in water, salt solutions, or dilute alcohol. Gluten is a mixture of 60 to 70 per cent. gliadin, and 30 to 40 per cent. glutenin. It is the gliadin which forms the sticky part of the dough and retains the gas, thus making bread light.

W. Mayer has discovered a very important ratio between the total phosphoric acid in wheat and corn generally, and the total nitrogen—1 part of phosphoric acid corresponding to 2 parts of nitrogen; the extreme variations do not appear to be more than from 1:1.83 to 1:2.19. Ritthausen, U. Kreuzler, and Pote, have, however, found that in wheats very rich in nitrogen, the proportion may be—phosphoric acid 1 to 1.31.

ANALYSIS OF FLOUR.

§ 84. The analysis of flour should in all cases be preceded by a careful microscopical examination, combined with measurement by a micrometer in order to detect any foreign starches, etc. Flour in this country is pretty well free from organic admixture; but cases do occur, and have occurred in other countries, in which there has been, from carelessness in the cultivation and reaping, some one or other of the following seeds:—*Melampyrum arvense* [Scrophulariaceæ], *Lychnis githago*, *Lotium temulentum*; or, in bad seasons, 'blighted' and 'ergotised' corn ground up with good corn.

The *Melampyrum arvense*, or *purple cow wheat*, is a not uncommon flower in cornfields. Its structure is unlike that of wheat, and may be discovered by the microscope. A chemical test is as follows:—About 15 grms. of the flour are made into a soft paste with acetic acid, diluted with double its volume of water. The paste is placed in a platinum dish, and the water and acid completely driven off by a gentle heat. The paste on section, should the melampyrum have been mixed with the flour, shows a coloration, violet or purple, according to the quantity.

¶ C. Hartwich found a rye bread which was of a violet colour. The rye flour from which it was made contained no less than 1.6 per cent. of the seeds of the melampyrum. An alcoholic extract of the flour showed an intense green colour, and sulphuric acid gave a blue play of colours. The seeds not only of *Melampyrum arvense*, but also of *M. cristatum*, *Rhinanthus hirsutus*, *Alectorolophus major* and *minor*, *Euphrasia odontidis*, and *Pedicularis palustris*, all give a violet colour to bread, and probably contain the same colouring-matter—rhinanthin.¹

¶ The *Agrostemma*, or *Lychnis githago*.²—The common corn-cockle of our fields is without doubt poisonous, containing a glucoside, 'saponin.' The seeds are in shape not unlike a rolled up caterpillar, and the surface is beset with regular rows of little warty projections. The microscopy of the seed is very characteristic (see fig. 27).

The surface of the testa shows very large (.1 to .6 mm. diameter) thickened cells, forming on the surface branching tubercles, beneath are

¹ *Archiv der Pharmacie*, 217, p. 280.

² The plant belongs to the nat. order *Caryophyllaceæ*, or clove-worts; the flower is large and purple; the stem dichotomous, from 2 to 3 feet high; the calyx is coriaceous, ribbed, with 5 linear lanceolate, constantly erect, patent, very long segments, styles downy, capsula 5-toothed.

two rows of a regular parenchyma resting on a thin epithelial membrane composed of flat cells, most of which exhibit a peculiar striation; next comes the endosperm, composed of ordinary large celled parenchyma, filled with very minute starch granules, and lastly, there is the embryo, which is

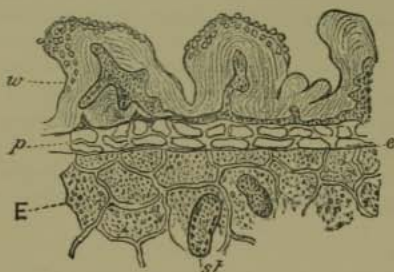


FIG. 27.—A section of the seed of the corn-cockle. *w*, the warty projections of the seed, consisting of an extraordinary thickened and convoluted cuticular layer; *p*, a loose parenchyma; *e*, the colourless outer membrane enclosing the endosperm (*E*) or seed proper; *st*, starch.

in no way peculiar. There are, however, bodies scattered among the endosperm with which every food-chemist should be practically acquainted; they consist of egg-shaped, or spindle-shaped, finely granulated grains, from .02 to .1 mm. in long diameter, and consist of masses of saponin, mucin, and starch. They decompose slowly in cold water, but more quickly on warming and in diluted alcohol; the starch is then set free, and exhibits the usual molecular motion. These bodies are found in other seeds belonging to the Nat. Order of Clove-worts but are different in size. F. Beneke (*Landw. Versuchsstat*, 1885, 6 Heft) has given the follow-

ing as the maximum sizes of those he has observed:—*Spergula* .030 mm., *beta* .057 mm., *spinacia* .64 mm., *agrostemma* .122 mm., hence over .07 mm. points to corn-cockle.

Should the microscope indicate the seeds of *agrostemma*, then an attempt may be made to separate saponin. At least 200 grms. of the flour are exhausted with 80 per cent. alcohol, boiled and filtered hot; the filtrate is now freed from alcohol by distillation, and from fatty matters by shaking up with ether; after separation of the ether, the liquid is concentrated to a small bulk and precipitated with saturated baryta water; if saponin be present, the precipitate will be composed of baryta saponin. This may be collected on a filter, washed with baryta water, and afterwards suspended in a little water, and the baryta separated by passing carbonic anhydride through the liquid.

It has also been noted that flour containing corn-cockle yields a larger percentage of oil to ether, and the ether extract has an acrid taste and is of a pronounced yellow colour.

Saponin ($C_{32}H_{54}O_{18}$?) is a white amorphous powder, with a pungent disagreeable taste; applied to the nostrils it excites sneezing. It is insoluble in ether, in benzine, and in absolute alcohol; it may to some extent be separated from its aqueous solutions by chloroform; it is soluble in water, and a very characteristic feature of the solution is that it froths like a solution of soap; solutions as weak as 1 per 1000 exhibit this peculiarity. Saponin is precipitated by basic acetate of lead and in concentrated solution, as above detailed, by baryta water. Saponin is levo-rotatory $[\alpha]_D = -7^{\circ}30$. Concentrated sulphuric acid dissolves saponin with first a red-yellow colour, changing into violet, and later into a more intense red. According to Schiaparelli¹ saponin has a remarkable power of dissolving salts insoluble in water, *e.g.*, a boiling aqueous solution will dissolve barium carbonate up to 10 per cent. 2 grms. given to an adult

¹ *Gazzetta Chimica Italiana*, xiii., 422-430.

will cause physiological symptoms, mainly consisting of nausea, diaphoresis, and diuresis; .5 gm. is a fatal dose for a kitten; the fatal dose for a human adult is not known.

The *Lolium temulentum*, or *Darnel*, has been found as an impurity in flour. Darnel cannot be identified microscopically by the starch granules, for they are not sufficiently characteristic; should, however, any of the husk be found, it may be readily distinguished from wheat, but possesses some similarity with that of the oat. The best distinction is that the darnel bran has lancet-shaped hairs, while those of the oat are more in shape like the upper bill of the vulture.

If flour contain a considerable quantity of darnel the characters of the alcoholic extract assist the diagnosis. The flour is digested in alcohol of 35%; if it is pure, the alcohol remains perfectly clear and limpid, or at the most takes a very pale-straw colour, from dissolving a little colouring-matter in the envelopes of the wheat, which may be in the flour, nor is the taste disagreeable. If, on the contrary, it contains darnel, the alcohol takes a greenish hue, which darkens gradually. The taste of the alcohol is acrid and nauseous, and on evaporation it leaves a yellowish-green resin.

Detection of Ergot in Flour.—It is most important to examine flour for grain matters damaged by mould, and especially ergot. A good preliminary test is that recommended by A. Vogel.¹ The flour is stained with aniline violet, and then examined microscopically: the damaged starch granules take up the colour intensely. This staining will take place with flour damaged by any fungus, and is not a special test for ergot. The best chemical method is that of Jacopy, as modified by J. Petri.² 20 grms. of the flour are placed in a proper exhausting apparatus, such as is described at page 49, and exhausted with boiling alcohol until the last alcohol is colourless. To the alcoholic solution 20 drops of cold diluted sulphuric acid are added, and the liquid is filtered and examined by the spectroscope in thinner or thicker layers, according to the depth of colour. If the flour is ergotised, the alcoholic solution will be more or less red, and show two absorption-bands in very dilute solution, one lying in the green near E, and a broader and stronger band in the blue between F and G. On mixing the original solution with twice its volume of water, and shaking successive portions of this liquid with ether, amyl-alcohol, benzine, and chloroform, the red colour, if derived from ergot, will impart its colour to each and all of these solvents. Other tests have been proposed from time to time; as, for example, a yellow colour developed when flour is treated with an alkaline solution, and the development of a smell of trimethylamine when the potash solution is heated. It may, however, be remarked that the yellow colour would not be conclusive as to the presence of ergot, for otherwise damaged flour will show this reaction; and as for the smell of trimethylamine, it may be noticed when certain gummy matters are decomposing, and though a flour producing such an odour with potash cannot be considered healthy, the odour in itself would not be conclusive in regard to the presence of ergot. The presence of ergot, and of the seeds mentioned, must be regarded, not as adulterations in the sense of a fraudulent addition, but as impurities. Nevertheless, an analyst would naturally certify, under the 'Sale of Food and Drugs Act,' if flour sold as good flour should be found to contain any of these substances.

The following substances have been fraudulently added to wheaten

¹ *Chem. Cent.*, [3 E.] x., 559.

² *Zeitschrift für anal. Chem.*, 1879, pp. 211-220.

flour:—Rye, rice-meal, barley-meal, potato starch, the flour from various Leguminosæ, linseed-meal, buckwheat, and some other starches.¹ It may once again be said, that in England all these adulterations of flour are of extreme rarity (with, perhaps, the exception of potato flour and ground rice); but there is good evidence that in times of scarcity, with bread at famine prices, all kinds of substances have been mixed with flour and sold. What has happened may occur again, and it is therefore well to know the chief chemical tests which have been recommended to detect even these uncommon admixtures. The general test recommended by A. E. Vogel may be useful: The suspected flour is extracted with 70 per cent. alcohol, to which hydrochloric acid has been added, in the proportion of 5 per cent. of the alcohol employed. If the flour is made of either pure wheat or rye, the alcohol remains colourless; it is of a pale yellow if either barley or oats should be present; orange-yellow with pea flour; purple-red with mildewed wheat; and blood-red with ergotised wheat.

Potato Starch.—So long ago as 1847, M. Donné proposed an excellent test for potato starch in wheat flour. The flour is examined in a very thin layer under the microscope, in the ordinary way, and then, while it is under observation, a weak solution of potash is added, when potato starch will begin to swell, and reach four or five times its volume, while wheat starch is scarcely affected. The test is best applied by putting a little of the flour on a stage micrometer; it is then easier to appreciate the alteration in size of any particular starch. When this method of detecting potato starch is combined with the subsidence process, proposed by Lecanu in 1849, so small a quantity as one part of potato starch in a thousand of wheat flour may be detected. The subsidence process is as follows: Any convenient quantity of flour, say 100 grms., is treated with 40 per cent. of its weight of water, and the gluten separated in the usual way; the washing water is well stirred, and passed through a sieve to retain the larger suspended matters, and then allowed to rest in a conical vessel until a deposit has formed. Without waiting for the supernatant water to become clear, it is decanted, and the deposit mixed by stirring with more water, and allowed again to deposit for a short time. The water is decanted, and the process again gone through. By this means the final and lowest deposit will consist almost entirely of potato starch, which, being of greater specific gravity than wheat, always subsides first. M. Robine, curiously enough, relies more upon a chemical identification of the apex of the cone of deposit than upon a microscopical, which latter is so much more decisive. The last deposit is recommended to be received on a lump of dry plaster, the apex cut off and triturated in an agate mortar—glass, porcelain, and Wedgwood mortars do not answer—and tested with iodine, which gives a blue colour with potato starch; but under these circumstances, not with wheat starch, the friction of the smooth agate not having been sufficient to tear the envelopes off the latter. M. Chevallier has also recommended a method for the detection of potato starch, based on the resistance which the wheat granules possess to the destruction of the outer membrane. Equal weights of flour and sand are

¹ To this list 'sawdust' may be added, a chemical test for which has been suggested by G. A. le Roy (*Ann. de Chim. anal.*, 1899, iv., 212-221), 1 gm. phloro-glucinol dissolved in 15 c.c. of 90-95 per cent. alcohol and 10 c.c. of phosphoric acid syrup. 1-2 c.c. of the above reagent is placed in a porcelain dish and triturated with a little of the sample. Sawdust assumes a rose tint, gradually becoming carmine. The cellulose of the flour only becoming coloured subsequently and then only slightly.

to be triturated with water until a homogeneous paste is formed, which is then diluted and filtered; to the filtrate is added a freshly-prepared solution of iodine, made by digesting for about ten minutes 3 grms. of iodine in 60 c.c. of water, and then decanting. If the flour is pure, this addition will give a pink colour, gradually disappearing; whilst if potato starch should be present, the colour is of a dark purple, only disappearing gradually; by comparing the reaction with flour known to be pure, this difference of behaviour is readily appreciated.

Detection of Leguminous Starches, etc.—As previously stated, the leguminous starches give no play of colours when examined by polarised light and a selenite plate, and are thus easily detected among the iridescent wheat starches. By treating the flour also under the microscope with a solution of from 10 to 12 per cent. of potash,¹ it is possible to dissolve the starch granules of the leguminous plants, and leave a characteristic reticular tissue, made up, for the most part, of irregular hexagons. The addition of lentils or vetches, on account of the brown colour of the seeds, can only take place in minute quantity, and then could only be added to dark flours of inferior quality.

Bean flour, haricot flour, or pea flour, may be mixed up to 5 per cent. without imparting any particular appearance, odour, or taste. Beyond that, all these characters are altered. Bean flour is said to give to the crust a more golden brown, which is agreeable to the eye. There is a principle in beans and vetches which, treated with nitric acid and ammonia, gives a red colour. The method to separate this colouring-matter from falsified flour is to exhaust any convenient quantity with boiling alcohol, and to evaporate the alcohol to a syrup. This syrup is freed from fatty matters by ether, and the insoluble residue exposed successively to the vapours of nitric acid and ammonia. An amaranth red colour denotes the presence of these substances. M. Biot has, however, stated that wheat from the Caucasus responds to this test although perfectly pure, so that, like many other reactions, it must not in itself be taken as conclusive. M. Marten proposed to separate *legumin*, and M. J. Lemenant des Chenais has modified Marten's original process as follows:—The gluten is separated in the usual manner, and to the liquid containing the starchy matters is added ammonia, which is a good solvent of legumin. The starch is allowed to deposit, the liquid is filtered, and to the filtrate a very dilute mineral acid is added, which precipitates legumin if present. The legumin is filtered, collected, dried, and weighed. According to M. Lemenant des Chenais, 9 of legumin in 100 grms. of flour represents an adulteration of 5 per cent.

The most scientific process, which embraces a fairly complete examination of flour for the leguminous constituents, is that of Lecanu:—The gluten is first separated in the usual way. The washing water, containing starch, soluble matters, and legumin, if present, is passed through a sieve to separate coarse particles in suspension, and then diluted sufficiently and allowed to deposit. The liquid is divided into two parts, and one part is allowed to putrefy or ferment spontaneously. With pure flours the lactic acid fermentation is most common; with flours containing legumin there is a putrid fermentation. The other portion is, after decantation

¹ A 1·8 per cent. solution of potash may be used for the detection of maize starch in flour. 100 mgrms. of flour are treated with 10 c.c. of such a solution for 2 minutes in a test tube and then nearly neutralised with dilute HCl; wheat starch is gelatinised, maize remains intact.

and filtration, concentrated until a yellowish scum forms on the surface ; it is allowed to cool, and separated from the albumen which all flours contain. Then legumin is precipitated by adding drop by drop acetic acid. Legumin is identified by its properties. It is without colour, taste, or odour. When dried it is of a horny consistence, insoluble in alcohol, not coloured by iodine, but very soluble in potash or ammonia water, from which solution it may be precipitated by the addition of an acid. The deposit is submitted to a careful microscopical examination, and tested with iodine to colour the starch and leave uncoloured the cellular tissue, or with potash in the way described on page 157. The suspended particles on the sieve are also examined microscopically, because they often contain large fragments of leguminous cellular tissue.

The leguminous starches contain more mineral matter than wheat flour—for example, pea flour contains, on an average, 2.65 per cent. of ash ; flour, .7. It hence follows that if pea flour be mixed with wheat flour in the proportion of 10 per cent., the ash would be .87 instead of .7, and it has been proposed to make this a test of the presence of such foreign starches, but, as the example just given shows, with moderate adulteration it would not be at all conclusive, and must only be considered one of the auxiliary means.

M. Rodriguez has ascertained that when pure flour is submitted to dry distillation in a stone retort, and the distillate is collected in a vessel containing water, the latter will remain perfectly neutral. But if bean, pulse, or pea meal has been added, the water will have an alkaline reaction. This test appears of doubtful value ; for, provided the distillate is alkaline, the alkalinity may, it is evident, have arisen from a variety of causes besides the addition of the substances mentioned. It has also been shown by Bussy that certain cereals yield on distillation an acid product.

Lassaigue (taking advantage of the fact that haricot beans, as well as beans, contain a tannin in their envelopes) adds a salt of iron, which, with pure flour, gives a feeble straw colour, but mixed with either of the two mentioned, or, of course, with any substance containing tannin, gives various shades, from orange-yellow to very dark green.

§ 85. *Detection of Alum and Mineral Matters generally in Flour.*—The most important test for the detection of mineral substances generally in flour is, without doubt, what is known as the 'chloroform' test—a test which, it would appear, was first proposed by M. Cailletet, a pharmacist of Charleville, in 1869, and was in England brought prominently before the notice of analysts by the researches of Dr. Dupré. The principle of the method is simple and obvious. The chloroform is of sufficient gravity to float the starchy substances and allow the alum, sand, sulphate of lime, or other mineral matters, to sink to the bottom. It, besides, has no very appreciable solvent action on alum, and none at all on the generality of mineral or saline substances. No solution made of sufficient specific gravity, by dissolving salts in water, or any other means, will answer the same purpose as chloroform, because, directly the flour is moistened with water, most of the alum is decomposed by the phosphate of potash present in the flour ; alum also forms an insoluble compound with the gluten.

The method is as follows :—The tube figured in the article on 'Beer' is taken, and a weighed quantity of the flour, from a quarter to half a pound, is placed in it, and sufficient methylated chloroform added to form a thin sort of paste ; the cylinder is closed by a stopper, shaken up once or twice, and allowed to stand over night. The next morning the 'rod-

stopper' is inserted and the cap removed; the latter will contain sand from the millstones, sulphate of lime, alum, or any other mineral powder of a greater specific gravity than chloroform, that happened to be in the flour; this fluid is placed in a burette, some more chloroform is added, and the matters allowed again to subside; lastly, the powder, with a little of the chloroform, is drawn off into a watch-glass, the chloroform evaporated, and the powder digested in warm water, the solution filtered into a clean watch-glass, and allowed to evaporate spontaneously. If alum were present crystals will be obtained, and these, if necessary, can be produced in court as a '*corpus delicti*.' Most of the chloroform used may be recovered by filtration, and purified by distillation.¹

The alum crystals may be easily identified by their form under the microscope, and by the reaction with gelatin and logwood. It may be a matter of some importance to be able to say whether the alum present is a potash or ammonia alum. The best method of detecting this is to take the smallest crystal, and having previously dropped a single drop of Nessler solution on a porcelain slab, stir the crystal into the Nessler; an immediate brown colour and precipitate is produced if the alum was an ammonia alum. Dr. Dupré has made some experiments as to the amount of alum which by this process it is possible to recover. Three mixtures were made, containing respectively 28, 10, and 2 grains of very finely powdered ammonia alum in 100 grains of a pure flour. On separation of the alum by the chloroform test, the residue or deposit obtained from the chloroform was dissolved in cold water, and precipitated by baric chloride, and the sulphate of baryta obtained calculated into ammonia alum; the result was that 27.1, 8.21, and 1.14 grains of alum were respectively recovered, instead of 28, 10, and 2 grains, which must be considered as fairly satisfactory. The sand and silica obtained by the chloroform process will be filtered off, and should be dried and weighed, more especially since there has been found to be a relationship between the silica present and the alumina in flour not existing as alum, but as clay, etc.

The Logwood Test.—A freshly-prepared tincture of logwood becomes blue when alum and certain other salts are added to it; an excellent and readily applied test has been proposed based on this reaction. The process usually adopted for flour is as follows:—

Fifty grms. of flour are weighed out and mixed by the aid of a glass rod with 50 c.c. of distilled water; to this is added 5 c.c. of recently prepared logwood solution, alkalisied by 5 c.c. of solution of ammonium carbonate. If $\frac{1}{10000}$ part of alum is present, the flour will become of a lavender-blue colour instead of pink. An approximate estimate of the quantity may be obtained by having a standard solution of pure alum, 1 gm. to the litre, and adding known quantities to exactly similar emulsions of pure flour, and testing as before with logwood, until an emulsion is obtained of very similar hue to the flour originally tested. If the cold extract gives a blue tint with the logwood test, or if the flour be submitted to dialysis, and the diffusate responds, alum is present as alum, and is not derived from dirt, clay, or from the millstones themselves.² The authors use little strips of gelatin to concentrate the alum on: a bit of gelatin

¹ Emmerling, instead of chloroform, uses a solution of zinc sulphate, 100 grms. of zinc sulphate dissolved in 72.5 grms. of water; such a solution has a gravity of 1.43 (*Zeit. f. anal. Chem.*, 1894, p. 46).

² The millstones are sometimes mended with an alum cement. This circumstance will of course, from time to time, be utilised for purposes of defence.

is soaked in the cold extract of the suspected flour for twelve hours, it is then taken out and steeped in the ammoniacal logwood; if alum is present the gelatin becomes of a beautiful blue colour; and the spectrum shows the shifting of the band as described on p. 73. The same blue colour is produced by the presence of magnesia, and clayey matters may also cause a bluish tint. Nevertheless, if a flour or bread does not respond to this test, it is certain that alum in any quantity is not present; on the other hand, if a blue colour is produced, there is likely to be either an adulteration with alum or some other admixture, and the sample should be more thoroughly examined.

Hermann W. Vogel¹ has shown that salts of alumina and magnesia can be recognised by their influence on the spectrum of purpurine. It is evident that here is a process by which the analyst may be assisted in his diagnosis of the cause of any blue colour imparted to flour. Pure purpurine gives, in saturated solutions, a spectrum extinguishing wholly the blue part. An alcoholic solution diluted until it is of a straw-yellow colour extinguishes the blue only partially, and shows two marked absorption-bands at F and *b* E (see fig. 16, p. 70). A diluted watery solution does not show these absorption-bands, but instead there appears a stronger absorption in the green between F and *b*, a weaker in the yellow from E. This reaction is dependent on a trace of alkali, for it is intensified by ammonia, whilst a slight excess of acetic acid colours the fluid yellow, and then there is only a weak absorption. The solution of purpurine should be prepared from purpurine which has been purified by sublimation, and it should be made very feebly alkaline. To test for alum, it is best to take the deposit from the chloroform process already described, and dissolve it in not more than 1 c.c. of water. 2 c.c. of water are now placed either in a test-tube or a little glass cell, and three drops of a saturated alcoholic solution of purpurine added, and then alkalisied by a drop of fourfold diluted ammonia water. On observing this solution by the spectroscope, it appears as curve No. 12, fig. 16, p. 70. A drop of the alum solution is next added: in dilute solutions two bands gradually appear; in the presence of half a milligramme of alum, the bands appear after the lapse of several minutes. Magnesia presents similar appearances, but is at once distinguished from alum by the fact that the bands are destroyed by the addition of acetic acid.

Proximate Analysis of Flour.

§ 86. The constituents of flour to be determined are—

- | | |
|-------------------------|--|
| (1) Water. | |
| (2) Fat. | |
| (3) Cold water extract. | } |
| | Sugar, gum, dextrin, and β amylan. |
| | Vegetable albumen. |
| | Phosphate of potash. |
| (4) Starch. | |
| (5) Gluten. | |
| (6) Ash. | |

(1.) The *water* is taken in the ordinary way; that is, by weighing carefully about 1 to 3 grms. in a tared dish, and exposing it to the heat of the water-bath until it ceases to lose weight.

(2.) The *fat*, according to the researches of Peligot, must be determined in the *perfectly dry* flour, error resulting in any other case.

(3.) The *cold extract* is obtained by digesting 10 grms. of flour in 500

¹ *Ueber eine empfindliche spectral analytische Reaction auf Thonerde und Magnesia.*

c.c. of water, and filtering and evaporating down 250 c.c. in a platinum dish. According to Wanklyn, 100 grms. of flour yield to water—

Sugar, gum, and dextrin,	Grms.
Vegetable albumen,	3.33
Phosphate of potash,	0.92
	0.44
	<hr/>
	4.69

On igniting the extract, the ash should consist entirely of phosphate of potash. When the weight of the ash is known, it may be dissolved in water, and the quantity of phosphoric acid estimated by titration with uranium solution; and if from this there is any discrepancy between the calculated phosphate of potash and that found, the ash should be carefully examined.

The determination of the sugar dextrin and β amylan may be made by the processes previously described; but it is usually sufficient to obtain merely the weight of the cold extract and the weight of its ash.

A method of estimating the value of flour by the amount of solid matter dissolved by acetic acid has been proposed by M. Robiné, who has taken advantage of the property which acetic acid, when properly diluted, has of dissolving the gluten and albumen, and leaving intact the starchy matters. The acetic acid solution increases in density according to the amount of solid substances it dissolves, and he has constructed an areometer, graduated in such a manner that each degree represents the value of the flour expressed in a loaf of 2 kilogrammes weight. A table is sold with the instrument, and without doubt, although not exact enough for the food-analyst, the process is of some value to the buyer of flour. The areometer is called '*Appréciateur des Farines.*' The acetic acid is diluted until the '*appréciateur*' sinks to 93° on the scale. 24 grms. of flour of the first quality are taken for the assay, but if the flour is of the second quality, then 32 are taken. This quantity of flour is washed successively with six quantities of the acid, each time using 31.25 c.c., and all the time triturating in a mortar. After ten minutes the whole is poured into a vessel, plunged in cold water of exactly 15° C., and allowed to remain at rest for an hour; the liquid is then decanted, and the '*appréciateur*' floated in it. By the number indicated, the number of loaves of bread 2 kilogrammes in weight which 150 kilogrammes of the flour will give is at once seen.

(4.) Determination of the starch may be made by O'Sullivan's method (*ante*, p. 136).

(5.) *The gluten or albuminoids* can only be approximately determined by the washing process described at p. 151; the usual method is to make a careful combustion of a small weighed portion of the flour with cupric oxide, and to measure the nitrogen obtained, and then to multiply the percentage of nitrogen found by the factor, 6.25.¹

(6.) *The ash* is best burned in a muffle, otherwise it is difficult to obtain a perfect ash. It has been proposed to mix the flour with nitrate of ammonia, then to heat carefully, and directly fusion commences, to withdraw the flame. Flour can certainly be burnt up very quickly in this way. If this method should be adopted, it will be necessary for the analyst to ignite a corresponding quantity of nitrate of ammonia in a platinum dish, and see whether any residue is left. Occasionally, nitrate

¹ But probably the factor 5.7 gives more exact results.

of ammonia may be met with which is sufficiently impure to cause an error in analysis. Flour may also be burnt up in a platinum trough in a combustion tube. In this case it is most convenient to begin the combustion in ordinary air, and then to finish in oxygen. A properly burnt flour ash should be below 1 per cent.; if it attains 1 per cent., mineral adulteration is probably present. (The method of estimating alumina and silica in the ash of flour is fully detailed at pages 167-169, and also the relationship between the silica and the alumina.)

Legal Case Relative to Flour.

§ 87. The following brief abstract will show the lines of defence likely to be adopted :—

In the month of February, 1880, the case of a miller summoned for selling adulterated flour was heard at the ECKINGTON sessions. The analyst deposed to having found alum in the proportion of 24 grains to 4 pounds of flour. He obtained the alum as alum by the chloroform process. He shook the flour with chloroform, which was a heavy liquid, the flour floated, and the alum sank to the bottom; it was from what sank that he obtained crystals in the characteristic form of alum; he tasted it, and it had the astringent taste of alum. It gave the logwood reaction such as alum gives. He placed about 30 grains of the flour in the chloroform, and the precipitate was probably one-eighth of a grain. He let the chloroform evaporate, and so obtained the crystals; alum crystallises in octohedra of the cubical system; the alum was in the fragmentary form until water was added to the deposit from the chloroform, and the liquid filtered and evaporated. Silica crystallised in hexagonal prisms, and could not be mistaken for alum, besides, it was insoluble in water. He had made an analysis for the purpose of estimating the quantity of alumina present, and found it was in the proportion corresponding to 30 grains of alum to 4 pounds of flour. On being asked whether clay and dirt might not account for the alumina, the answer was that clay and dirt might be present as a silicate of alumina, but it would be insoluble in water, and would not give the reaction with logwood. The defence was—

1. That the analyst was mistaken.
2. That alum was occasionally used in the mill for filling up the cracks in the stones.
3. That the defendant had made his flour lately from foreign grain on account of the bad quality of English wheat at the time, and there was nothing astonishing in finding 24 grains of alum in such wheat, although perfectly pure and unadulterated.

An analyst was called for the defence, who did not seem to be acquainted with the chloroform test, but had estimated the total alumina. The gist of his evidence was that he could not say positively whether there was alum or not in the flour, and that he thought that so small a quantity of alum as could be separated from 30 grains of flour could not be identified. The matter was then referred to Somerset House, and the Government chemists fully confirmed the presence of alum in the flour.¹

BREAD.

§ 88. The term Bread has been applied to any form of flour made into bread, but that made from wheaten flour can alone be treated of here. Wheaten bread is the flour of wheat made into a paste with water, and the paste is permeated by carbon dioxide, either by adding yeast, which causes a true fermentation with the production of alcohol and carbon dioxide, or the carbon dioxide is added in solution in water to the paste, as in Daughlish's system. The explanation of the bread-making process is not thoroughly worked out in all its details, but the following theory agrees fairly well with what is witnessed. On adding yeast to the dough, it is placed on one side, at a suitable temperature, and allowed to rise; that is, fermentation proceeds, and there is a continual evolution of gas; the starch in some degree becomes changed into sugar, which sugar is

¹ *Analyst*, 1880, pp. 72-86.

decomposed into carbon dioxide and alcohol. The gluten prevents, or rather retards, the escape of the carbon dioxide, and the tension of the warm gas expands little cells, and gives to the bread its familiar light spongy appearance. The alcohol mostly escapes, and although in large bread-making establishments it would seem to be feasible and economical to recover the alcohol, hitherto no really good appliance has been invented for this purpose, the apparatuses which have been tried interfering with the baking of good bread. The outside of the loaf, when placed in the oven, is raised to a temperature of from 210° to 212°, but the crumb is seldom much above 100°. The crust is to some extent caramelised, and, on analysis, shows, as might be expected, very much less water than the crumb. Thus, Rivot found in twenty-one samples of bread from 20.45 to 47.11 per cent. hygroscopic water in the crumb, and 16.40 to 27.44 per cent. in the crust. Tracing one by one the chief chemical changes which the flour undergoes under the influence of the yeast-fermentation and subsequent baking, we consider,

1. *Nitrogenous Matters*.—The soluble albumen and globulin become insoluble, and can no longer be separated from the starch. There is a partial union between the acid and the proteid bodies. There is a decrease of alcohol soluble nitrogen, an increase of water soluble nitrogen, hence probably some of the gliadin is rendered soluble.¹ In the crust there is a partial destruction of the nitrogenous substance. Thus, V. Bibra found—

	Wheaten Bread. Nitrogen per cent.	Rye Bread. Nitrogen per cent.
Crumb,	1.498	1.476
Crust,	1.363	1.293

2. *The Starch*, as already explained, is in part changed into soluble carbo-hydrates, a portion of which is converted by the yeast into carbon dioxide and alcohol, but, allowing for loss by fermentation, from 6 to 8 per cent. of the total insoluble starch is changed to soluble forms. The amount of carbon dioxide which is evolved during fermentation and baking averages 1 per cent. The average amount of alcohol which can be recovered is also 1 per cent., which is less than the theoretical amount (*Snyder*). There are also volatile acids, such as acetic and lactic, produced, and other organic volatile constituents, the nature of which has not been determined.

3. *The Fatty Matters* are in part volatilised.²

4. *The Ash* is not changed, save by the minute proportion of yeast ash which is added to it, an addition quite inappreciable. Further, any salt added by the baker increases a little its weight; but the ordinary method of burning bread volatilises very effectually chlorides of the alkalis, so that the ash of bread is still very small. It has been said that the alcohol escapes, which is true with regard to the bulk of the alcohol. Alcohol, however, has a wonderful property of adhering to organic substances, and Th. Bolas has shown that it can be detected in fresh bread in greater quantities than would *a priori* have been suspected. Thus, he found in six fresh samples of bread the following percentages of alcohol:—

Minimum,221
Maximum,399
Mean,313

¹ Studies in Bread and Bread-Making, by H. Snyder, U.S. Dept. of Agriculture, Washington, 1899.

² Losses in the Process of Making Bread, by L. A. Voorhees, M.A., U.S. Dept. of Agriculture, Washington, 1899.

In two of the samples a week old, he was able to detect .132 to .120 per cent. respectively. On keeping bread, there is a continual loss of water, and it becomes 'stale' from some peculiar molecular change. That this staleness is not due to the loss of water is proved by the simple experiment of re-baking a loaf, when it becomes for the time fresh again, but more rapidly after this process becomes stale and is notably drier. V. Bibra found that a bread cannot be made fresh again if it has lost 30 per cent. of water, but if the loss of water is below that, it then may be freshened by re-baking. V. Bibra found that wheaten bread lost the following percentages of water:—

After 1 day.	3 days.	7 days.	15 days.	30 days.
7.71	8.86	14.05	17.84	18.48

The mean composition of wheaten bread, from a large number of analyses collected by König, is as follows:—

	Minimum.	Maximum.	Mean for Fine Bread.	Mean for Coarse Bread.
Water,	26.39	47.90	38.51	41.02
Nitrogenous substances,	4.81	8.69	6.82	6.23
Fat,10	1.00	.77	.22
Sugar,82	4.47	2.37	2.13
Carbo-hydrates,	38.93	62.98	49.97	48.69
Woody fibre,33	.90	.38	.62
Ash,84	1.40	1.18	1.09

The ash of a properly burnt wheaten-flour loaf seldom exceeds 1.5 per cent., unless adulterated; anything beyond 2 per cent. would be certainly suspicious of a mineral addition. There has recently been an agitation on behalf of 'whole meal bread,' and analyses of the greater richness of such bread in azotised constituents are frequently quoted; but such a question cannot be decided by chemical analysis, or, at all events, by ordinary analysis, in which a few constituents are alone estimated. The question is rather a physiologico-chemical inquiry, and the proper way to solve the problem is to go on the lines of the well-known experiments of G. Meyer. A healthy individual is taken and fed on known weights of the substance experimented upon, and the amount of undigested substance recovered from the fæces is weighed. Meyer thus experimented on—

(1.) Horsford-Liebig bread, which is made without the addition of yeast or leaven, the carbon dioxide being developed by the action of bicarbonate of soda on phosphate of potash.

(2.) Munich rye bread, prepared from rye bread and coarse wheat meal and leaven.

(3.) White wheaten bread.

(4.) North German black bread (*Pumpernickel*) prepared out of whole rye meal, and with the use of leaven.

The amount of dry substance, etc., absorbed in percentages of these different breads was found to be as follows:—

	Dry Substance.	Nitrogen.	Ash.
1. The black bread,	80.7	57.7	3.4
2. Horsford-Liebig bread,	88.5	67.6	61.9
3. Rye bread,	89.9	77.8	69.5
4. White bread,	94.4	80.1	69.8

It is thus shown that of the black bread a person would have to eat very much more than of white bread. The white wheaten bread was nearly all absorbed. That this experiment was not made with whole

wheaten meal is true, but it still unmistakably casts some doubt on the question as to whether whole meal would be more nourishing than pure white flour.

Alterations of Bread by Moulds, etc.—Red, green, orange, and black spots occasionally appear on bread, and there are several instances on record of great damage and loss from such parasitic diseases.

In 1856, in France, M. Poggiale was commissioned to examine 22,000 rations served out to the French troops, the bread of which had turned a bluish-black. The bread had been made of inferior grain, but it also contained an enormous number of bacteria. Rather frequently, also, bread and other foods have been attacked by an orange-red growth, which has been attributed to a fungus, to which has been given the popular name of the *red bread fungus*, its scientific appellation being *Oidium aurantiacum*. The blood-red *Bacillus prodigiosus* has also occasionally caused trouble in bakeries.

The more common moulds of bread are the whitish *Mucor mucedo*, the green *Aspergillus glaucus*, and the black *Rhizopus nigricans*. It has not yet been established that any of the moulds or growths enumerated are in themselves injurious to health; but, as may be expected, they damage the bread, making it deficient in nourishment, and unpalatable.

§ 89. *Adulterations of Bread.*—The adulterations of bread enumerated by writers are sufficiently numerous, but those actually proved to exist are but few. Among organic additions,—rice flour, potatoes, bean flour, and pea flour are usually given; among mineral,—alum, borax, sulphate of copper, sulphate of zinc, chalk, and carbonate of magnesia.

In 1843 and 1847, some bakers in Belgium were convicted of adding sulphate of copper to their bread, and this fraud was repeated some years ago by a baker of Calais. There is, however, no reason to believe that English bakers are addicted to these practices, and, as a fact, no conviction has been obtained save for the use of alum. The detection of rice flour, bean flour, foreign seeds is to be undertaken in the same way as described in the sections on flour, save that here the chemical tests are more useful than the microscopical. It is an extremely difficult thing to detect and identify most starches when they have been swollen by heat and altered by fermentation. The only feasible course appears to be to make bread of flour adulterated with the substance suspected to be present, and examine sections and washings of such bread side by side with similar sections and washings of the suspected bread.¹

§ 90. *Alum in Bread.*²—Alum is added to bad or slightly damaged flour by both the miller and the baker. Its action, according to Liebig, is to render insoluble gluten which has been made soluble by acetic or lactic acids developed in damp flour, and it hence stops the undue conversion of starch into dextrin or sugar.

It will be found that generally the medical profession believes that alum even in small doses acts injuriously on the human animal organism. It is certainly true that a person may be poisoned by taking a sufficiently large dose of burnt alum or of the crystallised solid alum, or even a large dose in concentrated solution. It has also been satisfactorily established by Siem³ that if animals are treated by subcutaneous doses of alumina

¹ An exception may, perhaps, be made to this statement in the case of potato starch, which may be recognised tolerably easily even in bread.

² The use of alum is prohibited by the Bread Act, 6 & 7 Will. IV., c. 37.

³ Paul Siem, *Ueber die Wirkung des Aluminiums und des Berylliums*. Dorpat, 1806.

salts a peculiar nervous condition may be produced, similar to the disease known as bulbar paralysis. On the other hand, it is a question whether in the moderate doses in which alum is taken in pastry or bread, or cakes, the flour of which has been mixed with alum or an alum baking powder, it has the slightest appreciable influence on health. Christoph Schmitz¹ gave a dog for three weeks aluminium acetate mixed with sausage; the dog took in the 21 days 99 grms. of alum acetate, and gained in weight half a kilogram. To the same dog was given for 120 days, each day some 35 c.c. of a solution of aluminium acetate, the total amount taken being equal to 260 grms. of aluminium acetate, and the dog increased in weight 2 kilos. Schmitz himself took, for 31 days, 15 drops of the same solution, the total amount equalling 2.4 grms. of the solid acetate, and failed to find his health in any way affected.

Two young doctors took, for 33 days, 1 gm. of the tartrate of aluminium also without effect. Schmitz carefully examined the urine of his experimental dog, but could not obtain more than feeble and doubtful evidences of a trace of aluminium hydrate, and he comes to the conclusion that aluminium compounds in moderate doses are not absorbed by the human intestine.

We are, therefore, decidedly of opinion that alum in food in reasonable quantities is not injurious to health.

On the other hand, the addition of alum to bread is interdicted by law, and whether alum is injurious or not, does not touch the question of adulteration. Probably, in most cases, unless the purchaser is expressly informed that alum is in such and such a cake or such and such a loaf, its presence should be considered an adulteration; for no one should be unwillingly compelled to take anything concerning which he has a prejudice against, whether that prejudice be founded on just grounds or otherwise. In other words, an alumed article of food will generally fall under section 6, and not be of the nature and quality demanded.

In searching for alum, the crust and the crumb should be analysed separately; for many bakers use for the latter a flour technically called 'cones,' which is strongly alumed, and prepared from a fine species of wheat grown in the south of Europe, mixed with rice. This mixture is used for dusting the kneading trough and kneading boards; in point of fact, for 'facing' the sponge previous to baking it. To search for alum in the crust, there is no other method save burning to an ash, as shortly to be described; but with regard to the crumb of bread, the *qualitative* test is the same as for flour—viz., an ammoniacal tincture of logwood. From 300 to 400 grains of bread are crumbled in distilled water, and a slip of pure gelatin added, and the whole allowed to soak for twelve hours. On dissolving the gelatin in a little logwood, to which its own volume of a ten per cent. solution of ammonium carbonate has been added, if the bread is pure the solution will be reddish-pink; if the bread is alumed, the solution will be blue, and exhibit the spectroscopic appearances described at p. 73. This blue colour is not absolutely decisive of alum, for bread adulterated with magnesia carbonate exhibits the same reaction; but if such a colour is produced, the bread requires further examination.

The authors, in some special researches, have discovered that a certain portion of alum may always be washed out of bread as alum. The following process was employed: the bread is soaked in water for at least

¹ Christoph Schmitz, *Untersuchungen über die etwaige Giftigkeit des Aluminiums*. Bonn, 1893.

twenty-four hours (about 2 litres of water are used to 100 grms. of bread). The bread is separated by means of a sieve, and the mass afterwards pressed in a cloth, ultimate clear filtration being obtained when necessary by aid of the mercury pump. This extract may be concentrated in a platinum dish, and when cooled a slip of gelatin allowed to steep in a portion over night. The gelatin on being stained with logwood will exhibit a blue colour, if magnesia or alum is present. Another portion of the extract is dried and burnt up in the usual way, as in the process to be described, and the phosphate of alumina separated. The phosphate of alumina is now fused with sodic sulphate, the result of the fusion being sodic phosphate and alumina. The sodic phosphate is washed out with water; the alumina boiled with a drop or so of dilute sulphuric acid; to the sulphate of alumina thus obtained, a little solution of ammonia¹ is added, and the whole put in a watchglass to crystallise over sulphuric acid. To obtain crystals in this way is often very difficult, but that alum is really present can be readily proved by the reactions of the solution with reagents. By strictly following these directions, a very small quantity of alum can be detected. In a test experiment in a sample of bread in which 5 grains of alum had been added, it was found possible to obtain 1.5 in aqueous solution.

The *quantitative* method for estimation of the *total alumina* in bread, as originally proposed by Dupré, and slightly modified by Wanklyn, is as follows:—100 grms. of bread are incinerated in a platinum dish, until the ash does not exceed 2 grms. in weight. The ash is then moistened with 3 c.c. of pure strong hydrochloric acid, and 20 to 30 c.c. of distilled water added; the whole is boiled, filtered, and the precipitate (consisting of unburnt carbon and silica) well washed, dried, burnt, and weighed. To the filtrate containing the phosphates, 5 c.c. of strong solution of ammonia are added. If the bread has been alumed, the phosphates now precipitated are those of lime, magnesia, iron, and alumina, of which the latter (*viz.*, phosphate of iron and alumina) are insoluble in acetic acid, so that their separation is easy. The liquid is *strongly acidified* with acetic acid, boiled and filtered, and the phosphates of alumina and iron washed and weighed. Unless the liquid has been acidified sufficiently, phosphate of lime contaminates the precipitate and vitiates the results, so that this is an essential point. The last step is re-resolution of the precipitate in acid, and the estimation of the iron; this is usually best effected by a colorimetric process. A standard solution of metallic iron is made by dissolving a gramme of fine iron wire in nitro-hydrochloric acid, precipitating with ammonia, washing the peroxide of iron, and dissolving it in a little hydrochloric acid, and diluting accurately to 1 litre [1 c.c. = 1 mgrm. of metallic iron]. On now adding to an unknown very dilute solution of iron a known quantity of strong ammon. sulphide, a certain colour is produced, and this colour is exactly imitated in the usual way by a similar quantity of ammon. sulphide and the standard solution, the whole operation being conducted on the well-known principles of colorimetric estimation. The amount of iron in the precipitate being known, it is calculated into phosphate, and the phosphate of iron subtracted from the total weight of the precipitate gives the weight of the phosphate of alumina. From Mr. Wanklyn's experiments it would seem that in the case of bread ash, it is unnecessary to evaporate

¹ It is very easy to form a caesium alum, so that if, instead of ammonia, a solution of caesium chloride be added, the result is the formation of beautiful, well-marked crystals.

the hydrochloric solution to dryness, as is usually done, and that the separation of silica is complete by the method just detailed.

Another perfectly valid way of estimating alumina in bread or flour consists in a modification of the old Normandy process. The bread is burnt up as before, the ash powdered and treated with hydric chloride, diluted with water, boiled, and filtered. The filtered solution is again boiled, and whilst boiling poured into a very strong solution of sodic hydrate, the whole boiled, filtered, and washed. To the filtrate is added a few drops of disodic phosphate, it is then slightly acidified with hydric chloride, and subsequently rendered just alkaline by ammonia. The precipitate is collected, washed, and weighed as alumina phosphate.

The following table will be of use in the conversion of phosphate of alumina into alum:—

Phosphate of Alumina, $Al_2O_3 \cdot PO_5$ Parts.	=	Ammonia Alum, $NH_4Al_2SO_4 \cdot 12H_2O$ Parts.	=	Potash Alum, $KAl_2SO_4 \cdot 12H_2O$ Parts.
1	=	3.733	=	4.481
2	=	7.466	=	8.962
3	=	11.199	=	14.443
4	=	14.932	=	17.924
5	=	18.665	=	22.405
6	=	22.398	=	26.886
7	=	26.131	=	31.367
8	=	29.864	=	35.848
9	=	33.597	=	40.329
10	=	37.330	=	44.810

If it is desired to separate the phosphoric acid, the phosphates of alumina and iron may be treated with six times their weight of sodic sulphate, as before stated. Since, when operating in the usual way, the alumina is not recovered as alum, but as a salt of alumina, it is of importance to know whether alumina is contained in unadulterated flour, and if so in what quantity. It is certain that properly cleansed wheat contains no trace of alumina; but particles of clay from the ground, as well as sand from the millstones, do as a fact get into wheat flour, and there is no second-class flour in commerce which does not contain some small percentage of alumina. It might be expected that this adventitious alumina would have some sort of relationship to silica, for it may be presumed to exist as silicate of alumina. We fortunately possess a few analyses by Dr. Dupré, and an elaborate research of Mr. Carter Bell, which will very fairly settle the question. Dr. Dupré analysed twelve commercial samples of flour, none of which gave any reaction with the logwood test, and the results of the quantities of alumina and silica are as follows:—

	Alumina, Per cent. of Ash.	Silica, Per cent. of Ash.	Ratio.
Minimum,63	3.08	1 : 4.7
Maximum, . . .	3.72	26.91	1 : 7.2
Mean,	1.98	10.4	1 : 5.2

Mr. Carter Bell analysed no less than forty samples of flour, none of which contained alumina as alum, and the following are the main results:—

	Alumina.	Silica.	Ratio.
Minimum,003	.009	1 : 3.0
Maximum,011	.109	1 : 9.1
Mean,004	.034	1 : 8.6

Mr. Carter Bell also analysed thirty-two samples of bread, none of

which gave any reaction with the logwood test; the main results of these analyses are as follows:—

	Water.	Silica.	Iron Phosphate.	Alumina Phosphate
Minimum,	40.30	.010	.0005	.0022
Maximum,	49.50	.039	.0040	.0082
Mean,	45.56	.016	.0018	.0049

In these last researches with relation to bread, the ratio between the silica and alumina is 1 of silica to 7.1 of alumina. If the alumina is translated into alum, the important result is obtained that the number of grains of alumina, if calculated into alum, about equals the silica. Thus, in the mean of the thirty-two samples of flour, the alumina was 4 mgrms. Now .004 of alumina is equal to .035 of ammonia alum, and the silica is .034. Or, again, if the mean numbers of the silica and alumina of the thirty-two samples of bread are taken, there is .016 silica to .019 of the phosphate of alumina turned into alum. This, if calculated on the 4 lb. loaf, would be a little over 5 grains of alum. Hence from these researches it is clear that in cases in which the analyst finds the presence of alum in bread from tests detailed, and then burns a quantity of the bread up, and in the ash estimates the phosphate of alumina and the silica, it will be a perfectly fair calculation, *to allow for every part of silica found one part of alum, and this quantity is to be deducted as natural to the flour in the final calculation.*

TABLE XVII.—INFANTS' FARINACEOUS FOODS.

	Salts.	Fat.	Album- inoids.	Carbo-hydrates.	
				Soluble in Cold Water.	Insoluble in Cold Water.
I. Biscuit foods, made from baked wheat flour and condensed milk:—					
Nestlé's,	2.17	3.67	9.85	41.16	37.85
"	1.85	4.75	10.96	76.08	
"	1.70	...	9.50	78.72	
Gerber & Co., in Thun,	1.45	4.75	13.69	75.72	
Anglo-Swiss,	1.74	5.02	10.33	43.51	33.55
"	1.46	5.44	8.84	48.5	27.95
Giffey, Scheele & Co., in Rohrbach,	1.78	4.34	12.86	47.68	29.94
Faust and Schunster in Gottingen,	1.76	5.83	10.71	48.62	27.59
"	1.85	4.75	10.96	39.12	34.70
II. Other varieties of condensed infants' foods:—					
Gerber's lacto-leguminose,	2 to 3 (0.4 to .05 of SO ₃)	5 to 6	18 to 20	70	to 65
Liebig's children's soup,	1.71	0.82	8.41	48.61	
" lacto-leguminose,	3.01	1.34	20.47	16.25	49.41
French's children's flour, (according to the German patent made of sugar mixed with wheat flour.)	2.00 (.69 PO ₅)	4.26	16.80	71.09	
Ridge's food, (a mixture of various flours.)	1.13	1.95	9.65	8.12	75.47
Dr. Coffin's food, N. Y., (made chiefly from leguminose flours.)	3.02	1.59	17.15	35.12	34.82

INFANTS' FARINACEOUS FOODS.

§ 91. There are a great variety of farinaceous foods in commerce, most of them entirely unsuited to be the food of young infants. A child at the breast is more of a carnivorous than an omnivorous animal; and will digest all kinds of meat-broth, meat itself, and albuminous fluids with comparative ease; but if, instead of the natural milk of the mother, a large amount of starchy and saccharine food is given, so little may be digested that the infant is starved.

Some of the farinaceous foods, like, for example, "Polson's Patent Flour," consisting of the flour of Indian corn, are made entirely from one ingredient; the majority are, however, mixtures of starchy, saccharine, and albuminous powders.

A few examples of these foods are given in the preceding Table (XVII.).

OATS, OATMEAL.

§ 92. Of the various species of oat the *Avena sativa* and *Avena orientalis* are the two chief species now cultivated; but the varieties of



FIG. 28 is a section of the oat, $\times 190$: *a* is the outer layer corresponding to the bran of wheat; *b*, the cells of the inner covering of the seed; *c*, the gluten cells; *d*, starch-holding cells. The starch granules at *B* are multiplied by 350.

these two species, according to soil, method of cultivation, etc., are very numerous.

As met with in commerce, oats consist of the seeds enclosed in their paleæ or husk. The mean composition of the ground oats, or oatmeal, is as follows:—

	Per cent.
Water,	12.92
Nitrogenous matter,	11.73
Fat,	6.04
Sugar,	2.22
Dextrin and gum,	2.04
Starch,	51.17
Fibre,	10.83
Ash,	3.05

The nitrogenous substance is composed of gliadin and plant-casein. The 'gliadin' (according to H. Ritthausen and U. Kreuzler) has a much higher percentage of sulphur than the gliadin of wheat; the sulphur being 1.66 per cent. of the substance, while wheat gliadin has .85 per cent. of

sulphur. The plant-casein has the composition of legumin, but the properties of gluten-casein. Von Bibra also states that oatmeal has 1.24 to 1.52 per cent. of albumen. The oat possesses a greater proportion of fat than other cereals. The composition of the fat is as follows [*König*]:—

	Per cent.
Glycerin,	2.8
Oleic acid,	60.5
Stearic and palmitic acids,	36.7

Hence it follows that there is some free fatty acid.¹ The composition of the ash of oats is as follows:—

	Per cent.
Potash,	17.00
Soda,	2.24
Lime,	3.73
Magnesia,	7.06
Ferric oxide,67
Phosphoric acid,	23.03
Sulphuric acid,	1.36
Silica,	44.33
Chlorine,58

The richness of oats in oily matters and in protein compounds is an explanation of its great nutritive powers.

Adulteration.—The chief adulteration of oatmeal is with barley-meal, and more than one conviction has taken place in this country for quantities of from about 15 per cent. The method of detecting and estimating this adulteration is wholly by microscopical means, and is detailed at page 140. The defence which is to be expected in prosecutions for adulterated oatmeal is, that the barley has become mixed with the oats in an accidental manner. As a fact, genuine samples of oatmeal frequently contain other starches, such as barley, maize, etc. Should the foreign starches amount to above 3 per cent., such percentages have not been yet known to occur save as wilful or fraudulent admixture in oatmeal, and the analyst should not have the slightest hesitation in certifying and letting the case be tried upon its merits.

BARLEY.

§ 93. There are several species of barley under cultivation in this country, all of which may, however, be considered as varieties of the following species of hordeum:—*H. hexastichon*, *H. vulgare*, *H. zeocriton*, and *H. distichon*. It is used as a food in the form of 'barley-meal,' the grain being ground whole, and as pearl barley, the latter being the grain deprived of its coverings and rounded by attrition. Barley-meal in the time of Charles I. almost entirely took the place of wheat as the food of the common people, especially in the north of England. The composition of barley-meal is as follows:—

	Per cent.
Water,	15.06
Nitrogenous substances (albumen, 1.0 to 1.7 per cent.),	11.75
Fat,	1.71
Carbo-hydrates (Raffinose ² from .07 to 1.0 per cent.)	
(O'Sullivan, <i>Journ. Chem. Soc.</i> , xlix., 70),	70.90
Woody fibre,11
Ash,47

¹ There is some free acid, if the amount of glycerin is correct; but the saponification was by lead oxide, which gives a lower percentage of glycerin than when potash is used.

² Raffinose $C_{18}H_{32}O_{16}$ [α]; + 114.7° is a fermentable sugar, without action on

The nitrogenous substances are, it would appear, similar to those of wheat, comprising albumen, gliadin, gluten-casein, gluten-fibrin, etc. The constituents of the ash of barley are as follows:—

	Per cent.
Potash,	20·15
Soda,	2·53
Lime,	2·60
Magnesia,	8·62
Ferric oxide,	·97
Phosphoric acid,	34·87
Sulphuric acid,	1·39
Silica,	27·64
Chlorine,	·93

Barley-meal is used as an adulterant of various foods, but in itself it is little tampered with. The detection of adulterations is mainly microscopical, and the dimensions and appearances of barley-starch are described at page 143.

Barley Bread.—Barley bread, though but little used in England, is

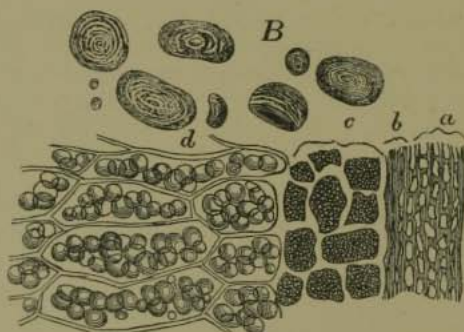


FIG. 29 is a section of barley, $\times 190$: *a* is the layer of cells forming the outer coat of the seed; *b*, the inner; *c*, the gluten cells; and *d*, the starch-holding cells. *B*, barley starch, $\times 350$.

eaten in some parts of the Continent. The mean of two analyses by Von Bibra is as follows:—

	Per cent.
Water,	12·39
Nitrogenous substances,	5·91
Fat,	·90
Sugar,	3·95
Carbo-hydrates,	71·03
Woody fibre,	5·63

RYE.

§ 94. The seed of the *Secale cereale*, in the form of rye-bread, was once a common article of diet in England, and it is now used as the daily bread

Fehling. Dilute H_2SO_4 reduces the plus rotation, and breaks it up into reducing sugars; according to O'Sullivan, the action of sulphuric acid results in substances with a rotation of $+43\cdot5$, and a K. of 81. Raffinose turns yellow at 120° and then reduces Fehling. It may be oxidised by nitric acid into mucic acid.

of the northern European nations. The mean composition of rye flour is as follows:—

	Per cent.
Water,	14·24
Nitrogenous substances,	10·97
Fatty matters,	1·95
Sugar,	3·88
Gum,	7·13
Starch	58·73
Woody fibre,	1·62
Ash,	1·48

The nitrogenous substances in rye are made up of albumen, mucedin, and gluten-casein, but gluten-fibrin and gliadin do not appear to be present. The fat extracted from the rye has, according to König, the following composition:—

	Per cent.
Glycerin,	1·30
Oleic acid,	91·60
Palmitic and stearic acids,	8·10

It, therefore, consists only in part of glycerides, some of the acids being in the free state.¹ The gum, according to M. H. Ritthausen, is soluble in alcohol, and has the ordinary composition of gum.

The ash of the rye-flour has, according to V. Bibra, the following composition:—

	Per cent.
Potash,	38·44
Soda,	1·75
Lime,	1·02
Magnesia,	7·99
Ferric oxide,	2·54
Phosphoric acid,	48·26
Sulphuric acid,
Chlorine,

The composition of fresh rye-bread, according to twenty-seven analyses, from various sources, collected by König, is as follows:—

	Minimum. Per cent.	Maximum. Per cent.	Mean. Per cent.
Water,	35·49	48·57	44·02
Nitrogenous matters,	3·49	9·22	6·02
Fat,	0·10	·83	·48
Sugar,	1·23	4·55	2·54
Carbo-hydrates,	32·82	51·13	45·33
Fibre,	·29	·39	·30
Ash,	·86	3·08	1·31

None of the cereals are so liable to become ergotised as rye. [See *Poisons*, by A. Wynter Blyth.] Roasted rye has been used to adulterate coffee, chicory, and other substances. It furnishes, by appropriate treatment, a good malt for the distillation of spirits, and is used in the manufacture of Hollands.

¹ The fat was saponified by lead oxide. (See the observations in the article on 'Olive Oil'.)

RICE.

§ 95. Rice is obtained from the *Oryza sativa*, and the term is popularly applied only to the seed denuded of husk and inner cuticle, the composition of which is as follows :—

	Per cent.
Water,	14.41
Nitrogenous substance,	6.94
Fat,51
Starch,	77.61
Woody fibre,08
Ash,45

The oil which is obtained from the rice embryo has a density of .924 at 15° C., and at 5° C. becomes thick and buttery ; it contains much olein and an albuminous substance.¹

The composition of the ash of rice is as follows :—

	Per cent.
Potash,	21.73
Soda,	5.50
Lime,	3.24
Magnesia,	11.20
Ferric oxide,	1.23
Phosphoric acid,	53.68
Sulphuric acid,62
Silica,	2.70
Chlorine,10

Rice is said to be adulterated from time to time with other starches, but it is in itself so cheap that it is more likely to be used as an adulterant

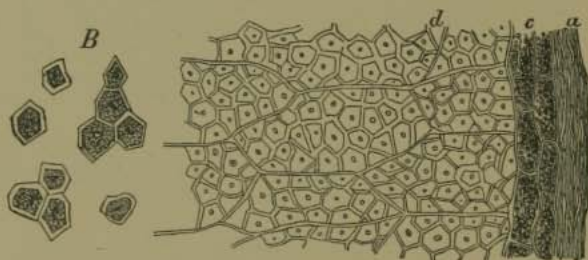


FIG. 30 represents the microscopical structure of rice. The figure is a section of the seed, $\times 190$. α is the outer husk ; c , the gluten cells ; and d , the starch-holding cells. B , the starch cells, $\times 350$.

than tampered with. The microscopical examination of a sample would easily detect any foreign matters. The size and characters of the little granules have been already described at page 144, and are entirely different from all the other starches. A chemical method of detecting the adulteration of rice has been proposed by M. Van Bastelaer. It appears that a saturated solution of picric acid does not cause the least precipitate in a cold watery extract of rice ; but if maize starch, leguminous starches, or other matters be present, there is a more or less abundant precipitate. The quantities recommended are 20 grms. of the powdered rice steeped for an hour in 100 grms. of water, and then the infusion decanted ; to this infusion the picric acid test is applied.

¹ A. Pavesi and E. Rotondi, *Gazetta Chimica Italiana*, iv., 192-195.

MAIZE.

§ 96. Maize, or Indian corn (*Zea Mays*), a native of tropical America, is extensively cultivated in America, Africa, Southern Europe, Germany, and other countries. It is ordinarily met with as the India corn meal of the shops, and forms the basis of many 'infants' foods'; its use appears to be on the increase. According to analyses of A. Riche and A. H. Church, maize has the following composition:—

	A. RICHE. Per cent.	A. H. CHURCH. ¹ Per cent.
Water,	17.1	12.5
Albuminoids,	12.8	9.5
Starch,	59.0	70.7
Dextrin and sugar,	1.5	
Oil,	7.0	3.6
Cellulose,	1.5	2.0
Ash,	1.1	1.7

According to König, the fatty matter of the maize contains 6.46 per cent. of glycerin, 79.87 per cent. of oleic acid, and 16.14 per cent. of



FIG. 31 represents a section of the seed, $\times 190$, and B the starch, $\times 350$. *a* is the outer husk; *b*, the inner; *c* is the gluten cells; *d*, the starch-holding cells.

stearic and palmitic acid. The mean composition of the ash from nine analyses of maize is as follows:—

	Per cent.
Potash,	21.73
Soda,	5.50
Lime,	3.20
Magnesia,	11.20
Ferric oxide,	1.23
Phosphoric acid,	53.68
Sulphuric acid,62
Silica,	2.74
Chlorine,10
	<hr/> 100.00

An aqueous decoction of maize gives with a little iodine a peculiar reddish-purple colour. On placing the iodised decoction in the dark, after some eight to twelve hours the precipitate becomes dirty white, and the supernatant fluid milky; if an excess of iodine is added the precipitate is then red, but it also becomes decolorised in the dark. Maize is said to

¹ The mean of several analyses of whole Indian grown maize. *Food Grains of India*, London, 1886, 65.

be occasionally adulterated with potato starches, etc.; these a microscopical examination will at once detect. M. Genin has given certain chemical reactions, based upon the different hues which mixtures of maize and potato starch assume when treated with iodine, and also a process based on the volume which a precipitate obtained by lead acetate in an alkaline extract occupies in pure and adulterated samples. These processes are, however, far too loose to be of any service.

MILLET.

§ 97. The millet seeds are derived from two species of panicum, *Panicum miliaceum* and *Panicum italicum*. It is extensively used among the Chinese and Eastern races as an article of diet, and its nutritive power is about equal to that of rice.¹ The average composition of millet deprived of its coverings, according to six analyses, is as follows (König):—

	Per cent.
Water,	11·26
Nitrogenous substance,	11·29
Fat,	3·56
Sugar,	1·18
Dextrin and gum,	6·06
Starch,	60·09
Cellulose,	4·25
Ash,	2·31

The composition of the ash of the millet deprived of husk is as follows:—

	Per cent.
Potash,	18·36
Soda,	3·82
Lime,
Magnesia,	21·44
Ferric oxide,	1·82
Phosphoric acid,	44·21
Sulphuric acid,	2·02
Silica,	8·33

POTATO.

§ 98. The chemical composition of the uncooked potato is, according to the analysis of some fair average tubers, as follows:—

	Per cent.
Water,	76·00
Starch,	19·68
Sugar,	1·20
Albumen, ²	·70
Gum,	·40
Asparagin,	·30
Fat,	·30
Solanin,	·05
Other nitrogenous substances,	·15
Insoluble matter,	·40
Ash,	·82

100·00

¹ In the 'Dictionary of Hygiene,' by one of the authors, will be found a remarkable experiment on the nutritive qualities of millet.

² The albumen, according to E. Schultze and E. Euplér (*Landw. Versuchs. Stat.* xxvii., 357), varies in different kinds from ·65 to 1·19 per cent.

A summary of seventy analyses, determining the principal constituents of the potato, is given by König as follows:—

	Minimum.	Maximum.	Mean.
Water,	68·29	82·22	75·77
Nitrogenous substances,	·51	3·60	1·79
Fatty matters,	·05	·80	·16
Starch,	12·05	26·57	20·56
Woody fibre,	·27	1·40	·75
Ash,	·42	1·46	·97

It is thus seen that, according to all analyses, some 95 per cent. of the potato is water and starch. The nitrogen of the potato, which, according to the old method of analysis, would be reckoned into albumen, or, at all events, into protein substance, is derived from albumen, asparagin, solanin, xanthine, leucin, and tyrosin. It has been calculated that about 56 per cent. of the total nitrogen is derived from asparagin and amido acids—a fact which must be remembered in diet calculations.

Besides the constituents enumerated, there are certain organic acids in the potato which may be extracted in small quantities by sulphuric acid and ether. Among these are citric and succinic acids, and possibly the presence of these organic acids in part accounts for the antiscorbutic power possessed by the potato. Siewert has also found from ·017 to ·057 per cent. of oxalic acid.

The composition of the ash of the potato, according to fifty-three analyses by E. Wolff, is as follows:—

	Minimum.	Maximum.	Mean.
Potash,	43·95	73·61	60·37
Soda,	16·93	2·62
Lime,	·51	6·23	2·57
Magnesia,	1·32	13·58	4·69
Ferric oxide,	·04	7·18	1·18
Phosphoric acid,	8·39	27·14	17·33
Sulphuric acid,	·44	14·89	6·49
Silica,	8·11	2·13
Chlorine,	·85	10·75	3·11
Percentage of ash in dried substance,	2·20	5·30	3·77

The potato is very subject to a fungus disease, the life-history of which has been very fully elucidated by various observers, and more especially by Worthington G. Smith. The fungus is named botanically *Peronospora infestans*, and the manner in which it grows, and its method of reproduction, is shown in the annexed woodcut (fig. 32).

The figure represents the very highly magnified section of a potato leaf, and the mycelium of the peronospora growing among the cells. A, A are the natural hairs of the potato leaf; B, B are the upper and lower layers of the healthy cells. The threads and bodies at C, D, E, F, and G belong entirely to the fungus. The fine thread at C is a direct continuation of the spawn or mycelium living inside, and at the expense of the leaf tissue. Emerging into the air, the thread is seen to bear two distinct species of fruit—one, D D, called simple spores or gonidia, while at E F are what are known as 'swarm spores.' The swarm spores, when moistened, set free fifteen or sixteen bodies, known as 'zoo-spores,' so named because they are endowed with spermatozoa-like motion, being furnished with two lash-like tails, which they move with great rapidity. A zoo-spore, when it falls on a leaf, has a surprising power of corroding the epidermis, and entering into the tissue. This action is probably due to some special

solvent secreted by the zoo-spore. When movement ceases, the tails disappear, and a minute

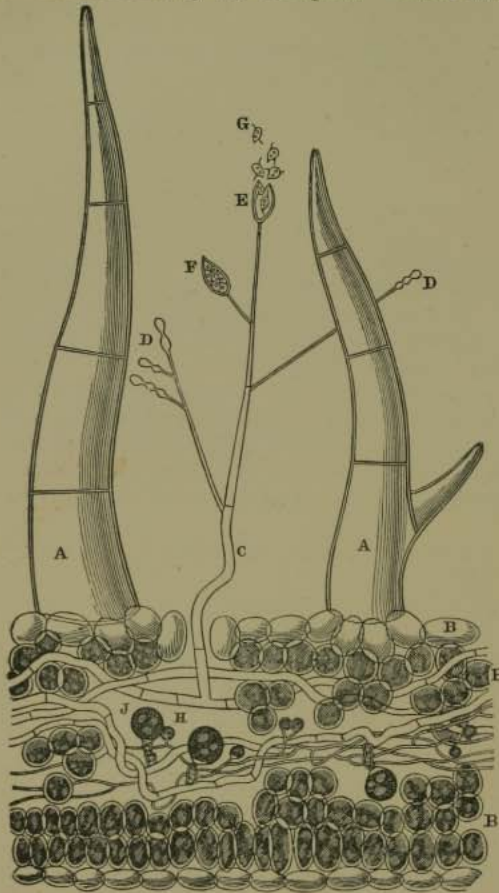


FIG. 32.

thread is protruded at one end, which develops into a network of mycelium. Both of these methods of production are asexual; but there is a third method which is sexual, and the more important, because, in this case, there are structures formed which resist frost and a variety of influences destructive of the former fragile structures. This third method is the production of oospores by the conjunction of two organs — the one named *antheridium*, and analogous to the anther of a flower, and the other *oogonium*, analogous to the ovary of a flower. The oospores are dark-brown in colour, reticulated, and covered with little warty prominences about $\cdot 001$ inch in diameter.¹

Analysis of the Potato.

— Since the potato is mainly composed of water and starch, a careful determination of those substances by the processes

already enumerated will be sufficient for most purposes. For those in

¹ The Peruvians make a national dish from frozen potatoes, which they call 'chuno.' The potatoes are steeped for a little while in water, then exposed for a few days to sharp frost, washed, and rubbed. By this method the peel becomes detached, and the starchy matters are dried in the sun or in an oven. The dried hard tubers are cut in thin slices and baked, and eaten with the addition of Spanish pepper. An analysis by Meisel (*Wagner's Jahresbericht der chemisch. Technol.*, 1881) is as follows:—

	Per cent.	
Water,	13·030	
Starch,	81·844	
Nitrogenous matter,	2·313	Total nitrogen, '4 %. Nitrogen soluble in water, '03 %.
Woody fibre,	1·133	
Fat,	'182	
Ash,	'356	
Soluble constituents in water,	1·142	'400 sugar. '141 asparagin. '601 soluble starch, dextrin, soluble ash constituents, etc.

which great accuracy is not required, it will be sufficient to take the specific gravity of the potato, and then refer to the following table. This table, for fairly good potatoes, will give results of from within .3 to .5 per cent. of the true value; but with regard to tubers poor in starch there may be a much larger error.

TABLE XVIII.—SHOWING THE PERCENTAGE OF STARCH AND DRY SUBSTANCE CORRESPONDING TO VARIOUS SPECIFIC GRAVITIES.

Specific Gravity.	Dry Substance.	Starch.	Specific Gravity.	Dry Substance.	Starch.
1.080	19.7	13.9	1.120	28.3	22.5
1.081	19.9	14.1	1.121	28.5	22.7
1.082	20.1	14.3	1.122	28.7	22.9
1.083	20.3	14.5	1.123	28.9	23.1
1.084	20.5	14.7	1.124	29.1	23.3
1.085	20.7	14.9	1.125	29.3	23.5
1.086	20.9	15.1	1.126	29.5	23.7
1.087	21.2	15.4	1.127	29.8	24.0
1.088	21.4	15.6	1.128	30.0	24.2
1.089	21.6	15.8	1.129	30.2	24.4
1.090	21.8	16.0	1.130	30.4	24.6
1.091	22.0	16.2	1.131	30.6	24.8
1.092	22.2	16.4	1.132	30.8	25.0
1.093	22.4	16.6	1.133	31.0	25.2
1.094	22.7	16.9	1.134	31.3	25.5
1.095	22.9	17.1	1.135	31.5	25.7
1.096	23.1	17.3	1.136	31.7	25.9
1.097	23.3	17.5	1.137	31.9	26.1
1.098	23.5	17.7	1.138	32.1	26.3
1.099	23.7	17.9	1.139	32.3	26.5
1.100	24.0	18.2	1.140	32.5	26.7
1.101	24.2	18.4	1.141	32.7	27.0
1.102	24.4	18.6	1.142	33.0	27.2
1.103	24.6	18.8	1.143	33.2	27.4
1.104	24.8	19.0	1.144	33.4	27.6
1.105	25.0	19.2	1.145	33.6	27.8
1.106	25.2	19.4	1.146	33.8	28.0
1.107	25.5	19.7	1.147	34.1	28.3
1.108	25.7	19.9	1.148	34.3	28.5
1.109	25.9	20.1	1.149	34.5	28.7
1.110	26.1	20.3	1.150	34.7	28.9
1.111	26.3	20.5	1.151	34.9	29.1
1.112	26.5	20.7	1.152	35.1	29.3
1.113	26.7	20.9	1.153	35.4	29.6
1.114	26.9	21.1	1.154	35.6	29.8
1.115	27.2	21.4	1.155	35.8	30.0
1.116	27.4	21.6	1.156	36.0	30.2
1.117	27.6	21.8	1.157	36.2	30.4
1.118	27.8	22.0	1.158	36.4	30.6
1.119	38.0	22.2	1.159	36.6	30.8

PEAS.

§ 99. The pea is, without doubt, the most important of all the leguminous plants. The garden pea is derived from the *Pisum sativum*, a native of the south of Europe, but long naturalised in this country. The field pea, grown for the purpose of feeding cattle, is the *Pisum arvense*.

Forty-one analyses collected by König give the following values:—

	Maximum.	Minimum.	Mean.
Water,	22.12	11.01	14.31
Nitrogenous substance, ¹	27.14	18.56	22.63
Fat,	3.30	.64	1.72
Nitrogenous free extractive matter,	59.38	41.90	53.24
Woody fibre,	10.00	2.22	5.45
Ash,	3.49	1.76	2.65

The 53.24 per cent. of non-nitrogenous soluble matter is composed of 36.03 starch, 5.51 dextrin, and 11.70 other substances, among which is some sugar. Cholesterin is also found in peas, but there have been no researches as to its exact quantity. The most important principle of the pea is 'legumin.' Its amount varies in different species. Thus, H. Ritthausen found in the green field pea, 3.95 per cent.; in the yellow, 9.45, and in the grey, 7.30 per cent.; in the garden pea, 5.40 per cent.

In the young unripe condition, peas contain much more water than the proportions given above. Thus, Grouven found in young unripe peas and beans the following:—

	Green Peas.	Green Beans.
Water,	79.74	91.34
Carbo-hydrates,	13.03	5.99
Albuminoids,	6.06	2.04
Salts,	1.12	.63

The albumen of peas (which may be obtained by boiling the solution after it has been freed from legumin) differs from ordinary vegetable albumen, both in elementary composition and in its behaviour to reagents. An analysis gives the following percentages:—

C, 52.94; H, 7.14; N, 17.14; S, 1.04; O, 21.74.

After coagulation it dissolves in potash water to a clear liquid, which is not the case with vegetable albumen.

The analysis of the ash of peas gives the following as the extremes and mean of twenty-nine analyses:—

	Minimum.	Maximum.	Mean.
Potash,	35.80	51.41	42.79
Soda,	3.57	.96
Lime,	2.21	7.90	4.99
Magnesia,	5.80	13.02	7.96
Ferric oxide,	3.83	.86
Phosphoric acid,	29.30	44.41	36.43
Sulphuric acid,	9.46	3.61
Silica,	3.02	.86
Chlorine,	6.50	1.54

Peas, when putrid, undergo some peculiar change not yet investigated, resulting in the formation of a poison, perhaps similar to the cadaveric poisons.²

¹ Some nitrogen determinations by C. Böhrer (*Versuch. Stat.*, xxviii., 247-262), in which ammonia, amido acids, and other products were differentiated, are as follows:— Total nitrogen in dried peas, 4.69 per cent.; nitrogen as albumen, 3.56 (= 22.5 per cent. albuminoids); nitrogen as ammonia, .020; nitrogen as amido-acid amides, .052 per cent.; nitrogen as amido acid, .361; nitrogen as peptones, .697 per cent.

² Some years ago a case of wholesale poisoning from this cause occurred in Salford. Many persons who had partaken of slightly decomposed peas exhibited symptoms of irritant poisoning. The peas were chemically examined, but contained neither arsenic, copper, lead, nor other metallic poison. [*Pharm. Journ.* (3), 294.] The

For the general analysis of peas, the water, the ash, and the amount of starchy matters are estimated by the processes already detailed. To separate the legumin, the peas must be powdered, or, if fresh, mashed into a paste, and treated with successive quantities of cold water, which may be advantageously feebly alkaline, but must not have the least trace of acid. The legumin may now be precipitated by acetic acid, the precipitate dissolved in weak potash, again precipitated, and then dried and weighed. Legumin is almost insoluble in cold or warm water; but since it may be extracted so easily from the fresh seeds, it is supposed to be in combination with phosphates of the alkalies when in its natural condition. But it is easily soluble in diluted alkaline liquids, and also readily dissolves in a solution of alkaline phosphates; if boiled it becomes insoluble in alkalies. Pure alkaline solution of legumin shows, with a little cupric sulphate, a beautiful violet colour. If impurities are present, such as gum or starch, the colour is blue. On boiling the alkaline solution, the legumin does not coagulate, but, as in the boiling of milk, a scum of altered legumin appears on the surface.

§ 100. *Preserved Peas.*—*Copper in Peas.*—Peas are preserved in several ways, sometimes by simply drying, when they form the well-known dried peas of the shops. But the more modern method is to heat the peas in a suitable tin capable of being hermetically sealed. The sealing is effected while the tin with its contents is at a high temperature. The rationale of the process is, that putrefying germs existing on the surface of the peas are destroyed, and fresh putrefactive agencies are prevented from gaining access by the exclusion of air. Peas so preserved may, as proved by analysis, be quite as nutritious as fresh peas. Preserved peas have often undergone a preparatory treatment by boiling in copper vessels, the object of which is to impart a fine green colour.

The reason why vegetables preserve their green colour when treated by copper is, according to Tschirch,¹ owing to the formation of a compound of copper with phyllocyanic acid ($C_{24}H_{36}N_2O_4$). Phyllocyanate of copper has the composition $(C_{24}H_{27}N_2O_4)_2Cu$, and contains 8.55 per cent. copper; it is insoluble in water, but soluble in strong alcohol and chloroform; it is not soluble in dilute acetic nor in dilute or concentrated hydrochloric acid. Its alcoholic solution gives an absorption spectrum characterised by four bands.

subject of the formation of new and poisonous substances in such an article of food would well repay investigation.

In Germany there has been used a condensed food made up of powdered and dried meat, incorporated with pea-meal, by strong pressure; it is scarcely necessary to say, that in this manner a food invaluable for the soldier is obtained, and one that contains in a very small compass all the essentials of nourishment. An analysis of these pea-meat tablets is as follows:—

	Per cent.
Water,	12.09
Nitrogenous matters,	31.18
Fat,	3.08
Carbo-hydrates,	47.50
Ash,	6.15

A condensed pea soup is also prepared. Two analyses of this condensed soup, given by König, are as follows:—

	1.	2.
Water,	7.58	8.08
Nitrogenous matters,	16.93	15.81
Fat,	8.98	24.41
Carbo-hydrates,	53.44	36.78
Woody fibre,	1.34	1.69
Ash,	11.73	13.23

¹ *Das Kupfer*. Stuttgart, 1893.

Tschirch has shown that although copper phyllocyanate is so insoluble in acids, yet when administered to animals it produces the same effects as equal quantities of copper given as the soluble tartrate; at the same time he considers that so small a quantity of copper added as will produce phyllocyanate only would probably not be injurious to health.

M. Guillemare and M. Lecourt have, however, now patented a process by which chlorophyll has been substituted for the objectionable coppering. The copper that has hitherto been found in tinned peas has amounted to about 2 grains to 2·6 grains in the pound tin, and the question arises whether the copper is injurious to health in this proportion or not. In the prosecutions which have been instituted, it is obvious that men of considerable scientific reputation have expressed strong opinions on the subject; nevertheless, the whole of the injurious action of coppered peas rests entirely on theory, and in no single instance (although the consumption of coppered peas has been very large) has any really definite case been brought forward of actual poisoning by peas coloured in this way. Legrif has found in the intestine of a healthy man ·036 to ·040 gm. of copper;¹ and Messrs. Paul and Kingzett have shown that, even when a soluble compound, like sulphate of copper, is ingested, most of it is excreted by the feces.

Some addition to exact knowledge as to the quantity of copper necessary to impart a green colour to peas, and also as to the action of coppered peas upon the human organism, has been made by Dr. F. W. Tunncliffe² for the Departmental Committee appointed in 1899 to inquire into the use of preservatives and colouring matters in foods.

After actually viewing the process of greening by copper in use by manufacturers, experiments were made by boiling definite quantities of copper sulphate with weighed quantities of peas, and the following results were obtained:—

1 part of copper sulphate boiled with 7950 of peas yielded peas containing on analysis 18·7 mgrms. copper per kilo.

1	part	boiled with	3975 ditto.,	38	mgrms. Cu.
1	„	„	1557 „	44	mgrms. Cu.
1	„	„	1987 „	72	mgrms. Cu.
1	„	„	774 „	90	mgrms. Cu.

So that usually there was some relation between the copper added and the copper absorbed by the peas. Tunncliffe concluded that 50 mgrms. of copper absorbed per kilo. was sufficient to impart a permanent and adequate greenness to peas.

This quantity is half of that permitted by the law of France and Italy (100 mgrms. per kilo.).

Experiments were also made upon a child as to the effect of the ingestion of coppered peas and as to how much of the copper was excreted. Since copper is normally present in other foods, the child was given a uniform diet during the whole period of observation (32 days).

The following table summarises the results obtained:—

A is a period of eight days during which the uniform diet was taken without peas or copper, save what copper may have been in the standard diet.

B. A period of two days, in which fresh uncoppered peas were taken in quantities of 200 grms. per day, in order to eliminate any effect on the intestines of peas as peas.

C. A period of two days, in which 200 grms. each day of peas were commercially greened with copper and previously analysed.

D. A period of two days, in which the same amount of sulphate of copper was added to fresh peas after they were boiled and just before they were eaten. This experiment was devised to ascertain any differences there might be between copper in the form of phyllo-cyanate and albuminate and copper mainly as a mineral salt.

e, *f*, and *g* were subsequent periods of eight, four, and six days respectively, during which no coppered peas were eaten, but in which the excreta were analysed and the copper estimated.

¹ Sonnenschein.

² Appendix No. 5, 304, 305. Appendix No. 7, 312, Departmental Committee on Preservatives. London, 1901.

TABLE XIX.—SHOWING THE RELATION BETWEEN THE INGESTION AND EXCRETION OF COPPER DURING THE VARIOUS PERIODS, EXPRESSED IN MGRMS. PER DAY.

	Mgram. Cu excreted.		Total excretion.	Total ingested with peas.	Mgram. Cu retained.	Per cent. Cu excreted in feces.
	Fæces.	Urine.				
A	0·29	...	0·29
B	0·72	trace.	0·72	0·35
C	10·79	0·03	10·82	14·0	3·18	77
D	7·45	0·03	7·48	22·06	14·59	34
e	1·42	0·08	1·48
f	0·57	0·08	0·65
g	0·35

The table shows that immediately after the ingestion of the coppered peas the excretion rose, and no less than 77 per cent. was excreted with the fæces, and therefore never entered the body at all. The remaining quantity was probably absorbed into the system, and stored up in the liver. That is to say, the small amount of 3 mgrms. was absorbed into the blood. On the other hand, when the mineral copper sulphate was so mixed with the food that very little of it could have combined with albuminous matters, then the excretion by the fæces was only 34 per cent., while over 14 mgrms. entered the blood.

These experiments, therefore, fully bear out the statements made in former editions of this work, viz., that there must be a distinction drawn between copper in organic combination and copper taken as a mineral salt.

Nevertheless, a majority of the Committee recommended "That the use of copper salts in the so-called greening of preserved foods be prohibited."

Dr. Tunnicliffe, a member of the Committee, dissented from this recommendation, and suggested that a limitation should be placed on the amount, that "the presence of copper in these preserved vegetables be in every case declared, and that its amount be restricted to half a grain of metallic copper per lb."

The method of detecting copper in peas is as follows:—A weighed quantity of the peas is burned to an ash in a muffle, the ash is dissolved in hydrochloric acid, the acid solution evaporated to dryness, and then taken up with nitric acid, filtered, and submitted with the precautions and under the conditions detailed at p. 112, to the galvanic current.

Tinned peas may contain traces of tin. The process for the detection of tin is as follows:—A sufficient quantity of the peas is incinerated in a platinum dish, the ash is heated with strong hydric chloride, and evaporated nearly to dryness; a little water is then added, boiled, and the solution filtered. This method of extraction is repeated once or twice. The solution is now saturated with hydric sulphide, and any yellow precipitate filtered off. This should present the characters of sulphide of tin. Tin has been found, according to Mr. Hehner,¹ generally in tinned goods to the amount of 10 mgrms. in the English pound, and it has been supposed, without adequate proof, to exist as a stannous hydrate, a tin compound which is poisonous.

No prosecution has hitherto taken place with regard to tin in preserved goods; and in such small quantities as have hitherto been found, it is very questionable—presuming the tin to exist as stannous hydrate—whether any injury would result.

¹ *Analyst*, 1880, p. 218.

CHINESE PEAS.

§ 101. A pea or bean, much used in China in the form of cheese, is the *Soya hispida*.¹ Its composition, according to G. H. Pellet, is as follows:—

	1.	2.	3.
Water,	9.000	10.160	9.740
Nitrogenous matters (coagulable nitrogen, 6.25),	35.500	27.750	31.750
Starch, dextrin, and sugar,	3.210	3.210	3.210
Cellulose,	11.650	11.650	11.650
Ammonia,290	.274	.304
Sulphuric acid,065	.234	.141
Phosphoric acid,	1.415	1.554	1.631
Chlorine,036	.035	.037
Potash,	2.137	2.204	2.317
Lime,432	.316	.230
Magnesia,397	.315	.435
Substances insoluble in acids,052	.055	.061
Not estimated mineral substances,077	.104	.247
Different organic matters,	15.289	25.539	24.127

LENTILS.

§ 102. The lentil is the seed of the *Ervum lens*, one of the Leguminosæ. Lentils are grown and eaten in all parts of the civilised world, and are highly nutritious. They contain, according to H. Ritthausen, 5.9 per cent. of legumin, and their general composition is as follows:—

	Per cent.
Water,	12.51
Nitrogenous substances,	24.81
Fat,	1.85
Carbo-hydrates,	54.78
Woody fibre,	3.58
Ash,	2.47

The general composition of the ash is as follows:—

	Per cent.
Potash,	34.76
Soda,	13.50
Lime,	6.34
Magnesia,	2.47
Ferric oxide,	2.00
Phosphoric acid,	36.30
Chlorine,	4.63

¹The pea-cheese is considered, in China and Japan, a very important food. The peas (*Soya hispida*) are soaked in water for about 24 hours, then strained; they are next ground to a thin paste with some of the water which has been put on one side. The grinding is effected by a mill. The matters are filtered, and the filtrate is concentrated by heat; and after skimming once or twice is cooled, the casein coagulated by plaster, and a salt, which appears to be chloride of magnesium, added. The cheese is greyish-white, and has the following general composition:—

	Per cent.
Water,	90.37
Fatty matters,	2.36
Nitrogen,78
Ash,76

—M. Stanislaus Julien et M. Paul Champion
"Industries de l'Empire Chinois."

BEANS.

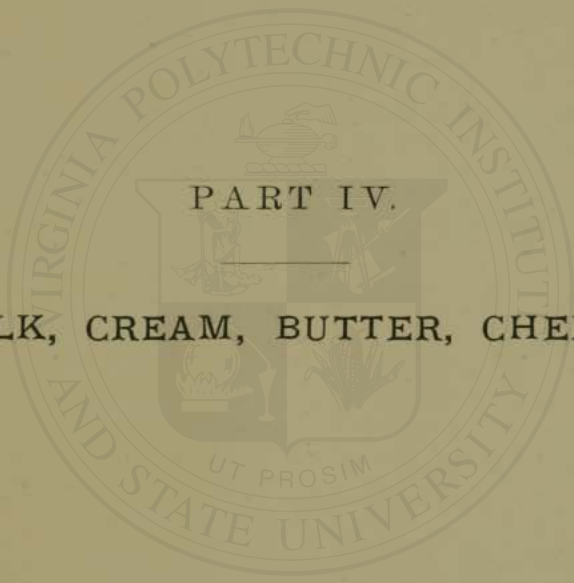
§ 103. The beans eaten in this country are mostly the kidney bean, *Phaseolus vulgare*, and the broad bean, *Vicia faba*. The following is the average composition of these vegetables :—

	Broad bean.	Kidney bean.
Water,	14·34	13·60
Nitrogenous substances,	23·66	23·12
Fat,	1·63	2·28
Carbo-hydrates,	49·25	53·63
Woody fibre,	7·47	3·84
Ash,	3·15	3·53

§ 104. The percentage composition of the ash of these different beans has the following composition :—

	Broad bean.	Kidney bean.
Potash,	42·49	44·01
Soda,	1·34	1·40
Lime,	4·73	6·38
Magnesia,	7·08	7·41
Ferric oxide,	·57	·32
Phosphoric acid,	38·74	35·00
Sulphuric acid,	2·53	4·05
Silica,	·73	·57
Chlorine,	1·57	·86

From both the broad and the kidney bean a small quantity of cholesterin can be separated. According to Ritthausen, the legumin of the kidney bean has a composition different from that of other legumins; for while the percentage of nitrogen in pea and millet legumin amounts to 16·77 per cent., that of kidney bean legumin has only 14·71 per cent.



PART IV.

MILK, CREAM, BUTTER, CHEESE.

PART IV.—MILK, CREAM, BUTTER, CHEESE.

MILK.

HISTORICAL INTRODUCTION.

§ 104. Before the birth of experimental philosophy, the origin rather than the composition of substances was the subject of inquiry, and of fanciful and more or less ingenious conjecture. Milk to the ancient, as well as to the modern world, was a fluid of great virtue. Aristotle affirmed, '*Lac est sanguis concoctus, non corruptus,*' which may be translated, *Milk is elaborated, not decomposed, blood*—an opinion identical with that held by nineteenth-century philosophers.

Averroes, Avicenna, and others, reasoning in part from the difficulty with which many females conceive while suckling, held that milk was altered menstrual blood. Avicenna, indeed, formularised this doctrine by declaring that the menstrual blood of the pregnant was divided into three parts—part going to nourish the fœtus, part ascending to the breasts, and the remainder being an excrementitious product. These opinions may be traced to writers of a much later, almost modern epoch. The ancients were acquainted with only three constituents of milk—viz., butter, with which they used to anoint their infants; casein, which they precipitated with vinegar; and the whey from which the curd and butter had separated, and this, up to the early part of the sixteenth century, constituted the whole of what was known as to the composition of milk. Placitus enumerates no more constituents than Avicenna, but devotes several pages to the then all-important question as to whether milk was hot, cold, or moist, and concludes that animal milk, as compared with that of human, is cold, human with that of animal, hot. Placitus¹ was an upholder of the menstrual theory. Panthaleon² similarly cites with approval the dictum that *milk is a fluid superfluity, twice concocted in the breasts*, and gravely discourses, as stated, whether it is hot or cold.³ He recognises three parts only in milk—viz., serum, butter, and curd. His treatise is mainly

¹ Sexti Placiti Papyriensis: *De Natura et Usu Lactis*, MDXXXVIII. It would appear, according to this author, that the Germans in his time used the milk of all animals, for he enumerates the milk not only of cows, mares, and goats, but also of pigs.

² *Summa Lacticinorum*, 1528.

³ There are several other treatises on milk about this epoch, but they nearly all, as, for example, that of Gesner (*Libellus de Lacte et Operibus Lactarius*, auth. Conrad Gesnero, Medico), consist of commentaries on the opinions of older writers, and are of no value.

composed of references to the ancients, and the usual disputations as to whether milk is hot or cold. The first mention of a fourth constituent of milk occurs in a curious work by Bartoletus, published in 1619. Bartoletus¹ called it the 'manna' of milk, or '*nitrum seri lactis*.' In his days sulphur, mercury, and a saline principle, were considered as the three active essences of all things, and as existing in all things; hence, Bartoletus, from the yellow colour of butter referred it to a sulphur principle, the whey, doubtless from its mobility, to quicksilver, and the curd to a saline element. He then compares milk with blood, also composed of a sulphurated, saline, and mercurial principle.² The discovery of Bartoletus for a long time was not known beyond Italy. A French apothecary, named Bartholomew Martin, writing in 1706,³ enumerates the constituents of milk as three—butter, analogous to sulphur, serum to mercury, and cheese to salt; but was not acquainted with milk-sugar, although eight years before Ludovico Testi⁴ had written an entire treatise on it, calling it by the name it now bears.

In the early part of the eighteenth century, Leeuwenhoek discovered the microscopical characters of milk. He saw that it was a fluid containing many globules. Some, which he judged to be of a buttery nature, rose to the top of the liquid; and others, again, rather sank to the bottom, and were evidently different in composition.⁵ Some twenty years later, A. Donné, in his *Cours Microscopique*,⁶ published some beautiful plates of several kinds of milk, fresh and sour, human and animal, exhibiting the globules, etc., drawn to scale with wonderful accuracy.

§ 105. In the early part of the eighteenth century flourished the school of the illustrious master Boerhaave, who laid the foundations of animal chemistry. Boerhaave saw in milk the most perfect food, and to him it was a fluid containing, wrapt up in mystery, all the elements of the body. Hence he laid the greatest stress on the importance of its study, and without doubt his example and teaching were the immediate cause of the numerous experiments carried out by his disciples, Vullyanoz, Doorschodt, and others. Boerhaave says⁷—

"An animal is composed of matter which was not that animal before, but is changed into it by the vital power of the animal. . . . Milk, therefore, appears to be the first thing to be examined, for this is a true chyle, and much less diluted with the lymph than the chyle when poured into the subclavian vein, and therefore approaches nearer to the aliment. It has flowed through the veins, the heart, the lungs, and the arteries, and therefore has been mixed with all the juices, and being afterwards separated by the particular structure of the breasts, it may be collected and examined apart. Milk is a liquor prepared from the aliment chewed in the mouth, digested in the stomach, perfected by the force and juices of the intestines, and elaborated by means of the mesentery and its glands and juices, and the juices of the thoracic duct; it has undergone some actions of the veins, arteries, heart, lungs, and juices, and begun to be assimilated,

¹ Bartoletus was an Italian physician, a professor at Bologna and Mantua, b. 1586, d. 1630. His work is entitled, *Encyclopadia Hermetico-Dogmatica sive Orbis Doctrinarum, Physiologica, Semiotica, et Therapeutica*. Bononia, 1619, 4to. The quarto is little over 300 pages, and is divided into five parts, viz., (1.) Physiology, (2.) Hygiene, (3.) Pathology, (4.) Semiotics, and (5.) Therapeutics.

² "*Enim in lacte videre est, in quo serosa portio mercurialiliquori, butyrosa sulphurea, casciosa vero saline substantie respondet. Ita in sanguine alia sulphurea, alia saline, alia mercurialia substantie proportionaliter respondet.*"

³ *Traité du Lait*, par Barth. Martin, Apothicaire, Paris, 1706.

⁴ *Relazione concernente il Zucchero di Latte*, 1698.

⁵ *Letters*, tome ii., 4to edition, 1722.

⁶ *Cours de Microscopie*, 8vo, Paris, 1844; *Atlas*, in folio, Paris, 1845.

⁷ "The Practice of Chemistry," translated from Boerhaave's *Elementa Chemicæ*. By Peter Shaw, M.D. 2nd ed. Lond. 1741, 2 vols., 4to.

yet may be had separate and discharged out of the body. And thus, by their own milk prepared from the proper matter of the chyle, all the known animals that have milk are nourished, both male and female; for milk is always prepared from the chyle as well in men as in women, as well in virgins and barren women as in mothers and nurses. Whence every such animal consists, is nourished, and lives on its own proper milk, and from this alone prepares all the other parts, both the solid and fluid, by means of the vital actions. It is now certain that men may live for years upon milk alone, and perform all the actions of life, and have all the solid and fluid parts of their bodies perfectly elaborated thereby. The serum, therefore, the blood, the lymph, the spirits, bones, cartilages, membranes, and vessels, proceed from milk, and if a man may live for years upon milk alone, milk must contain in itself the matter of all the parts of the human body."

Boerhaave appears to have tested milk with a great variety of reagents, and found that it was curdled by all acids, whether nitric, acetic, hydrochloric or sulphuric, or by acid vegetable juices. He also distilled milk, and found that it gave no spirit on distillation. "It also appears not to contain any trace of saline matter, being inodorous and perfectly insipid, and causing no pain if dropped into the eye." On boiling milk with alkalies, Boerhaave was the first to notice the yellow colour caused by the decomposition of the sugar. He thought that a similar change took place in fevers, for he notices the yellow milk of feverish women, and warns the physician that he must not suppose the yellowness to be caused by an acid, but rather by an alkaline tendency, and by too much heat. Boerhaave paid particular attention to the state of the milk in fevers and infectious diseases; "and in the last contagion among the cows, whilst their meat remained in the stomach, and was neither discharged upwards by ruminating nor expelled downwards, and therefore truly putrefied with the violent degree of heat, so that the stomach was almost scorched with heat, as we explained the thing Then the milk grew sharp, yellow, somewhat foetid, and thin in the dug, and in this form was either milked out or dropped spontaneously." He also condemned the use of milk from heated or improperly fed animals, or those suffering from fever, and remarked that it would be found of a foetid urinous odour, yellow in colour, thin, of a saline ungrateful taste, and acquiring, after a time, an odour of rancid cheese.¹

§ 106. Boerhaave, so far as is known, made no quantitative determination of any of the constituents of milk; but a very early attempt is found in a research undertaken by Geoffroy, published in 1737.² This experimenter took 12 lbs. of milk, and after coagulating the fluid, heated it gently over the fire, in order to separate the coagulum more completely. The liquid was now filtered, and the serum and coagulum both weighed. The serum weighed 8 lbs., the coagulum 2 lbs. 7 ozs. The serum was then evaporated to dryness, and left a residue weighing 7 ozs. 24 grains; in other words, it amounted to 5·2 per cent.; and since it must have been mainly composed of milk-sugar and salts, the determination is almost as exact as that of any analysis of the present day. He now appears to have distilled the residue, and obtained empyreumatic products, and a 'caput mortuum,'³ from which he extracted soluble salts by lixiviation, and among these salts he recognised chloride of sodium by its cubical crystals.

Doorschodt⁴ experimented on milk, possibly under the immediate superintendence of Boerhaave; for he distilled it, and noticed that the distillate

¹ A work by Dumonchaux, about the period of the Boerhaave school, *De Lacte Mammarum et Pinguidine*, Petrus J. Dumonchaux, Duaci, 1754, contains nothing new about the composition of milk, but merely cites the opinions of others.

² *Commercium Literarium ad Re Medicæ et Scientiæ Naturalis Incrementum Institutium*, etc., 1737.

³ The *caput mortuum* was the name of any residue left after distillation in the retort.

⁴ Henricus Doorschodt: *De Lacte*, 1737.

was neither acid nor alkaline, concluding, hence, that water alone was condensed, and that there was no other volatile principle. He also boiled the milk with alkalies, and details with great precision the successive changes of colour. He appears to have been the first to notice that alcohol coagulates milk, and also that it may be preserved by borax and other antiseptics.

§ 107. M. Vullyanoz, another disciple of Boerhaave, published a tract,¹ in 1756, on the essential salt of milk, which tended greatly to spread a knowledge of the substance discovered by Bartoletus, and described so fully by Testi the Italian. It would appear from his treatise that sugar of milk was then an article of commerce, but that there was great difficulty in preparing it white and pure. "There is in Switzerland a chemist named Creuzius who has composed the salt admirably, but unfortunately he will not impart his secret to any one. This is the more vexatious, because the salt he is proprietor of is infinitely finer than the others; it is whiter, sweeter, and dissolves better on the tongue." The method used in the time of Vullyanoz was simple evaporation, but he complains that the product was often 'sour,' and was not the same as the Swiss sugar. Vullyanoz established the fact that all herbivorous animals, as well as women, gave sugar of milk; he also investigated the solubility, and found it insoluble in hot alcohol, in spirits of ammonia, and in very pure aqua fortis, etc. Noticing that it effervesced with nitric acid, he made experiments which proved it to be a neutral salt, and thence drew an analogy between milk-sugar and soap, concluding that the latter contained an oil and also an acid, that it could be fermented, and that on distillation it yielded an acid, and was decomposed by sulphuric acid.²

§ 108. The next important paper on milk in order of time, is that of Voltelenus,³ important because his experiments were quantitative. He took 42 ozs. of cows' milk and distilled it. The process was conducted very carefully, and occupied many days, and, as may be expected, was very troublesome, from the irregular bursts of ebullition. By the fifth day he obtained 29 ozs. of distillate '*Aqua Lactis Destillata*'; in other words, his determination of water was 69 per cent., much below the truth. The residue in the retort weighed 2 ozs. 3 drms., and effervesced with alkalies. He now increased the fire, and obtained an unctuous oil, weighing 14 drms., mixed with what he calls an oily spirituous matter, acid, acrid, and like the spirit from guaiacum wood. The carbon in the retort weighed 10 drms., but on burning to an ash it weighed 3. The ash, boiled with water, left 2 drms. insoluble. In other words, he determined the ash to be '89 per cent., and the soluble portion '31 per cent. Hence, Voltelenus most certainly made a correct determination of the amount of saline matters in milk, and was probably the first who did so. Voltelenus next made a similar experiment with women's milk, taking 32 ozs., from which, in thirteen days, he had distilled over 31 ozs. 6 drms. of odourless liquid. Here, unfortunately, his retort broke; but he concluded that human milk is resolved by fire into much water and spirit; a double oil, a double salt, fixed and warm alkali and earth, to which may be added a '*spiritus sui generis*.' He refers to sugar of milk, and

¹ "Sur le sel essentiel de Lait." Par M. Vullyanoz, Docteur en Médecine à Lausanne. "Recueil Périodique, Observations de Médecine, Chirurgie, Pharmacie," etc. Par M. Vandermonde, 1756.

² Subsequent to the work of Vullyanoz appeared a treatise on milk-sugar, "Abhandlung vom Milch-Zucker," Braunschweig, 1772, by G. R. Lichenstein, who considered it an earthy salt, and called it *terra-oleosum sal mediam*.

³ Floris Jacobi Voltelenii: *De Lacte Humano ejusque cum Asino et Ovillo Comparatione*, etc. Lipsiæ, 1779.

affirms that he has separated a similar substance from human milk. The same process was applied to asses' milk, 32 ozs. being distilled over a sand-bath in three days. On the first day a lactescent distillate came over, in quantity amounting to 1 oz. 17 drms. 1 scr.; on the second day, a more limpid liquid, amounting to 19 ozs. 4 drms.; and on the third day there came over 6 ozs. 1 drm. 1 scr. of a feebly acid liquid; by the fourth day he had to increase the heat, and obtained a black opaque oil, which separated on standing into three parts—a thick substance, a thinner, and what he calls a spirit. The carbon in the retort was weighed and then burnt. The ash weighed 3 drms., and on lixiviation the insoluble portion weighed 2 drms. 1 scr. He made precisely similar experiments on the milk of the sheep—identified salt, determined the amount of ash, etc. He thus came to the conclusion that all milk had the same constituents.

§ 109. Schoepff, in a very learned paper,¹ containing full references to the works of his predecessors, was the first who noticed the yellow colour of the whey—'liquidem colore diluti citrinum.' He crystallised milk-sugar, and determined its amount with fair accuracy; but did not know exactly what it was, for the crystals were of a yellow colour, and reddened syrup of violets; hence they were probably contaminated with lactic acid and colouring-matter.² One of the last workers on the chemistry of milk, prior to the nineteenth century, was Scheele, who discovered lactic acid, and established that phosphate of lime was always present in the casein. He considered, in fact, that the casein formed with lime a true combination, the proportion between the two being from 1 to 1·5 per cent. of calcium phosphate to every 100 parts of dried casein.³ Experiments similar to those recorded were undertaken by Hoffman, who determined the total solids of cows' milk to be 13·5 per cent.; of asses, 9·5; goats about 10 per cent.; and of human, 9. He exhausted the total solids by water, and evaporated and weighed the soluble matter thus extracted, but no accurate result followed; and, indeed, it is very difficult to dissolve out milk-sugar and salts fully from the milk solids, unless they have been previously deprived of their fat. Caspar Neumann repeated and enlarged the experiments of Hoffman; he made out that cows' milk contained 14 per cent. of total solids, and he also distilled milk as well as butter. From 16 ozs. of fresh butter, distilled in a retort, at first over a sand-bath, and afterwards over an open fire, there arose 1 oz. of liquor of no remarkable smell or taste; 1 oz. and half a drm. of a reddish acidulous liquor, which smelt like burnt butter; 1 drm. of a brownish-yellow oil; 3 ozs. 3 drms. of a yellow oil; 1 oz. 6 drms. of a white, and 5½ drms. of a yellowish-brown oil—all of a thick butyraceous consistence, and a volatile smell like that of horse-radish; and 1 oz. 6 drms. of a thin empyreumatic oil, which smelt like the *Oleum philosophorum*, that is, old olive oil distilled over from bricks. There was not the least mark of any volatile alkali in the whole process. The *caput mortuum* weighed 3½ drms.⁴

¹ *Specimen Inaugurate Chemico-medicum de Variis Lactis Bubuli Salibus aliisque Substantiis in ejusdem parte Aquosa Contentis, etc.* Ludovicus Augustus Schoepff, 1784.

² Previous to Schoepff, Beaumé appears to have made an accurate determination of the amount of salt in milk, saying that the third evaporation yielded crystals of sea salt, in the proportion of 7 to 8 grains per pint. Beaumé: *Dict. de Chimie*, ii., 1778, 498. Rouellium denied that the crystals were those of sea-salt, but considered them "salis febrifugi sylvii."

³ *De Lacte ejusque Acidis: Nova Acta Acad. Reg. Sued. Anni 1780; Opuscula Chemica*, vol. ii., pp. 101-118.

⁴ "The Chemical Works of Caspar Neumann," abridged and methodised, by Wm. Lewis. Lond. 1773.

THE COMPOSITION OF COWS' MILK.

§ 110. Up to the present time the milk of the mammalia alone has been fully analysed. This has been found to consist of water, a peculiar sugar, albuminous bodies, a small amount of saline matter, and an emulsified fat. The milk of every class of animals has not, however, yet been examined completely; and although it may be presumed, on physiological grounds, that all milks contain qualitatively identical or analogous ingredients to those of the cow, yet this has been by no means proved. Cows' milk may be considered first, as its composition is more completely known¹ than that of any other milk.²

Cows' milk consists of matters partly in solution,³ and partly in suspension; sugar of milk, casein, peptones, lactochrome, saline matters, and minute quantities of several other substances are dissolved in water, whilst milk-fat is apparently emulsified, and a portion of the casein is in the form of extremely fine granules, which can only be arrested by filtration through porous earthenware, or a similar filtering medium. When a very thin layer of healthy milk is examined by a microscope, the milk-fat alone is visible, and appears in the form of innumerable globules, the number of globules depending on its richness in fat. Thus M. E. Bouchut found in different samples of milk the following varying numbers:—

Globules in a cubic millimetre.	Specific gravity.	Fat per litre.
1102500	1022	24
1820000	1021	21
1925500	1030	26
2105000	1028	29
2205000	1032	37
2305000	1028	29
2205000	1032	37
2400000	1030	37
2407000	1033	34
2692000	1030	29
3700000	1030	34

It would hence appear that good milk contains from about two to three and a half millions of globules in every cubic millimetre. It has hitherto been taught that the globules are surrounded by a thin pellicle or membrane, and, as a proof of the existence of this membrane, the fact is usually cited that, if you shake up milk with ether, scarcely any of the fat dissolves unless a little acetic acid is first added, which is supposed to act by dissolving the hypothetical membrane; but it may be shown that the fat can be extracted from milk by shaking with ether, provided that the volume of ether to that of the milk be excessive. Further, the globules are coloured by aniline red,⁴ and their behaviour with moderate quantities of ether may be ascribed to the acids destroying the emulsifying property

¹ The milk of the buffalo (*Bos bubalus*) has, however, been investigated somewhat minutely by A. Pappel and H. Droop Richmond, *Journ. Chem. Soc.*, lvii., p. 752.

² The chemistry of the milk-secreting glands has scarcely been investigated. Bert (*Gaz. hebdom.*, 1879, N. 12) states that the gland contains a peculiar body which easily splits up with the production of a sugar by boiling with dilute sulphuric acid, or even simply with water.

³ The specific gravity of the whey of milk is from 1·0280 to 1·0302 at 15°·5 (*Vieth*).

⁴ De Siney: *Arch. de Physiol.*, 1874, 497.

of the milk. Nor can one understand, on the 'membrane' theory, how—when milk in thin layers is dried at the ordinary temperature of the air, and under conditions which involve no destruction of the membrane, should this exist—to account for the fact that ether so readily dissolves the butter fat. Hoppe-Seyler¹ has indeed, by estimation of the proportion existing between the water and casein in cream, considered that a casein layer exists round fat globules; yet this must be so thin as not to be capable of estimation by weight. From these various facts the existence of the membrane is still doubtful.

§ 111. *Amphioteric Reaction of Milk.*—Milk when tested immediately after its removal from the cow, has a peculiar action on litmus and turmeric paper, turning litmus blue and turmeric brown,—the so-called '*Amphioteric Reaction.*' On this point alone, although of no great importance, there is a most voluminous literature.² The amphioteric reaction of milk is similar to that shown by a solution of magnesium-hydric phosphate to which a little acid has been added, and is probably due to the acid phosphate of the alkalies existing in milk. It must also not be lost sight of, that there is a continuous development of CO₂ in milk, which gas in solution is always present; and this being the case, its feeble acid reaction must have an influence on the total reaction derived from other substances. Milk ultimately becomes decidedly acid, and has a constant tendency to acidity.³

§ 112. *Total Solids of Milk.*—The amount of solid matter in milk varies within considerable limits, and is much influenced by all circumstances that affect the health and nutrition of the cow, certain cows secreting double and treble the normal amount of fat. The remark just made refers to the entire residue *minus* the water; but if we subtract the water as well as the fat, then the percentage of solid matter varies but little, and in healthy, fairly-fed cows does not, save in exceptional cases, fall below 8·5 per cent. This very important fact, formerly much disputed, has been (or ought to have been) set at rest by the results obtained in the experiments of so many chemists, that it is scarcely worth while reviewing the evidence on which it is based. The numerous analyses of Wanklyn, Carter Bell, Vieth, and others tend rather to show that the true lowest percentage of milk solids, *minus* fat and water, is 8·9 per cent. The highest amount of the same solids which the authors have yet found in the secretion from healthy animals reaches to about 11 per cent., so that at most there is an extreme fluctuation between 8·5 and 11 per cent.—a remarkable fact, which *a priori* would have been pronounced improbable, considering the complex nature of milk.

§ 113. *Milk-Fat.*—Pure dry milk-fat is at ordinary temperatures a solid fatty substance, with an agreeable taste, of specific gravity '9223 to '9377 at 15° C., '91200 to '91400 at 37°·7 C. [100° Fahr.]; its melting point is 35°·8 C. Milk-fat, under the form of butter, is constantly tinted more or less yellow from dissolved lactochrome; but it may, by the use of suitable solvents, be obtained almost colourless.

¹ *Archiv für path. Anatom.*, bd. xvij., s. 417, 1859.

² e.g., Kritisches u. Thatsächliches über die Reaction der frischen Milch, by J. Schlossberger, *Annal. der Chem. u. Pharm.*, lxxvii., 317, 1852; *Idem.*, xcvi., 76. Also a paper by Vogel, *Journ. für prakt. Chem.*, 1874, viii., 137, "Ueber das Verhalten der Milch zum Lackmus Farbstoffe."

³ According to C. Arnold, *Arch. Pharm.* [3], xix., 41, 42, fresh unboiled milk gives a blue colour to tincture of guaiacum; sour milk gives the same reaction, but not milk which has been boiled.

According to Wigner, from 1000 volumes pure fat expands at $37^{\circ}7$ C. [100° Fahr.] to 1,047.2 at 100° C. [212° Fahr.], its average expansion being .00076 for every degree Centigrade.¹ This is, however, not perfectly accurate for the degrees between $65^{\circ}5$ C. [150° Fahr.] and $87^{\circ}7$ C. [190° Fahr.], the expansion being slightly in excess of the average rate; but the abnormal deviation between these degrees does not appear to be peculiar to milk-fat.² 1 grm. of the fat requires for saturation 227.3 mgrms. of potassium hydrate (KHO). This observation (originating with Dr. Koettstorfer³) has been utilised by the food-analyst in the distinguishing between butter and other fats.

§ 114. Milk-fat is essentially an intimate mixture of the glycerides of the fatty acids—palmitic, stearic, and oleic—not soluble in water, and also of the glycerides of certain soluble volatile fatty acids, of which butyric is the chief, and caproic, caprylic, and capric acids minor constituents.

Palmitin, or *Tripalmitin*, $C_3H_5(C_{16}H_{31}O_2)_3$, is a white solid fat, little soluble in cold, but readily soluble in hot alcohol or ether. A mixture of stearin and palmitin crystallises in little needle-like tufts, and was at one time considered a definite single fat, and called *margarin*. On saponifying palmitin by means of an alkali and subsequent decomposition, it yields glycerin, and 95.28 per cent. of its weight of palmitic acid.

Palmitic Acid ($C_{16}H_{32}O_2$) has a melting point of about 62° C. It may be obtained in quantity from palm oil, and also from the saponification of spermaceti. When purified by repeated crystallisation from alcohol, it is a tasteless white fat, crystallising in tufts of needles.

Stearin, or *Tristearin*, $C_3H_5(C_{18}H_{35}O_2)_3$, is a white solid fat, melting at about 66° C., and is a special constituent of fats with high melting points. On saponification and subsequent decomposition of the soap by a suitable acid, 95.73 per cent. of stearic acid may be obtained.

Stearic Acid, $C_{18}H_{36}O_2$.—This acid is to be found in nearly all animal fats as well as in a few vegetable fats. Stearic acid is an article of commerce, and made upon a large scale, especially in the manufacture of stearin candles. For this purpose, animal fats are saponified by hydrate of lime; the lime compound is subsequently decomposed by dilute sulphuric acid, and the mixture of oleic, palmitic, and stearic acids submitted to strong pressure; by this means, the oleic acid is separated, and a mixture of palmitic and stearic acids obtained, which in commerce is known as stearin. From this commercial stearin, stearic acid may be obtained by solution in alcohol and fractional precipitation by acetate of lead or barium, the stearate of lead or barium, as the case may be, separating before the palmitate. On decomposing the salt with sulphuric acid, and dissolving the acid in boiling alcohol, stearic acid crystallises, as the solution cools, in white glistening needles or leaflets, which appear under the microscope as elongated lozenge plates. The melting point of stearic acid is $69^{\circ}4$ C. If impure, it crystallises in needles. It is without odour or taste, does not feel greasy to the touch, and dissolves in all proportions in boiling alcohol or ether, from which it separates on cooling.

Olein, or *Triolein*, $C_3H_5(C_{18}H_{33}O_2)_3$.—Over 40 per cent. of milk-fat consists of olein, which is a combination of oleic acid with glycerin, and is at all

¹ Allen gives the difference as .00062.

² On the Ratio of Expansion by Heat of Butter-fat, by G. W. Wigner, F.C.S., *Analyst*, No. 43, p. 183.

³ New Method for the Examination of Butter for Foreign Fats, by Dr. Koettstorfer, *Analyst*, No. 39, 1879, p. 106.

ordinary temperatures a fluid oil, solidifying about 5° C., at first colourless, but soon becoming yellow from absorption of oxygen. It has the power of readily and copiously dissolving palmitin and stearin, and is soluble in absolute alcohol or ether. On decomposition, olein yields 95.70 per cent. of oleic acid, $C_{18}H_{34}O_2$.

Pure oleic acid is difficult to obtain, since it so readily oxidises. When perfectly pure, it is without colour, taste, or smell, and has all the appearance of a colourless oil; at a low temperature (4° C.), it crystallises in needles; on destructive distillation, among a variety of gaseous and liquid products, it yields an acid known as sebacic acid ($C_{10}H_{18}O_4$), which is a constant product when any oil containing oleic acid is destructively distilled. Oleic acid forms two classes of salts, normal and acid. The normal oleates of the alkalies are soluble in water, but the other salts of oleic acid are insoluble in water, no exception being found even in the case of the acid salts of the alkalies. The oleates of lead and copper are soluble in ether, as well as in cold anhydrous alcohol. The analyst takes advantage of this fact to separate the oleates of lead and copper from the stearates and palmitates, which are insoluble in ether.

Butyrin, Caproin, Caprylin and Rutilin have not yet been separated in a pure state; they yield on saponification butyric, caproic, caprylic, and rutilic acids respectively.

Butyric Acid, $C_4H_8O_2$.—There are two butyric acids, one, normal butyric acid, C_3H_7COOH , boiling point 163.4° C., specific gravity .9817 at 0°; the other, isobutyric acid, $C(CH_3)_2H.COOH$, boiling point 154° C., specific gravity .8598 at 0° C. The latter has a less offensive odour than normal butyric acid. Butyric acid is found in several plants, such as the locust bean, the fruits of the *Sapindus saponaria*, in the *Tamarindus indica*, the *Anthemis nobilis*, the *Tanacetum vulgare*, *Arnica montana*, the fruit of the *Gingko biloba*, and probably several other plants. Butyric acid is the characteristic fatty acid of butter, and butter fat contains from 3 to 4 per cent. of it. Butyric acid is volatile, and may be distilled unchanged; it is also soluble in all proportions in water, alcohol, and ether. Most of the salts are soluble: baric butyrate crystallises in long prisms with four atoms of water; zincic butyrate in anhydrous pearly tables, which are remarkably soluble; cupric butyrate is bluish-green, and but sparingly soluble. Calcic butyrate is a very characteristic salt, for it is more soluble in cold than in boiling water, consequently, when a solution is boiled some of the salt is precipitated. The most characteristic reaction of butyric acid is its easy etherification by treatment with sulphuric acid and alcohol. In this way is formed butyric ether, which has a powerful smell resembling that of pine-apples. Its specific gravity is .902, and boiling point 119° C.

Caproic Acid, $C_6H_{12}O_2$.—There are two caproic acids, the one, normal caproic acid, $C_5H_{11}COOH$, boiling point 205° C.; the other, isocaproic acid, $C_3(CH_3)_2H_5COOH$, boiling point 199° to 200° C. Caproic acid occurs in a very large number of plants, and has been found in human perspiration and in cheese. It is almost insoluble in water, is volatile, and may be distilled unchanged. The caproate of silver is in large thin plates, almost insoluble in water, and but slightly sensitive to light. The caproate of barium is soluble.

Caprylic Acid, $C_8H_{16}O_2$.—fusing point 58° C., boiling point 236° C. The amount of caprylic acid in butter is very small. It is slightly soluble in boiling water.

Rutilic Acid, $C_{10}H_{20}O_2$.—A white crystalline solid, fusing about 29.5° C.

This acid, in combination with glycerin, also occurs in very minute quantity in milk-fat. It is even less soluble than caprylic acid.

§ 115. *The Albuminoids of Milk.*—The albuminoids of milk comprise at least three principles,—viz., casein, albumen, and nuclein. To these a fourth substance used to be added—viz., lacto-protein; but this, as the authors have shown,¹ is not a simple substance, and it is probable that it is a mixture of peptones. Casein appears but little, if at all, different from alkali-albuminate, the minor differences which exist being, with probability, ascribed to impurities. It is true that when milk is filtered through a porous cell, casein, for the most part, is left behind; while, if a solution of alkali-albuminate is similarly treated, it passes through. Experiment has, however, shown that a solution of alkali-albuminate shaken up with butter fat behaves exactly like casein; and similarly, Soxhlet has proved that a concentrated solution of sodic carbonate precipitates both casein and alkali-albuminate, provided they are under the same conditions, and that it does not (as asserted by Zahn) leave alkali-albuminate in solution.²

Hoppe-Seyler³ does not adopt altogether this view; for although he states that without doubt no other albuminoid, in its properties, stands so near casein as alkali-albuminate, yet the notable difference in its power of rotating a ray of polarised light, and its behaviour to rennet, separates casein as a distinct substance. The casein of either cows' or goats' milk, not only in acid fluids, but in perfectly neutral solutions, is coagulated by gastric juice or a watery extract of the stomach. This coagulation takes place slowly at common temperatures, rapidly on warming; and the cause of it, according to Hammersten, is a body distinct from pepsin. Hammersten finds that the casein of cows' milk, in the absence of lime salts, is coagulated by the addition of an acid, but not by rennet. A solution of casein which has been precipitated by an acid, run into lime water neutralised by very dilute phosphoric acid, quickly coagulates on the addition of rennet, but without rennet neither on the addition of an acid, nor on boiling. Hammersten considers that the chemical change produced in the coagulation of casein by rennet, is the splitting up of the casein into two bodies, one of which is precipitated, and an albuminoid, which remains in solution, and is neither precipitated by boiling, nor by any of the following reagents—acetic acid, potassic ferrocyanide, or nitric acid; but is precipitated by mercuric chloride, and also by Millon's reagent.

Casein is precipitated by a variety of substances—lead acetate, cupric sulphate, alum, mercuric chloride, tannic acid, rennet, sulphate of magnesia, and mineral acids, if not too dilute; but none of these precipitate casein in a pure state, the precipitate usually containing fat, nuclein, and phosphate of lime, the latter, as already stated (p. 193), in the proportion of from 1 to 1.5 per cent. of casein. The best precipitant is sulphate of magnesia, which leaves the nuclein to a great extent in solution. The fat may then be extracted by ether; but the phosphate of lime is in true combination with the casein, and only a portion of it can be removed. A solution of casein in combination with sulphate of magnesia, and freed from fat, turns a ray of polarised light in dilute alkaline solution, $[a]_D - 87^\circ$; in strong alkaline solution, $[a]_D - 91^\circ$. Pure casein is a perfectly white,

¹ Composition of Cows' Milk in Health and Disease, *Journ. Chem. Soc.*, 1879.

² Beiträge zur physiologischen Chemie der Milch, by Dr. F. Soxhlet. *Journal für praktische Chemie*, vol. vi., p. 1, 1872.

³ Hoppe-Seyler, *Physiologische Chemie*, p. 930.

brittle, transparent substance, insoluble in water, but soluble in very dilute acid solution, as well as in very dilute alkaline solution; in each case there is little doubt that a true chemical combination is formed. The presence of phosphate of soda in a solution of casein (as, for example, in the milk itself), prevents the precipitation by simple neutralisation by an acid, the casein not falling down until the acidity of the liquid is decided. It has been shown by Schutzenberger that, on sealing up casein in a tube and heating with baryta water, it behaves like albumen, and is resolved into the following substances:—The elements of urea (ammonia and carbon dioxide) traces of sulphurous acid, of sulphuretted hydrogen, of oxalic and acetic acids, tyrosin, $C_9H_{11}NO_3$, the amido-acids of the series $C_nH_{2n+1}NO_2$, corresponding to the fatty acids, $C_nH_{2n}O_2$, from amido-cenanthylic acid to amido-propionic acid—leucin, $C_6H_{13}NO_2$, betain, $C_5H_{11}NO_2$, and amido-butyric acid, $C_4H_9NO_2$, with a few less known or identified products. Many of these substances may be identified in putrid milk.

The amount of casein in milk is fairly constant, being about 3.0 per cent.; and the authors have never known it exceed 5 per cent.

Serum-albumen occurs in milk, in no respect differing from the albumen of the blood. By careful addition of an alkali, this albumen may be changed into alkali-albuminate—that is, into casein; therefore, according to this view, the albumen in milk may be considered the residue of an incomplete reaction. Albumen is not precipitated by acetic, carbonic, phosphoric, or tartaric acids. A small quantity of a dilute mineral acid does not precipitate; with a larger quantity of concentrated mineral acid the solution becomes turbid, and the deviation of a ray of polarised light increased; a still larger quantity of acid precipitates it as acid albumen. The best method to obtain a solution of pure albumen is to precipitate a solution by basic acetate of lead, pass carbon dioxide through the mixture, separate the carbonate of lead by filtration, and, lastly, pass through it hydric sulphide, to remove the trace of lead still existing. Albumen is then in solution, but on adding a little acetic acid, and evaporating, it may be obtained in the solid state contaminated slightly with acetic acid.¹

Another method of obtaining albumen pure is by dialysis. The physical characters of solid albumen differ according to the method of separation. Albumen obtained by dialysis is in the form of a yellow transparent mass, specific gravity 1.314; but albumen separated in the ordinary way from milk, for the purpose of quantitative determination, is in yellowish flakes, brittle, without taste or smell, insoluble in water, alcohol, and ether, soluble in dilute caustic alkali, if gently warmed, and from this alkaline solution precipitable by an acid. The amount of albumen in milk is really fairly constant, and averages .4 per cent. In healthy cows it is a very constant quantity, the chief deviation occurring directly after calving, when the amount may rise as high as 3 per cent., but this is always accompanied by a corresponding rise in the casein. According to the authors' experience, the albumen preserves very constantly the above relation to the casein; so that if either the amount of casein or albumen is known, the one may be calculated from the other with great accuracy.

¹ Meggenhofen appears to have been one of the first who detected the presence of albumen in milk. He estimated the amount in cows' milk as .59 per cent. *Dissertatio Inauguralis sistens indagatorem Lactis Muliebris Chemicam*. C. Aug. Meggenhofen. Frankfort, 1826.

Nuclein.—Nuclein is the organic phosphorus compound of milk, containing, according to Miescher, 9.6 per cent. of phosphorus. Its formula is $C_{29}H_{49}N_9P_5O_{22}$. It is by no means peculiar to milk, but has been found in the blood, in pus, in the yolk of eggs, in the liver cells, and in yeast cells. When freshly precipitated, it is a white amorphous body, somewhat soluble in water; freely soluble in ammonia, soda solution, and phosphate of soda. The special test distinguishing nuclein from other albuminoids is the presence of phosphorus, and the production of no violet colour, either by Millon's reagent, or by a copper salt, added to a solution of nuclein alkalisied by soda lye: it forms a very definite compound with lead, the lead and phosphorus being in the proportion of Pb to P.

The method adopted by Hoppe-Seyler¹ to separate nuclein from pus, was isolation of the pus cells by Glauber's salts, washing with very dilute hydrochloric acid and much water; then extracting the nuclein by the aid of a very weak alkaline solution of caustic soda, and filtering (which in this case proves a troublesome operation), and precipitating by a mineral acid. The precipitate is again dissolved in weak alkaline solution, and again precipitated, and the process repeated until the nuclein is supposed to be in a fairly pure condition. Nuclein may be separated from milk on the same principles, first exhausting the solids by alcohol and ether to remove fat.

§ 116. *Milk-Sugar*, $C_{12}H_{22}O_{11}H_2O$.—Milk-sugar, so far as is known, is only found in human milk, the milk of the herbivora, and of the bitch. It is easily distinguished from other sugars; its specific gravity is 1.53; and its solution turns a ray of polarised light to the right at 20° C. $[\alpha]_D = +52.53^\circ$, but with anhydrous milk-sugar $[\alpha]_D = +55.3^\circ$.

The separate researches of Erdmann² and Schmoeger³ have shown the existence of four modifications of milk-sugar, exhibiting a different rotation to normal sugar, viz.:—1. Crystallised milk-sugar exhibiting in solution strong bi-rotation. 2. Anhydrous milk-sugar obtained by dehydrating crystallised sugar at 130°, showing in solution also strong bi-rotation. 3. Anhydrous milk-sugar obtained by quickly evaporating a solution of milk-sugar in the presence of sand, or other finely divided substance, so as to ensure a large surface; the solution shows slight bi-rotation. 4. Anhydrous milk-sugar evaporated down from a solution quickly, but without the addition of sand or other substance. All these modifications are at once transformed into milk-sugar of normal rotation by boiling their solutions, or gradually, without the application of heat.

Milk-sugar is soluble in six parts of cold, and 2.5 parts of boiling water; it is insoluble in absolute alcohol and in perfectly dry ether, but in dilute alcohol and commercial ether it is slightly soluble, the solubility in amount depending mainly on the percentage of water which the ether contains. At 150° C. it loses an atom of water without further decomposition; its watery solution is perfectly neutral, and has a sweet taste: the sweetening power of milk-sugar as compared with cane sugar is but feeble.

It reduces Fehling's copper solution in a proportion different from that of grape sugar (see § 141). Milk-sugar undergoes lactic fermentation readily (see § 179), but alcoholic with some difficulty. Milk-sugar is precipitated by acetate of lead and ammonia; neutral acetate of lead, even at a boiling temperature, neither precipitates nor changes it. The oxides of copper, of bismuth, and silver are reduced by solutions of milk-sugar, and indigo is decolorised; these latter reactions are similar to those of grape sugar. When oxidised by nitric acid, milk-sugar yields mucic acid, acetic

¹ *Med. Chem. Untersuch.*, Hoppe-Seyler. Berlin, 4 Heft.

² *Bericht d. d. chem. Gesells.*, xiii., 2180-2184.

³ *Ibid.*, xiii., 1915-1931.

acid, and tartaric acids, and on further decomposition oxalic acid may be obtained.

By boiling milk-sugar for several hours with 4 parts of water and 2 per cent. sulphuric acid, neutralising with carbonate of lime, evaporating the filtrate to a syrup, a different sugar from lactose may be obtained in microscopical crystals. To this altered milk-sugar, the name of galactose has been given. Its action on polarised light is $[\alpha]_D + 81.4^\circ$ to 81.7° at 10° to 15° C.; it is a fermentable sugar, and yields, on oxidation with nitric acid, twice as much mucic acid as milk-sugar.

The amount of milk-sugar in normal milk preserves a very constant relation to the percentage of proteids and ash, the ratio according to Vieth being 13 : 9 : 2. Richmond gives the ratio as 52.8 : 37.8 : 8.3.

§ 117. *Mineral Constituents of Milk.*—The mineral constituents of milk have been fully and early investigated, and the following may be considered a very close approximation to their actual amount and character:—

Potassium oxide, K_2O ,	18.82
Sodium oxide, Na_2O ,	11.58
Calcium oxide, CaO ,	22.97
Ferric oxide, Fe_2O_3 ,06
Chlorine, Cl,	16.23
Magnesium oxide, MgO ,	3.31
Phosphoric pentoxide, P_2O_5 ,	27.03

Four analyses of milk ash by R. Weber and Haidlen give the following:—

	Minimum.	Maximum.	Mean.
Potash,	17.09	33.25	24.67
Soda,	8.60	11.18	9.70
Lime,	17.31	27.55	22.0
Magnesia,	1.90	4.10	3.05
Ferric oxide,33	.76	.53
Phosphoric acid,	27.04	29.13	28.45
Sulphuric acid,30
Chlorine,	9.87	16.96	14.28

The chlorine is in combination with the alkalis, the iron and the earths occur as phosphate, as well as the potassium oxide. So that the mineral constituents of cows' milk are, phosphate of potash, phosphate of lime and magnesia, common salt, and a trace of phosphate of iron. Other mineral inorganic constituents have been found in small quantity. If sufficient milk be used, it is not difficult to obtain a fluorine reaction, and since fluorides form an essential constituent of the teeth, it is easy to see their importance. A minute quantity of sulphuric acid as sulphates exists in milk, averaging from .05 to .08 grm. per kilogramme; and it has also been asserted by G. Musso, that milk contains a sulpho-cyanate. This assumption was based on the following experiment:—15 litres of milk, freed from casein, fat, and albumen, were neutralised by baryta water, and evaporated to a syrup, and the syrup extracted with absolute alcohol; the alcoholic extract dissolved in water and treated with zinc and sulphuric acid, yielded some hydric sulphide; and subsequent treatment yielded from 6 to 21 mgrms. of barium sulphate per kilogramme.¹ The experiment appears to the authors as hardly conclusive of the presence of a sulpho-cyanate, and requires further investigation.

Nadler has ascertained that neither cows' nor goats' milk contains any iodine: he used for the research 6 litres of cows' milk and 3 of goats'

¹ *Berichte der deutschen chemischen Gesellschaft*, xi., p. 154, 1878.

milk.¹ Minute traces of copper have been found in milk ; but lead, arsenic, and all other metals, save iron, are absent.

§ 118. *Other Constituents of Milk.*—In 1864, E. Millon and Commaille, after coagulating and separating the casein and albumen, obtained a precipitate from the yellow whey by means of a solution of mercury nitrate. This precipitate was white, amorphous, and became slightly red on drying ; it was insoluble in water, alcohol, and ether. The precipitate was washed with water, then with alcohol, and finally with ether, and after drying weighed.

	grms.
Cows' milk yielded	2·9 to 3·4 per litre, ²
Goats' milk "	1·52 "
Sheeps' milk "	2·53 "
Asses' milk "	3·28 "
Woman's milk "	2·77 "

To this body they ascribed the following formula— $C_{30}H_{31}N_5O_{18}$, $HgO + HgO$, NO_3 , and gave it the name of *Lacto-proteine*. In 1879, one of the authors studied this body and decomposed it, and came to the conclusion that lacto-proteine, as a single definite substance, had no existence ; but that the mercury precipitate was composed of two substances to which the names of *galactin* and *lactochrome* respectively were ascribed. With these substances are precipitated small portions of albumen, which may have escaped precipitation, and traces of urea.

The method of separation is as follows : The casein and albumen are separated in the manner described in § 142 ; the yellow whey is then precipitated by a solution of nitrate of mercury, of about the same strength as that used for estimation of urea ;³ the dense flocculent precipitate is then, after suitable washing, suspended in a very little water, and decomposed by hydrogen sulphide. The liquid is filtered, and to the filtrate a slight excess of acetate of lead in solution is added, when a dirty-white precipitate falls, which is collected, decomposed by hydrogen sulphide, and recomposed by acetate of lead until it is obtained perfectly white. On combustion, numbers are obtained agreeing with the following formula, $11(PbO)_2PbO_2C_{54}H_{78}N_4O_{45}$.

Galactin, as obtained by decomposing the lead salt by hydrogen sulphide, presents the appearance of a white (or, if slightly impure, a fawn-coloured), brittle, neutral, tasteless, non-crystalline mass, soluble in water, insoluble in strong spirit. It presents some of the characters of a peptone, and can be separated from the yellow whey of milk by the general alkaloidal reagents of Sonnenschein and Scheibler.⁴ After the galactin has been removed from the liquid, an alkaloidal colouring-matter (for which one of the authors proposed the name of *lactochrome*) remains in solution, and may be precipitated by means of nitrate of mercury. The simplest formula for this appears to be $HgOC_6H_{18}NO_6$. *Lactochrome*, as obtained

¹ Ueber den angebliehen Iodgehalt der Luft und verschiedener Nahrungsmittel. *Journal für praktische Chemie*, xcix., p. 198.

² Nouvelle Substance contenue dans le Lait. Extrait d'une Note de MM. E. Millon et Commaille. *Comptes Rendus*, lix., p. 301, 1864.

³ The ordinary solution for the estimation of urea is made by dissolving 100 grms. of pure mercury in half a litre of nitric acid ; a further quantity of acid is added until no red fumes are evolved ; the solution is evaporated to a syrup, and after adding enough nitric acid to prevent the formation of a basic salt, it is made up with distilled water to exactly 1400 c.c., each c.c. = '01 urea.

⁴ Sonnenschein's reagent is a nitric acid solution of phospho-molybdic acid. Scheibler's reagent is a solution of phospho-tungstic acid.

by careful decomposition of the mercury-compound, is in the form of bright red, orange, resin-like masses, softening at 100° C., freely soluble in water, very soluble in hot alcohol, but partially separating as the liquid cools. There appears little doubt that lactochrome is the cause of the yellow colour of milk whey, and also the colouring-matter of butter. Thudichum described some years ago,¹ under the name of butyro-luteine, the spectroscopic appearances of the colouring-matter of butter.

§ 119. Two bitter principles, possibly derived from substances eaten by the cow, were separated from the milk of Devonshire cows by one of us in 1879. A commercial gallon of milk, measuring 3800 c.c., was freed from casein, albumen, and alkaloidal matters by successive treatment in the way indicated in § 142, and the whey precipitated by solution of mercury nitrate, the mercury precipitate separated by filtration, and the excess of the mercury nitrate thrown out by hydrogen sulphide; the liquid was then made alkaline by ammonia, and lastly precipitated by tannin. The precipitate was washed, dried, and triturated with litharge and alcohol; and, finally, the mixture was exhausted by boiling alcohol, filtered, and the filtrate evaporated to dryness. The dry residue was now dissolved in water, digested with animal charcoal, filtered, and evaporated to dryness. A dark sticky extract was the result of these various and successive operations. On treatment by absolute alcohol, about 20 mgrms. of minute white crystals were obtained, which, on combustion with cupric oxide, were found to be non-nitrogenous, and (so far as a small quantity can be trusted) gave a formula closely agreeing with $C_2H_6O_{10}$. The larger portion of the extract was of a dark brown colour, very hygroscopic, and becoming quite fluid on exposure to the air; soluble in water in all proportions, insoluble in strong alcohol, and reducing copper solution on boiling, as well as chloride of gold at the ordinary temperature; it also rapidly reduced nitrate of silver on warming. The taste was woody and feebly bitter; the reaction was neutral. Two strictly concordant analyses gave numbers agreeing with the formula $C_{18}H_{18}O_{24}$.

Kreatinine appears to exist in normal milk, for we have separated, in the ordinary way, .059 of the chloride of zinc compound of kreatinine from 310 c.c. of milk; this corresponds to .0115 of kreatinine in 100 c.c. of milk. Commaille had already discovered that in a sample of milk which remained some time in a partially closed flask, appropriate treatment would give crystals yielding with chloride of zinc, or oxide of mercury, kreatinine reactions; and the presence of this substance in small quantities as a normal (perhaps constant) constituent of milk is now placed beyond a doubt. Urea has been found in almost all animal fluids, and in such small quantities as a milligramme per 100 c.c., it is rarely absent, but any marked percentage of urea is, of course, abnormal.

§ 120. Milk has a peculiar sweet odour, and the odoriferous principle may be separated by agitation with three or four times its volume of petroleum ether. MM. Millon and Commaille have recommended bisulphide of carbon for this purpose, but it is not quite so convenient a solvent.² In neither case is any of the milk-fat dissolved. Lactic acid is almost invariably present in cows' milk, and gradually increases in quantity. The amount of lactic acid obtained from milk which has been drawn from the cow within the hour is from .01 to .02 per cent.

Citric acid is also a normal constituent of milk.

¹ "Chemical Physiology," by J. L. W. Thudichum, M.D., London, 1872, p. 149.

² Analyse du Lait, by MM. Millon et Commaille. *Compt. Rend.*, t. lix., 396.

§ 121. Putting in one view the constituents of milk, so far as is known at the present time, they are as follows:—

AVERAGE COMPOSITION OF HEALTHY COWS' MILK.		Parts per cent. by weight.	
Milk-fat,	Butyrin,15	} 3.90
	Caproin,14	
	Caprylin,62	
	Caprin,07	
	Laurin,29	
	Myristicin,79	
	Palmitin,	1.00	
	Stearin,07	
	(Olein,	1.37)	
Casein,		3.00	
Albumen,		0.40	
Milk-sugar,		4.75	
Ash derived from the following salts of which the percentage composition accord- ing to Soldner =	Sodium chloride,	10.62	} 75
	Potassium chloride,	9.16	
	Mono-pot. phosphate,	12.77	
	Di-pot. phosphate,	9.22	
	Pot. citrate,	5.47	
	Di-mag. phosphate,	3.71	
	Magnes. citrate,	4.05	
	Di-calcium citrate,	7.42	
	Tri-calcium phosphate,	8.90	
	Calcium citrate,	23.55	
	(Lime combined with proteids,	5.13)	
Water,		87.20	

Besides the above there are the colouring-matter of milk (lactochrome), odorous principles, bitter principles (probably derived from the plants eaten), small quantities of other proteid bodies, kreatinine, urea, alcohol, and fluorides.

GASES OF MILK.

§ 122. One of the authors has investigated the gases contained in milk. Various samples of milk were clamped on to a mercurial pump, and the whole of the gas which they yielded pumped out and received in tubes, whence the gas was transferred to a gas apparatus and analysed. A litre of new milk, while fresh and warm from the cow, connected in this way to the pump, yielded 1.83 c.c. of gas, which on analysis had the following composition:—

	c.c.	Per cent.
Carbon dioxide, CO ₂ ,08	3.27
Nitrogen, N,	1.42	77.60
Oxygen, O,35	19.13

The proportion of oxygen to nitrogen was therefore nearly as 1 : 4. Another litre of good Devon commercial milk, on being subjected to the same process, yielded 3.468 c.c. of gas, the percentage composition of which was:—

	Per cent.
Carbon dioxide, CO ₂ ,	60.47
Nitrogen, N,	30.21
Oxygen, O,	9.30

This sample had been standing at a temperature of 15° C. for some hours; hence the diminution of oxygen and the increase of carbon dioxide. Various

other similar experiments were made, with the result of establishing the fact that a litre of fresh milk yields to the Sprengel pump from 1 to 3 c.c. of gas, in which there is always a certain percentage of carbon dioxide, and in which the relation of the nitrogen to the oxygen is very similar to the relation that exists in the air dissolved in water; but that fermentation, at any temperature in which fermentation is possible, at once commences, when the lactic ferment begins to use up the oxygen, and ultimately carbon dioxide is the only gas which can be obtained. This evolution of carbon dioxide is slow but continuous. As an example of this fermentation, one of the experiments may be cited:—100 c.c. of milk in which fermentation had begun, were suitably clamped to a Sprengel pump: on the first day there was a small percentage of nitrogen and a little oxygen; and on the second day a trace of oxygen; but on the succeeding days the gas consisted wholly of carbon dioxide, as follows:—

1st day,	1.123 c.c. of CO ₂
2nd day,	5.086 " "
3rd day,	19.540 " "
4th day,	7.621 " "
5th day,	7.370 " "
6th day,	9.023 " "
7th day,	1.780 " "
15th day,	21.350 " "
19th day,	4.370 " "

Giving a total of 77.263 c.c. of carbon dioxide in nineteen days, the temperature ranging between 14° and 19°.5. Milk previously deprived of dissolved air by the Sprengel pump, then confined over mercury and submitted to an atmosphere of oxygen, rapidly absorbs the oxygen, the place of which is taken by carbon dioxide, provided the temperature is a fermentation one—that is, above 9° and below 60°. This continuous absorption of oxygen was well shown in an experiment, in which a litre of milk was submitted to the action of a Sprengel pump, and in which it was found there was a continuous slow diffusion of air through the india-rubber connections. It has long been shown by Graham, that air thus finding its way through the minute pores of thick rubber is very highly oxygenised; yet all oxygen rapidly disappeared from the gas, and after the second day pure nitrogen and carbon dioxide could alone be obtained:—

1st day,	6.732 c.c. of gas.
	Percentage composition.
Carbon dioxide,	55.392
Nitrogen,	33.780
Oxygen,	10.828
	Ratio of oxygen to nitrogen as 1 to 3 nearly.
2nd day, total gas,	7.2 c.c.
Carbon dioxide,	49.73
Nitrogen,	49.73
Oxygen,54
3rd day,	4.863 c.c.
Carbon dioxide,	61.06
Nitrogen,	38.94
4th day—	
Carbon dioxide,	87.98
Nitrogen,	12.02
5th day—	
Carbon dioxide,	91.52
Nitrogen,	8.48

'FORE' MILK.

§ 123. If an animal is fractionally milked—that is, the whole of the milk received into three or four different vessels—it will be found that, on analysis, the several portions exhibit some difference of composition, more observable in the last and the first, than in the intermediate portions. This difference mainly affects the fat, the first portions of the milk yielding, as a rule, but little fat, while the latter portions, called 'strippings' (in speaking of cows' milk), contain an excess of milk-fat. Thus, in a Devon cow milked in this way for the purpose of analysis, the writers found the two extreme portions to have the following composition:—

	Fore Milk.	Strippings.
Specific gravity,	1·0288	1·0256
Milk-fat,	1·166	5·810
Casein,	2·387	4·304
Albumen,	1·830	·975
Peptones,	·381	·545
Milk-sugar,	3·120	3·531
Ash,	·797	·895
Water,	90·319	83·940
Common salt in ash,	·340	·267

In another experiment a Guernsey cow yielded the following:—

	Fore Milk.	Strippings.
Specific gravity,	1·040	1·023
Milk-fat,	·357	5·946
Casein,	4·708	3·435
Albumen,	·451	·860
Peptones,	·267	·156
Milk-sugar,	4·943	5·280
Ash,	·874	·929
Water,	88·400	83·394
Common salt in ash,	·100	·098

Dairymen are perfectly aware of the poorness of fore milk in fat, and more than once fraudulent milkmen have endeavoured to defend themselves by having recourse to the strange expedient of partially milking a cow before such functionaries as aldermen or policemen, and delivering with all formalities the sample to be analysed. The analyst, not knowing its history (for in such cases it is transmitted as an ordinary commercial milk), and finding it on analysis deficient in fat, certifies accordingly, and until the matter is explained suffers in reputation. Such tricks were once rather common, but have been so fully exposed that they are not likely to re-occur.

This difference in the first and last milkings is not confined to cows' milk, but has also been observed in the milk of other animals. Peligot had an ass milked in three successive portions, and found as follows:—

	1	2	3
Milk-fat,	·96	1·02	1·52
Milk-sugar,	6·50	6·48	6·50
Casein,	1·76	1·95	2·95
Total solids,	9·22	9·45	10·97
Water,	90·78	90·55	89·03

Reiset¹ has also found a considerable difference in the percentage of total

¹ J. Reiset: *Annales de Chimie et de Physique*, 3 ser., xxv., 1849.

solids in human milk in fractions taken before the child was applied to the breast and after.

	Before Suckling. Total Solids per cent.	After Suckling. Total Solids per cent.
1.	10.58	12.93
2.	12.78	15.52
3.	13.46	14.57

It has been considered that this difference is merely due to the effect of a physical cause; that, in short, as regards cows' milk, the milk already secreted is in the same state as if it stood in a vessel, and the fat rising to the top is, of course, drawn last. This explanation cannot be altogether true, for the same phenomenon is observed in human milk, and here the breasts are horizontal, or nearly so. It is more probable that during the act of milking secretion goes on, and it would seem that the fatty contents of the milk-producing cells are set free before the more watery and albuminous. Hence, the strippings are, as the most recent portions of the whole secretion, rich in fat. This view is supported by an experiment of Reiset, in which it was proved that the longer the time elapsing between the partial milkings, the less the percentage of solids.

HUMAN MILK.

§ 124. Woman's milk (1) has been long an object of research, and numerous analyses of it are scattered through scientific literature. These analyses, in their quantitative results, show considerable discrepancies, so that we must either adopt the supposition that human milk is very variable, or, what is more probable, that the samples taken did not represent the average secretion. From experiment, the authors have come to the conclusion that it is impracticable with any mechanical appliances to obtain a complete sample of human milk. In civilised life the nervous system assumes such a high and delicate state of organisation, that the secretion is far more dependent on the presence and contact of the offspring than among animals. Hence, samples of human milk taken by breast pumps, or other exhaust apparatus, can only be considered partial samples; and a study of partial sampling in the case of cows' milk (p. 206) has taught us how very widely the quantities of the fatty constituents in such samples differ from one taken from the whole bulk. Woman's milk contains milk-fat which has not been obtained in quantities sufficient for accurate investigation, and a knowledge of its exact composition is still a desideratum. It, however, certainly contains butyric acid, for one of the authors has succeeded in isolating a sufficient quantity of butyric acid from saponified human milk-fat to identify it satisfactorily. Milk-sugar,¹ casein, albumen, peptones, and a colouring-matter, with mineral substances, are also constituents of woman's milk.² The casein, like that of the ass, is peculiar in not separating in flocculent masses by the processes recommended in § 142, and the analyst is under the necessity of adopting a different process. This

¹ Carter and Richmond state that the sugar has a specific rotatory power of $[\alpha]_D = 48.7^\circ$. They are of opinion that more than one sugar is present.

² Human milk on being shaken up with ether parts with its fat, the globules dissolving in the ether, the fluid therefore clears up and separates into two layers—an upper ethereal containing the fat in solution, and a lower layer consisting of a solution of the casein, albumen, and salts. This peculiarity is not shared by the milk of the herbivora. See P. Radenhausen, *Zeit. f. physiologische Chemie*, 5, 13-30.

difference is all-important ; for in artificial feeding with cows' milk, as soon as the milk reaches the stomach, the milk, in popular language, 'curdles,' and is often rejected by vomiting.

One of the earliest exact analysis of human milk was made by Meggenhofen¹ in 1826. His treatise scarcely appears to be known, yet it contains pretty well all that is known of the composition of human milk. The total solids of human milk Meggenhofen determined from twelve samples, the highest of which is 13·38, and the lowest 9·25 per cent., the mean being about 12 per cent. Probably for the first time Meggenhofen determined the albumen separately from the casein, and also weighed *materiæ animalis tinctura gallarum animalis precipitata*. His view of the composition of human milk may be fairly stated thus—

	Per cent.
Milk-fat,	2·90
Casein,	2·40
Albumen,	·57
Albuminoid precipitated by gallic acid,	·10
Sugar,	5·87
Ash,	·16
Water,	88·00

With regard to other constituents, urea is often present ; there is also an odorous principle. Human milk decomposes similarly to cows' milk, and yields similar gaseous and other products.

§ 125. *Milk of the Ass* (2).—One of the authors has investigated the milk of the ass. Milk was obtained under his personal superintendence from asses kept and fed in London dairies for the purpose of supplying the demand that still exists for asses' milk. The animals were fed on a uniform diet of bran, hay, and oats. The yield of each milking was carefully noted, and the ass in each case milked dry. It would appear that the milk of the ass under these circumstances has a very uniform composition, the differences observed being quite unimportant. The yield for commercial purposes appears not to exceed 3 pints, and to average about 2½ pints daily. More than this is doubtless secreted, but some of it is used by the foal. In no case did a single milking yield half a litre (three-fifths of a pint), but usually between 300 and 400 c.c.

The fat contains 5 per cent. of butyric acid, equal to 6·6 per cent. of butyrin ; it is probably very similar to butter-fat. After the precipitation of casein, albumen, lactochrome, and peptones, there yet remain principles precipitable by tannin. As in human milk, the casein is not readily precipitated, but remains suspended in a state of fine division, however far lactic fermentation may have progressed.

§ 126. *Milk of the Goat* (3).—The milk of the goat, as a rule, contains more cream than that of the cow, and rather less albuminous matter.

§ 127. *Milk of the Mare* (4).—The milk of the mare closely resembles in its constituents the milk of the cow : the casein, the sugar, and the fat being very similar, if not identical. M. J. Duval² asserts that he has discovered in the milk of the mare a new acid, to which he has given the name of equinic, and which crystallises in groups of little needles ; it is not volatile without decomposition, in odour fragrant. It is combined with a base volatilised by heat, which the author considers a base of the ammonia type. Its reactions with silver nitrate, ferric chloride and auric chloride distinguish it from hippuric acid. No analyses are, however, given.

¹ *Dissertatio Inauguralis Indagationem Lactis Muliebris Chemicam*. C. Aug. Meggenhofen. Frankfort, 1826.

² *Comptes Rendus*, lxxxii., 419, 1876.

The mean composition of asses' milk and the milk of various other animals is given in the table:—

	Specific gravity.	Total solids.	Solids not fat.	Fat.	Sugar.	Casein.	Albumen.	Aash.	Peptones.		
1. Human, average,	1·0313	13·267	9·137	4·131	6·936	1·995	·201	..	Leeds. ¹		
" maximum,	1·0353	16·66	12·09	6·89	7·92	4·86	·37	..	"		
" minimum,	1·0268	10·91	6·57	2·11	5·40	·85	·13	..	"		
" mean compos.	11·8	..	3·3	6·8	1·5	·2	..	Richmond. ²		
2. Ass,	8·83	7·81	1·02	5·5	1·09	·7	·42	·1	A. W. Blyth.	
3. Goat,	12·46	8·26	4·2	4·0	3·0	·62	·56	·08	..	
" rich,	1·0357	17·98	10·96	7·02	5·28	4·87	..	1·01	..	Voelcker. ³	
" poor,	1·030	15·52	9·41	6·11	4·68	3·94	..	·79	..	"	
" colostrum,	1·030	16·49	9·15	7·34	5·19	3·19	..	·77	..	"	
4. Mare,	9·94	7·85	1·09	6·65	1·89	..	·31	..	Vieth.	
"	11·2	8·7	2·5	5·50	2·19	·42	·5	·09	A. W. Blyth.	
5. Sheep,	1·040	17·73	12·43	5·3	4·2	6·1	1·00	1·00	·13	"	
6. Camel,	1·042	13·06	10·16	2·9	5·66	3·84	·66	Dragendorff.	
Llama,	10·45	7·30	3·15	5·6	·90	·8	Doyere. ⁴	
7. Hippopotamus,	9·02	4·51	4·51	4·40	small	·11	Gunning.	
8. Sow,	15·96	11·41	4·55	3·13	7·23	1·05	König.	
9. Bitch,	1·035	23·40	13·83	9·57	3·19	5·53	4·38	·73	
10. Cat,	18·377	15·044	3·333	4·911	9·548	·583	Commaille.	
Mule,	8·50	6·91	1·59	4·80	1·64	·38	5	
Rabbit,	30·50	20·05	10·45	1·95	15·54	2·56	5	
Elephant,	32·15	12·58	19·57	8·84	3·09	·65	5	
Porpoise,	61·59	13·09	48·50	1·33	11·19	·57	5	
Whale,	51·33	7·66	43·67	..	7·11	·46	5	
Gamoose or water-buffalo,	5·56	5·41	3·26	·60	1·03	Richmond and A. Pappel.
Buffalo,	17·07	..	7·46	4·21	4·59	·81	Fleischmann.	

¹ A. R. Leeds (*Chem. News*, iv. pp. 263-267, 280, 281) from eighty-four samples.
² Richmond (*Dairy Chemistry*, 1899, p. 325) deduces these figures from Leeds', Pfeiffer's, Luff's, Johanssen's, Lehmann's, Carter's, and his own figures.
³ *Bied. Centr.*, 1881, p. 858.
⁴ *Ann. de l'Inst. Agron.*, 1852, p. 251.
⁵ Richmond, *Dairy Chemistry*, 1899, p. 323.

§ 128. *Milk of the Sheep* (5).—Sheep's milk ¹ is remarkable for its high specific gravity, and the large amount of solid matter which it contains; the specific gravity ranges from 1·038 to 1·041, and the total solids may rise as high as 19 per cent.

The casein behaves similarly to the casein of cows' milk, and separates easily by dilution, acidulation with acetic acid, etc. (see p. 227). The fat yields 5 per cent. of its weight of butyric acid, and is probably of similar composition to the milk-fat from cows' milk.

§ 129. *The Milk of the Camel* (6).—Chatin has analysed the milk of the camel. He describes ² it as perfectly white in colour, and possessing globules smaller but more numerous than those in cows' milk, the diameter being on an average one-half. Specific gravity, 1·042. It appears to be rather richer in milk-sugar and casein than cows' milk. Dragendorff ³ has also analysed camels' milk, and gives the figures in the table.

¹ An analysis, by Voelcker, of rich samples of ewes' milk shows that the fat may attain 12·78 per cent. *Bied. Centr.*, 1881, 858.
² Sur le Lait de la Chamelle à deux Bosses, par M. Chatin. *Journal de Pharmacie et Chimie*, t. i., 4 ser., p. 264.
³ *Zeit. f. Chemie*, 1865, s. 735.

§ 130. *Milk of the Llama*.—The figures given in the table are from three analyses by Doyère.

§ 131. *Milk of the Hippopotamus* (7).—There are few opportunities of analysing the milk of this enormous animal, as it is fierce when it suckles its offspring. A sample of the milk was, however, investigated by Gunning.¹ He describes it as of an acid reaction, and under the microscope showing larger globules than that of other animals. The young hippopotamus sucks under water, and can remain there for a much longer time than the adult animal. The secretion of milk is excessive in quantity, and escapes from the distended teats in streams, which make the water around the animal quite opaque.

§ 132. *Milk of the Sow* (8).—The mean of eight analyses collected by König² of sow's milk, is given in the table.

There are also two analyses of the milk of a sow investigated by Filhol and Joly; the animal was fed on horse-flesh, a diet far from natural; under this diet was secreted a highly albuminous fluid, containing but little sugar. Specific gravity 1·044.

	Per cent.	
Albumen,	12·89	21·0
Fat,	6·6	5·4
Sugar,	0·5	1·2
Extractives and salts,	3·01	4·3
Water,	77·0	68·1
Total solids,	23·0	31·9
Solids not fat,	16·4	26·5

§ 133. *Milk of the Bitch* (9).—The milk of the bitch is highly charged with albuminous solids, and is of a specific gravity ranging from 1·034 to 1·036. It has been investigated by Simon, Dumas, Filhol and Joly, Talmatescheff, Bensch, Scubotin, and others.

§ 134. *Milk of the Cat* (10).—The milk of the carnivora generally has the peculiarity of having the milk-sugar almost entirely replaced by lactic acid, and hence the milk invariably possesses an acid reaction. An analysis of the milk of a cat by Commaille is given. The milk was taken twenty-four hours after kitting; it was feebly acid.

§ 135. *Milk-like Secretions of Birds and Plants*.—It is usually held that mammals alone secrete milk, but this is by no means certain; for during the latter portion of the incubation-period, as well as more profusely for a little while after the young birds are hatched, the pigeon secretes a nutritious albuminous fluid in her crop, which is supposed to be used for the purpose of feeding the young birds. According to Lecomte's analysis this secretion contains,

Casein and salts,	23·23
Fat,	10·47
Water,	66·30

Such milk-like secretions are by no means confined to the internal mucous membranes of birds. Jonge³ has made a most valuable research on the secretion of the glands known to anatomists as *Glandula uropygii*, situated at the tail of the common goose. The secretion was obtained in sufficient quantity for a complete qualitative and quantitative analysis, and

¹ *Gazetta Chim. Italiana*, 1871, p. 255.

² *Op. cit.*

³ "Ueber das Secret der Talgdrüsen der Vögel und sein Verhältniss zu den fetthaltigen Hautsecreten der Saugthiere, insbesondere der Milch," von D. de Jonge. *Zeitschrift für physiol. Chemie*, von F. Hoppe-Seyler. Strasburg, 1879.

although the analysis was not quite so complete as if a larger quantity had been obtainable, it fairly shows that there is a considerable analogy between milk and this secretion, the most marked difference being that no trace of milk-sugar could be found. The analyses of two samples were as follows:—

	1.		2.	
Total solids,		391·93		415·34
Water,		608·07		584·66
Albumen and nuclein,	179·66		127·63	
Compounds insoluble in absolute ether,	186·77		247·08	
Alcoholic extract,	10·90		18·31	
Water extract,	7·53		11·31	
Ash,	7·07	{ Sol. 3·71 }	11·07	{ Sol. 7·71 }
		{ Insol. 3·36 }		{ Insol. 3·36 }
<i>In ether extract,</i>	1.		2.	
Cetyl-alcohol,	74·23		104·02	
Oleic acid,	6·48		...	
Lower acids,	3·73		14·84	
Lecithin,	2·33		...	

In the vegetable kingdom, numbers of trees or plants yield a white fatty secretion, popularly called milk, though, as a rule, such fluids have no right to this title, being totally different in composition and properties. A very remarkable exception to this assertion is, however, met with in the 'milk tree' (*Brosimum galactodendron*), to be found in Central America.

This tree, on incision, yields an abundance of a thickish feebly acid fluid, coagulating on exposure to the air. M. Boussingault has analysed this juice, and considers it perfectly analogous to ordinary milk, since it contains a fatty principle, an albuminous principle, a sugar, and phosphates. The exact composition of these different matters has, however, not been determined. Boussingault's general analysis is as follows:—

Fatty saponifiable matters,	35·2
Sugar, and substances analogous,	2·8
Casein, albumen,	1·7
Earths, alkalies, phosphates,	·5
Substances not estimated,	1·8
Water,	58·0

This milk is used largely as a food in the regions where the tree grows.

ABNORMAL MILKS.

§ 136. Milk which deviates from the natural secretion, the animal suffering from no disease, and milk secreted under unnatural conditions, may be conveniently classed as 'abnormal.' (Milk derived from the unhealthy will be considered in another section.) Instances of *healthy* cows giving milk differing essentially from ordinary milk are very few. One such, however, is recorded by Mr. Pattinson, who analysed the milk of a roan cow, which only gave 2 per cent. of albuminoids, and yielded no less than 4 grms. per litre of common salt. The animal is stated to have been in good health. Instances of marked deviations from the ordinary standard are to be found in *The Analyst* of Jan. 1, 1893. (See also page 231.)

The newly-born human infant almost constantly secretes a fluid in the mammae, and adult males have not only secreted milk, but that in abundance enough to suckle.

Females also, both human and animal, occasionally secrete milk without having been previously pregnant. With regard to the milk secreted by infants, there is some doubt about its real nature. Kölliker does not view it as a true milk, but considers its appearance connected with the formation of the mammary glands. Sinety, on the other hand, upon anatomical grounds, considers it a true lacteal secretion. It is probably imperfect milk loaded with leucocytes, and this is the more likely, as Billard (*Traité des Maladies des Enfants nouveaux nés*, 3me édition, 1837, p. 717) notices that it frequently ends in abscess.

Schlossberger gives an imperfect quantitative analysis of a sample of milk¹ obtained by squeezing the breasts of a newly-born infant—a male. In the course of a few days about a drachm was obtained. The following was the result of the analysis:—

	Per cent.
Water,	96·75
Fat,	·82
Ash,	·05
Casein, sugar, and extractives,	2·83

Sugar reaction strong.

The most complete analysis we yet possess of such milk is one by V. Gesner, which is given in the following table with other less perfect analyses:—

	1 ²	2 ³	3 ⁴
Milk-fat,	1·456	·82	1·40
Casein,	·557	...	2·80
Albumen,	·490	...	
Milk-sugar,	·956	...	6·40
Ash,	·826	·05	
Water,	95·705	96·30	79·40
Total solids,	4·295	3·70	10·60

Joly and Filhol have recorded the case of an old lady, 75 years of age, who suckled successfully her grandchild.⁵ Similar instances have been recorded in dogs, and we fortunately possess one or two analyses which show that the fluid is certainly milk. Thus Filhol and Joly give the following analysis of the milk derived from a bitch which had no connection with a male:—

Specific gravity,	1·069
Total solids,	29·00
Fat,	2·20
Sugar,	·32
Albumen,	23·20

The ash, on analysis, gave the following percentages in 100 parts:—

Chloride of sodium,	65·10
Chloride of potassium,	3·88
Calcic phosphate,	27·75
Sodic phosphate,	1·40
Sodic carbonate,	1·87

Traces of magnesian and other phosphates.

Men before now have suckled children. Humboldt⁶ relates the case of Francisco Lozano, whom he saw, and whose case he carefully investigated; and it appears established that this man did secrete from his breasts a nutrient fluid on which his infant son lived for many months, it is said, indeed, a whole year. The curious in such matters may consult the references given in the footnote for additional cases.⁷

¹ Untersuchung der sogenannte Hexenmilch, J. Schlossberger, *Annalen der Chemie u. Pharmacie*, b. 87, 1852.

² *Jahrb. f. Kinderkrankheiten*, N. F., Bd. ix., § 160.

³ Schlossberger u. Hauff, *Ann. Chem. Pharm.*, Bd. xcvi., p. 68.

⁴ Gabler u. Quevenne, *op. cit.*

⁵ "Recherches sur le Lait," iii., Bruxelles, 1856.

⁶ Humboldt: "Voyage aux Régions Equinoxiales du Nouveau Continent."

⁷ Robert, Bishop of Cork: Letter concerning a Man who gave Suck to a Child, *Phil. Trans.*, 1741, No. 461, t. xli., p. 813. Franklin: "Narrative of a Journey to the Shores of the Polar Sea," 1819, p. 157. Cobbold: Milk from the Male Mamma, *Monthly*

Instances have also been known of a like kind among animals. Schlossberger has analysed the milk derived from a he-goat (*Annalen der Chemie u. Pharmacie*, 1844):

Milk-fat,	26.50
Casein, with salts soluble in alcohol,	9.60
Sugar, with salts soluble in alcohol,	2.60

The ash was .782 per cent.—viz., .325 soluble in water, .457 insoluble. Occasionally the female mamms after confinement have continued to yield milk, although the infant has either been dead or nourished otherwise. In such cases the milk deviates from its normal composition, and is, for the most part, highly albuminous. In a case of this kind recorded by Filhol and Joly,¹ three analyses of the milk were made as follows, at different dates, about a week apart:—

	1.	2.	3.
Specific gravity,	1.039	1.025	1.023
Total residue,	21.50	18.30	18.63
Milk-fat,	5.00	6.15	7.80
Sugar,	2.19	1.27	3.50
Albumen,	12.96	9.00	5.65
Extractives and salts,	1.35	1.88	1.68
Water,	78.50	81.70	81.37

Casein was entirely absent.

The composition of the ash was as follows:—

	Per cent.
Chloride of sodium,	73.10
Chloride of potassium,	traces.
Calcic phosphate,	23.40
Sodic phosphate,80
Sodic carbonate,	1.89
Magnesia and ferric phosphates,81

THE GENERAL EXAMINATION AND ANALYSIS OF MILK.

§ 137. The general examination and analysis of milk may be conveniently divided into three sections—

- A. The microscopical and biological examination.
- B. The analysis of the normal constituents of milk.
- C. The analysis of milk for the purpose of detecting adulteration.

A. MICROSCOPICAL AND BIOLOGICAL EXAMINATION OF MILK.

§ 138. A mere chemical analysis is incomplete and insufficient in itself, and should in all cases be preceded or supplemented by a careful and painstaking microscopical examination. Normal milk, viewed under the microscope, presents for the most part a multitude of fat globules floating in a clear fluid. The globules of human milk measure in diameter from .002 to .005 mm.; those in the milk of the cow, from .00062 to .00039 inch [.0016 to .01 mm]. These fat globules are of two kinds. By far the most numerous are evidently drops of fluid fat; but there are occasionally to be seen others which would appear to consist of solid fat, for they are rougher on the surface, and less soluble in ether, characteristics which they lose on warming, becoming like liquid-fat globules. In human milk, and, to a certain extent, in cows' milk, there are also as normal constituents, but in sparse quantity—

- (1.) Fatty drops having a half moon-shaped, finely granular substance;

Journal of Med. Science, 1854; t. xviii., p. 271. Morgagni: *Adversaria Anatomica Omnia* (V. *Animadversio*, i., p. 3).

¹ *Comptes Rendus*, t. xxxvi., p. 571, 1853.

(2.) Clear cells enclosing one or two fatty drops, and an eccentric nucleus;

(3.) Round clear bodies, easily coloured by eosin and picrocarmine. These last Heidenhain considers to be free nuclei.¹

In the colostrum, or milk drawn the first few days after parturition, there are present other elements—viz., the so-called ‘colostrum cells.’ Some of these consist of a number of small and large fat globules, held together by a hyaline tissue or membrane, swelling on the addition of acetic acid or alkalis, and only slowly coloured by aniline red. There are other granular cells coloured at once by the same reagent. If the milk is taken fresh and warm, and a minute drop examined on a Strecker’s warm stage,² and kept at a temperature of 38°, the corpuscles will exhibit amœboid movements, perfectly similar to those which have been noticed in the white corpuscles of the blood. Indeed, it is almost certain that the colostrum cells are no other than the white corpuscles of the blood, infiltrated with milk-fat, for Heidenhain, having injected into the dorsal lymph vessel of the frog 1 c.c. of fresh milk, after 48 hours found the white corpuscles loaded with milk-fat, and in no respect distinguishable from colostrum cells. In abnormal milk may be detected pus or blood, or sometimes both. If the pus is derived from inflammations within the mammæ, and has been mixed with the milk before milking, the pus cells become infiltrated with milk-fat, and are difficult to distinguish from colostrum granules; but if derived from ulcers on the teats, they have the usual appearance of pus cells. The pus cells, like the colostrum cells, and the mucus corpuscles, are all different forms of white blood-corpuscles [leucocytes], and when placed on the warm stage exhibit amœboid movements. Pus cells, as usually observed, are spheroidal, granular, and colourless, measuring from about 1-2500 to 1-3000th of an inch in diameter. On treatment with dilute acetic acid, the cells clear up, and show two, three, or four nuclei. Blood, in small quantity, gives a pinkish colour to milk; if a large amount be present, it sinks to the bottom in red flocculent masses, which soon, from being deoxidised by the milk, acquire a tint varying from a red more or less dark, to a shade almost black. In small quantities reliance must be placed on the microscopic appearance of the blood-discs, which are wholly unlike any cell found in normal milk. The red blood-discs of the cow are like those of the human subject—little circular, biconcave, flattened discs, measuring on an average 1-4000th part of an inch. Human blood-discs have an average diameter of 1-3500th inch. By the aid of the micro-spectroscope, the absorption-bands may also be seen. These are, in oxidised blood, two bands between D and E, the one close upon the red being narrower, darker, and better defined than the one nearer to the green; with deoxidised blood, only one band is seen, between D and E. On treating the blood with oxygen, or shaking it up with air, the two bands re-appear.

The biological examination of milk is treated of under the sections dealing with milk secreted by the unhealthy and with the decomposition of milk.

¹ R. Heidenhain : “Handbuch der Physiologie.” Herausgegeben von Dr. L. Hermann. Leipzig, 1880.

² In default of Strecker’s stage, a plate of copper, having a central aperture and a thick straight wire, some inches in length, may be used. The plate is kept at the desired temperature through heating the wire by means of a spirit lamp.

B. THE ANALYSIS OF THE NORMAL CONSTITUENTS OF MILK.

§ 139. In the analysis of the normal constituents of milk, the following determinations should be made:—

- | | |
|---|--|
| <p>a. Sp. gravity at 15.5° C.
 b. Total solids.
 c. Fat.
 d. Lactose or milk-sugar.</p> | <p>e. Proteids, { Casein and
 { albumen.
 f. Mineral matters or ash.</p> |
|---|--|

In addition to the above, it may sometimes be desirable to further examine the fat and proteids of the milk, and to make an examination for substances such as citric acid, and for others which cannot be said to be normal constituents of milk, *e.g.*, alcohol, urea, metals, and so forth.

§ 140. One of the first accurate processes for the general analysis of milk was published in 1853, by MM. Vernois and A. Becquerel.¹

A small quantity [30 grms.] was taken, dried, exhausted with ether, burnt up to an ash; the sugar obtained *à saccharimètre* from the whey, the casein being first separated by coagulation by acetic acid, and then estimated by difference.

Mr. Wanklyn, by his work on milk analysis,² revived the more accurate method of using comparatively small quantities for analysis, thus avoiding very considerable error, from the risk of large quantities decomposing by prolonged heating. He advocated the use of platinum dishes, and supported strongly the doctrine of the fairly constant character of the non-fatty constituents of milk, dividing the total milk solids into two divisions: the one, *milk-fat*, the other, *solids not fat*.

Whatever modifications have been since introduced in the methods for the analysis of milk, the general process is still on the principles advocated by Mr. Wanklyn.

(a.) *Specific Gravity*.—The specific gravity is first taken. This may be done with fair accuracy by an hydrometer, and still more correctly by a Westphal's balance or a Sprengel tube.

It is to be noted that milk when drawn from the udder contains many air bubbles, and the specific gravity is low, consequently the specific gravity should not be taken until at least one hour has elapsed. This phenomenon was first observed by Recknagel, and has been confirmed by Vieth, Bourcart, and Richmond. The specific gravity of milk must be corrected to 15.5° C.

Vieth has constructed a useful table (see Table XVIIIa.) for this purpose.

(b.) *Total Solids*.—The specific gravity having been obtained, exactly 10 c.c., or better 5 c.c., are transferred by means of a pipette to a platinum dish, and submitted to the heat of a water-bath, until the contents cease to lose more than 1 mgrm. in one hour; this usually takes from three to five hours. When the residue is perfectly dry, it is at once weighed, and the results expressed in percentage by weight. The weight of the 10 c.c. is known from the specific gravity already taken. Thus, supposing a milk of 1.032 specific gravity to give a total residue from 10 c.c. of 1.423 grm.: since the specific gravity has shown that 100 c.c. of the milk weighed 103.2 grms., it follows that 10 c.c. must weigh 10.32 grms. Hence, 10.32 grms. have yielded a residue of 1.423 grms., which is 13.78 per cent.

Some analysts, *e.g.*, Allen, prefer the use of flat porcelain dishes, the

¹ *Comptes Rendus*, xxxvi., p. 187, 1853.

² "Milk-Analysis." By J. A. Wanklyn. Lond. 1874.

TABLE XVIIIa.—FOR CORRECTING THE SPECIFIC GRAVITY OF MILK ACCORDING TO TEMPERATURE.

		DEGREES OF THERMOMETER (<i>Fahrenheit</i>).																				DEGREES OF THERMOMETER (<i>Fahrenheit</i>)										
DEGREES OF LACTO-METER.	DEGREES OF LACTO-METER.	45	46	47	48	49	50	51	52	53	54	55	56	57	58	59	60	61	62	63	64	65	66	67	69	70	71	72	73	74	75	
20	19.0	19.0	19.0	19.1	19.1	19.2	19.2	19.3	19.4	19.4	19.5	19.6	19.7	19.8	19.9	19.9	20.0	20.1	20.2	20.2	20.3	20.4	20.5	20.6	20.9	21.0	21.1	21.2	21.3	21.5	21.6	20
21	19.9	20.0	20.0	20.1	20.2	20.2	20.3	20.3	20.3	20.4	20.5	20.6	20.7	20.8	20.9	20.9	21.0	21.1	21.2	21.3	21.4	21.5	21.6	21.8	22.0	22.1	22.2	22.3	22.4	22.5	21	
22	20.9	21.0	21.0	21.1	21.2	21.2	21.3	21.3	21.3	21.4	21.5	21.6	21.7	21.8	21.9	21.9	22.0	22.1	22.2	22.3	22.4	22.5	22.6	22.8	23.0	23.1	23.2	23.3	23.4	23.5	22	
23	21.9	22.0	22.0	22.1	22.2	22.2	22.3	22.3	22.3	22.4	22.5	22.6	22.7	22.8	22.9	23.0	23.1	23.2	23.3	23.4	23.5	23.6	23.7	23.8	24.0	24.1	24.2	24.3	24.4	24.5	23	
24	22.9	23.0	23.0	23.1	23.2	23.2	23.3	23.3	23.3	23.4	23.5	23.6	23.7	23.8	23.9	24.0	24.1	24.2	24.3	24.4	24.5	24.6	24.7	24.9	25.0	25.1	25.2	25.3	25.4	25.5	24	
25	23.8	23.9	24.0	24.0	24.1	24.1	24.2	24.2	24.3	24.4	24.5	24.6	24.7	24.8	24.9	25.0	25.1	25.2	25.3	25.4	25.5	25.6	25.7	25.9	26.0	26.1	26.2	26.4	26.5	26.6	25	
26	24.8	24.9	25.0	25.0	25.1	25.1	25.2	25.2	25.3	25.4	25.5	25.6	25.7	25.8	25.9	26.0	26.1	26.2	26.3	26.4	26.5	26.6	26.7	26.8	27.0	27.1	27.2	27.3	27.4	27.5	26	
27	25.8	25.9	25.9	26.0	26.1	26.1	26.2	26.2	26.3	26.4	26.5	26.6	26.7	26.8	26.9	27.0	27.1	27.2	27.3	27.4	27.5	27.6	27.7	27.8	28.0	28.1	28.2	28.3	28.4	28.5	27	
28	26.7	26.8	26.8	26.9	27.0	27.0	27.1	27.1	27.2	27.3	27.4	27.5	27.6	27.7	27.8	27.9	28.0	28.1	28.2	28.3	28.4	28.5	28.6	28.7	28.8	29.0	29.1	29.2	29.3	29.4	28	
29	27.7	27.8	27.8	27.9	28.0	28.0	28.1	28.1	28.2	28.3	28.4	28.5	28.6	28.7	28.8	28.9	29.0	29.1	29.2	29.3	29.4	29.5	29.6	29.9	30.0	30.1	30.2	30.3	30.4	30.5	29	
30	28.6	28.7	28.7	28.8	28.9	29.0	29.1	29.1	29.2	29.3	29.4	29.5	29.6	29.7	29.8	29.9	30.0	30.1	30.2	30.3	30.4	30.5	30.7	30.9	31.1	31.2	31.3	31.5	31.6	31.8	30	
31	29.5	29.6	29.6	29.7	29.8	29.9	30.0	30.1	30.2	30.3	30.4	30.5	30.6	30.8	30.9	31.0	31.1	31.2	31.3	31.4	31.5	31.7	31.8	32.0	32.2	32.4	32.5	32.6	32.8	33.0	31	
32	30.4	30.5	30.5	30.6	30.7	30.9	31.0	31.1	31.2	31.3	31.4	31.5	31.6	31.7	31.9	32.0	32.2	32.3	32.5	32.6	32.7	32.9	33.0	33.2	33.3	33.4	33.6	33.7	33.9	34.0	32	
33	31.3	31.4	31.4	31.5	31.6	31.8	31.9	32.0	32.1	32.3	32.4	32.5	32.6	32.7	32.9	33.0	33.2	33.3	33.5	33.6	33.8	33.9	34.0	34.2	34.3	34.5	34.6	34.7	34.9	35.0	33	
34	32.2	32.3	32.3	32.4	32.5	32.7	32.8	33.0	33.1	33.2	33.3	33.5	33.6	33.7	33.9	34.0	34.2	34.3	34.5	34.6	34.8	34.9	35.0	35.2	35.3	35.5	35.6	35.8	36.0	36.3	34	
35	33.0	33.1	33.2	33.3	33.4	33.6	33.7	33.9	34.0	34.2	34.3	34.5	34.6	34.7	34.9	35.0	35.2	35.3	35.5	35.6	35.8	35.9	36.1	36.2	36.4	36.5	36.7	36.8	37.0	37.3	35	

advantages being that the heat is not so intense when burning to an ash, and that they are more constant in weight than platinum.

Richmond points out that 1 grm. of milk spread over a large surface dries rapidly and completely.

The Association of Official Agricultural Analysts of America have adopted the following two methods:—

(1.) Heat to a constant weight at 100° C. one to two grms. of milk in a tared flat dish, not less than 5 centimetres in diameter. If desired, from 15 to 29 grms. of pure dry sand may be previously placed in the dish.

(2.) *Babcock Asbestos Method.*—A hollow perforated metal cylinder, 60 mm. long, 20 mm. in diameter, and closed 5 mm. from one end by a disc of the same material, is loosely filled with from 1.5 to 2.5 grms. of freshly-ignited woolly asbestos, free from fine or brittle material. The perforations are about 0.7 mm. in diameter, and the same distances apart.

The weight is taken of the metal tube thus packed. A definite weight, say 4 grms. of milk, is taken and introduced on to the asbestos. The whole is dried to constant weight, and the residue may be conveniently used for the determination of fat by exhaustion with volatile solvents.

Vacuum Processes.—The authors find that total solids may be estimated in the following way:—5 c.c. of milk are placed in a small flask; this flask is connected with a second, containing strong sulphuric acid. The flasks are made vacuum by means of a good mercury pump; the milk, on gently heating, loses all water at as low a temperature as 49°. The solids are dry under two hours, but the weight is generally 0.5 per cent. higher than by the ordinary processes.

A modification of this process is the *Direct Determination of the Water.*—In most analyses the water is inferred from the loss; it may, however, occasionally be necessary to estimate it directly. This can be readily done as follows:—5 c.c. of milk are placed in a small flask; a piece of tubing is sealed at one end, and graduated into c.c.'s; it will only be necessary to mark it at 4.5 and at 5.5, graduating it, between these numbers, into 10ths. The tube is now bent twice at right angles, and connected by a caoutchouc cork to the flask; another narrow tube goes to the mercury pump; the flask is now exhausted of air; and by applying a flame to the connecting tube, the tube is drawn out and sealed. By now plunging the limb of the graduated sealed tube into ice and salt, and gently warming the flask, the milk boils, the water is all condensed in the limb, and the amount can be seen by simple inspection. A somewhat similar method of analysing milk, by a special apparatus, has been patented in Germany, by J. Petri and R. Muencke.¹

§ 140a. (c.) Fat—

Solvents for Fat.—The solvents for milk-fat are petroleum-ether, ether, bisulphide of carbon, etc. Bisulphide of carbon has some advantages when acid milks are analysed, since it has no solvent action on the lactic acid. In quite fresh milks, however, the quantity of lactic acid is so small, that ether may be used. 25 to 50 c.c. of milk are evaporated in a flat dish with constant stirring and breaking-up of the caseous films by a glass rod, until the whole is reduced to a rather coarse granular powder. This powder may be transferred to any simple apparatus in use for the exhaustion of substances by volatile solvents; as, for example, Soxhlet's, described at page 49,—the results are a little low.

(a.) *Addition of Sand, or Sulphate of Lime, or other Powder.*—

¹ German Patent, No. 7477.

Numerous experiments by English analysts on the extraction of milk-fat by solvents acting on dried milk have fully proved that the greatest accuracy is only obtained by subdividing minutely the dry milk solids; this may be done either by mixing gypsum, sand or powdered pumice stone; the amount of indifferent material used must be at least equal in weight to the milk taken, and it is even advantageous to double the quantity, for instance—20 grms. of gypsum to 10 grms. of milk. The material is stirred into the milk, dried, and thoroughly well pulverised, the powder being then transferred to a Soxhlet's apparatus, and exhausted by solvents in the usual way.

(β.) *Adams' Method of Extracting Milk-Fat.*—The Society of Public Analysts appointed in 1884 a committee to investigate the best methods of milk analysis. The members of the committee analysed by different methods a large number of samples. The results were of considerable interest, especially with regard to extraction of the milk-fat.¹ The committee clearly established that milk evaporated to dryness in the ordinary way retains with tenacity a small quantity of fat, and therefore the Wanklyn method is always a little low. The results of the Wanklyn method are better with milks rich in fat than with poor or skimmed milk; in the latter, the longer the total solids are dried, the less are they amenable to the influence of ether. By finely dividing the milk, as for example by mixing it into a paste with plaster of Paris, drying, and then exhausting in a Soxhlet, much higher results were obtained. "Milks so highly skimmed that they yielded nothing practically by Wanklyn's method, give upwards of .6 per cent. by the plaster of Paris method;"² but the best results of all were obtained by an ingenious process invented by Dr. Adams—this consists in spreading a measured or weighed quantity of the milk over a small coil of specially prepared blotting- or filter-paper. Special paper cut into strips can now be bought. 5 c.c. of the milk are very gradually dropped on to the coil, distributing it as evenly as possible; it is then dried at 100° C. and exhausted by ether in a Soxhlet. Sour milks, which Dr. Adams has proposed to treat with ammonia so as to liquefy them, admit of analysis by means of this coil without such addition. In the latter case a quantity can be weighed out in a tared dish, and poured on to the coil, the dish being rinsed out by the least quantity of water possible; the washing being added to the coil.

The general result of the coil process is that it gives results about .5 per cent. higher than the Wanklyn method, and .2 per cent. higher than the plaster of Paris method. To meet the obvious objection that the coil method might perchance yield something to ether other than fat, careful analyses were made by the committee of the ether-extracts, both from the plaster of Paris method, and from the coil method; but in both, fat, and fat only, was found to be extracted.

(γ) *Estimation of Milk-Fat by Centrifugal Machines.*—One of the earliest, 'the lactocrite,' was patented by Carl G. P. de Laval (1885, No. 8610). The milk-fat is separated from the milk by strongly acidifying and rotating the milk in a special tube in a centrifugal machine.

The tube is thus described in the specification—the test vessel consists of a cylindrical silvered metallic box (a), into which a plug, likewise silvered, (b) fits accurately. This plug is turned and bored hollow from the bottom, tapering towards the top into a fine hole communicating with a narrow tube (c) fixed to the plug, and either graduated or not graduated. This

¹ *Analyst*, Jan., 1886.

glass tube is fixed upon the plug by the nut (*d*) received into the socket or holder (*e*) secured to the plug and surrounding the tube, and provided with opposite openings so that the degrees on the tube may be read. The

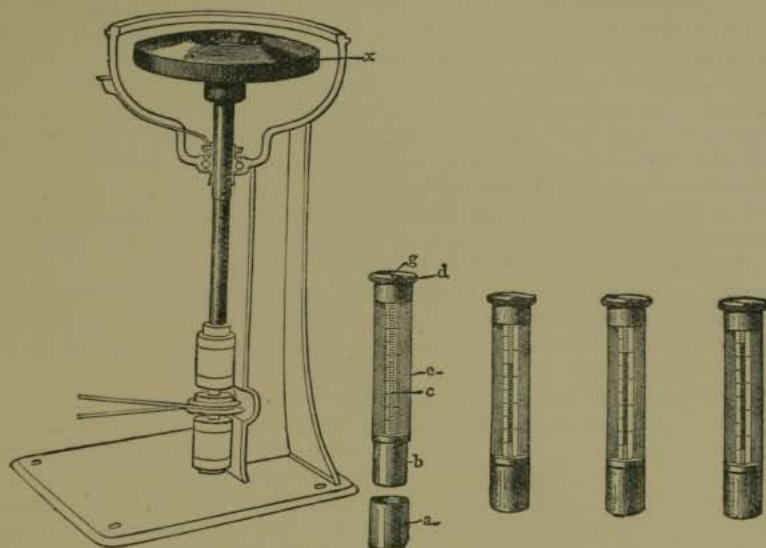


FIG. 33.

nut is provided with a hole (*g*), so that the plug and the tube constitute conjointly a vessel or channel open at both ends (fig. 33).

TABLE XVIIIb.—COMPARISON OF CENTRIFUGAL DETERMINATIONS OF MILK-FAT WITH ADAMS' PROCESSES.¹

No. of Samples	Specific Gravity.	Total Solids.	Fat.			Solids not Fat.			Ash.
			Lac-tocrite.	Adams'	Calculated by Clausnizer's Formula.	Lac-tocrite.	Adams'	Calculated by Clausnizer's Formula.	
1.	1033.0	12.90	3.40	3.44	3.42	9.50	9.46	9.48	.80
			3.50			9.40			
2.	1031.0	14.12	4.55	4.69	4.62	9.57	9.43	9.50	.80
			4.65			9.47			
3.	1032.0	13.12	3.60	3.57	3.62	9.52	9.55	9.50	.77
4.	1030.5	13.07	3.90	3.99	3.89	9.17	9.08	9.18	.74
5.	1032.0	12.98	3.80	3.76	3.51	9.18	9.22	9.47	.76
6.	1031.5	14.27	4.90	4.84	4.63	9.37	9.43	9.64	.76
7.	1032.5	13.84	4.20	4.26	4.08	9.64	9.58	9.76	.78
8.	1030.5	13.00	3.70	3.69	3.84	9.30	9.31	9.16	.76
9.	1031.0	13.51	4.05	4.09	4.14	9.46	9.42	9.37	.80
10.	1034.0	11.76	2.10	2.07	2.12	9.66	9.69	9.64	.76
11.	1035.0	9.99	.45	.502	.518	9.54	9.488	9.472	.86

The milk is prepared as follows:—An equal volume of milk and a mixture of acetic and sulphuric acids (20 vol. of glacial acetic and 1 vol.

¹ See also Table XVIII*d*.

conc. sulphuric acid) are placed in a test tube provided with a cork, through which a fine glass tube (to allow for expansion) is carried, and the tube with its contents is then heated in a water-bath for five or six minutes; the special tube is then charged and transferred to a recess on a metallic disc (*x*), previously brought by hot water to the temperature of 50° C. The disc is usually fitted with twelve such recesses, so that twelve samples of milk may be operated upon at one and the same time. The disc is rotated at a speed of 6000 revolutions per minute; after from three to five minutes the milk-fat will have risen in a well-defined layer; the reading is done at once while the tubes are warm. The divisions are in tenths, thus 33.5 is equal to 3.35 per cent. of fat in the milk.

The lactocrite is not much used now, its place being taken by various centrifugal machines, the most favoured being the Leffman and Beam and the Lister-Babcock. The disc in these does not require to be kept specially warm, the casein is brought into solution by strong hydrochloric and sulphuric acids, and the separation of the fat aided by a certain proportion of amyl alcohol. There are printed directions supplied by the makers of each instrument.

(8) *Soxhlet's Process*.—F. Soxhlet¹ is the author of a very ingenious process of milk analysis. This process is based on the fact, that if a

measured quantity of milk, alkalisied by caustic potash, be shaken up with ether, the ether fully extracts the milk-fat, and, on standing, collects in a clear layer. A small, quite constant, proportion of ether remains in solution in the milk, without retaining any of the fat, and without affecting the result. The amount of the fat dissolved in the ether may be determined by the specific gravity of the ether; the higher the specific gravity the greater the proportion of milk-fat. The details of Soxhlet's method are as follows:

The apparatus figured (see fig. 34) is used. C is a measuring flask of 300 c.c. capacity, provided with a doubly perforated cork, and connected, on the one hand, with the caoutchouc elastic bulbs figured, which are furnished with suitable valves, and, on the other, with the tube D, which is provided with a water-jacket. This tube carries an areometer, E, which bears a delicate scale of from

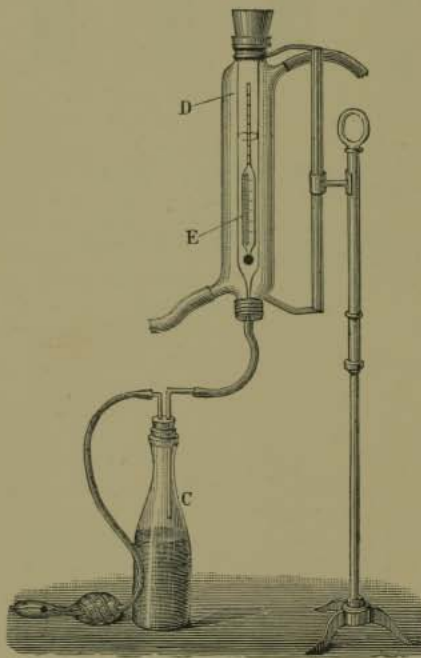


FIG. 34.

·766 to ·743, and it has also a thermometer, divided into thirds of a degree. 200 c.c. of milk are measured by means of a pipette, and run into the flask; 10 c.c. of potash and 60 c.c. of ether, which has been saturated with water at from 16°.5 to 18°.5 C., are then added, and the

¹ *Zeitschrift der Landwirthsch. Ver. Bayern.* 1880.

whole shaken up in the properly closed flask for a quarter of an hour. The fluid is then allowed to repose until the ether rises in a clear layer to the surface. By gently working the caoutchouc bulbs, a sufficient quantity of the fat-laden ether may now be blown up into the tube D, to float the areometer. (It should be mentioned that the tube going through the perforated cork of C, is so arranged as to dip well into the ether, but does not touch the surface of the milk.) The areometer must float freely, and for this purpose there are little prominences on the inside of D, about the middle, for the purpose of keeping the instrument from adhering to the side. When sufficient ether has thus been blown up, it is retained by a pinch-cock, and a cork is fitted into the tube D, to prevent loss by evaporation. The water in the jacket must be of any temperature between $16^{\circ}\cdot5$ and $18^{\circ}\cdot5$ C. The specific gravity once obtained in this way, and the temperature of the areometer noted, the content in fat of the milk is determined by the annexed table (p. 222).

If the temperature is exactly $17^{\circ}\cdot5$ C., then no correction is required; but if it is above that temperature, for every degree, a degree must be added to the gravity; if below, for every degree, a degree must be subtracted. Thus, supposing the areometer marks 58·9 at $16^{\circ}\cdot8$ C., then, as the difference between $16^{\circ}\cdot8$ and $17^{\circ}\cdot5$ is ·7, subtract ·7 from 58·9, equalling 58·2; or if the observation be 47·6 at $18^{\circ}\cdot4$ C., then the corrected value is 48·5 (*i.e.*, specific gravity 74850).

Afterwards the ether is allowed to run back into the flask, and D is washed out with a little ether. The authors think that the apparatus might be improved by adopting the principles used in the tube figured at p. 223, and floating the ether layer by means of mercury.

(e) *The Werner-Schmidt method of fat estimation* gives accurate results. A test tube of 50 c.c. capacity is calibrated in cubic centimetres and tenths; into it are placed 10 c.c. of milk, or a weighed quantity (say 5 grms.) of cream, 10 c.c. of strong hydrochloric acid added, and the whole boiled, with shaking until the liquid forms a dark brown solution. The tube, with its contents, is rapidly cooled by placing it in cold water; a known volume of ether is added by filling it up to one of the divisions, some 25 to 30 c.c. of ether being necessary. The contents are now vigorously shaken for half a minute, and the tube is allowed to stand for five minutes. At the end of that time the layer of ether is carefully read, and an aliquot part pipetted off, evaporated to dryness, and weighed.

The majority of analysts now prefer to exhaust the contents of the tube with several quantities of ether and so remove the whole of the fat.

If a measured quantity of ether is to be used, then Stokes' tube (see fig. 34a) is the most convenient.

If the whole of the ether with its contained fat is to be removed, one of us has invented the following apparatus, which will be found convenient. The apparatus consists of a small mercury reservoir attached by means of a caoutchouc tube to a glass bulb of convenient capacity, which terminates in a two-way stop-cock connected with two very narrow right-angled tubes (see fig. 34a). The prepared milk is sucked in through one right-angled tube by lowering the reservoir; the vessel in which the milk was heated is washed with ether which is sucked into the bulb, the mercury is nearly all run out, the taps turned off and the vessel shaken, then allowed to stand; the ether is then expelled from the other tube by raising the reservoir carefully; in this way the whole of the ether which has risen to the top

may be removed. The last drops of ether remaining in the side tube may be removed by admitting air through one tube and expelling it through the other. Three or four extractions will suffice to deprive the sample of all milk fat.

TABLE XVIIIc.—CORRECTION OF SPECIFIC GRAVITY OF THE ETHEREAL SOLUTION OF FAT (p. 221).

Specific gravity.	Fat. Per cent.	Specific gravity.	Fat. Per cent.	Specific gravity.	Fat. Per cent.	Specific gravity.	Fat. Per cent.	Specific gravity.	Fat. Per cent.
43	2.07	47.7	2.61	52.3	3.16	56.9	3.74	61.5	4.39
43.1	2.08	47.8	2.62	52.4	3.17	57	3.75	61.6	4.40
43.2	2.09	47.9	2.63	52.5	3.18	57.1	3.76	61.7	4.42
43.3	2.10	48	2.64	52.6	3.20	57.2	3.78	61.8	4.44
43.4	2.11	48.1	2.66	52.7	3.21	57.3	3.80	61.9	4.46
43.5	2.12	48.2	2.67	52.8	3.22	57.4	3.81	62	4.47
43.6	2.13	48.3	2.68	52.9	3.23	57.5	3.82	62.1	4.48
43.7	2.14	48.4	2.70	53	3.25	57.6	3.84	62.2	4.50
43.8	2.16	48.5	2.71	53.1	3.26	57.7	3.85	62.3	4.52
43.9	2.17	48.6	2.72	53.2	3.27	57.8	3.87	62.4	4.53
44	2.18	48.7	2.73	53.3	3.28	57.9	3.88	62.5	4.55
44.1	2.19	48.8	2.74	53.4	3.29	58	3.90	62.6	4.56
44.2	2.20	48.9	2.75	53.5	3.30	58.1	3.91	62.7	4.58
44.3	2.22	49	2.76	53.6	3.31	58.2	3.92	62.8	4.59
44.4	2.23	49.1	2.77	53.7	3.33	58.3	3.93	62.9	4.61
44.5	2.24	49.2	2.78	53.8	3.34	58.4	3.95	63	4.63
44.6	2.25	49.3	2.79	53.9	3.35	58.5	3.96	63.1	4.64
44.7	2.26	49.4	2.80	54	3.37	58.6	3.98	63.2	4.66
44.8	2.27	49.5	2.81	54.1	3.38	58.7	3.99	63.3	4.67
44.9	2.28	49.6	2.83	54.2	3.39	58.8	4.01	63.4	4.69
45	2.30	49.7	2.84	54.3	3.40	58.9	4.02	63.5	4.70
45.1	2.31	49.8	2.86	54.4	3.41	59	4.03	63.6	4.71
45.2	2.32	49.9	2.87	54.5	3.43	59.1	4.04	63.7	4.73
45.3	2.33	50	2.88	54.6	3.45	59.2	4.06	63.8	4.75
45.4	2.34	50.1	2.90	54.7	3.46	59.3	4.07	63.9	4.77
45.5	2.35	50.2	2.91	54.8	3.47	59.4	4.09	64	4.79
45.6	2.36	50.3	2.92	54.9	3.48	59.5	4.11	64.1	4.80
45.7	2.37	50.4	2.93	55	3.49	59.6	4.12	64.2	4.82
45.8	2.38	50.5	2.94	55.1	3.51	59.7	4.14	64.3	4.84
45.9	2.39	50.6	2.96	55.2	3.52	59.8	4.15	64.4	4.85
46	2.40	50.7	2.97	55.3	3.53	59.9	4.16	64.5	4.87
46.1	2.42	50.8	2.98	55.4	3.55	60	4.18	64.6	4.88
46.2	2.43	50.9	2.99	55.5	3.56	60.1	4.19	64.7	4.90
46.3	2.44	51	3.00	55.6	3.57	60.2	4.20	64.8	4.92
46.4	2.45	51.1	3.01	55.7	3.59	60.3	4.21	64.9	4.93
46.5	2.46	51.2	3.03	55.8	3.60	60.4	4.23	65	4.95
46.6	2.47	51.3	3.04	55.9	3.61	60.5	4.24	65.1	4.97
46.7	2.49	51.4	3.05	56	3.63	60.6	4.26	65.2	4.98
46.8	2.50	51.5	3.06	56.1	3.64	60.7	4.27	65.3	5.00
46.9	2.51	51.6	3.08	56.2	3.65	60.8	4.29	65.4	5.02
47	2.52	51.7	3.09	56.3	3.67	60.9	4.30	65.5	5.04
47.1	2.54	51.8	3.10	56.4	3.68	61	4.32	65.6	5.05
47.2	2.55	51.9	3.11	56.5	3.69	61.1	4.33	65.7	5.07
47.3	2.56	52	3.12	56.6	3.71	61.2	4.35	65.8	5.09
47.4	2.57	52.1	3.14	56.7	3.72	61.3	4.36	65.9	5.11
47.5	2.58	52.2	3.15	56.8	3.73	61.4	4.37	66	5.12
47.6	2.60								

N.B.—The numbers in the specific gravity column correspond to those on the areometer scale, the number 7 being omitted on account of the narrowness of the stem of the instrument; thus 46 really means .7460, and so on with the rest.

(ζ) *Babcock Asbestos Method.*—This has been already mentioned. The cylinder containing the dry solids is extracted by anhydrous ether and the fat weighed after distilling off the ether.

(η) *Ritthausen's Method.*—10 c.c. of milk are diluted to 100 c.c. with distilled water, 5 c.c. of a 6 per cent. solution of copper sulphate are added and nearly neutralised by sodium hydrate. The precipitate is washed, dried, and extracted with ether.

(θ) *Calculation Methods.*—An indirect method of estimating milk-fat has been proposed by F. Clausnizer and A. Mayer. They assume that every percentage of "milk solids not fat" raises the specific gravity .00375,

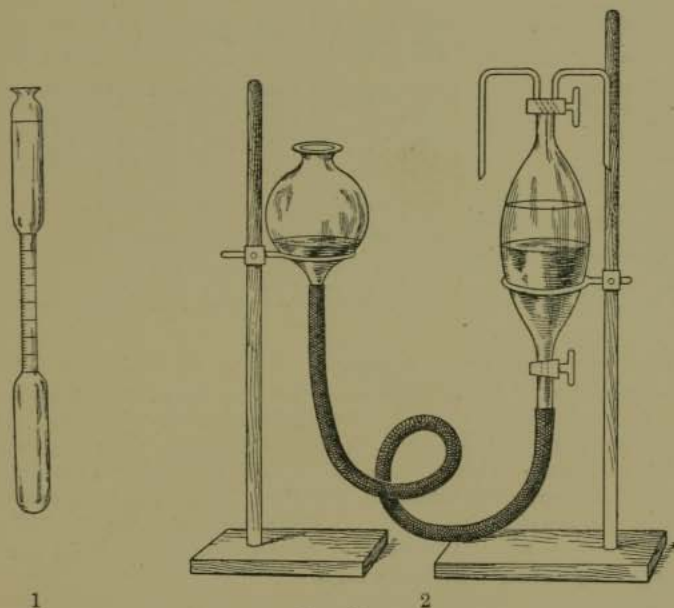


FIG. 31a.
1, Stokes' tube; 2, M. W. Blyth's extraction bulb.

whilst every 1 per cent. of fat lowers the specific gravity .0010; by therefore first making an accurate determination of the specific gravity, and then determining either fat or total solids, the equation can be easily solved.

$$\begin{array}{l}
 x = \text{fat.} \\
 s = \text{specific gravity.} \\
 t = \text{total solids.}
 \end{array}
 \quad
 x = \frac{t \cdot 00375 + 1 - s}{\cdot 00475}$$

Richmond suggests the following formula :—

T = total solids; Q = last two figures of sp. gr. (water = 1000); F = fat; then,

$$T = \frac{Q}{4} + \frac{6}{5}F + \cdot 14.$$

Richmond has calculated out a table (see Appendix) giving percentage of fat if total solids and sp. gr. are known. A milk scale, known as Richmond's milk scale, is a useful instrument and much used.

(c) *Somerset House Method*.—See *Analysis of Sour Milk*.

The various methods described have been critically examined by Richmond.¹ His results are contained in the following Table XVIII*d*.:—

TABLE XVIII*d*.—ANALYSIS OF NINE SAMPLES OF MILK BY THREE DIFFERENT ANALYSTS, EACH USING VARIOUS METHODS.

Mean Results (to indicate variation between methods).

Method.	A.	B.	C.	D.	E.	F.	G.	H.	I.	Mean.
Total Solids:										
Society of Public Analysts,	13.16	13.90	9.68	10.91	12.19	12.36	11.34	16.28	13.35	12.58
Asbestos,	13.33	14.06	9.73	10.92	12.27	12.37	11.35	16.11	13.37	12.61
Somerset House,	13.25	14.02	9.92	11.12	12.46	12.51	11.55	16.13	13.48	12.72
Fat:										
Adams (dry ether),	4.25	4.60	.23	2.65	3.12	3.47	3.09	8.21	4.26	3.76
Adams (commercial ether),	4.25	4.60	.28	2.68	3.13	3.45	3.05	8.21	4.33	3.78
Werner Schmidt,	4.20	4.78	.32	2.72	3.18	3.52	3.09	8.72	4.33	3.88
Somerset House,	4.24	4.55	.28	2.71	3.16	3.46	3.03	8.26	4.21	3.77
Centrifugal,	4.22	4.53	.18	2.65	3.20	3.52	3.05	7.90	4.24	3.72
Centrifugal (R.),	4.25	4.50	.20	2.70	3.15	3.50	3.05	7.90	4.15	3.72
Solids not Fat:										
Somerset House,	9.02	9.46	9.63	8.41	9.33	9.05	8.52	7.56	9.27	8.95
Society Public Analysts—Adams Commercial,	8.91	9.30	9.40	8.23	9.06	8.91	8.29	8.07	9.02	8.80
Society Public Analysts—Werher Schmidt,	8.96	9.12	9.36	8.19	9.01	8.84	8.25	7.56	9.01	8.70
Asbestos—Adams (dry),	9.04	9.47	9.54	8.26	9.18	8.95	8.30	7.90	9.16	8.87
Cale (H. & R.),	8.70	9.10	9.35	8.10	9.00	8.80	8.05	7.50	8.90	8.61

§ 141. (d.) *Lactose or Milk Sugar*—

(a) *Extraction of the Milk-Sugar*.—On now exhausting a fat-free milk residue by weak boiling alcohol, filtering the alcoholic fluid, and evaporating to dryness, the milk-sugar, with mineral matters dissolved out by the alcohol, is obtained. This evaporation is best effected in a platinum dish. On drying very carefully, weighing, and then burning the sugar away, the ash is left, and must be weighed and subtracted from the original residue of milk-sugar and ash. The amount of sugar is thus obtained with fair accuracy, always being a little too high.

(β) *Polarimetric Methods*.—The first step is to prepare a clear whey. An excellent paper on this subject was published in 1887 by Dr. W. Wiley, the chief chemist for the U.S. Department of Agriculture. He used the following reagents for the precipitation of the albuminoids:—

- (1.) Saturated solution of basic lead acetate.
- (2.) Nitric acid solution of mercuric nitrate diluted with an equal volume of water.
- (3.) Acetic acid, specific gravity 1.040.
- (4.) Nitric acid, specific gravity 1.197.
- (5.) Sulphuric acid, specific gravity 1.255.
- (6.) Saturated solution of sodium chloride.
- (7.) Saturated solution of magnesium sulphate.
- (8.) Solution of mercuric iodide in acetic acid, a litre of the solution being made with 161 grms. HgCl₂, 395 grms. KI, and 228 c.c. of acetic acid.

Wiley ultimately gives the preference to the following process:—A solution of mercuric nitrate is prepared by dissolving mercury in twice its weight of nitric acid (sp. gr. 1.42) and diluting with four volumes of water. To 60 c.c. of the milk are added 10 c.c. of the nitrate solution; the mixture is diluted to 100 c.c., well shaken, then filtered through a dry filter and its rotation at once observed. The volume of the sugar solution must be

¹ *Departmental Committee on Milk Regulations, 1901, Appendix xxiv.*

corrected by deducting the weight of fat in grammes multiplied by 1.075 and the weight in grms. of the proteids multiplied by 0.8.

For example, a sugar-containing liquid from 60 c.c. of a milk containing, say, 3 per cent. of fat and 3.5 per cent. of proteids, would be, after dilution,

$$\begin{aligned} & 100 - (1.8 \times 1.075 + 2.1 \times 0.8) \\ & = 100 - (1.935 + 1.68) \\ & = 96.38 \text{ c.c.} \end{aligned}$$

Wiley takes the specific rotation of milk-sugar as $(a)_D = 52.5^\circ$; the percentage of sugar is then calculated in the ordinary way.

$$\text{Milk-sugar} = \frac{100 \times d}{52.5 \times e}$$

d = angular rotation found, and e = length of tube.

The result must then be multiplied in the case given by $\frac{96.38}{100}$ to correct for volume.

This will give the hydrated sugar in 60 c.c. of milk, or if anhydrous sugar is required \times by 0.95, which will give weight of anhydrous sugar in 60 c.c. of milk or in 60 grms., if the volume is multiplied by the specific gravity.

The whole calculation may be expressed thus:—

$$\text{Anhydrous milk-sugar by weight} = \frac{100 \times d}{52.5 \times l} \times \frac{100 - (1.075 F \times 0.8 P)}{Q} \times \frac{.95}{\text{Sp. Gr.}}$$

Where Q = quantity of milk taken.

F = Weight of fat in quantity, Q , of milk.

P = Weight of proteids in quantity, Q , of milk.

l = Length of tube.

Vieth's Method.—This is nearly the same as Wiley's. Vieth, however, uses mercuric nitrate, prepared by dissolving mercury in twice its weight of nitric acid (sp. gr. 1.42) and adding an equal bulk of water.

There is some difficulty in obtaining accurate estimates of the milk-sugar in human milk, the filtrate with ordinary precipitates being still turbid. P. Thebault¹ recommends a preliminary precipitation with picric acid and glacial acetic acid (picric acid 10 grms. per litre, acetic acid 25 c.c. per litre).

Richmond and Carter found the results of estimation by Fehling and polarisation unsatisfactory, and estimate the sugar by difference.

(γ) *Gravimetric Methods.*—The proteids and fat must be removed; this may be effected by copper sulphate. 25 c.c. of the sample are diluted with 400 c.c. of water and 10 c.c. of a solution of copper sulphate (34.64 to 500 c.c. of water) are added to the milk made almost neutral with sodium hydrate. If O'Sullivan's or Wein's method is to be used, the original 25 c.c. of milk must be diluted to 250 c.c. The precipitate is filtered off through a dry filter.

Wein's Method.—60 c.c. of Fehling are mixed with twice their volume of the filtrate obtained as above; before mixing, both solutions should be heated in the water bath. The mixture is boiled for exactly four minutes

¹ *Journ. Pharm.* (6), iv. 5.

and filtered through a Soxhlet tube, previously plugged with asbestos and weighed. The filtered oxide is well washed with water and reduced to metallic copper in a current of hydrogen gas at a gentle red heat; or the copper may be dissolved in nitric acid and deposited electrolytically on a platinum dish. The milk-sugar may then be calculated from Wein's table.

TABLE XVIIIc.—WEIN'S TABLE FOR CALCULATING THE WEIGHT OF MILK-SUGAR FROM THE WEIGHT OF COPPER REDUCED.

Copper.	Milk-sugar.	Copper.	Milk-sugar.	Copper.	Milk-sugar.
120	86.4	215	158.2	310	232.2
125	90.1	220	161.9	315	236.1
130	93.8	225	165.7	320	240.0
135	97.6	230	169.4	325	243.9
140	101.3	235	173.1	330	247.7
145	105.1	240	176.9	335	251.6
150	108.8	245	180.8	340	255.7
155	112.6	250	184.8	345	259.8
160	116.4	255	188.7	350	263.9
165	120.2	260	192.5	355	268.0
170	123.9	265	196.4	360	272.1
175	127.8	270	200.3	365	276.2
180	131.6	275	204.3	370	280.5
185	135.4	280	208.3	375	284.8
190	139.3	285	212.3	380	289.1
195	143.1	290	216.3	385	293.4
200	146.9	295	220.1	390	297.7
205	150.7	300	224.4	395	302.0
210	154.5	305	228.3	400	306.3

Soxhlet's method only differs from Wein's in minute details. O'Sullivan's method is also similar, and has been already described at p. 112.

(δ) *Volumetric Methods*.—These have been already described at pages 114–115. Most of the methods are applicable to the determination of milk-sugar.

§ 142. (e.) *Total Proteids*—

Kjeldahl's Method.—The nitrogen is determined in 5 grms. of milk by Kjeldahl's method (see p. 97). The sulphuric acid is added directly to the milk, which need not be evaporated.

According to Munk,¹ milk contains a sensible proportion of non-proteid nitrogen, from .022 to .034 per cent. in cow's milk and from .014 to .026 per cent. in human milk. The American Association of Official Agricultural Chemists use the factor 6.25 to convert nitrogen into milk proteids instead of the usual factor 6.38.

Ritthausen's Process.—To 10 c.c. diluted to 100 c.c. are added 5 c.c. of 6 per cent. sulphate of copper solution. The solution is nearly neutralised by half normal soda solution, but it must remain slightly acid; hence the acidity is determined carefully on another sample of the milk, using phenolphthalein as indicator before adding the copper sulphate, so that the exact quantity of sodium hydrate to be added is known. The precipitate is washed once or twice by decantation, passing the washings through a tared filter; the precipitate is next transferred to the same filter and well washed. The nitrogen in the filter and its contents may

¹ *Journ. Chem. Soc.*, 1894, xi., 196.

be determined by Kjeldahl's method, or the filter with contents may be dried at 130° C. The weight found is that of proteids, fat and mineral matter. On burning to an ash and subtracting, the result is total proteids and fat. The fat having been already obtained by other methods, the proper quantity of fat is subtracted from this weight, which then equals the proteids.

Or the wet filter may be washed with alcohol, extracted with ether in a Soxhlet apparatus, then dried and weighed, and lastly burned to an ash; on subtracting the ash the total proteids are obtained.

Estimation of Casein and Albumen.—100 c.c. of milk are divided into three equal portions. One of these portions is diluted to about four times its volume, and acidified with dilute acetic acid until the casein coagulates in a flocculent condition; a current of carbon dioxide is now passed through, and the precipitate allowed to subside. The whey is then carefully syphoned off on to the second portion of the milk: more acid, if necessary, is added; the same operation repeated; and this second whey similarly added to the third portion of milk. Finally, the whole of the casein is collected on a filter, and washed. The result of the process is, that the albumen from 100 c.c. of milk is held in solution in about 250 to 300 c.c. This solution is now raised to the boiling point, and gently boiled for a few minutes. The whole of the albumen falls down, and is easily collected on a previously dried and weighed filter. This easy separation of the casein and albumen by acetic acid and carbon dioxide only applies to the milk of the cud-chewers; with human milk, the milk of the horse or of the ass, the process gives no good result: when treated in the same way, it is true that the casein appears to coagulate, but is in a state of such fine division that nearly all of it remains suspended in the liquid, and filtration through paper becomes impossible. From all these a clear filtrate may, however, be obtained by filtration under pressure through one of the Pasteur or Berkefeldt candles, or the globular-shaped vessels formed of kaolin and quartz, as described by W. Pukall. The globular shape is somewhat better than the cylindrical. In all these cases the filtration is upwards; by means of an angle tube and caoutchouc-pressure tubing, the filter is attached to a mercury or water pump. The milk is diluted, acidified with acetic acid, and saturated with carbon dioxide as before; the filter is then totally immersed in the dilute acid milk, which for this purpose is placed in a tall beaker; a good vacuum is maintained, and ultimately the whole of the whey passes through, and the casein may be washed two or three times with water.

This method has been modified by Ritthausen.¹ 10 grms. of milk are diluted with 90 c.c. of water at 42° to 43° C. and 1.5 c.c. of a 10 per cent. solution of acetic acid added; after 5 minutes a precipitate of casein settles, the albumen remaining in solution.

The precipitated casein is treated with alcohol and ether, weighed and burnt up to an ash as in the estimation of total proteids. The albumen is obtained by neutralising the filtrate with sodium hydrate and adding 3 c.c. of a 10 per cent. acetic acid solution, boiling for 15 minutes, and collecting on a tared filter; or the N in the albumen may be estimated by Kjeldahl's process and the N found multiplied by 6.38.

Sebelien's Method.—Leffmann and Beam have slightly modified Sebelien's original process as follows:—10 c.c. of milk are mixed with a saturated solution of magnesium sulphate, and the powdered salt added to saturation.

¹ *Ber. Chem. Gesell.*, xxvi., 1159-1172.

The mixture is washed into a graduated measure with a small amount of the saturated solution, and made up to 100 c.c. with the same saturated solution, mixed, and allowed to stand until separation takes place. As much as possible of the clear solution is drawn off with a pipette and passed through a dry filter. Of this filtrate an aliquot portion is taken, the albumen precipitated by tannin solution (prepared by dissolving 4 grms. of tannin in 190 c.c. of 50 per cent. alcohol, and adding 8 c.c. of acetic acid of 25 per cent. strength).¹ The nitrogen in the tannin precipitate is determined by Kjeldahl's process. Phospho-tungstic acid² may be used as a precipitant instead of tannin.

Lehman's Method.—This method has the advantage over others in the fact that no reagent which may cause chemical change in the proteids is added. 5 grms. of milk are poured on to the centre of a porous unglazed porcelain plate, previously wetted with water. In about an hour and a half the serum is absorbed by the plate, the casein and fat remaining behind. The residue is scraped off the plate by means of a spatula, the fat extracted with ether, and the remaining casein dried and weighed.

§ 143. (f.) *The Ash*—

The ash is estimated in a quantity of milk, which should not exceed 25 c.c. It will be found that larger quantities do not at all conduce to accuracy, as the large amount of carbon, with even great care, develops too much heat, and the phosphates are liable to fuse, enclosing little particles of charcoal extremely difficult to burn. With small quantities, however, the milk rapidly burns to an almost white ash. It may be further analysed on the principles laid down at p. 93.

If porcelain evaporating dishes are used there will be little fear of overheating. A good method is to char the ash well, extract with water and evaporate the filtered aqueous extract to dryness and weigh, after igniting at a just visible red heat. This gives 'soluble ash.' The insoluble ash will remain mixed with carbon, but containing no volatile chlorides. This part may now be burned up to a white ash without fear of loss. Allen adds to the black char a few drops of strong sulphuric acid and one drop of nitric acid. On ignition the ash will be obtained white; the weight found must be multiplied by '67 to correct for chemical action of sulphuric acid on the mineral constituents.

§ 144. *Citric Acid in Milk.*—Various observers, such as Henkel,³ Scheibe,⁴ Vaudin,⁵ have separated citric acid in quantities of from '05 to '11 per cent. from cow's milk; these observations have found satisfactory confirmation by a special research of Alfred Wöhlk.⁶

The best method of separating citric acid from milk is probably to coagulate the milk proteids by acid mercuric nitrate, using only a slight excess of the precipitant. After filtering off the proteids, the clear whey is neutralised by soda solution, and heated on the water bath for some time; a white precipitate falls, which consists of mercury and calcium phosphate

¹ Almen's reagent.

² To prepare phospho-tungstic acid 100 grms. of sodium tungstate are dissolved in a litre of water and 100 grms. of phosphoric acid solution of specific gravity 1.13 added. The mixture is boiled and acidified with HCl. After twenty-four hours the whole is filtered; the filtrate is then ready for use.

³ *Jahresbericht der Thierchemie*, 1889, § 94.

⁴ *Landwirthschaftliche Versuchs-Stationen*, 1891, § 244, 164.

⁵ *Annales de l'Institut Pasteur*, viii., 502.

⁶ Ueber die Einwirkung von Brom und Kaliumpermanganat und Citronensäure (Stahre's Reaction) and den Nachweis von Citronensäure in Milch, von Alfred Wöhlk. *Zeits. f. analytische Chemie*, 1902, 79.2

and citrate. The mercury is got rid of by acidifying with hydrochloric acid, and passing a current of SH_2 through the liquid. After filtering off the mercuric sulphide, the liquid is cooled, a solution of calcium chloride added and exactly neutralised by soda solution, using decinormal phenolphthalein as an indicator. The calcium phosphate falls, the calcium citrate remains in solution. On filtering off the phosphate and boiling, calcium citrate is deposited.

To identify the salt, a little of it may be dissolved in dilute sulphuric acid and permanganate solution added until the liquid is of a pink colour. The permanganate oxidises citric acid to aceton-di-carbon acid, which gives with Denigès reagent (5 grms. of mercuric oxide dissolved in 20 c.c. of sulphuric acid and made up to 100 c.c. with water) a crystalline precipitate, $\text{SO}_4\text{Hg}_2\text{O}_2\text{HgCO}_2(\text{CH}_2\text{COO})_2\text{Hg}$.

When added to the same permanganate solution, a few drops of bromine water produce a crystalline precipitate of penta-bromacetone (melting point, 73°C .)

An acid organic substance which, on boiling in neutral solution, yields with calcium chloride a precipitate, which gives, when oxidised with potassic permanganate, a crystalline precipitate with Denigès reagent, and, on carrying the oxidation a step forward, forms penta-bromacetone with bromine water, must be citric acid.

To estimate citric acid, the simplest method is to convert the calcium citrate by ignition into calcium carbonate, and titrate with deci-normal acid; or, rather, to dissolve the carbonate in a known volume of the acid, and titrate back with standard alkali, using methyl-orange as the indicator; 2 c.c. of d.n. acid equals 6.4 mgrms. of citric acid.

§ 145. *Estimation of Urea*.—The estimation of urea is of some importance, since any disorder interfering with the action of the kidneys throws (as has been well ascertained) an excess of urea on all the secretions of the body. A known quantity of milk, which should not be less than half a litre, is evaporated with constant stirring in very large flat dishes to a granular condition. The fat is next extracted by dry ether, and from the fat-free solids the urea is extracted with other substances by absolute alcohol. The alcoholic solution is evaporated to dryness; and from this dry residue the urea may be further purified by dissolving in alcohol. A litre of milk in its normal state yields about 10 mgrms. Urea must be identified by its properties as follows:—

It is crystalline, crystallising in quadratic prisms, and polarising with a gentle blue colour under the microscope. The crystals should be heated with a little hydrate of baryta in a closed tube to 200° for some hours, when a very definite reaction ensues, ammonia and carbon dioxide being produced. The liquid may be distilled, and ammonia identified by the Nessler test. Carbonate of baryta will appear as a precipitate, and may be readily examined, converted into sulphate, and weighed; 1 part of barium sulphate = 257.4 urea. A convenient method of identifying urea is also to dissolve the crystals in the smallest possible quantity of water, and then to add a drop of dilute nitric acid; the nitrate of urea is precipitated, and can be identified by its microscopic characters. Nitrate of urea crystallises in the rhombic system. The most common appearance is that of large plates, many of which lie one upon the other.

§ 146. *Volatile Acids*.—Volatile acids may be separated by careful distillation in a vacuum, first acidifying the milk by tartaric acid. Acetic acid in small quantities is invariably present in fermented milks; but the dis-

tillate of normal quite fresh milk is neutral. In milks already undergoing decomposition, it is best to dilute the milk slightly, and filter through an earthenware cell, and then distil. Under these circumstances, it is not necessary to distil *in vacuo*.

§ 147. *Estimation of the Total Acidity of Milk—Estimation of Lactic Acid.*—100 c.c. of milk are placed in a beaker and titrated with $\frac{N}{10}$ alkali, using phenolphthalein as an indicator until a faint pink is obtained. It is usual to call each c.c. of alkali used 1° of acidity. Normal milk examined twelve hours after milking usually has an acidity of 20°.

Estimation of Lactic Acid.—It may be of great importance to make an accurate estimation of lactic acid in milk. The following method may be employed:—The milk is dried, exhausted of fat by carbon disulphide, then treated with an alcoholic solution of oxalic acid, filtered, and an excess of hydrated oxide of lead added. Any lactic acid now contained in the fluid will be present in solution as a lactate of lead. The liquid is filtered, saturated with SH_2 , and again filtered, concentrated by evaporation, and boiled with oxide of zinc; on filtration, evaporation, and standing, crystals of lactate of zinc are produced. There are four isomeric lactic acids; that which is obtained from milk is fermentation lactic acid, also termed 'ethylene lactic acid.' The zinc salt has the composition $2(\text{C}_3\text{H}_5\text{O}_3)\text{Zn} + 3\text{H}_2\text{O}$. It crystallises in four-sided prisms; it is soluble in 6 parts of boiling, 58 of cold water. It is nearly insoluble in hot or cold alcohol. 100 parts of the salt contain 25.8 of zinc oxide. Lactic acid itself may be obtained in a very pure state by decomposing the zinc-salt with hydrogen sulphide, when the acid presents itself as a colourless, strongly acid liquid. A drop of this acid, placed in the subliming cell,¹ and heated very gradually above 200° C., gives a white sublimate of lactide, $\text{C}_3\text{H}_4\text{O}_2$, a very characteristic reaction. If the heat is not gradual, this sublimate is not obtained, for it then decomposes into carbon dioxide and aldehyde.

§ 148. *Detection of Metals in Milk.*—In rare cases it may be necessary to test for such metals as copper, zinc, lead, mercury, arsenic, or antimony in milk.

Non-volatile metals, such as copper or lead, are conveniently sought for by the ordinary chemical reactions in the ash. Volatile metallic poisons, such as arsenic or antimony, may be looked for as described in testing beer for arsenic. Mercury is best detected by Reinsch's test.

C. THE ANALYSIS OF MILK FOR THE PURPOSE OF DETECTING ADULTERATION.

§ 149. The various methods by which milk may be adulterated are very numerous. The analyst may have to detect and make quantitative estimations when possible of:—

- (1) The addition of water.
- (2) The abstraction of fat.
- (3) The addition of separated milk.
- (4) " " " condensed milk, boiled milk, or sterilised milk.
- (5) " " " gelatin.
- (6) " " " starch, dextrin, cane sugar, glucose, or invert sugar.
- (7) " " " preservatives.
- (8) " " " colouring matters.

¹ See *Poisons, their Effect and Detection*, by A. Wynter Blyth.

(1 and 2.) *Added Water and Abstraction of Fat.*—Providing the milk is fresh and in good condition the analyst will have to determine (a.) specific gravity, (b.) fat, (c.) total solids, (d.) solids not fat.

(a.) The specific gravity may be taken as already described and corrected to the normal temperature by the table on page 216. (b.) The fat may be determined by one of the rapid methods, such as Leffmann and Beam's. (c.) The total solids may be calculated from Richmond's table (see Appendix). (d.) The 'solids not fat' may be determined by subtracting the percentage of fat from the percentage of total solids.

With many milks these determinations will be sufficient to decide whether the sample is genuine. If there be any doubt, the total solids and ash must be directly determined, care being taken that no added mineral in the ash is overlooked; the fat must be determined by the Werner-Schmidt method, or by means of an Adam's coil. On another portion of the sample, the total proteids and milk-sugar must be estimated.

The direct determination of the total solids should correspond closely to the total solids calculated. The milk-sugar, proteids, and ash should be in the average ratio calculated by Vieth, *i.e.*, 13 : 9 : 2.

If this relationship be much disturbed, or if the ash be above the normal (*i.e.*, 0.75 per cent.) the milk is probably abnormal. In abnormal milks the ash is often high, the milk-sugar usually low. Provided that the milk be not abnormal, the analyst has now figures from which to calculate the percentage of added water and the percentage of fat abstracted.

The percentage of added water is calculated upon the percentage of 'solids not fat.'

$$\text{Percentage of added water} = 100 - \frac{s \times 100}{y}$$

where *s* = 'solids not fat' in the sample, and *y* = the 'solids not fat' in the original milk. As the solids not fat in the original milk are but rarely known, some standard of comparison must be taken.

In taking a standard of comparison a clear distinction must be drawn between the *average composition of milk* and the *minimum amount of solids not fat and fat* contained in genuine milk.

The *average composition* of milk as deduced by Vieth and Richmond from 200,000 analyses is:—

	Per cent.
Water,	87.10
Fat,	3.90
Milk sugar,	4.75
Casein,	3.00
Albumen,40
Ash,75

The average composition of milk produced for sale in this country is according to the *Report of the Departmental Committee on Milk and Cream Regulations, 1901*:—

	Per cent.
Total solids,	12.5-12.8
Fat,	3.7- 4.0

The minimum amount of solids not fat found in the mixed milk of a herd of cows, such as are kept in this country, is best illustrated by Richmond's figures. He has shown¹ that only in 2.19 per cent. of the samples are the 'solids not fat' likely to fall below 8.5 per cent. ; in only .3

¹ *Dairy Chemistry, 1901, p. 122.*

per cent. of the samples are they likely to fall below 8.4 per cent., and even in these cases, at least .5 per cent. of nitrogen and .7 per cent. of ash will be found.

The minimum amount of fat in the mixed milk of a herd of cows will be rarely found to fall below 3 per cent.

The milk of individual cows will naturally present greater variations than the mixed milk of many cows, and before deciding whether the milk of an individual cow has been watered or skimmed, the analyst will have to take into consideration the circumstances discussed later on.

The minimum amount of 'solids not fat' contained in milk for the purposes of the Sale of Food and Drugs Act has been fixed in this country at 8.5 per cent., so that the formula for calculating the added water becomes

$$\text{added water} = 100 - \frac{s \times 100}{8.5}$$

If, however, it is necessary to calculate the probable amount of water added, it is better to divide by 8.9 than by 8.5.

Richmond considers that a truer estimate of the real percentage of added water is obtained by using the following formula—

$$\text{added water} = 100 - \frac{G + f}{34.5} 100,$$

where G equals degrees of gravity in whole numbers, less 1000: thus 1032 = 32 and f = percentage of fat.

M. Lescoeur has proposed the following method of detecting added water:—

The milk is coagulated by adding to it a trace of rennet, the whey filtered off and collected.

The specific gravity at 15° C. and the total solids of the whey are estimated.

The specific gravity of normal milk whey varies from 1.029 to 1.031, and the total solids from 6.7 to 7.1 per cent.

If the specific gravity falls below 1.027 and the total solids below 6.7 per cent. he considers the sample watered.

A. J. Woodmann¹ proposes a method similar to the above. To 100 c.c. of milk 2 c.c. of acetic acid (sp. gr. 1.035) are added. The mixture is heated to 70° C. for 20 minutes, then placed in cold water for ten to fifteen minutes, filtered, and the sp. gr. determined. Pure milk serum has a sp. gr. of 1.029; every 10 per cent. of added water causes a decrease of .0031 in sp. gr.

The percentage of fat abstracted is calculated on the percentage of fat contained in the sample.

Per cent. of fat abstracted = $100 \frac{f_1 - f_2}{f_1}$ where f_1 = fat in original milk and f_2 = fat in sample.

The fat in the original milk is not usually known, so, as already explained, some standard of fat is taken.

The standard for milk fat in this country has been fixed for the purposes of the Sale of Food and Drugs Act at 3 per cent., so that in calculating fat abstracted for the purpose of certifying under this Act, f_1 must be taken as 3.0.

¹ *Journ. Amer. Chem. Soc.*, xxi., 503-508.

If the amount of fat that has probably been abstracted is required it will be better to take f_1 as 3·9 instead of 3·0.

It must, however, be remembered that there are considerable variations of fat in genuine milks, and that the amount of milk fat is often much higher than 3·9

§ 150. (3.) *Adulteration with Separated Milk*.—The adulteration of whole milk with milk from which a large portion of the fat has been abstracted, is the most common of all adulterations. To what an extent this is done may be seen by comparing the milk as supplied by farmers with the milk sold by retailers in any large town.

This form of adulteration can only be detected when the original composition of the milk is known, or when the fat in the milk falls below the standard taken.

§ 151. (4.) *The Adulteration of Fresh Milk with Unsweetened Condensed Milk, Boiled Milk, or Sterilised Milk*.—This has been studied by Faber, and also by Messrs. Droop Richmond and Boseley.¹ The original suggestion of Faber, that this might be detected by the diminished amount of soluble albumen in milk, has been confirmed by Richmond and Boseley. They find the percentage of soluble albumen in fresh milk to vary from 0·41 to 0·45 per cent., as estimated by Sebelien's method of separating casein and albumen (see page 227).

C. H. Stewart gives the following table, showing the percentage of soluble albumen in milk which has been raised to various temperatures:—

Time of Heating.	Soluble Albumen in Fresh Milk.	Soluble Albumen in Heated Milk.
10 minutes at 60° C.,	·423	·418
30 " " " "	·435	·427
10 " " 65° C.,	·395	·362
30 " " " "	·395	·333
10 " " 70° C.,	·422	·269
30 " " " "	·421	·253
10 " " 75° C.,	·380	·07
30 " " " "	·38	·05
10 " " 80° C.,	·375	none
30 " " " "	·375	none

The following test will also be found useful :—If to 5 c.c. of milk about ¼ gramme of paraphenylene-diamine be added, and then two drops of 10 per cent. H₂O₂ solution, fresh milk gives a blue colour; pasteurised milk, faint blue; sterilised milk no colour.

(5.) *Gelatin*, see page 270.

(6.) *Starch—Dextrin*.—These will be readily detected by their characteristic reactions with iodine.

Cane Sugar—Glucose—Invert Sugar—See *Condensed Milk* (p. 264).—For the simple qualitative detection of cane sugar in milk, Cotton's method may be used:—10 c.c. of the milk to be tested are mixed with 5 grms. of powdered ammonium molybdate and 10 c.c. of dilute HCl (1 in 10). A second tube containing milk free from cane sugar is treated in the same manner. The two tubes are placed in a water bath and gradually heated. At about 80° C. the milk, if it contain cane sugar, becomes blue, the control tube showing but little colour.


¹ *Analyst*, July, 1893.

§ 152. (7.) *Preservatives*.—The substances most frequently added to milk in order to preserve it for a short time are borax and boracic acid. Usually mixtures of these two substances are sold to farmers and dairymen under trade names, such as 'Arcticanus,' 'Conservine,' 'Glacialine,' 'Preservitas,' and others. The majority of these contain 10 to 20 per cent. of borax ($\text{Na}_2\text{B}_4\text{O}_7 \cdot 10\text{H}_2\text{O}$) and 70 to 80 per cent. of boric acid (H_3BO_3).

Next in importance to boron compounds is a 40 per cent. solution of formic aldehyde, known as 'Formalin,' and sold as such or under various trade names, such as 'Lactic fluid,' 'Steryl,' etc.

Other preservatives which have been used from time to time are:—

Salicylic acid and sodium salicylate, sodium chloride and sodium carbonate, potassium nitrate, sodium fluoride, sodium fluosilicate, fluoboric acid and fluoborates, β -naphthol, glycerin, sodium benzoate.

Richmond¹ describes a curious preservative labelled thus  consisting of 12½ per cent. free boric acid, 13 per cent. of salicylic acid, combined with soda and magnesia in glycerin solution, a bitter principle being also present.

§ 153. *The Detection and Estimation of Preservatives in Milk*.—A method applicable to all preservatives and capable of dealing rapidly with a large number of samples is as follows:—²

Measure out 10 c.c. of each sample of milk into wide clean test tubes. Add to one test tube as a control 10 c.c. of milk known to be free from preservatives. Then to the control and each of the others add 1 c.c. of a strong litmus solution; examine the colour of all the tubes, and if they are not the same shade of blue make them so by adding drop by drop a half normal solution of sodium carbonate, heat in boiling water for 10 minutes, allow to cool, then inoculate each tube with .5 c.c. of a very dilute solution of sour milk in water. Place the tubes, plugged with cotton-wool, in an incubator at about 20° C. The control milk will become white in about 24 hours, while those tubes containing preservatives will remain blue or pink, the shade varying with the quantity present.

§ 154. *Borax—Boric acid—Detection*.—Evaporate down 5 or 10 c.c. of the milk previously made alkaline, thoroughly char the residue, boil the char with a little water, then acidify with HCl and dip in either ordinary turmeric paper, or turmeric paper which has been dipped in oxalic acid solution and dried; dry the turmeric paper over a flame; it will become pink changed to olive green or blue by alkalis, if boric acid be present. The test with oxalic acid turmeric papers is a modification of a test proposed by Cassal and Gerrans,³ and will be found far more delicate than the ordinary turmeric method. Or to the ash a little glycerin may be added, the mixture stirred with a glass rod, and a portion on the loop of a platinum wire introduced into the Bunsen flame, and examined by the spectroscope, when the bands peculiar to boracic acid will be seen, and may be compared with the spectrum of pure boracic acid. Or the ash may be decomposed by pure sulphuric acid, a little alcohol added; on heating and setting the vapour alight a transitory green flame is observed.

*Quantitative Estimation of Boric Acid*⁴ (Moissan's method).—The borate

¹ *Departmental Committee on Food Preservatives, etc.*, 1901, Appendix xxxi.

² Meredith Wynter Blyth, *Analyst*, vol. xxvi., 148.

³ *British Food Journal*, October 1902.

⁴ H. Moissan, *Comptes Rendus*, 1893, cxvi., 1087.

is put into a small flask and heated by means of a calcium chloride bath with 1 c.c. of nitric acid. The mixture is distilled to dryness. (The distillate is best received in strong ammonia solution.) The flask is cooled; 10 c.c. of methyl alcohol added, 1 c.c. of water, and 1 c.c. of nitric acid, and the mixture again distilled to dryness; this operation is repeated three or four times until the complete distillation of the methyl borate is rendered certain by testing a drop of the distillate. The distillate is poured on to from 8 to 10 grms. of calcium oxide (made by igniting calcium nitrate) evaporated to dryness on the water bath, and then the crucible ignited in a muffle or before the blowpipe, so as to convert it into calcium oxide and calcium borate; the gain in weight is B_2O_3 . The residue should in all cases be tested for calcium carbonate; this may easily be present from insufficient ignition.

Hegner prefers to distil directly into a measured volume of standard sodium phosphate solution, which is then evaporated and ignited. The increase in weight is due to boric acid.

A better method¹ probably is that which is based upon the volatility of ammonium-boron-fluoride. The borate (Reischle) is mixed with six times its bulk of sublimed ammonium fluoride in a platinum crucible and warmed for some time so as to volatilise the greatest part of the ammonium-boron-fluoride. After cooling, sulphuric acid is added, and the excess of sulphuric acid with the remnant of fluoride got rid of by heat. In this way an ash is obtained containing sulphates, but free from boric acid. To a milk ash it would be advisable to add some caustic lime before adding the ammonium fluoride.

Parmentier² divides a solution of boric acid in either hydrochloric or sulphuric acid in two equal parts, and estimates the acidity in the one by using helianthin as an indicator, in the other by litmus, with soda, since boric acid is insensible to helianthin; the difference between the two titrations he considers equals the contents of boric acid.

R. T. Thomson³ has also proposed a method of titrating boric acid, utilising the fact that an aqueous solution of boric acid is neutral to methyl-orange and slightly acid to phenolphthalein, and this will be found the best method to use in ordinary cases. We have made a large number of determinations by this method, and find that with care the estimation is very accurate. Make 100 c.c. of the milk distinctly alkaline with caustic soda, and evaporate to dryness in a platinum dish. Char the residue, heat with 20 c.c. of water and add hydrochloric acid drop by drop until only carbon is left undissolved. Transfer the liquid, which should not measure more than 50 c.c. to an 100 c.c. flask. Add about .5 gramme of dry calcium chloride. Add a few drops of phenolphthalein solution, then run in a 10 per cent. solution of caustic soda until a permanent pink colour is obtained, then add 25 c.c. of saturated lime water. Make up the total bulk to 100 c.c.; shake well, filter through a dry filter into a 50 c.c. flask. Treat this 50 c.c. as follows:—Add normal H_2SO_4 until the pink colour disappears, then a few drops of methyl-orange, then the acid until the yellow colour just changes to pink. Add very carefully $\frac{N}{5}$ NaOH until the yellow just returns. Boil, cool, add as much glycerin as will give about 30 per cent. of this substance. Titrate with

¹ *Zeit. f. anal. Chem.*, 1894, 373.

² *Comptes Rendus*, cxiii., 41.

³ *Journ. Soc. Chem. Ind.*, xii., 432. *Analyst*, July, 1893.

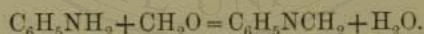
$\frac{N}{5}$ NaOH until a permanent pink colour is obtained. Each 1 c.c. $\frac{N}{5}$ NaOH = .0124 boric acid, or .007 B₂O₃.

§ 155. *Formaldehyde in Milk.*—'Formalin,' a 40 per cent. solution of formaldehyde, is used at the present time as an antiseptic; 15,000 gallons of formalin are stated to have been sold to milk vendors in 1894. The amount added is about 5 ozs. of formalin to a gallon of milk, equalling in a litre to about 21 mgrms. of formaldehyde; extended experience has shown that it preserves milk for several days with but little change. R. T. Thomson found that a sample of milk, to which 35 grains of formalin per gallon had been added, contained at the end of eight days 0.07 per cent. lactic acid, and at the end of eleven days 0.10 per cent. of lactic acid, the same milk untreated yielded 0.68 and 0.71 per cent. lactic acid respectively.

Formalin must be looked for in the sample directly it arrives, as it disappears rapidly from milk (see also *Colouring-Matter*, p. 241). The best test for formalin is Hehner's:—Take 10 c.c. of milk in a test tube, add one drop of ferric chloride solution, and dilute the milk to about 30 c.c.; pour concentrated sulphuric acid down the side of the tube, so as to form a layer at the bottom of the milk. A violet-blue ring will be formed at the junction of the liquids, if formic aldehyde is present. The following confirmatory tests may be applied to a distillate from the slightly acidified milk:—

To the fluid supposed to contain formaldehyde is added 0.5 c.c. dimethyl-aniline, acidified with a few drops of sulphuric acid, and after shaking heated on the water-bath for half an hour. The solution is then alkalisied and boiled until the smell of dimethyl-aniline has disappeared. The liquid is filtered through a small filter, and the filter washed a few times with water; it is then opened, spread on the bottom of a porcelain dish, moistened with acetic acid, and finely-powdered peroxide of lead added; a blue colour will appear if formaldehyde is present.

For both the detection and quantitative estimation of formaldehyde the reaction with aniline can be utilised. The solution of formaldehyde is dropped slowly into aniline solution (0.3 per cent. strength) and a precipitate forms of anhydro-formaldehyde-aniline, C₆H₅NCH₂, according to the reaction



After forty-eight hours, the precipitate is filtered off, dried at 40° C., and weighed. Although the precipitate forms in solutions of so feeble strength as 1 : 20,000, and the compound is 3.5 times heavier than the original formaldehyde, it is doubtful whether the minimal quantities of formaldehyde added to milk can be satisfactorily estimated by this or any other process which entails actual weighing.

Richmond and Boseley have proposed to test the milk distillate by diphenylamine. A solution of diphenylamine in water is made by the aid of a little sulphuric acid. The distillate is added to this solution and boiled; in the presence of formaldehyde a white flocculent precipitate is deposited, often coloured green if the acid contained nitrates. Schiff's reagent is useful for testing for formalin; it is a solution of magenta bleached by sulphurous acid; excess of sulphurous acid must be avoided. In presence of an aldehyde Schiff's reagent produces a pink colour. If, therefore, to a distillate made acid with one or two drops of hydrochloric

acid, Schiff's reagent produces a pink colour, it is a sign that a volatile aldehyde is present.

Mr. Bevan¹ has shown that quite small quantities of formalin increase the total solids of milk. In cases where only a few drops of formalin have been added, the increase will not amount to more than 0·2 per cent., but large quantities would appear to increase the weight considerably; thus Mr. Bevan evaporated 5 c.c. of a mixture of equal parts of milk, sugar, and albumen, and obtained a residue of 7·59 total solids per cent. On evaporating down the same quantity with the addition of 1 c.c. of formalin, the total solids rose to 9·29 per cent., a difference of 1·7. Part of the increase may be due to a polymer, and part to conversion of milk-sugar into galactose.

Formaldehyde is, without a doubt, poisonous, and the question has to be discussed as to whether it is permissible to preserve articles of daily food, such as milk, by its addition. The Departmental Committee on Food Preservatives, etc., 1901, recommend that the addition of formalin to milk shall be absolutely prohibited.

Trillat and Berlioz² have shown that 0·8 grm. injected in a single dose subcutaneously into guinea-pigs produces rapid death; poisonous effects, but not fatal, are produced by quantities of from 0·53 to 0·66 grm., while 0·038 grm. produces no apparent symptoms. It is evidently excreted by the kidneys, for the urine of animals thus treated does not putrefy like other urine.

If it affects man in anything like the same proportion as guinea-pigs, a man weighing 68 kilos. (150 lbs.) would require to take in a single dose about 17 grms. in order to produce any appreciable effect, and probably even more than this, for organic poisons generally act less energetically when swallowed than when introduced directly into the circulation or beneath the skin. It follows then that from 20 to 30 mgrms. per litre of formaldehyde are not likely to be poisonous; but whether an antiseptic of this kind taken daily, even in small doses, will render the milk less easily assimilable or nutritious is not known. Many interesting experiments on formic aldehyde in food will be found in the *Appendix to Report of Departmental Committee on Food Preservatives, etc.*, 1901.

§ 156. *Estimation of Formic Aldehyde.*—Commercial milks contain, when preserved with formic aldehyde, from ·0002 per cent. to ·006 per cent.; ·001 per cent. is considered as the maximum in the Government Laboratory, but as milks are examined there late, by the time the Government chemists analyse the milk much of the aldehyde must have disappeared.

The estimation must be made as rapidly as possible, owing to the destruction of the aldehyde by micro-organisms. The following process³ may be used:—

A bulb of about fifty cubic centimetres capacity is blown on a piece of soft glass tubing of about $\frac{1}{4}$ -inch bore; one end of the glass tube is drawn out close to the bulb into a capillary tube and turned at right angles to the bulb; the tube at the other side of the bulb is also turned at right angles to the bulb (see fig. 34b).

Ten cubic centimetres of the milk to be examined are introduced into the bulb and made slightly acid, if the milk is not already sour. The end

¹ *Analyst*, July, 1895.

² *Comp. Rend.*, t. cxiv., 1892.

³ "The Detection and Estimation of Preservatives in Milk," by Meredith Wynter Blyth, *Analyst*, xxvi., 503. Meredith Wynter Blyth and W. L. Alton, *British Sanitarian*, vol. i., 9.

of the capillary tube is now sealed up. The bulb is completely immersed in a paraffin bath, and the open tube connected with a very short rubber connection with a series of bulbs containing each five cubic centimetres of water. The bulbs are placed in a bath containing cold running water. The paraffin is then heated to 120° C., and the milk distilled nearly to dryness; frothing up the tube when it occurs can easily be prevented by warming the tube with a burner. When the milk is nearly dry the capillary tube is broken at the end, and a slow and steady current of air aspirated through the apparatus by means of a water pump, the temperature of the paraffin is allowed to rise until it reaches 200° C., and kept at this temperature for at least 15 minutes. The bulbs are then disconnected, and the second bulb tested for formic aldehyde; none will be found unless the original solution contained a very large amount; should there be any present it must be estimated, and the third bulb tested.

The first bulb is washed out with distilled water, the whole bulk

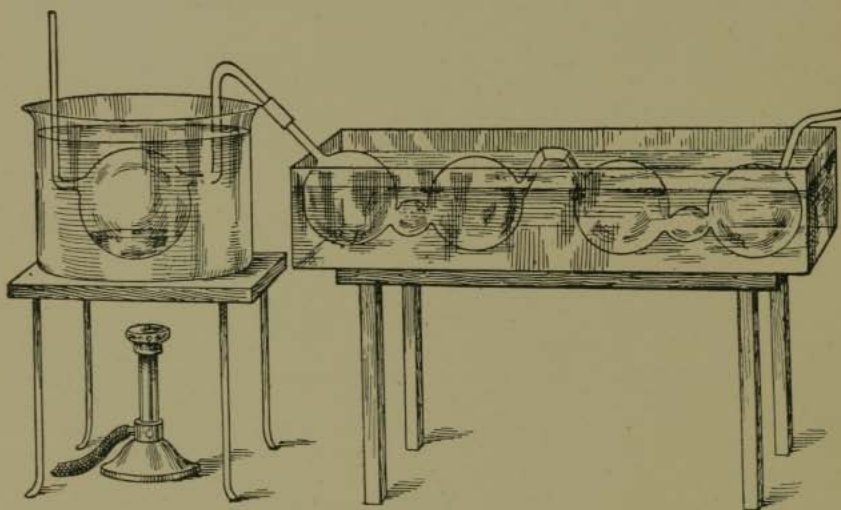


FIG. 34b.

should not be more than twenty cubic centimetres, two drops of litmus are added, and a few drops of decinormal sodium carbonate solution until neutral.

A weak standard solution of formic aldehyde is now prepared.

This is best effected by standardising a fairly strong solution of formic aldehyde either by Romijn's iodine method or his cyanide process;¹ the latter is as follows:—

Ten cubic centimetres of a decinormal solution of silver nitrate are mixed with ten cubic centimetres of a slightly under decinormal solution of potassium cyanide, containing a few drops of 50 per cent. nitric acid, the solution is made up to 50 cubic centimetres and filtered through a dry filter, and the excess of silver nitrate estimated, in 25 cubic centimetres of the filtrate, by means of a solution of ammonium sulphocyanate. A measured quantity of formic aldehyde solution is now added to 10 cubic centimetres of the cyanide solution, and the process repeated in exactly

¹ *Zeit. f. analytische Chem.*, 1897, xxxvi., 18-24.

the same way; the difference in the two readings gives the formic aldehyde present, formic aldehyde and cyanide uniting in molecular proportions.

The advantages of this method are that the solution of silver can be easily standardised, that it can be used in the presence of acetone and other organic compounds, which entirely vitiate the results of any oxidation process. It can also be used in the presence of acetaldehyde, if the titration is carried out at once.

Control tubes are made up with fresh or sterilised milk and the standard solution of aldehyde, so that the several tubes contain (1) .008 per cent., (2) .005 per cent., (3) .002 per cent., (4) .001 per cent., (5) .0008 per cent., (6) .0006 per cent., (7) .0003 per cent., of formic aldehyde.

Various quantities of the neutral milk distillate are also injected in duplicate into portions of the same milk; the best quantities to take are two test tubes containing 5 c.c. of milk and 5 c.c. of the distillate; two tubes containing 7 c.c. of milk and 3 c.c. of distillate and two tubes of 8 c.c. of milk and 2 c.c. distillate. All these tubes are treated with litmus exactly as described on page 234; control tubes 1 to 3 and one set of the other tubes are placed in the warm incubator at 38° C., the rest of the tubes are placed in the cool incubator at 22° C. They are examined after 24 hours and from time to time. From a comparison of the colours the amount of preservatives may be judged. It is best to distil sufficient, in the first place, to make a second experiment, if necessary; care must be taken not to shake the tubes after they have once been put to incubate, as the action of the milk is to reduce by abstracting oxygen, so that if the tubes be shaken they will take up oxygen and become red.

§ 156a. *Glycerin in Milk.*—A process of preserving milk by glycerin exists, and occasionally it is found in milk. To detect glycerin the casein, fat, and albumen must be separated by dilution, acetic acid, carbon dioxide, and heat, as described at p. 227. The sugar is then estimated in one portion of the yellow whey by copper solution; the remainder is first neutralised, and then evaporated to dryness, and freed from any trace of fat by exhaustion with pure ether. The glycerin is now dissolved out by a mixture of alcohol and ether, the alcohol-ether evaporated off, and the glycerin identified by its physical characters and the production of acrolein fumes when heated with sulphuric acid.¹ Use may also be made of the fact that glycerin sets free boracic acid from borax. A little borax, therefore, may be moistened with the syrupy drops supposed to be glycerin, heated in a Bunsen flame, and examined before the spectroscope for the boracic acid bands.²

Salicylic acid is used occasionally as a preservative of milk, and it is easily detected by shaking up milk whey (first acidified by hydrochloric acid) with ether. The ethereal solution on evaporation leaves the acid in a pure enough state to permit the successful application of reagents. The best test for salicylic acid is the beautiful violet colour which it gives with a neutral solution of ferric chloride. Besides this test, a minute portion may be placed in the subliming cell, when a well-marked sublimate is obtained at about 100° C. The crystalline form of this sublimate may be

¹ A test for glycerin has been proposed by E. Donath and J. Mayshofer (*Zeitschrift für anal. Chem.*, xx., 79). The supposed glycerin is heated to 120° with two drops of phenol and the same quantity of concentrated sulphuric acid. The whole is then treated with a little water, and the insoluble portion rendered slightly ammoniacal. Under these circumstances glycerin gives a beautiful red colour.

² A. Senier and A. J. G. Lowe: *Chem. Soc. Journal*, clxxxix., Sept., 1878.

compared with one obtained from a known pure sample of salicylic acid. The melting point of pure salicylic acid is 155° to 156° C.

Benzoic acid is also occasionally used, and it may be detected as follows:—200 c.c. or more of milk are alkalisied with baryta water and evaporated down to one-fourth; the thickened residue is next mixed with calcic sulphate to a paste and dried on the water-bath. The mass is finely powdered, moistened with dilute sulphuric acid, and extracted with cold 50 per cent. alcohol. The alcoholic extract is neutralised with baryta water, and concentrated by evaporation to a small volume. The liquid is now acidulated with dilute sulphuric acid and extracted with ether. On separating and evaporating the ether, any benzoic acid is left sufficiently pure to respond to the usual tests. That with ferric chloride succeeds best if to an aqueous solution of the acid a little sodic acetate is first added. For a quantitative estimation of the acid, it is best to sublime the acid, and weigh the sublimate, checking this weight by ascertaining the loss the residue has experienced.

§ 157. *Sodium Carbonate* will be detected by the effervescence of the ash when treated with a drop of HCl.

Leon Padé¹ states that the soluble ash from 10 c.c. of genuine milk only requires one drop of d. n. acid for neutralisation, but if sodic bicarbonate is present more will be required. Part of the carbonate may have been converted into phosphate, therefore soluble phosphate must be estimated and calculated into NaHCO_3 .

§ 158. *Sodium Fluoride, Sodium Fluosilicate, Fluoboric Acid, and Fluoborates*.—The ash from 25 to 50 c.c. of milk is treated in a platinum dish, with strong sulphuric acid, the dish is warmed to about 98° C., and in the vapour is suspended a cold watch glass coated with paraffin, from which the paraffin has been scratched off in a few places. The vapour is allowed to act for an hour; if a fluoride be present the glass will be etched in the parts unprotected by paraffin; this etching, where traces of fluorine are present, will only be evident by breathing on the glass.

Richmond suggests placing a drop of water on the paraffin away from the lines scratched on it, for the purpose of detecting fluosilicates; if the latter be present, a white film of silica will be formed on the surface of the drop.

Fluo-borates give the reactions of both boric acid and fluorides.

§ 159. *β -naphthol*.—Several compounds of β -naphthol are now used as preservatives. Leffmann and Beam² describe one known as 'Abrastol' or 'Asaprol.' Professor Thorpe³ has met with β -naphthol-sulphonate of soda which is said to be used on the continent. But these substances, so far as known, are not used in this country. Leffmann and Beam give the following test:—200 grms. of the sample are acidified with sulphuric acid and steam distilled until 150 c.c. of distillate are obtained. This liquid is shaken with 20 c.c. of chloroform, the latter withdrawn, rendered alkaline with potassium hydroxide and heated almost to boiling for a few minutes. Colour changes occur as follows:—

Salol . . .	light red.
Phenol . . .	light red—to brown—to colourless.
β -naphthol . . .	deep blue—to green—to brownish.

¹ *Ann. chim. anal. appl.*, 1886, i., 33, 8.

² *Select Methods of Food Analysis*, 1901, 84.

³ *Report of Departmental Committee on Food Preservatives, etc.*, 1901. Appendix iii., 363.

§ 160. (8.) *Artificial Colouring Matter in Milk.*—The artificial colouring of milk is a practice which has increased enormously during recent years, so much so indeed that it is now the exception to find the milk as sold in the large towns of England uncoloured. At the time of writing there is no regulation prohibiting the use of harmless colouring matters in milk, but the Departmental Committee on Food Preservatives, etc., 1901, in their report recommend that the use of colouring matters in milk offered for sale in the United Kingdom shall constitute an offence under the Sale of Food and Drugs Act.

The colouring matters said to have been used at various times in milk include: coal-tar dyes, annatto, turmeric, saffron, carrot, marigold, caramel, and chromates, but the only substances used to any extent in the United Kingdom are annatto and certain sulphonated-azo-dyes and mixtures of these substances. In America caramel is also used occasionally. The amount of colouring matter added is about 1 part to 200,000 parts of milk.

The colouring matters at present used may be detected as follows:—

(1.) The fresh milk gives a pink colour with hydrochloric acid: presence of a *sulphonated-azo-dye*.

Confirm by extracting as below, and apply the usual tests.

(2.) A piece of filter-paper soaked for twenty-four hours in the milk made alkaline with sodium carbonate takes a brown stain which is changed to pink by hydrochloric acid: presence of *annatto*.

Confirm by extracting as below, and applying the following tests:—(α) A drop of the colouring matter dissolved in water and made alkaline with potash gives an orange stain on filter-paper, changed to pink by stannous chloride solution. (β) A little of the residue is dissolved in water containing a little alcohol and a drop of ammonia. A bundle of white cotton fibres is introduced, and the liquid evaporated nearly to dryness. The fibre is then immersed in a solution of citric acid. It will be coloured rose-red if annatto is present.

(3.) *Caramel.*—Coagulate 10 c.c. of the milk by means of acetic acid, collect the curd by straining through linen, then place it in a white porcelain dish, and just cover with strong HCl. Treat a control sample known to be free from caramel in the same manner. A blue violet colour indicates the presence of caramel.

Other colouring matters may, however, be present, or the original presence of certain coal-tar dyes may be masked by the sample being partly decomposed, since one of the authors has shown¹ that certain coal-tar dyes are reduced very rapidly in decomposing milk under the influence of nascent hydrogen set free by the action of anaerobic organisms. In the latter case it is possible to isolate their decomposition products.

For the isolation of colouring matters and their decomposition products, the following method may be used:—Take at least 60 c.c. of the milk, carefully add to it weak soda or strontia solution until it is just alkaline to delicate litmus paper. Evaporate the solution to a thin paste on the water bath.

(a.) Thoroughly extract the paste with ether. This will remove the fat, and, if the milk was sour, the decomposition products of those dyes reduced by nascent hydrogen. Evaporate off the ether, and shake the fat with warm water. Separate the water from the fat, and evaporate the water to dryness in a white porcelain dish; note the colour of the residue. (Pure milk yields no coloured residue.) To the coloured residue add

¹ "The Detection of Artificial Colouring Matters in Fresh and Sour Milk," by M. Wynter Blyth. *The Analyst*, vol. xxvii., p. 146, 1902.

various reagents as in the following table, which gives the reactions of a few dyes :—

WATER EXTRACT FROM THE FAT.

Note the Colour and apply various Reagents to the Dry Residue.

Colour of Residue.	Probable Original Colouring Matter.	Add a Drop to Ferric Chloride.	To the Ferric Chloride add Strong H_2SO_4 .	Other Reactions.
Brown	Acid yellow	Dark green	Yellow, green on dilution	
Brown	Butter yellow	Dark blue-green	Yellow, green on dilution	To a slightly acid solution of the colouring matter add a few drops of H_2S solution, then ferric chloride, heat—magenta colour.
Brown	Aniline yellow	Yellow	Yellow	Strong H_2SO_4 , or HCl , gives a delicate violet.
Yellow	Martius yellow	Red	Yellow	Strong HCl gives a beautiful rose red colour.
Yellow	Victoria yellow	Red	Yellow	Same as Martius yellow.
Rose red	Methyl orange	Fugative scarlet	Yellow	Treat as with butter yellow, the solution becomes a beautiful blue.
Brown-red	Orange IV.	Green	Scarlet, green on dilution	Treat as butter yellow, the solution becomes dirty violet.
Yellow	Unreduced dyes, soluble in ether from alkaline solution			

(b.) Extract the fat-free residue with boiling alcohol, filter and evaporate to dryness in a white porcelain dish. A portion of the residue if yellow or orange may be taken up with a little d. n. acid, and shaken with ether; this will divide the possible dyes into two groups, *i.e.*: (a) the natural colouring matters (annatto, turmeric, saffron, etc.), the acid coal-tar dyes, and the non-sulphonated-azo-dyes, which all colour the ether to a certain extent. (β) The basic dyes and the sulphonated-azo-dyes, which do not colour the ether at all. The sulphonated-azo-dyes will be readily recognised by striking brilliant colours with acids, and the other dyes may be recognised by the ordinary methods.

(9.) *Detection of Nitrates in Milk.*—Pure milk contains no nitrate, hence the detection of nitrates in milk would be fair presumptive evidence of watering. A test recommended by J. Szilasi¹ is as follows :—1 c.c. of a solution of diphenylamine sulphate is put into a small porcelain dish, and a few drops of milk are dropped in; if nitrates are present, a blue colour will gradually appear. Soxhlet tests milk for nitrates as follows :—The milk is coagulated by a solution of calcic chloride free from nitrate, the serum is now treated with a solution of diphenylamine in concentrated

¹ *Repert. anal. Chemie*, xxxiii., p. 212.

sulphuric acid in the same way as with the ferrous sulphate test. The nitrate test will be found of great utility in those cases in which an impure strongly nitrated well-water has been added to milk, but with ordinary waters containing .01 grm. and under of nitric anhydride per litre, the test is not sufficiently delicate to be of much utility.

§ 161. *Sour Milk.*—*Government Laboratory Method.*—The analysis of the milk is conducted by what is known as the 'maceration' process, and the weight of the non-fatty solids and fat is independently ascertained in duplicate, whilst as a control the direct determination of the total solids is made on a third portion of milk. Before proceeding with the analysis, the total contents of the bottle are transferred to a suitable vessel and thoroughly mixed by means of a wire whisk.

From ten to twelve grammes of the sample are weighed out into flat-bottomed platinum capsules, each of which has been tared along with a short glass rod with flattened ends. The weighed portions are next neutralised with decinormal strontia solution, with phenolphthalein as indicator. Decinormal soda answers the purpose perfectly well when the degree of acidity is not greater than 10 c.c. n/10 solution for 10 grammes of milk, but above that amount it is of great advantage to use the strontia solution.

The milk is then evaporated over the water bath until the residue attains the consistency of dry cheese, and while so warm as to ensure that the fat shall be still in the melted condition, 20 c.c. of dehydrated ether (specific gravity .720) are poured over the milk solids, which are then carefully stirred for some time with the glass rod. The ether containing the dissolved milk fat is passed through a Schleicher's filter of 10 cm. diameter, which has previously been dried and weighed in a weighing bottle. The maceration of the milk is continued with eight successive additions of 10 c.c. of ether, at which point it has been found that the fat has been wholly separated from the non-fatty solids. The filter-paper having its edges cut down is well washed with boiling ether, and the fractions of the filter-paper are replaced in the weighing bottle and dried at 100° C. The increase of weight, which with careful work should not exceed a few milligrammes, is added to the weight of the non-fatty solids. At the conclusion of the maceration process, the non-fatty solids should be in a fine state of division, resembling the precipitated chalk of pharmacy. The capsules are next dried overnight in the water oven and the first weighing is taken in the morning. Usually the weight is constant by this time, as shown by the weighings taken during the day. As from the conditions of the analysis the anhydrous salts of sodium or strontium are present in the dried solids, the correction for the added alkali is in the proportion of .0022 gramme per 1 c.c. of n/10 soda used and .0042 gramme for each c.c. of the strontia solution required.

The ether solution of the milk-fats contained in tared beakers is evaporated and the weight of the dried fat ascertained.

In determining the nature and amount of the loss in non-fatty solids consequent on keeping, the alcohol present is by far the most important item. When the milk sample under analysis measures 170 c.c., that is nearly 6 ounces (and it should not be much less than this), 75 grammes of the milk can be spared for distillation, which is carried out in a glass still, with glass spiral condenser.

The acidity of the milk having been previously ascertained, the portion for distillation is neutralised with soda up to one-half of the total acid present. If nearly neutralised, ammonia may pass into the distillate and

so vitiate the result. The first distillate, which usually contains a little acid, is re-distilled with the addition of 0.5 c.c. n/10 NaHO, and, after having been made up to original bulk, the specific gravity at 15.5° C. (60° F.) is taken in a 50 gramme or in a 1000 grain weighing bottle. Supposing that the distillate gave a specific gravity of 999.67, or 0.33 less than 1000, then this difference multiplied by 1.16 gives the amount of proof spirit by volume present in the milk. The product multiplied by .842 equals the actual weight of anhydrous milk sugar which has been converted into alcohol.

The correction for free volatile acid, reckoned as acetic acid, is ascertained as follows:—

10 grammes of the milk are neutralised to the extent of one-half the total acidity with n/10 NaHO, and a little phenolphthalein added. The mixture is then evaporated to dryness on a water bath with frequent stirring, and after treatment with 20 c.c. of boiling distilled water so as to break up and thoroughly detach the milk solids from the capsule, a further addition of n/10 NaHO is made, until the neutral point is reached. The difference between the original acidity of the milk and that of the evaporated portion is regarded as acetic acid. The number of c.cs. of soda shown, when multiplied by .425, gives the percentage of loss by weight not recovered in the change of milk sugar into alcohol and thence into acetic acid.

Example:—

Acidity of milk,	11.6 c.c. n/10 NaHO.
" " evaporated portion,	9.2 " "
Difference,	2.4

$2.4 \times .0255$	= .061 per cent. by weight lost,
or	
$2.4 \times .006 \times 10 \times .425$	= .061 " " "

The loss arising from the change in the casein is very small.

To estimate it, two grammes of the milk are made up to 100 c.c. with distilled water and filtered to a clear solution. 10 c.c. of the filtrate increased to 50 c.c. by the addition of distilled water are Nesslerised against NH_4Cl solution, equivalent to .01 mgr. of NH_3 in each c.c. As the Nessler colour produced in presence of milk differs somewhat from that of pure saline ammonia, the blank experiment is carried out with the addition of 10 c.c. of the filtrate from two grammes of new milk slightly acidified, and diluted to the same extent as the sour milk. The quantity of test ammonia required varies between 0.5 c.c. and 4 c.c.

In the case of a milk containing ammonia equal to 2.6 c.c. of the test solution, the loss of weight is calculated as follows:—

$$\begin{aligned} & .00001 \text{ mgm.} \times 2.6 \times 500 \times 5.2 = .067 \text{ per cent. casein} \\ \text{or} \quad & \frac{2.6 \times .052}{2} = .067 \quad " \quad " \end{aligned}$$

It is evident that any other degree of dilution may be conveniently adopted according to circumstances, or the proportion of ammonia which might be indicated in the milk.

According to the three instances given, the loss of solids, or, in other words, the addition to be made to the non-fatty solids, is as follows:—

Loss from alcohol,322	per cent.	} Total. .45 per cent.
„ acetic acid,061	„	
„ ammonia,067	„	

THE CHIEF INFLUENCES WHICH TEND TO MODIFY THE COMPOSITION OF COWS' MILK.

§ 162. Although for the purposes of the Sale of Food and Drugs Act the analyst will only be required to determine if the 'solids not fat' fall below 8.5 per cent., the milk-fat below 3.0 per cent., and that the milk is of normal composition; yet most chemists will be called upon from time to time to decide questions with regard to the watering and skimming of cows' milk which have no connection with the Sale of Food and Drugs Act. To these a few facts as to the causes of variations observed in the composition of cows' milk will be useful.

As before stated, a marked distinction must be drawn between the milk of individual cows and that from a mixed herd of cows. The milk of individual cows often varies greatly from the normal. Richmond records the following extreme variations in 'solids' and 'fat,' the highest fat being from Bannister's determination:—

	Fat. per cent.	Solids not fat. per cent.
Maximum,	12.52	10.60
Minimum,	1.04	4.90

The period of lactation, the breed of cow, and other circumstances naturally influence the composition of the milk of individual cows to a much greater extent than the milk from a mixed herd. The chief influences tending to modify the composition of cows' milk are the following:—

(1) The breed, (2) period of lactation, (3) season of year, (4) conditions as to milking (morning and evening milking, foremilk, strippings, etc.), (5) climate, (6) land, (7) feeding.

(1) *Breed*.—Table XVIII_f. will give a good idea of the considerable variation in the quality of the milk from cows of different breeds.

Breed is now recognised as the most important factor influencing the quality of milk.

As a rule, the richer the quality, the less the quantity of milk. Thus Jersey cows give a small quantity of rich milk, Shorthorns a large quantity of medium quality milk.

In this country the non-pedigree Shorthorn is the cow most usually kept for producing milk for ordinary consumption, but many dairymen keep a few animals, such as Channel island cows, yielding a richer milk so as to enhance the quality of the mixed milk.

According to Mr. George Barham, in Scotland and the eastern counties—Ayrshires; in Norfolk and Suffolk—Redpoles; and in Ireland—Kerries are the cows commonly kept.

Pure bred Dutch cows are not now found in England; it is true that there is a herd in Cheshire, and a few in other places, but the milk

is not sold for ordinary consumption. In Essex there are also a number of cross bred cows derived from Dutch and Shorthorns.

TABLE XVIII.—AVERAGE COMPOSITION OF MILK OF DIFFERENT BREEDS OF CATTLE.

Breed.	Total Solids. Per cent.	Milk-fat. Per cent.	Solids not Fat. Per cent.	Authority.
Jersey,	14.89	5.66	9.23	Vieth.
"	14.34	4.78	9.56	Jersey State Exp. Stn.
"	14.65	5.43	9.22	Bell.
"	14.64	5.31	9.33	Lloyd.
"	15.40	5.61	9.79	American Exp. Station.
Guernsey,	14.48	5.02	9.46	Jersey State Exp. Stn.
"	14.46	5.16	9.30	Bell.
"	13.89	4.77	9.12	Lloyd.
"	14.60	5.12	9.48	American Exp. Station.
Welsh,	14.15	4.91	9.24	Vieth.
"	13.55	4.40	9.15	Bell.
Sussex,	14.18	4.87	9.31	Vieth.
Kerry,	13.70	4.72	8.98	"
"	13.68	4.70	8.98	Bell.
Red Poll,	13.22	4.34	8.88	Vieth.
"	12.63	3.63	9.00	Lloyd.
Ayrshire,	13.46	4.24	9.22	Bell.
"	13.28	4.13	9.15	Lloyd.
"	12.70	3.68	9.02	Jersey State Exp. Stn.
"	13.06	3.57	9.49	American Exp. Station.
Devon,	13.77	4.15	9.62	" " "
N. Devon,	13.11	3.43	9.68	Bell.
Shorthorn,	12.90	4.03	8.87	Vieth.
"	12.45	3.65	8.80	Jersey State Exp. Stn.
"	12.93	3.91	9.02	Lloyd.
"	12.78	3.93	8.85	Bell.
Pedigree Shorthorn,	12.86	4.03	8.83	Vieth.
Dutch,	12.40	3.75	8.65	Bell.
"	11.91	3.23	8.68	Fleischmann.
American Holderness,	12.63	3.55	9.08	American Exp. Stn.
Holstein,	12.12	3.51	8.61	Jersey State Exp. Stn.
" Frisian,	12.39	3.46	8.93	American Exp. Station.
Montgomery,	12.61	3.59	9.02	Vieth.
German,	12.25	3.40	8.85	Fleischmann.

§ 163. *Period of Lactation.*—The period of lactation, mainly affecting the milk of individual cows, has also to be taken into account in the consideration of the mixed milk of a herd. The greater number of cows calve in the spring and early summer, and to this fact the seasonal variations in the quality of milk are due.

The cow after calving first yields 'colostrum' (see page 214); it is not usual to sell any milk yielded on the first four days after calving. The milk for the remainder of the first week is usually rich in fat, the quality then drops rapidly to a minimum during the second week, the quality then improves until about the week before drying up, when it rapidly drops both in quantity and quality.

According to Kühne the casein and mineral matter increase with the fat, while the milk sugar diminishes. Richmond observes that the majority of abnormal milks are obtained from cows which are getting dry.

Seasonal Variations.—The period of calving and the difference of food at

different seasons of the year account for seasonal variations in the quality of the milk. Richmond summarises these differences as follows:—

- (1) November, December, January, milk rich in fat and 'solids not fat.'
- (2) February, March, and April, fat less in quantity, but 'solids not fat' about the same as during the three previous months.
- (3) May, June, July, and August, the fat is low, with a tendency to rise at the end of the period. In July and August 'solids not fat' are below the average.
- (4) September and October, an improvement both in fat and in solids not fat is noticeable.

Conditions as to Milking.—In Holland, where cows are milked three times a day at regular intervals, the quality of the three milkings is about the same, but where cows are milked at irregular intervals the quality of the milk generally differs. In this country, cows are milked twice a day, and there is a difference between the morning and the evening milk; the evening milk containing, as a rule, from .2 to .4 per cent. more fat than the morning milk. The 'solids not fat' do not appear to be affected. A number of tables proving this will be found in the *Report of the Departmental Committee on Milk and Cream Regulations*, 1901.

The difference of foremilk and strippings has already been noticed (see p. 206). A fair sample of milk is only obtained if the cow is milked dry. The composition of milk may be modified by the cow 'holding up its milk,' owing to a strange milker, novel surroundings, or nervousness from some cause or other.

Climate.—An excessively dry summer is liable to cause milk to be poor in 'solids not fat.' The average of nearly 100 determinations made at the Wisconsin Creamery during a protracted drought in 1895 gave but a trifle over 8.5 per cent. of 'solids not fat.'¹ Although the effect of drought on the amount of fat is not yet decided, the tendency would seem to raise the percentage.

An excessively wet season tends to render milk of poor quality: excessive heat or cold also affects the quality of the milk.

Land.—Cows kept on certain land would appear to yield a poor milk. This evidently is due to the quality of the food raised on the land.

§ 164. *Feeding.*—The effect of feeding is a matter on which there is much doubt and discussion. The evidence points strongly to the view that decreasing or increasing the food of a healthy cow *for a limited time* has no effect on the quality or quantity of the milk, the first effect of poor feeding being rather to decrease the weight of the animal than to alter the milk.

On the other hand, high or low feeding carried on *for a length of time* affects the milk. Lawes and Gilbert have shown that high nitrogenous feeding increases both the yield and richness of the milk.

The milk of half-starved cows is, as might be expected, of poor quality. J. Campbell Brown² and J. Carter Bell³ give some interesting analyses of the milk from half-starved cows, Bell's average for half-starved cows being 11.42 per cent. total solids, fat 2.87 per cent., and solids not fat 8.55 per cent.

§ 165. The colouring and the alkaloidal and active principles of plants impart their distinctive properties somewhat readily to milk. Thus it has long been noticed that browsing on certain plants affects the colour of milk: *Caltha palustris*, saffron, and rhubarb colour it yellow; rhubarb, opuntia, and *Rubia tinctorium*, red; *Myosotis palustris*, polygonum, and *Anchusa equisetum*, blue. Purgative vegetables, such as rhubarb, or even

¹ Löffmann and Beam, *Select Methods of Analysis*, 1901, 198.

² *Chemical News*, xxxi., 226.

³ *Analyst*, 1881, 53.

the juices of acid fruits, taken by a suckling woman almost invariably affect the infant. There are instances of milk becoming poisonous from containing the active principles of plants. In June 1875 an epidemic of diarrhoea occurred in the Rhone Gorge, and was traced to goats' milk, the goats having browsed in fields where the meadow saffron was growing. Professor Ralti isolated colchicine from the milk. Similar outbreaks, caused by the animals having fed on poisonous shrubs, have been recorded in the Western States of America and Australia.

§ 166. Experiments have been made with the object of ascertaining whether metallic compounds would be excreted by milk. Arsenic passes readily enough in minute quantities, and the same may be said of lead and oxide of zinc. Antimony also, if administered, appears in the milk. The statements with regard to mercury are conflicting, but the balance of evidence leads, on the whole, to the conclusion that it is not excreted, even in minute quantities, by the mammary glands. Bismuth, when administered, was detected in milk by Marchand, Lewald, Chevallier, and Henry. Lewald gave 15 grms. of potassium iodide to a cow, and its presence for four days afterwards was detected in the milk; 21 grms. were then given, and the drug could be detected so long as seventy-two hours afterwards. On administering it again, it was found in the milk for eleven days.

THE QUANTITY OF MILK GIVEN BY THE COW, THE METHOD OF FEEDING, ETC.

§ 167. The capacity for milk of the udder of the cow is usually estimated at about 3 litres [$\cdot 66$ of a gallon, or a little more than 5 pints]. The quantity of milk secreted is about three times this amount, but varying in individual cows, and depending on circumstances, such as the breed, the health, the size of the cow, the time after parturition, and the nature and quantity of the food given.

§ 168. The breeds in England most approved of are the Alderney, Ayrshire, Holderness, Kerry, and Suffolk. In Germany, the Swiss, Allgäuer, and Dutch cows appear to be the favourites. Some careful estimates of the amount yielded by different breeds of Continental cows have been published as follows:—

TABLE XVIII_g.—AVERAGE YIELD OF MILK.¹

	Litres per year.	Gallons per year.	Average yield in gallons per day.
Ausbacher,	1284	283·07	·77
Mürzthaler,	1500	330·70	·90
Voigtlander,	1600	352·75	·95
Simmenthaler,	1690	372·59	1·01
Saxony,	2023	446·01	1·22
Walzthaler,	2272	500·90	1·36
Pinzgäuer,	2338	515·45	1·40
Swiss,	2625	578·70	1·60
Allgäuer-Montafaner,	2697	594·60	1·62
Allgauer,	2710	597·47	1·62
Oldenburger,	2751	606·51	1·65
Dutch,	2906	640·68	1·74

¹ *Abl. Centr.-Bl. f. Agric. Chemie*, 1877, 236.

These breeds, then, are not superior to our own. The favourite cow of the London dairymen appears to be the Yorkshire cow, essentially a short-horn. The average yearly yield is from 600 to 700 gals., 15 of these cows giving about 10,000 gals. of milk yearly, or 1·7 gal. per day; individual cows, of course, occasionally exceed this. A cow has been known to give daily for some time as much as 5 gals. of milk.

The time elapsing before and after calving causes, as might be expected, considerable variation in the mammary secretion, the quantity augmenting during the first two or three weeks, and diminishing towards the end of the third or fourth month. Towards the seventh month the quantity sinks to one-half, and in the ninth and tenth months it is often reduced to three-quarters of the quantity secreted at first. On the approach of calving, the milk ceases altogether.

The age of the animal has some influence, very young cows secreting less than mature adult cows. It is also found that, *cæteris paribus*, the larger the cow the greater the yield of milk. Mr. Ockle of Frankenfeld took four Dutch milking cows, two weighed 2112 lbs., and two others only 1537 lbs.; he fed them on the same food, and submitted them to similar conditions for sixteen days. The results of this experiment are embodied in the following table¹ :—

No.	Weight at commencement of experiment. lbs.	Weight at end. lbs.	Green Lucerne consumed. lbs.	Produce of milk. lbs.
Two heavy cows, .	2112	2102	4921	68
Two light „ .	1537	1537	3859	48
			Produce in milk per 100 lbs. Green Lucerne. galls. pints. ozs.	Lucerne consumed per 100 lbs. live weight.
Two heavy cows,	1 3 16	14·6 lbs.
Two light,	1 11 16	16·0 lbs.

§ 169. The feeding of milking-cows varies somewhat according to local circumstances. In town dairies brewers' grains are much in use, and one to two bushels are given daily, besides mangolds, hay, and meal to each cow. A very common course of home-feeding is as follows² :—At 4 a.m. the cow receives two or three pecks of grass, immediately after being milked, then 4 to 5 lbs. of hay; at 9 a.m., from 20 to 25 lbs. of chopped mangolds, and another 3 to 4 lbs. of hay; at 1 p.m. there is a second milking; another similar feed follows, and she is given plenty of water. If oil cake is used, 3 to 4 lbs. a day are given either with the mangolds or in a gruel with the grain. In this country the chief dependence is placed on hay, mangolds, barley meal, and bean or Indian flour; in the summer abundance of green food is given, such as clover, vetches, cabbage, etc.

THE MILK SECRETED BY THE UNHEALTHY.

§ 170. The result of the analyses and cases shortly to be quoted shows, —(1) That in the case of the cow, in certain diseases only, the milk constantly deviates from the normal standard; (2) that the most marked changes are found in *local diseases of the udder or mammary glands*; (3) that the animal may be labouring under a most mortal and virulent

¹ "On Milk," by Dr. A. Voelcker, *Journ. R. Agricult. Soc.*, xxiv., 1863.

² T. Carrington: *Journ. R. Agricult. Soc.*, xxxix., 1878, p. 670.

malady, and yet secrete milk which, although differing from the same milk secreted by the same animal when in health, yet, considered in itself, in no way *chemically* differs from healthy milk; (4) that it is only by biological methods of experiment that such diseased milk can be detected. These remarks apply only to the *composition* of the milk; but if we also regard the *quantity* secreted, then there is in all cases a remarkable difference, for whenever an animal suffers from a sufficient amount of disease to affect its health materially, the diminution in the total quantity of milk is almost invariable.

I. HUMAN MILK.

§ 171. With regard to the milk secreted by women in various maladies, the same remarks apply only to a certain extent; for the human mammary secretion is so dependent on mental influences, that its composition appears readily affected. Vogel gives the following analysis of milk derived from a woman suffering from hysteria, the sample being taken directly after the attack:—

	Per cent.
Milk-fat,	5.14
Casein,	5.000
Sugar,	3.492
Ash,	1.010
Water,	89.984
Specific gravity,	1.032

Deveux¹ found the milk of a woman who suffered from nervous attacks, when taken in the seizure, to be a transparent viscid secretion like albumen. J. F. Simon² examined the milk of a woman who was suffering from the effects of passion. The secretion was apparently the cause of violent convulsions and diarrhoea in an infant. The milk was acid, and had acquired a peculiar odour, and after a little time developed hydrogen sulphide; or, in other words, the milk had commenced to undergo lactic acid and putrefactive changes in the breast itself. Local affections of the breast, as might be anticipated, interfere with the healthy action of the milk-producing cells. For example, Schlossberger gives the following as the composition of a sample of milk taken from a woman whose breast was considerably enlarged; the fluid was white and thick, and without odour, specific gravity, .98 to .99 at 15° C. :—

	Per cent.
Fatty matter,	8.54
Lactine and extractives,75
Casein,	8.74
Ash,41
The fat fused at 33° and solidified at 26°.	

II. COWS' MILK.

§ 172. *Aphthous Fever*.—One of the few affections in which it is possible for the investigator to discover an abnormal condition of the milk, and even from the appearance of the fluid to know what particular malady the cow is suffering from, is *foot-and-mouth disease*, or *aphthous fever*. This is a febrile, highly infectious disease, which has caused at certain times great ravages

¹ *Crell's Chemische Annalen*, vol. i., p. 369.

² J. F. Simon's "Animal Chemistry," Syd. Soc., 11, 58.

among our herds; its most obvious signs are ulcers on the mouth, feet, and teats. Unless the fever is high, the milk is secreted during the whole course of the disease. It presents different (one might almost say, opposite) appearances in different cases; in those where there are ulcers on the teat, either externally or just inside, the pus from these ulcers mixes with the milk, and the analyst finds a high fatty residue, from which cholesterin, nuclein, lecithin, and milk-fat may be separated. If, on the contrary, there are no ulcers and no local affection of the udder, the milk in the more severe cases may be deficient in solids, and especially in milk-fat; nor does it recover its normal composition until about the seventh and eighth days, when the cow begins to improve.

A special micro-organism is, according to Klein,¹ always to be found associated with apthous fever; it is a micrococcus, which either occurs singly, as dumb-bells or as streptococci; the chains of the streptococcus sometimes grow to a considerable length. The individual elements of the diplococci and streptococci are spherical, and have an average measurement of 0.006 to 0.008 mm. The micro-organism is readily cultivated in nutrient gelatin, agar-agar, or broth; it grows slowly in sterilised milk, the milk remaining fluid. Milk, according to Klein, preserves the micro-organism in a wonderful manner; infected milk kept months in tubes still yielded successful cultures. If the local affection is at all severe, blood-cells, and occasionally a considerable quantity of blood, may be found in such milk.

The following Table (XIXa.) (over) gives the composition of milk obtained from cows suffering from various diseases.

(1.) *Mammitis*.—This disease, as its name implies, signifies an acute inflammation of the mammae. Theoretically, milk secreted by an inflamed organ should be altered much in quality; but, in the case of a heifer suffering from this disease, milk taken the second day after calving did not appear to differ essentially, either in microscopical appearances or in chemical composition, from normal milk.

(2.) *Parturient Apoplexy*.—Urea was absent; there was much lactochrome. No abnormal elements detected by a microscopical examination.

(3.) *Pneumonia*.—This is the only milk in which the writer found an estimable quantity of cholesterin. The microscopical results were negative.

(4.) *Tubercle*.—The whole quantity of the fluid did not exceed 70 c.c. It was of a dirty amber colour, with the casein partially separating.

This is phthisical milk in its most intense form, and one never likely to be found in commerce, but admixture of such a fluid with genuine milk is possible. See pp. 254–257.

It is essentially an albuminous serum, containing urea, common salt, and just sufficient casein and milk-sugar to show its origin from a much-diseased milk gland.

(5.) *Retention of Foetal Membrane*.—The milk was pink in colour, and contained about a twentieth of its bulk in blood; it was perfectly fresh when examined, but rapidly putrefied. The blood was separated by subsidence as much as possible. The reaction was feebly acid.

(6.) *Typhus*.—The milk of cows suffering from typhus has been analysed by Husson,² who states that from the commencement of the malady, the azotised principles augment, and that there are often found bloody and purulent fluids admixed. The analysis given is an average

¹ *Fifteenth Annual Rep. Loc. Gov. Bd.*, supplement containing report of Med. Officer for 1885.

² *Comptes Rendus*, t. lxxiii., 1871, p. 1339.

TABLE XIX.—MILK OF UNHEALTHY COWS.

	Milk-Fat.	Milk-Sugar.	Casein.	Albumen.	Peptones.	Urea.	Ash.	NaCl. in Ash.	Cholestrin.	Specific Gravity
(1) Milk from a heifer suffering from mammitis,	2.8	5.54	4.02	.56	0.92	0.11	...	1.036
(2) Parturient apoplexy.—A cow suffering from parturient apoplexy. Pulse imperceptible; temp. 99.4° F., third day after calving; sp. gr. 1.037,	3.75	not estimated.	4.02	1.14	0.98	0.10	...	1.037
(3) The milk of a cow suffering from pneumonia, fourteen days after calving. Pulse 82; temp. 102.4° F.,	2.96	3.88	3.86	0.43	.09	.005	0.8	0.48	0.58	...
Engorgement of rumen and congested liver. Pulse 68; temp. 101° F.; sp. gr. 1.032,	6.05	4.49	4.79	1.06	0.113067	0.09	...	1.032
Phthisis.—A cow 5 years old, with extensive tubercular deposit in right lung. The dam was also scrofulous. A. Dec. 1878; B. Feb. 1879,	2.77 3.83	2.82 3.34	3.65 5.4	0.867 0.365	0.866 0.770	0.09 0.15	...	1.0297 1.034
Phthisis.—A cow 2 years old, in advanced stage of phthisis. A. Jan.; B. Feb.,	2.60 3.28	2.89 4.10	3.0 3.98	...	?	...	0.91 0.78	0.10 0.15	...	1.0329 1.0335
(4) A sample of milk drawn from an udder actually infiltrated with tubercular deposit,	0.49	0.47	1.21	2.38	none	.039	0.76	0.43	...	1.018
(5) Retention of fetal membrane.—From a heifer two days after calving, a portion of the udder much inflamed,	4.4	9.8162	.269	...	1.16	1.0313
(6) Typhus,	1.49	3.14	...	2.06	1.85

sample of milk from cows suffering from a not too severe form of typhus.

§ 173. *The Propagation of Disease through Milk.*—Modern researches on zymotic diseases have for long been converging to the one conclusion, that these diseases are all produced by germs; and that, consequently, such diseases are only special forms of fermentation or putrefaction in living tissues, the disease-zymads growing and multiplying at the expense of the tissues.

Now, if the composition of milk and of the tissues be compared, it will be seen that milk, although physically a fluid, yet resembles in its chemistry a tissue, and contains all that is necessary for the nourishment and growth of a zymad. Hence it is, that if a scarlet-fever zymad, or a typhoid zymad, fall into milk, for all practical purposes it is immersed in a tissue; and cultivation experiments have shown that milk is an excellent medium or soil for the multiplication at suitable temperatures of pathogenic micro-organisms.

The Relation of Milk to Scarletina.—In December, 1885, an outbreak of scarlatina, traced by the senior author to a particular milk supply, was very fully investigated by the Loc. Gov. Board,¹ and it was proved as clearly as such a matter is capable of proof, that the milk was not infected by any human agency, but that the cows were at the time suffering from a general feverish disorder, the chief external evidence of which was vesicles on the teats and udder rapidly passing into ulcers. The disease is readily propagated by inoculation among calves, and the pathological signs are in many respects strikingly similar to those of human scarlatina.

According to Dr. Klein's researches the malady is intimately connected with the multiplication and growth of a micro-organism, which probably gains access to the milk with the discharges from the diseased teats. The micro-organism consists of spherical micrococci, arranged as diplococci, and as shorter and longer, straight, wavy, or curved chains—streptococcus—the latter sometimes of great length (see Plate, fig. 1). When inoculated into gelatin the growth is slow (see Plate, fig. 2), at the end of a fortnight the streak of inoculation showing up as a white line, made up of smaller and larger droplets. It does not liquefy the gelatin: it is very similar in appearance and manner of growth to the streptococcus of foot-and-mouth disease, but Dr. Klein thinks the two may be readily distinguished by their action on milk, the foot-and-mouth micro-organism causing no apparent change in sterilised milk; the scarlatinal streptococcus, on the other hand, turning it after two days' incubation into a solid mass. A similar organism has been isolated by Dr. Klein from the blood of persons suffering from human scarlatina.

Drs. Jamieson and Edington² have also investigated scarlatina by biological methods. Dr. Edington has cultivated the blood drawn from the finger at different dates, and also the epithelial scales thrown off from the skin, taking all possible precautions to avoid aerial contamination. By cultivating a drop of the blood in Koch's jelly, diffusing a minute quantity of this culture through a large body of distilled, sterilised water, and then cultivating drops of this water, the various organisms in the blood were isolated; and from the isolated colonies, pure cultures made; lastly, their effect on animals was ascertained by inoculation.

¹ *Fifteenth Annual Report Loc. Gov. Board*, containing Report of Med. Officer for 1885.

² *Brit. Med. Journal*, June 11, 1887.

The organism, which was invariably found in the blood of patients during the first three days of the fever, and in the desquamation after the twenty-first day, is described under the name of the *Bacillus scarlatinae* (see Plate, fig. 3). It is in the form of motile rods measuring $\cdot 4 \mu$ in thickness, and $1\cdot 2$ to $1\cdot 4 \mu$ in length, generally forming very long jointed and curved leptothrix filaments; it liquefies the jelly (see Plate, fig. 4). Infected into broth and incubated its growth is very rapid, and a coherent, parchment-like film, which is not easily broken, is formed on the surface; later the film becomes deeply wrinkled.

A calf inoculated with a culture, and also given some in milk to drink, developed fever in six hours, and died within twenty-four. The pathological appearances were those found in the human subject in rapid cases of scarlatina with early death. A second calf was inoculated with a pure culture, and developed fever within twelve hours, and there was a vivid rash followed by desquamation; the throat was severely affected. In six days the temperature was normal. When guinea-pigs and rabbits were submitted to inoculation from cultures, very similar results were obtained.

Other organisms isolated from milk, giving either negative or insignificant results, were *Sarcina lutea*, three forms of micrococci, and three of bacilli, to which provisional names have been assigned.

The essential difference between Edington's and Klein's researches is, that Edington has concentrated his attention on human scarlatina—Klein on a disease of the cow. Both agree that scarlatina is communicable from the cow to man, and they agree also that milk is an excellent medium for the growth of *Scarlatinal contagium*; but Klein identifies the cause of the disease as a micrococcus, Edington as a bacillus.

With regard to the cultivating powers of milk, Edington remarks: "If desquamation be placed in milk kept at a gentle heat for twenty-four hours, the bacilli can be recognised by their motility and the pellicle which they soon form. Thus it is evident that, if milk be taken as food containing infective desquamation, the spores will have a perfect nidus and suitable temperature afforded them for development."

More recently¹ Dr. Mervyn Gordon has investigated the intimate pathology of scarlatina. He has shewn that a fatal result in this disease is to be attributed to invasion of the blood and organs by streptococcus. He further has come to the interesting conclusion that the infectious malady scarlatina is to be referred to *Streptococcus scarlatinae*, whereas the dangerous phases of the disease frequently result from supplementary invasion of the blood and tissues of the patient by *Streptococcus pyogenes*.

The Relation of Milk to Phthisis and Tubercular Maladies.—Villenin² was one of the first to attempt to ascertain, by direct experiment, whether tubercle was transmissible or not. He inoculated the rabbit, the sheep, the dog, and the cat with human tuberculous matter, always in very small quantities, with more or less success. The experiments of Villenin were repeated, with infinite variation, by most of the leading pathologists of Europe; but their deductions were quite different from those of Villenin, for, on putting various substances (other than tuberculous matter) in the subcutaneous tissue of guinea-pigs, they produced a febrile disorder, and found after death products which, they declared, were not to be distinguished from tubercle. Hence, tuberculosis was considered for a long

¹ 29th and 30th Annual Reports of Local Government Board, containing Reports of Medical Officer for 1899-1900 and 1900-1901.

² "Cause et Nature de la Tuberculose."

Fig. 1



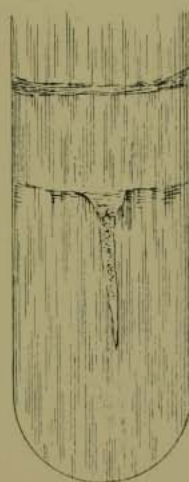
Fig. 2



Fig. 3



Fig. 4



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time as due to a mechanical irritation, chiefly set up in the delicate cells lining the lymphatic channels. While animals alone were the subject of the French, German, and English experimenters, a doctor in Greece—Zallonis of Syra—actually inoculated the human subject—a man affected with gangrene of the foot—with tuberculous sputa. In thirty-eight days the man died with unmistakable tuberculous signs, which an autopsy confirmed. The opinion¹ that the introduction into the tissues of a great variety of foreign matters will cause this malady is not now held, there having been of late years some most valuable experiments on the subject, more especially those by Bollinger, Cohnheim,² and Koch.

The most striking of Cohnheim's experiments were those in which he introduced tuberculous matter into the anterior chamber of the eyes of kittens; the animals generally became infected after a well-marked period of incubation of from fourteen to twenty-one days. Cohnheim, associated with Solomons, has also proved the possibility of aerial infection, having produced the disease in animals by causing them to inhale tuberculous dust. The pathological changes thus produced they compared side by side with those produced by mere irritants, whether breathed or introduced into the system by other channels; and they declare, as the result of such comparison, that the products of the latter are entirely different, and not to be confounded with tubercle. Direct experiments with the milk from tubercular cows have been made by Gerlach, Klebs, Bollinger, Sidney Martin, and Sims Woodhead and others. Gerlach fed two calves, two pigs, one sheep, and two rabbits for three weeks with the unboiled milk of a phthisical cow; the whole of the animals became affected with tubercular disease. Klebs made a similar successful experiment with nine guinea-pigs. The accidental infection of a large St. Bernard dog, which, having come across the milk designed for one of the experiments, drank it, and became tuberculous, is perhaps more striking than a formal experiment. The experiments of Bollinger were on pigs, as follows:—Two young pigs were fed with the unboiled milk of a cow which in life exhibited symptoms of lung disease, and which a *post-mortem* examination showed to have suffered from phthisis. This experiment was negative. The pigs enjoyed good health, and on being killed proved to have all their organs in a sound condition, with the single exception of some slight infiltration of the glands of the neck in the case of one. In a second experiment, the milk of a highly tuberculous cow was given for ten weeks to four healthy young pigs three weeks old. During this time a general enlargement of the lymphatic glands of the neck was observed; at the end of four and five months the animals were killed, when tubercular infiltration of the lungs, liver, spleen, etc., was fully established. Another experiment was made on a young pig, fed with the same milk for fourteen days. On killing the animal three weeks afterwards, there was found cheesy inflammation of the large intestine, an exquisite miliary infiltration of the lungs, and a slight cheesy deposit in the bronchial glands. The milk in all the above experiments had been given unboiled. In another experiment, however, in which six pigs were taken, four were fed with the milk of the same cow (two with the unboiled milk, two with the boiled milk), and two were fed on ordinary diet to control the experiment. After a few months the last

¹ "The Transmissibility of Tuberculosis," by G. Fleming, *Med. Chir. Review*, Oct., 1874.

² "Die Tuberculosen vom Standpunkte der Infectionslehre," von J. Cohnheim. Leipzig, 1880.

two were healthy ; the two fed on unboiled milk highly tuberculous ; one of the pigs fed on the boiled milk, on being killed, was found perfectly healthy ; the other, killed a little later, was tuberculous.¹

On the other side, there are numerous failures, and many observers have failed to propagate the disease. E. Perroncito, of Turin, records the case of a whole family (consisting of a man, his wife, and two children) drinking for eight days the milk of a cow most decidedly tuberculous, as proved by an autopsy ; and yet they remained well. Negative results are, however, of little value, unless extremely numerous. If the disease can be propagated by milk, it does not necessarily follow that every animal experimented upon will become infected ; for there are numbers of facts proving that some human beings and some animals have a great resisting power, and do not, with any readiness, take such diseases. It has hitherto been almost universally taught that bovine tuberculosis has nothing essentially different in its course or pathology from human tuberculosis. This view has, however, been contested, Dr. Charles Creighton, in a very able paper,² giving a series of cases (eight in number) in which he contends that there was more analogy to "*Perlsucht*, bovine tuberculosis," than to human, and moreover, that this form is a distinct form, quite as distinct as glanders—the salient points in the cases cited being, (1.) the occurrence of tumour-like embolic infarcts in the lungs ; (2.) the implication of the bronchial, or of the mesenteric and portal lymphatic glands ; (3.) the characters of the eruption in the serous membranes, and its relative frequency ; (4.) the microscopic appearances ; (5.) the elements of obscurity in the cases viewed as cases of ordinary or autochthonous tuberculosis.

The experiments of Dr. Sidney Martin, carried out for the Royal Commission on Tuberculosis, 1900, all tended to show that the lower animals were very susceptible to bovine tuberculosis, and to a less degree to human tuberculosis. His experiments were carried out upon guinea-pigs, rabbits, pigs, and calves, the tuberculous milk and other matter either being mixed with the food of the animal or else administered by subcutaneous or intra-peritoneal injection. The result of his experiments was not absolutely conclusive, as he himself says, "It is evident that in the case of tuberculous sputum we are dealing with material which is less infective to calves than bovine tuberculous material." Still further doubt has been thrown on the identity of bovine and human tuberculosis by Dr. Robert Koch, the discoverer of the tubercle bacillus,³ who in his address to the British Congress on Tuberculosis, 1901, stated that he had come to the conclusion (1) *That human tuberculosis differs from bovine, and cannot be transmitted to cattle* ; (2) that the susceptibility of man to bovine tuberculosis is not absolutely decided, but if such susceptibility really exists, the infection of human beings is but a very rare occurrence.

Dr. Koch's conclusions were chiefly based upon the following facts :—

(1.) Cattle free from tubercle were infected in various ways with pure cultures of tubercle bacilli taken from cases of *human tuberculosis*. No trace of tubercle was observed in the cattle during life or when killed six to eight months after infection.

¹ *Aerztlich. Intelligenz Blatt*, No. 47, 507.

² An Infective Variety of Tuberculosis in Man, identical with Bovine Tuberculosis, *Perlsucht*, by C. Creighton, M.D., *Lancet*, June 19, 1880.

³ *Deutsche Med. Woch.*, 1881, and *Mitth. aus d. K. Gesundheitsamte*, ii.

TUBERCLE BACILLI IN MILK, AND TUBERCULOSIS OF LYMPH GLAND.

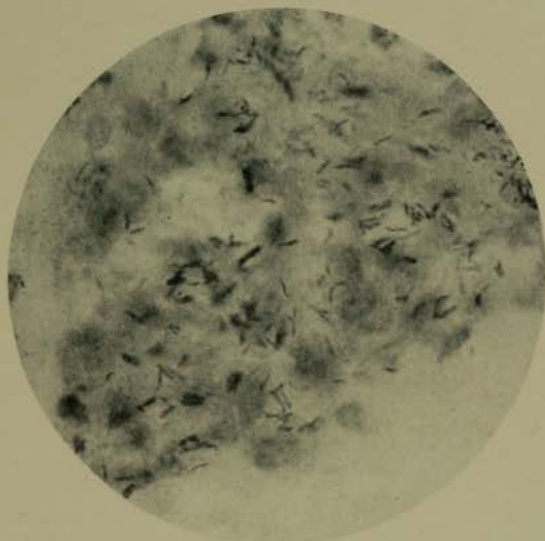


FIG. A.—A photograph¹ of a mass found in the milk of Cow with tuberculous disease of the udder, $\times 750$ diameters. The mass is composed of amorphous matter, a few degenerating cells with nuclei still intact, and very numerous tubercle bacilli, most of which showed the characteristic beading very well. Beaded tubercle bacilli are frequently met with in tuberculous milk side by side with the unbeaded rods seen in tuberculous lesions.

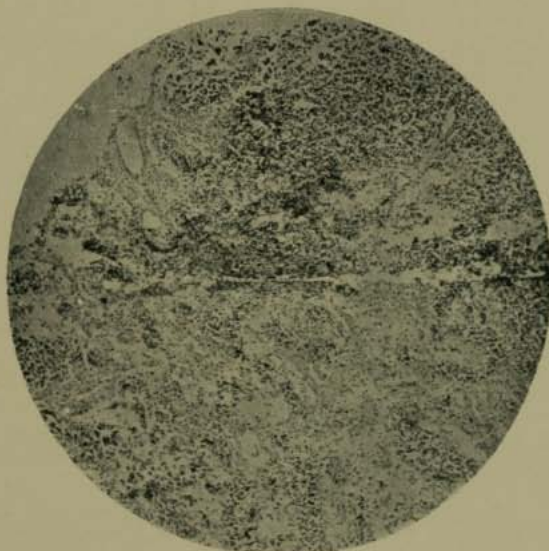


FIG. B.—A photograph¹ of a section of a lymphatic gland showing very early tuberculosis, $\times 35$ diameters. The gland was the right supramammary gland of a Cow, and showed no naked-eye tubercle. At one part of the figure to the left the gland structure appears condensed, and at this spot tubercle bacilli were found. The figure is intended to demonstrate that tuberculosis of a lymph gland may be present when undetectable by the naked eye.

¹ From Report of Royal Commission on Tuberculosis, 1895.

TUBERCLE BACILLI IN MILK, AND TUBERCULOSIS OF LYMPH GLAND.



FIG. C.—Section of tubercular udder from a Cow,¹ slaughtered at Camden Town, the udder of which is now at the museum of the Royal Veterinary College. This section was made through a part of the udder in which the tubercular process was well advanced. The nuclei still stain distinctly, but between them there is a homogeneous-looking mass in which tubercle bacilli are scattered in considerable numbers. Some of the nuclei in this specimen are evidently disintegrating and undergoing degenerative and atrophic changes, whilst at one or two points the bacilli appear to be actually within the degenerating cells. ($\times 700$.)

¹ From *Report of Royal Commission on Tuberculosis*. 1895.

(2.) The result was the reverse when pure cultures of tubercle bacilli from *bovine tuberculosis* were used.

(3.) Similar results were obtained, showing a marked difference between human and bovine tuberculosis, when experiments were made on swine, asses, sheep, and goats.

(4.) That infected food is the cause of tuberculosis can only be assumed when primary tuberculosis of the intestine is found. Koch maintains that primary tuberculosis of the intestine is extremely rare in spite of the enormous amount of tuberculous milk, meat, and butter that is consumed.

Koch's conclusions have been hotly contested by many pathologists whose opinion must be regarded as of equal value to his own, and the whole question is at present under investigation in England by a Royal Commission appointed in August, 1901.

Detection of the Bacillus of Bovine Tuberculosis in Milk.—This is most satisfactorily done by injecting the suspected milk into susceptible animals.¹ Only in very few cases can it be found by microscopical examination. For as Klein points out, not only may the tubercle bacillus be missed when it is present, but also it may be thought to be present when absent, for milk often contains bacilli which simulate with regard to staining power the true tubercle bacillus. If, however, a microscopical examination is made, the best method is to whirl 10 c.c. of the milk in a sterile tube, the cream and upper portion of the milk is removed, leaving a deposit at the bottom of the tube. Thin cover slip preparations are made from this deposit; these are dried, flamed, placed in absolute alcohol for a few minutes, the fat removed by washing in ether, the ether removed by alcohol, and finally they are stained by the Ziehl-Neelsen method.² The slides are then carefully examined. According to Pakes,³ the following may be observed: (1) Tubercle bacilli may be present. (2) No tubercle bacilli, but (a) a fairly large number of some other bacterium, or (b) a great excess of leucocytes, often amounting to pus, or (c) a considerable amount of vegetable matter and débris. (3) Nothing but a few leucocytes. The same author has examined many milks containing a large number of streptococci in the deposit, and he is inclined to think that streptococcal enteritis in children may be caused by such milk. The presence of a large number of leucocytes may disguise the presence of tubercle bacilli, and must be looked upon with suspicion.

§ 174. *Cholera, Typhoid*, and any water-borne disease may be contracted through drinking milk which has become contaminated with the specific organisms of these diseases. The attempt to isolate the specific bacilli may be made in the same manner as if dealing with a contaminated water. See chapter on Water (*post*).

§ 175. *Diphtheria and Follicular Tonsillitis.*—The Klebs-Löffler bacillus has been found in milk, and there is no doubt that diphtheria has been caused by drinking contaminated milk.

The bacillus may sometimes be demonstrated by whirling a small portion of the milk, and either making coverslip preparations from the sediment, or else inoculating good inspissated blood serum with very small quantities of the sediment, taking care that the colonies shall grow as separately as possible. After ten hours' incubation at 38° C., the tubes

¹ See Sheridan Delépine, *British Medical Journal*, Sept. 23, 1893.

² See any recent work on Practical Bacteriology.

³ "The Application of Bacteriology to Public Health," by W. C. C. Pakes, *Public Health*, March, 1900, 430.

must be examined. The bacilli should be stained by Gram's method, and counterstained with eosin. Curtis¹ calls attention to the fact that epidemics of acute follicular tonsillitis sometimes occur, apparently dependent on the presence of large numbers of strepto- and staphylo-cocci in the milk.

§ 176. *Diarrhœa*.—*Bacillus enteritidis sporogenes* (Klein) has been isolated from milk, butter, and cheese.

Klein originally isolated this spore-forming anaerobe from the evacuations of patients suffering from a severe epidemic of diarrhœa which broke out in St. Bartholomew's Hospital in 1895. It may be isolated from milk, as described in the chapter on Water (*post*).

§ 177. *Apthous Fever*.—The physical and chemical characters of apthous milk have already been described. In certain stages of the disease, the milk acts upon young calves like a virulent poison, the calf dying from apnoea with great suddenness. After death few marked changes are noticed, save intense pulmonary congestion, and a somewhat injected patchy tongue. The milk has been given to pigs with a fatal result, and even cats have suffered indisposition from it; nor is there wanting the strongest evidence to show that it may convey the apthous disease to man. This transmission to man has been observed for some time, for in the middle of the eighteenth century Michael Sagar² described the apthous epidemic, which prevailed in Moravia, 1763-64, and related how the milk propagated the disease to animals and men. In 1834 three German veterinarians³ (Hertzig, Mann, and Villain) made on themselves some very conclusive experiments, each taking a pint of the warm milk drawn from an infected cow for four consecutive mornings. On the third day M. Hertzig had feverish symptoms; by the sixth, the mucous membrane of the mouth was swollen; by the seventh, there was a well-marked eruption on the edge of the tongue, the lips, and the internal surface of the cheeks; by the tenth, the vesicles continually increasing in size, had burst; and by the twenty-fourth day the ulcers had dried, and there was some desquamation. MM. Mann and Villain were also affected in the same way, but to a less degree. This experiment is supported by a number of instances of partial epidemics in the human kind, which could be satisfactorily traced to apthous milk. It would appear certain that such milk after boiling is harmless. For example, Boulay records that foot-and-mouth disease, when imported into the Commune of Morchier by a pig-dealer, extended in a few days to over a hundred head of cattle, but spared the calves, which were fed on boiled milk and water, and not allowed to suck their mothers.

§ 178. *A Form of Febrile Disease Associated with Milk*.—The milk from a dairy near Aberdeen appears to have been the propagating agent of a peculiar and entirely new malady. This remarkable outbreak has been investigated and described with great ability by Dr. Ewart.⁴ Twenty persons were attacked, and there were three deaths. The symptoms consisted essentially of fever, with one or more relapses, and swelling of the cervical glands, frequently ending in suppuration. The connection of the epidemic with the milk-supply was established by the

¹ *Essentials of Practical Bacteriology*, by H. J. Curtis, 1900, 267.

² *Libellus de Aphthis Pecoris*, Vienna, 1765.

³ "Nouveau Dictionnaire de Médecine Vétérinaire."

⁴ On a New Form of Febrile Disease associated with the Presence of an Organism distributed with Milk from the Oldmill Reformatory School, Aberdeen, by J. Cossar Ewart, M.D., *Proc. Roy. Soc.*, 1881, xxxii., 492.

fact of the illness being confined to those who drank the milk, as well as by the microscopical appearances of the milk, and some well-devised experiments on animals. The microscopic appearances of the milk showed:—

1. Numerous micrococci, some free, others in groups or chains.
2. Numerous spores and cells of the yeast-plant.
3. Spores similar to *B. anthracis*.

Some pus from the neck of one of the patients was found to contain bacilli and spores apparently identical with those found in the milk, and such pus caused fatal illness when injected into small animals subcutaneously. These elements were submitted to cultivation, and a variety of experiments on rats were instituted with the suspected milk, side by side with control-experiments with healthy milk, the main result being to prove satisfactorily a direct connection between the bacilli and the disease; the evidence pointed to a contamination of the water supplying the dairy, and the author concludes that the organism producing this new fever was morphologically not unlike the anthrax organism in its mode of development and life-history; and, further, that it was introduced into the milk after it had left the udder.

§ 178a. Klein has recently isolated a pathogenic blasto-mycete from milk. A detailed description of this organism will be found in the 30th Annual Report of the Local Government Board, containing the Medical Officer's Report for 1900-1901.

THE DECOMPOSITION OF MILK.

§ 179. The decomposition of milk is due to the action of numerous bacteria. These bacteria are, however, not secreted with the milk, providing the animal is perfectly healthy, but are derived from the air, from contamination of the teats and ducts of the teats, the hands of the milker, the pails, etc. Milk, when freshly drawn under cleanly conditions, contains but few bacteria; it is, however, such an excellent medium for their growth that they increase with extreme rapidity, and after a few hours may amount to several millions per cubic centimetre.

Ordinary commercial milk as delivered to the consumers in London contains, according to the authors' estimations, rarely less than 2,000,000 bacteria per c.c., capable of growing on nutrient gelatine.

In order to isolate any particular organism, it is necessary to dilute the milk with a large volume of sterile water, so that the bacterial colonies may grow well separated. A convenient dilution will be found to be 1 part of milk to 10,000 parts of water. Plates, etc., may be set as in dealing with water (see *post*).

Besides the disease-producing bacteria which have already been noticed, and which are but rarely present in milk, there are bacteria which (1) act upon the milk-sugar, and the proteids, and are the true organisms of decomposition; and (2) certain bacteria of great importance to the dairyman, as by their growth in milk they may cause 'Milk Faults,' which render the milk unfit for use.

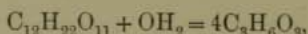
1. (*a.*) *Lactic Fermentation*.—Nearly all milk, if allowed to stand in a warm place, becomes sour, owing to the formation of lactic acid by the alteration of the milk sugar under the influence of bacteria.

The aerobic bacteria which bring about this change in milk are very numerous, and include not only bacilli, but streptococci, micrococci, etc.

About 35° C. is the optimum temperature for the growth of the majority of the lactic ferments, and they cease to grow at about 9° C.; they are readily killed by heat, since they are, as a rule, non-spore forming. The sugar is never wholly converted into lactic acid, because the acid formed checks and finally stops the growth of the bacteria; if, however, the milk is kept neutral by the addition of chalk, nearly all the sugar may be converted into lactic acid.

When the lactic acid reaches .207 per cent. according to Thorner,¹ the milk coagulates on warming. Rideal² estimates the quantity necessary to bring about coagulation as somewhat higher than this.

The milk sugar is not simply converted into lactic acid, according to the equation



for there is always a certain amount of CO₂ produced, and in ordinary milk very often alcohol and butyric acid. The alcohol produced must be regarded as a bye-product of lactic and other fermentations, and may finally become converted into acetic acid.

This *alcoholic fermentation* under ordinary conditions takes place to only a slight extent, but by introducing the right organism (this organism would appear to be the yeast found in Kephir grains) nearly the whole of the sugar may be converted into alcohol.

(b.) *Butyric Fermentation.*— Sometimes milk develops an alkaline reaction, becomes bitter, and the coagulum at first formed is redissolved with the formation of butyric acid. This change is brought about by the butyric bacillus and other allied organisms. Both the casein and milk sugar undergo decomposition, and it is at present impossible to give any formula approximately representing the change. Analysts who receive milk in this state will be unable to form any accurate idea of the original composition of the milk.

(c.) *Proteid Ferments.*— Many bacteria, both aerobic and anaerobic, decompose the proteids of milk. Their action is very complicated and little understood. Some of them would appear to act in a similar manner to pepsin and trypsin, producing peptone, leucine, tyrosine, and butyric acid.

O. Kalischer³ has named those aerobic sporing organisms which are not destroyed at 90°–95° C. 'peptonising bacteria,' and according to him they form ammonia, and cause a diminution of lactose, at the same time converting the casein into peptone, leucine, trypsin, and hydroxy acids.

II. *Milk Faults.*—*Red Milk.*— This may be caused by the growth of *Bacillus lactis erythrogenes* or *Sarcina rosea*. *Micrococcus rosaceus* and *Micrococcus prodigiosus* are also said to have caused trouble in milk. These organisms are all slow growing.

Blue Milk.— This is caused by *Bacillus cyanogenus*. Patches of a blue colour appear upon the surface of the infected milk after from about twenty-four to seventy-two hours. The blue colour is only produced in the presence of lactic acid; in alkaline milk a slate colour is produced. The organism occurs in many shapes:—(1) cocci-like bodies; (2) short rods, sometimes collected into zooglœa masses; (3) somewhat larger rods having a spore in the middle or at the end; (4) other irregular patches. The organism is mobile, except in the zooglœa patches.

¹ *Chem. Zeit.*, 1891, p. 1108.

² *Departmental Committee on Food Preservatives, etc.*, 1901. App. xxiv.

³ *Arch. Hygiene*, 1900, 37, 30–53.

Yellow Milk is said to be caused by the *Bacillus synxanthus*, *Violet Milk* by the *Bacillus violaceus*, *Bitter Milk* by a number of different bacteria, such as Weigmann's bitter milk bacillus and Conn's bitter milk micrococcus.

Slimy or Ropy Milk is milk which becomes thick and slimy; such milk is not uncommon. It is caused by a very large variety of micro-organisms. It may be produced artificially by immersing the stem of the butterwort (*Pinguicula vulgaris*) in the milk, and in Norway is used as an article of diet.¹

For all these faults, cleanliness and disinfection will be found efficient remedies.

THE PRESERVATION OF MILK.

§ 180. The preservation of milk is a problem of great practical importance.

The commercial methods may be divided into three, *i.e.*, the application of (1) Cold, (2) Heat, (3) Preservatives.

To these may be added methods which are of value to the food analyst, but which render the milk unfit for use as a food.

(1) *Cold*.—The majority of bacteria cease to be active at about 4° C., so that milk will keep for a very long time at low temperatures. In the milk trade great and increasing use is made of this method of preservation. It is to be noted that if portions of the milk are actually frozen, unless care be exercised in remelting and mixing, the ice portion will have much the same composition as watered milk. This method does not destroy disease germs, if present.

(2) *Heat*.—(a) If milk be *pasteurised*, *i.e.*, heated to about 70° C. for 20 minutes, the majority of the non-sporing bacteria will be killed, and the milk will keep for a considerably longer time than un-pasteurised milk. This method usually destroys all disease germs, with the exception of spores. (b) If milk be heated to 120° C. for 2 hours, or to 130° C. for 30 minutes, all or nearly all bacterial life will be killed. This heating to a high temperature has many disadvantages, as the albumen is altered, calcium citrate is deposited, there is a certain amount of caramelisation of the lactose and an aggregation of the fat globules. Heating milk to any temperature over 70° C., according to Duclaux, imparts to it a permanent cooked taste.

(c) Intermittent pasteurisation, *i.e.*, heating to 70° C. for 20 minutes, on several successive days, and incubating at 37° C. in the intervals, will eventually sterilise milk.

To these methods may be added evaporation processes either with or without the addition of sugar, as in the preparation of condensed milks and milk powders.

It is interesting to note that many of the so-called sterilised milks on the market contain bacterial life, especially in the form of anaerobic spores.²

(3) *Preservatives*.—The numerous substances which are added to milk in order to preserve it for a short time have already been described (page 234). It should be widely known that the small quantities of preservatives used in commercial milks do *not* kill the specific organisms of typhoid, cholera, and other pathogenic organisms, as we have ascertained by numerous experiments. Such preserved milks, far from being safer than

¹ Aikman, *Milk: its Nature and Composition*, 1895, p. 88.

² A. Weber, *Arb. aus dem Kaiser. Gesundheitsamte*, 1900.

ordinary milks, are in our opinion far more dangerous, since they often come from those dairies which rely more on preservatives than they do on thorough cleanliness.

(4) *Methods of Use to the Analyst.*—It is often of great importance to preserve milk for a long time in a fit state for analysis.

Many methods have been proposed, none of them being altogether satisfactory. Cold or heat may be used, but the first is not, as a rule, available to the ordinary analyst. Heat has the disadvantage of altering the composition of the sample to a slight degree, and causes the cream to clot at the surface. Of the ordinary preservatives, 'Formalin' is the best to use for preserving milk for a short time. Bevan¹ has shown that the total solids are increased by the addition of formalin. If, however, 3 drops of 40 per cent. formic aldehyde solution are added to an 8-oz. bottle of milk, the correction for total solids will only be in the second place of decimals.

Richmond² advocates the addition of $\frac{1}{2}$ c.c. of ordinary hydrofluoric acid to 100 c.c. of milk. This curdles the milk, and the bottle must be well shaken before taking a sample. The ash is slightly increased by the acid attacking the glass. This is a very good method when samples are to be kept for a long time. The addition of alcohol, chloroform, and solid preservatives we have not found satisfactory.

SKIM-MILK.—SEPARATED MILK.

§ 181. Skim-milk is milk skimmed by hand; separated milk is milk which has been 'whirled' in a centrifugal apparatus; by this latter process a far more perfect separation of the milk-fat is effected. By the old method of setting in open pans from '8 to 1'0 per cent. of milk-fat remained; by the modern centrifugal method, only from '1 to '5 per cent. of milk-fat is left. The composition of separated-milk compared with the milk in its original state is well seen in the following analyses by W. Fleischmann:—

	Fresh Milk.	Cream.	Separated-Milk.
Fat,	3'64	67'63	'46
Casein,	2'73	1'17	2'88
Albumen,	'68	'25	'49
Sugar,	4'69	2'25	5'34
Ash,	'71	'12	'72
Water,	87'55	28'58	90'11
Total solids,	12'45	71'42	9'89
'Solids not fat,'	8'81	3'79	9'43

Thus, by the operation the proportion of 'solids not fat' has been somewhat raised, and this is constantly observed, and, of course, the specific gravity is also raised, gravities as high as 1'037 and above being met with.

CONDENSED MILK.

§ 182. Condensed milk is prepared by concentrating whole or separated milk. The product may or may not contain added sugar. As a rule, the sugar added is cane sugar, but invert sugar may be present.³ There are

¹ *Analyst*, xx., 152.

² *Dairy Chemistry*, 1899, p. 144.

³ *Select Methods of Analysis*, by Leffmann and Beam, p. 225.

also in the market several forms of condensed milk mixed with other matters, specially prepared for the feeding of infants.

Analysis of Condensed Milks.—In the analysis of condensed milks it is not advisable to treat the condensed milk direct with solvents, even if the milk is presented as a dry powder, for the sufficient reason that it is found difficult to ensure equal admixture of aliquot parts of the product. It is better to mix as thoroughly as possible the whole contents of the sample submitted, to weigh out from 10 to 50 grms., dilute with water to a definite bulk and to take aliquot portions of this. A convenient strength is 10 grms. of the sample in 100 c.c. of water.

§ 183. *Total Solids.*—10 c.c. of a 10 per cent. solution are run into a weighed platinum dish and dried first over the water bath and then in an air bath at 110° C. until loss of weight practically ceases. Any difficulty experienced in drying may be dealt with by the use of a little asbestos in the platinum dish.

§ 184. *Ash.*—The residue from the total solids is burned in a muffle and weighed.

§ 185. *Estimation of the Milk-Fat.*—Accurate estimation of milk-fat in sweetened condensed milk is not easy; the ordinary Adams method, for example, gives erroneous results, because the cane sugar dries in a hard mass on the filter coil and renders the extraction of the fat difficult. J. F. Geisler, however, believes he has attained fair results by using for the extraction petroleum spirit or a mixture of this with 15 per cent. anhydrous ether.

Centrifugal Methods.—Pearmain and Moor¹ obtained what they considered fair results by the Leffmann-Beam's process, using 1 gm. of the original sample and adding H₂SO₄ of 85 per cent. strength.

A better process, however, is that of Leach.² Twenty-five c.c. of 40 per cent. solution, corresponding to 10 grms. of the original milk, are measured into the ordinary test bottle for milk-fat determination used in the Babcock machine. The bottle is then filled nearly to the neck with water, the contents shaken and 4 c.c. of a solution of copper sulphate of the approximate strength of Fehling's copper solution is added. The bottle is then well shaken and whirled in the centrifuge until the precipitate, which consists of the proteids and the fat, has settled out. The clear supernatant liquid, which contains the sugar in solution, is drawn off by means of a pipette having sufficient capacity, a small wisp of absorbent cotton being lightly twisted about the lower end of the pipette to serve as a filter. If a suction pump is available, it will be found convenient for this purpose. After the liquid is drawn up into the pipette, the cotton wad is removed from its lower end by rubbing against the inside of the neck of the bottle, and the pipette is withdrawn. The precipitate is washed twice as above by decantation, taking care to thoroughly mix the settled precipitate with the wash water. If the precipitate cakes down hard, it may be desirable to use a platinum wire as a stirrer. After the final washing, sufficient water is added to amount to about the normal volume of 17.6 c.c. (the quantity of milk usually measured for the Babcock test). The contents of the bottle are thoroughly mixed, and the regular amount of sulphuric acid is added, viz., 17.5 c.c.

From this point on, the usual Babcock centrifuge method is followed. Multiplying the final reading by 1.8 we get the percentage of fat. In

¹ *Analyst*, 1895, 270.

² *Fifth Annual Report Massachusetts State Board of Health*.

working Leach's method we have found that unless a high speed machine is used the proteids settle with difficulty, and that this settling is aided by standing the bottle in ice-cold water. A convenient substitute for the pipette and cotton-wool is a short glass tube constricted about $\frac{1}{8}$ th of an inch from the end, the end being plugged by a small piece of well teased asbestos, the filtration being assisted by attachment to a pump. A centrifugal machine is not absolutely necessary, as the proteids settle on long standing.

Werner Schmidt Method.—This method is interfered with by the carbonisation of the cane sugar.

By washing the ethereal extract with cold water, until no more colouring matter is removed, or, better still, by the use of petroleum ether, the fat is obtained practically pure (*Allen*).¹

Leffmann and Beam use anhydrous ether, which precipitates the caramel on to the side of the flask.

We have used successfully the following modification:—The proteids and fat are precipitated with copper sulphate, as in Leach's process; the precipitate is washed with cold water four times, the proteids dissolved by HCl, and warming, the solution transferred to a separating tube, the original bottle well washed with ether, the ether added to the liquid and the solution extracted in the ordinary way with anhydrous ether.

The Ritthausen method may also be used, the precipitated proteids and fat being extracted with anhydrous ether.

§ 186. *Estimation of the Total Proteids in Condensed Milk.*—10 c.c. of a 50 per cent. solution of condensed milk are diluted with water and 5 c.c. of a 6 per cent. solution of sulphate of copper added; the fluid is then treated with d.n. soda solution until nearly neutral, and the precipitate dealt with as described in Ritthausen's process, p. 226.

§ 187. *Estimation of Lactose in Condensed Milk.*—The best method is to precipitate the proteids by copper sulphate and sodium hydroxide and determine the reducing sugar in the filtrate gravimetrically. Volumetric methods may also be used when only approximate results are required.

§ 188. *Estimation of Cane Sugar in Condensed Milk.*—An approximate estimation of the cane sugar may be made by deducting the proteids, ash, fat, and milk sugar from the total solids. Such a method is useful as a check on more exact methods of analysis.

Leffmann and Beam's method consists essentially of precipitating proteids by mercuric nitrate and taking a polarimetric reading, then, after nearly neutralising the filtrate and adding a little thymol, inverting by invertase at 35° C. to 40° C., and allowing it to stand for twenty-four hours. The cane sugar is inverted, the milk sugar remaining unaffected. The polarimetric reading is taken after clearing with aluminium hydroxide. The difference in the reading will give the cane sugar. The usual corrections for precipitated proteids and fat must be made.

Stokes and Bodmer determine first the reducing power of a clear filtrate. This is due to lactose, and can be calculated as such. Next, the filtrate is boiled with citric acid solution (1 per cent.), and submitted to Fehling, an increased K is observed, which obviously is due to both invert sugar and lactose. The amount of lactose being known, it is easy to calculate the reduction due to cane sugar.

W. D. Bigelow and K. P. MacElroy use the following method to determine the carbohydrates of condensed milk:—

¹ *Commercial Organic Analysis*, iv., 229.

The following solutions are required:—

Acid Mercuric Iodide.—Mercuric chloride, 1.45 grms.; potassium iodide, 3.32 grms.; glacial acetic acid, 2 c.c.; water, 64 c.c.

Alumina Cream.—A cold saturated solution of alum is divided into two unequal parts; to the larger portion is added a slight excess of ammonium hydroxide, the remainder is mixed until there is only a faintly acid reaction.

The entire contents of a can are mixed thoroughly in a porcelain dish. A number of portions of 25 grms. each are taken and placed in 100 c.c. flasks. To two of the flasks water is added, and the solutions boiled. The flasks are cooled, the contents clarified by the mercuric solution, and the alumina cream filtered, made up to a definite volume, and a polarimetric reading taken. Other portions of the milk are heated in the water bath to 55° C., and inverted by means of invertase (or brewers' yeast), the invertase being allowed to act for five hours. The sample is then clarified, filtered, and polarised. The difference of the two readings serves as a basis for calculating the percentage of cane sugar. A correction for the volume of precipitated proteids and fat must be made.

The total reducing sugar is estimated by Fehling in one of the portions; if the sum of it and the amount of cane sugar obtained by inversion is equal to that estimated by the polarimetric reading before inversion, no invert sugar was originally present in the sample. But if the reducing sugar is too great, the amount of milk sugar must be redetermined as follows:—

250 grms. of the condensed milk are dissolved in water, the solution boiled, cooled to 80°, and a solution of about 4 grms. of glacial phosphoric acid added; the mixture is kept at 80° C. for a few minutes, then cooled to the ordinary temperature, made up to a definite volume, shaken and filtered. It may be assumed that the volume of the precipitate is equal to that obtained by mercuric iodide solution. The free acid is nearly neutralised by sodium hydroxide, and sufficient water added to compensate for the volume occupied by the solids precipitated by phosphoric acid. The whole is now filtered, and the filtrate measured in portions of 100 c.c. into flasks of 200 c.c. capacity. A solution containing 20 mgrms. of potassium fluoride and half a cake of compressed yeast is added to each flask and the mixture allowed to stand for 10 days, at a temperature between 25° and 30° C. In the presence of a fluoride the invert and cane sugar are fermented by the yeast, while the milk sugar remains unaffected. The remaining milk sugar is therefore determined by the polariscope. The invert sugar and lactose have already been determined by a copper solution on the original unfermented solution; and, by subtracting the copper due to lactose, the invert sugar (if present) may be calculated.

§ 189. *Composition of Original Milk.*—The best method is to calculate to a standard, such as the average amount of non-fatty solids in ordinary milk, or the average total proteids. The average non-fatty solids may be taken as 8.9 per cent., the average proteids 3.4 per cent.

The fat of the original milk is then calculated as follows:—

$$\text{Fat of original milk} = \frac{\text{Fat of condensed milk} \times 8.9}{\text{Non-fatty solids of condensed milk}}$$

or

$$\text{Fat of original milk} = \frac{\text{Fat} \times 3.4}{\text{Proteids of condensed milk}}$$

The non-fatty solids of condensed milk will include only milk sugar,

proteids, and mineral substances, and will, for the purpose of calculation, exclude cane and invert sugars and added preservatives. The total solids of the original milk may be arrived at by adding 8.9 to the percentage of fat calculated for the original milk. The ash of the original milk may be calculated on the same principles, thus:—

$$\text{Ash of original milk} = \frac{\text{Ash of condensed milk} \times 8.9}{\text{Non-fatty solids of condensed milk}}$$

§ 190. *Degree of Concentration.*—The degree of concentration is obtained by dividing the total milk solids of condensed milk by the calculated milk solids in the original milk, thus:—

$$\text{Degree of concentration} = \frac{\text{Total milk solids of condensed milk}}{\text{Total milk solids of original milk}}$$

Example:—Viking Brand :

	Per cent
Total milk solids,	35.53
Non-fatty solids,	25.1
Fat,	10.43

$$\text{Fat of original milk} = \frac{10.43 \times 8.9}{25.1} = 3.7.$$

$$\text{Total solids of original milk } 8.9 + 3.7 = 12.6.$$

$$\text{Degree of concentration by weight} = \frac{35.53}{12.6} = 2.82.$$

The meaning of the 'degree of concentration' is, taking the above example, that every grm. of condensed milk must be made up to 2.82 grms. with water in order to produce a fluid containing 3.7 per cent. of fat and 12.6 per cent. of total solids; hence, in the above case, to every 100 grms. of milk 182 grms. of water must be added.

If the volume of water required to dilute a given volume of condensed milk to its original composition is wanted, it will be necessary to know the specific gravity of the condensed milk.

The degree of concentration by volume will equal the degree of concentration by weight multiplied by the specific gravity of the condensed milk, or

Degree of concentration by volume

$$= \frac{\text{Total milk solids of condensed milk} \times \text{sp. gr. of condensed milk}}{\text{Total milk solids of diluted milk}}$$

From this value deduct 1 to get the volume of water to be added to every 1 c.c. of the condensed milk, in order to produce a fluid having the same amount of milk solids as the original milk.

§ 191. *Adulteration of Condensed Milk.*—The most common adulteration is the removal of fat from the original milk before concentration. This may be detected by calculating in the manner already given the composition of the original milk.

It has been proposed to judge the quality of condensed milk by the relationship between the proteids and fat, *i.e.*, condensed milk should contain as much fat as proteids. The average amount of proteids in genuine milk is 3.4 per cent., and it is the least variable of all the constituents. Hence, according to this standard, the fat would have to be 3.4 per cent.; since,

however, the limit in this country is 3 per cent. for milk fat, no higher richness in fat can be insisted upon as regards milk used for condensing, so that it is preferable to calculate the composition of the original milk either on a standard of 8.9 per cent. solids not fat or on a standard of 3.4 per cent. total proteids; if, then, the fat of the original milk falls below 3 per cent. the condensed milk may be judged deficient in fat.

Cane Sugar.—Cane sugar may be found in samples of milk stated to be free from such addition; it may be tested for by Cotton's method (p. 233), and estimated by the processes already given.

*Glucose.*¹—According to Richmond,¹ glucose is sometimes substituted for cane sugar. Its presence may be detected by the high reducing power before inversion and by the fact that the reducing sugar bears no relation to the amount of total proteids in the sample. In ordinary condensed milk, the milk-sugar is about 1.44 times the quantity of total proteids.

Starch has been found in ordinary condensed milk, and it is sometimes found in condensed milks 'prepared for infants'; it is easily detected by the addition of iodine solution.

Glycerin is stated (*Allen*) to have been found as an adulterant of condensed milk.

Preservatives may be looked for in the same manner as in milk.

CREAM.

§ 192. Milk on being allowed to rest for some time becomes covered with a yellow fatty layer, known as cream. In composition it fairly agrees with ordinary milk, save that it contains a large percentage of fat, and that there is also a somewhat relatively higher percentage of casein and albumen. The albuminoids have a tendency to separate partially,² and

¹ *Dairy Chemistry*, p. 146, 1899.

² Richmond considers this to be due, in the case of *clotted cream*, such as Devonshire cream, to the evaporation of water from the surface during heating. In the case of ordinary cream he denies that the ratio of solids not fat to water is any higher than in milk. He considers that other observers have fallen into this error through not taking into account: (1) the evaporation of water from the surface of the cream, (2) methods of analysis, *i.e.*, either the fat has been imperfectly extracted, or has increased in weight through overheating. He has also worked out the following table, which enables the fat to be judged with considerable accuracy from the total solids of ordinary cream but *not* from clotted creams:—

TABLE XX.—RATIO OF FAT TO TOTAL SOLIDS IN CREAM.

Total Solids.	Fat.	Solids not Fat.	Total Solids.	Fat.	Solids not Fat.	Total Solids.	Fat.	Solids not Fat.
60	55.8	4.2	49	43.7	5.3	38	31.6	6.4
59	54.7	4.3	48	42.6	5.4	37	30.4	6.6
58	53.6	4.4	47	41.5	5.5	36	29.3	6.7
57	52.5	4.5	46	40.4	5.6	35	28.2	6.8
56	51.4	4.6	45	39.3	5.7	34	27.1	6.9
55	50.3	4.7	44	38.2	5.8	33	26.0	7.0
54	49.2	4.8	43	37.1	5.9	32	24.9	7.1
53	48.1	4.9	42	36.0	6.0	31	23.8	7.2
52	47.0	5.0	41	34.9	6.1	30	22.7	7.3
51	45.9	5.1	40	33.8	6.2	29	21.6	7.4
50	44.8	5.2	39	32.7	6.3			

mechanically adhere to the fat; for example, the authors found the average composition of Devon cream as follows:—

	Per cent.
Milk-fat,	65·011
Casein,	3·530
Albumen,	·521
Peptones,	·050
Lactochrome,	Undetermined
Milk-sugar,	1·723
Water,	28·675
Ash,	·490
Chlorine in Ash,	·013
Calcic Phosphate,	·373

It will thus be seen that the milk has thrown up casein with the fat, for if we allow that ordinary milk contains 87·2 of water and 3·0 per cent. of casein, then the amount of casein in the cream, if none were separated, would be—

Water.	Casein.	Water.	Casein.
87·2	: 3·0	: 28·675	= ·98

But the cream, instead of containing ·98 per cent., actually contains 2·55 in excess of this quantity.

The amount of albumen strictly follows the casein, for the ratio of casein to albumen in milk being as 3·0 is to ·4, the theoretical yield of albumen in this particular case would be ·69, the amount actually found being about ·1 per cent. lower than this estimate.

If the composition of the Devon cream shows clearly that there is some considerable separation of the casein, the milk-sugar follows very closely the proportions one would expect to find from the amount of water; for, taking the average of 4·7 of milk-sugar dissolved in 87·2 of water, we get in the present case—

Water.	Sugar.	Water.	Sugar.
87·2	: 4·7	: 28·7	= 1·5

which does not deviate very considerably from the numbers actually obtained, viz., 1·723.

Devon cream is of the consistence of a soft paste, and is covered with a skin-like layer of partially dried caseous and fatty matter. This cream is produced by keeping the milk in large pans, at a gentle heat, for many hours. The temperature is always far under boiling point, yet probably sufficiently high to arrest fermentation. This application of a moderate heat during a lengthened time causes the fat to coalesce and rise more rapidly than the ordinary method. Such cream is preserved in some degree from the infection of the lactic ferment, and will keep perfectly sweet many days, even in warm weather, provided always that the layer on the top is not destroyed nor disturbed; as when once this is done, especially if the top portion be mixed thoroughly with the rest, lactic fermentation is very rapid.

Ordinary, or raw cream, is either cream raised in the ordinary way, that is, by allowing the milk to rest undisturbed at the ordinary temperature; or it is separated by more modern and scientific processes, such as the action of cold and centrifugal machines. Cream, as thus produced, is a thickish, yellowish liquid, containing a variable amount of milk-fat, for which, unfortunately, there is no legal standard, and it is difficult to say what is the minimum proportion of fat a cream may contain, and yet be

properly designated cream. Without a doubt, this is either a matter which should be regulated by law, or the dairies should charge so much for every percentage of fat; it is the authors' opinion that any rich milk sold as cream with less than 25 per cent. of fat is not *cream*, properly so called. There was a great diversity of opinion amongst those witnesses who suggested a limit for fat in cream before the Departmental Committee on Milk and Cream Regulations, 1901, the limits varying from 10 per cent. to 50 per cent. The Committee finally decided "That the weight of evidence was not favourable to the establishment of official limits of fat for cream, nor does it appear that any substantial difficulty arises in selling cream in different qualities, at corresponding prices, to meet the requirements of the public. . . . The Committee consider, therefore, that no regulations are required with regard to the amount of fat in cream."

The following are the monthly averages of a very large number of analyses of cream examined by Dr. Vieth.¹ It will be seen that the lowest average is 31·8 per cent.; the highest, 51·1 per cent.; and the mean of the whole, 41·0 per cent., and this may be taken as fairly representing the composition of good cream:—

	1883.	1884.	1885.	1886.
	Fat.	Fat.	Fat.	Fat.
January,	32·8	32·6	35·9	42·1
February,	34·7	33·6	37·8	43·5
March,	32·8	34·2	37·1	45·4
April,	34·9	33·7	37·9	45·7
May,	38·1	36·4	40·0	45·0
June,	40·5	37·0	41·9	42·6
July,	37·6	39·0	42·8	41·7
August,	41·1	37·7	45·2	43·3
September,	36·2	36·3	47·8	45·1
October,	33·4	35·6	48·4	46·0
November,	32·1	33·6	51·1	45·3
December,	31·8	33·9	44·6	44·2
Yearly averages per } the whole, }	35·5	42·1	42·5	44·2

The *analysis of cream* is conducted on the same principles as that of milk. Trouble will be found in drying the cream in order to estimate the water, unless, for this purpose, quantities so small as a gramme are taken. This, spread out in a thin layer on a platinum dish, dries easily enough at the temperature of 100° C. Should larger quantities for any reason be taken, it will then be necessary to treat the cream, after partial drying, with petroleum or ether; extract the fat, and then dry the fat and the 'solids not fat' separately—the fat at about 105° C., the 'solids not fat' at 100° C.; the loss is then considered as water. The fat of ordinary cream can be determined by the Adams' method, or, if diluted, by a centrifugal method or by that of Werner-Schmidt.

Adulteration of Cream—Preservatives.—The most common adulterant is borax, but the preservatives found in milk should all be looked for. The Departmental Committee on Food Preservatives, etc., 1901, recommended that no preservatives other than borax compounds should be used in cream, the amount not to exceed ·25 per cent. boric acid, and the nature and amount of preservative to be declared.

§ 193. *Gelatin.*—This is a common adulterant of cream. The Departmental Committee on Milk and Cream Regulations, 1901, recommended

¹ *Analyst*, 1884, 1885, 1886, 1887.

that the artificial thickening of cream by any addition of gelatin or other substance shall raise a presumption that the cream is not genuine.

For the detection of gelatin in cream Stokes' method may be used:—To 10 grammes of cream add 25 c.c. of water and 2 c.c. of Wiley's acid mercuric nitrate solution, and shake well; filter through a dry filter. In the presence of much gelatin the filtrate cannot be obtained clear, and it is not essential that it should be so. On adding a saturated solution of picric acid in water, a yellow precipitate will come down if gelatin be present. If the quantity of gelatin be small, the precipitate will not form for some minutes.

Richmond describes a substance known as 'viscogen,' consisting of lime in cane sugar syrup, of which he says a small quantity has a remarkable effect in increasing the thickness of cream.

Dextrin and *Starch* have been used.

Albumen has been found by one of us; it may be detected by estimating the casein and albumen in the cream; they should have the same approximate ratio to the water as they have in milk.

Colouring Matters.—Cream is very often artificially coloured, the colours most frequently used being annatto and coal-tar yellows. The colours may be detected as in milk.¹

KOUMISS.

§ 194. Koumiss is an alcoholic drink made by the fermentation of milk; it is prepared by the nomad population of Asia (especially by the Tartars) from the milk of the mare and that of the camel; it is also manufactured from cows' milk. The preparation of koumiss by the Tartars is very simple: ten parts of fresh warm milk, with a little sugar, are added to one part of milk which is already sour—that is, which contains lactic ferment—and the whole is allowed to rest for two or three hours with repeated stirring. The chemical changes taking place seem to be a partial decomposition of the sugar into lactic acid, the development of carbon dioxide and alcohol, and possibly certain changes in the albuminoids, changing them partly into peptones. The composition of koumiss, since it may be derived from such different sources, is variable. A few analyses are as follows:—

	Mean of ten analyses. König.	Koumiss from mares' milk. W. Fleischmann.	Koumiss from cows' milk. W. Fleischmann.	Koumiss 48 hours old. J. A. Wanklyn.
Water,	87·88	91·53	88·93	87·32
Milk-sugar,	3·76	1·25	3·11	6·60
Lactic acid,	1·06	1·01	·79	
Casein,	2·83	1·91	2·03	2·84
Milk-fat,	·94	1·27	·85	·68
Alcohol,	1·59	1·85	2·65	1·00
Carbonic acid,	·88	·88	1·03	·90
Ash,	1·07	·29	·44	·66

In the koumiss from cows' milk, Fleischmann separated ·166 per cent. of glycerin.

BUTTERMILK.

§ 195. Buttermilk is the thin whey left behind when the fat has been extracted in the process of butter-making. It is never fat-free; it contains all the constituents of milk, but a great portion of the sugar has been changed into lactic acid. It is then essentially a dilute, poor acid milk.

The average composition of fresh buttermilk is:—

Water,	90·62
Casein,	3·78
Fat,	1·25
Milk-sugar,	3·38
Lactic acid,	·32
Ash,	·65

The lactic acid tends to increase, so that samples which have stood a little time will contain more lactic acid than the proportion above given; besides the ordinary salts of milk it frequently contains the common salt added to preserve the butter. It is not an article of commerce, and from its occurrence merely as a bye-product differs in composition considerably.¹

BUTTER.

§ 196. *Constituents of Butter.*—In the manufacture of butter the cream is violently agitated in a churn or other suitable apparatus, and in this manner the thin membrane² enclosing the fat globules is supposed to be ruptured. The free fat then coalesces, entangling with it some casein and serum; the butter is well pressed together to free it as much as possible from moisture, and salt added to assist its preservation. Butter, therefore, is composed principally of milk-fat, with a small and variable quantity of water, casein, milk-sugar, and ash, the latter consisting chiefly, but not entirely, of the salt added.

The fat of butter may be shown, by careful cooling, to consist of about 45·5 per cent. of butter oil and 54·5 per cent. of solid fat;³ it is usually stated to consist of a mixture of the glycerides of the fatty acids—palmitic, stearic, and oleic—not soluble in water; and also of the glycerides of certain soluble and volatile fatty acids, principally butyric, with small quantities of caproic, caprylic, and capric acids. It is the association of about 7·8 per cent. of the triglycerides of these volatile acids with the glycerides of the insoluble acids, which gives to butter-fat its peculiar and distinctive characters; but it is probable that stearin, palmitin, butyrin, and caproin do not exist in butter, their place being taken by more complicated glycerides, the glycerin being combined with two or three different acids. A crystalline glyceride,⁴ $C_3H_5 \begin{matrix} \diagup O \cdot C_4H_7O \\ -O \cdot C_{16}H_{31}O_2 \\ \diagdown O \cdot C_{15}H_{31}O_2 \end{matrix}$, has, indeed, been isolated from butter.

The different constituents, as well as the physical characteristics, of butter- or milk-fat have been already described at p. 196 *et seq.*

¹ Some analyses by Dr. Vieth (*Analyst*, 1884) may be thus summarised:—

	Total Solids, per cent.	Fat, per cent.	Solids, not Fat, per cent.	Ash, per cent.
Maximum,	10·70	2·51	10·16	1·32
Minimum,	8·13	·49	7·13	·64
Mean,	9·43	·7	8·56	·75

² Reasons for doubts as to the existence of this membrane are given at page 194.

³ A. Wynter Blyth and Robertson, *Journ. Chem. Soc.*, 1889 (Proceedings), 5.

⁴ A. Wynter Blyth and Robertson, *Op. cit.*

The general composition of butter-fat, as usually stated, is as follows:—

GLYCERIDES EQUAL TO FATTY ACIDS.

Olein, . . .	42·21	=	Oleic Acid, . . .	40·40	
Stearin and	} 50·00	=	{ Stearic and Pal-	mitic Acids, . . .	47·50
Palmitin, . . }					
				87·90	Total insoluble solids.
Butyrin, . . .	4·67	=	Butyric Acid, . . .	3·49	
Caproin, . . .	3·02	=	Caproic, . . .	2·40	
Caprylin and	} ·10	=	{ Caprylic and Rutic	Acids, . . .	·80
Rutin, . . . }					
				94·59	Total acids. ²
100·00 ¹ =					

Pure, dry butter-fat melted at a heat not exceeding 100° F., has at that temperature a specific gravity ranging from ·91079 to ·91400; its fusing point, taken in the manner to be described, ranges from 30°·5 to 36°·5; average specific gravity at 15° is ·93072.

The relative proportions of fat, casein, and salt, in butters, may be gathered from the following table:—

Per cent.	Danish. Richmond.	English. Richmond.	Irish Salt Firkin. Tich- borne.	Australian. Beamish Harrington and Forrest.	Swedish. Harrington	Italian.
Fat, . . .	83·41	86·85	63·58-88·60	85·21	82·55	84·12
Curd, . . .	1·30	0·59	0·46- 4·16	1·30	1·16	·65
Salt (Ash), . . .	1·87	1·02	1·02- 7·02	1·68	2·70	·18
Water, . . .	13·42	11·54	8·12-30·84	11·81	13·59	15·05

MARGARINE.

§ 197. *Margarine*.—The manufacture of margarine³ is an important industry in the United States, and is also largely produced in Holland and

¹ The theoretical percentage of C, H, and O, corresponding to these glycerides, is as follows:—C 72·8, H 13·3, O 13·9.

² E. Duclaux (*Compt. Rend.*, cii., 1,022, 1,077) by a process of fractional distillation has examined a series of prize Normandy butters, in special relation to the proportions of Caproic and Butyric acids; his results are contained in the following table:—

	1.	2.	3.	4.	5.	6.	7.	8.
Water,	12·40	13·36	12·28	10·72	13·34	11·62	14·00	13·03
Fat,	86·71	85·48	86·76	88·30	86·01	86·52	85·31	88·33
Milk-sugar,	0·16	0·20	0·17	0·13	0·20	0·30	0·20	0·11
Casein and salts,	0·73	0·96	0·79	0·85	0·45	1·56	0·49	0·53
<hr/>								
Caproic acid (per cent.),	100·00	100·00	100·00	100·00	100·00	100·00	100·00	100·00
Butyric acid (per cent.),	2·10	2·18	2·17	2·23	2·26	2·00	2·08	2·19
Sum of the acids,	3·55	3·52	3·53	3·60	3·65	3·38	3·52	3·46
Ratio,	5·65	5·70	5·70	5·83	5·01	5·38	5·60	5·65
Ratio,	2·1	2·0	2·0	2·0	2·0	2·1	2·1	2·0

The mean of the eight determinations gives Caproic 2·15 and Butyric Acid 3·52 per cent.

³ Mège Mouries, a chemist, appears to have been the first who proposed the manufacture of artificial butter. It came first into commerce about the year 1872. See Appendix.

in Belgium. As manufactured in Chicago, it would appear to be, at all events, a cleanly article. Animal life is plentiful in the States; American technical operations are on a gigantic scale, and too much capital is involved in the matter to allow of the use of any process likely to disgust the consumers. The chief constituent used is beef-fat, which consists for the most part of stearin, margarine (so-called), and olein. The olein and margarine melt at a much lower temperature than the stearin. Mutton-fat contains more stearin than beef-fat; hence, in summer, the softness of beef-dripping as compared with the solidity of mutton-fat. This is obviously the reason why the manufacturer prefers beef-fat. The process of manufacture is briefly as follows:—The beef-fat, freed first as much as possible from fibre, passes in a very finely-divided state from a sort of mincing-machine, technically called a ‘hasher,’ to large tanks, where it is melted by means of water-jackets applied to the tanks, and heated to a temperature never allowed to exceed 39° C. The result of this process is, that the fat melts to a clear yellow oil, the water and *débris* sinking to the bottom, and a thin scum of impurities rising to the surface. The latter is skimmed off, and the yellow oil run into wooden cars, in which the stearin, after a little time, begins to deposit in a more or less crystalline or granular condition; the refined fat is then put in a press-room, and kept at a temperature of from 26°·6 to 32°·2 C. The oleo-margarine is filtered through cotton cloths, and ultimately pressed; the result of which is, that the stearin is left behind as a white cake, and is ultimately disposed of to the candle-maker. The oleo-margarine, at this stage, is quite tasteless, and has no flavour of butter. This flavour is given by churning it with milk; lastly, the product is coloured with annatto, and rolled with ice, after which it is either made up into pounds, or packed into kegs for export. Arrived in this country, it is either sold honestly as ‘margarine’ at the price of about eightpence per pound; fraudulently at a higher price, as butter; or it is used as an adulterant of butter. The chemical proportions of the artificial butter vary according to the fats used in the manufacture and the details of the process employed; but they all agree in this, that when the butter is saponified and the acids set free, there is a great deficiency of soluble fatty acids, as compared with those of true butter-fat.

A recent return shows that during one month the following ingredients were used in the manufacture of margarine in the United States, the figures indicating the percentage each ingredient bears to the whole. Neutral fat 31·88, olive-oil 28·84, cotton-seed oil 4·34, salt 6·83, milk 16·13, butter and butter-fat 7·10, cream 4·02, and small quantities of sesamé oil, colouring, sugar, stearine, glycerine, and glucose.

The proximate analysis of commercial margarine is as follows:—

Water,	12·01
Palmitin,	18·31
Stearin,	38·50
Olein,	24·95
Butyryn, Caproin, and Caprylin,	·26
Casein,	·74
Salts,	5·23 ¹

¹ It has been asserted that artificial tributyrin has been added to oleo-margarine. Such an admixture may be readily dissolved out of the butter-fat by strong alcohol, while the natural tributyrin is not so easily separable from butter-fat by any solvent.

ANALYSIS AND ADULTERATION OF BUTTER.

§ 198. The only common adulterations of butter are the substitution or admixture of fats other than butter,¹ of water, the latter being either left in the butter in undue proportion through faulty manufacture, or fraudulently added, of colouring-matters, and of boracic acid or borax. The addition of mineral substances, flour, and other articles enumerated by different writers, is at the present day rare. The analysis of butter naturally divides itself into—(1) The general examination and analysis; and (2) the investigation of the fat.

§ 199. 1. *The General Examination and Analysis of Butter.*—The colour, taste, and odour of the sample should, of course, be noted. It will also be useful to make a microscopical examination of the sample.

The Microscopical Examination of Butter.—A very small piece of the butter is placed on a slide and just covered with a drop of olive oil, a cover glass is gently pressed down upon it until the film is sufficiently transparent. The preparation should be viewed both by ordinary and by polarised light. Coarse adulteration, such as starch, will be at once observed. Ordinary well-made butter will have a characteristic appearance, the water globules varying in size within narrow limits; under polarised light it will show no bright patches or crosses, except where definite crystals of salt or other preservative can be observed. Butter that has been heated and cooled, as, for instance, *process or renovated butter*, butter that has been *mixed with water* at a high temperature or churned at too high a temperature—butter mixed with *margarine*, butter that has become *rapidly stale*, may also show bright appearances under polarised light. Butter made from *pasteurised cream* is said to sometimes give the same appearance.

*Milk blended butter*² under ordinary light has a very much more uniform appearance than ordinary butter; the water globules being all small and of the same size, it shows no bright places under polarised light. Butter mixed with an excess of water at a high temperature often shows very large water globules.

Margarine has a very different microscopical appearance to butter, the ground substances consisting of a coarsely granular substance containing some very large water globules; under polarised light it shows many bright places, but margarine may, under certain conditions, be so mixed with butter as not to give any evidence of crystalline structure when viewed under polarised light.

The Proximate Analysis of Butter—that is, the separation of butter into mineral matters, curd, butter-fat, and water—is very readily performed.

10 to 20 grms. of the butter are weighed into a counterpoised porcelain dish, and melted over a low gas flame, keeping the butter at any temperature between 105° and 110° C., with constant stirring, until all effervescence has ceased. (By using a thermometer as a stirrer this is easily effected.) The weight of the dish and its contents when cool, subtracted from the first weight, gives the loss equalling the water, and it may be worked into

¹ A. Mayer (*Landw. Versuchs.-Stat.*, 29, 215-232) has made experiments on a man and a boy as to the relative nutrient power of natural and artificial butter. He found on the average that 1.6 per cent. less of the artificial butter was absorbed than of the natural.

² See page 276.

percentage. Keeping the butter for a considerable time at 100° C., or above, must be avoided, for it increases the weight.

The above method has the merit of expedition, and it is tolerably accurate; but the water may also be estimated by placing about 1 gm. of the butter in a large platinum dish, so that the fat forms a thin layer, and then exposing it to the heat of the water-bath with frequent agitation until it ceases to lose weight. The salt is best determined in a separate sample; from 5 to 10 grms. of the butter, made up from different portions of the sample, are shaken up with hot water in a separating funnel, the water poured away from the fat, and the chlorine estimated by silver nitrate and potassic chromate, as described in the article on Water Analysis. The *milk sugar* may be estimated on a portion of this water extract by any of the usual methods.

The *fat* may be estimated by extracting it from the dried residue used for the water determination, by boiling ether, benzene, etc.; the residue left in the dish dried and weighed will give the *solids not fat* and *salt* and any added mineral matter; these two latter and the ash subtracted from the 'solids not fat' will give the *curd*. The actual *proteids* are best estimated by making a nitrogen determination. The fat is usually most easily determined by subtracting the percentage of water, curd, and minerals from 100.

The general analysis finished, it remains to consider the results:—

§ 200. WATER.—(See Sale of Butter Regulations, page 46.)

The quantity of water in well-made butters usually lies between about 9 per cent. and 15 per cent., the average being about 12 per cent. Salt butter, with the exception of Irish pickled butter, contains, as a rule, a little less water per cent. than fresh butter, although salt butter has a moister appearance.

Butter may contain an excess of water by reason of (1) faulty or bad methods of manufacture, (2) the fraudulent addition of water.

(1) If butter is not properly made it may contain 30 per cent. or more of water, which cannot be properly got rid of by subsequent washing and working, or by the addition of salt; of the many factors which go to make up the process of churning, the temperature of the cream or milk is the most important; this temperature has been found to lie between 15° and 20° C. for milk and 12° and 18° C. for cream, the exact temperature depending on the kind of churn, the sourness of the cream, etc. If the proper temperature is exceeded, the butter will be soft and greasy and will contain an excess of water.

The method at present used in certain parts of the south-west of Ireland, which consists in pressing into the butter a saturated solution of salt, must be considered a faulty method of manufacture, since it yields a butter containing, as a rule, a large excess of water. The brine is worked into the butter either at the ordinary or at higher temperatures.

This latter method is, however, according to several authorities, only used for the purpose of fraudulently adding water to the butter.

The valuable experiments of Messrs. R. H. Beamish, W. B. Harrington, and T. A. Forrest, on the composition of Irish dry salted butters and brine salted butters made with brine at various temperatures, clearly showed that properly made dry salted butters need never contain more than 16 per cent. of water, and that dry salt can be mixed much more effectually with the butter than can brine; they further found that the difference in the amount of curd in dry salted and pickled butters was very small and

not sufficient to influence the keeping qualities of the butters. The following table summarises their results:—

TABLE SHOWING THE QUANTITY OF WATER WHICH CAN BE ADDED TO BUTTER AT VARIOUS TEMPERATURES.

Fresh and Dry Salted Butters.	Water. Per cent.	Curd. Per cent.	Mineral Salts. Per cent.
From the Munster f Average,	14·06	·91	·84
Dairy School, \ Extreme variations,	16·09-9·7	3·5-0·58	2·04-0·09

Brine Salted Samples—

Temperature of Brine.	Water. Per cent.	Curd. Per cent.	Mineral Salts. Per cent.
58- 60° F. Average,	21·31	0·78	3·76
70° F. " "	27·38	0·71	5·30
90° F. " "	31·18	0·84	7·05
95° F. " "	32·03	0·62	9·46
105° F. " "	29·80	0·56	8·15
58-105° F. Extreme variations,	42·62-17·86	1·09-0·20	9·46-1·91

It is evident that brine salted butters may be recognised by their high water, high salt, and low curd content.

(2) Water may be fraudulently added to butter at the original churning, by working the butter at a high temperature, or by using hot brine, or by working water or milk into the butter after manufacture. This latter process was practised at one time to a considerable extent on the continent, and a large amount of milk blended butter is produced in London at the present time (1902). The addition of soda, peptone, and other substances to the mixture of butter and milk or water is said to admit of a much larger incorporation of water than without these substances.

Benno Martiny records the analyses of many so-called peasant-made butters in Germany, containing quantities of water varying from 21·36 to 49·02 per cent., and one butter from Hamburg, in 1892, containing no less than 60 to 65 per cent.

The milk blended butter sold in England may be recognised by its pale creamy, greasy appearance, by its microscopical characters, its high water and high proportion of curd to fat.

As a rule, butters containing fraudulently added water have a dry granular appearance, and do not show any visible excess of moisture as do butters which have been badly manufactured.

The Solids not Fat.—These consist of proteids, milk-sugar and mineral substances derived from the original milk. The amount of these constituents left in the butter will vary according to the nature of the butter. Storch gives the following figures for butters from fresh cream:—Proteids ·64 per cent., milk-sugar ·35 per cent., ash ·14 per cent.; from ripened cream, proteids ·84 per cent., milk-sugar ·39 per cent., ash ·16 per cent.

The amount of proteids and milk-sugar left in butters is of great importance, as the larger the quantity left in, the more quickly will the butter

go rancid; thin butters, like milk blended butter, with an excessive amount of proteids and milk-sugar, have to be preserved by the addition of boron or other preservative.

§ 201. THE ASH.—*Fresh and Salt Butter*—

The ash should consist of common salt and a small amount of salts derived from the original milk.

Butters containing less than 2 per cent. of common salt are usually classed as *fresh* or *mild cured*; those with more than 2 per cent. of salt, as salt butters. The quantity of salt usually found in salt butters is 4 per cent.; it may, however, be as much as 9 per cent. in brine pickled butters. The ash will also contain borax, sodium silicate, or other added minerals.

§ 202. *Preservatives*.—The most common preservative added to butter is borax, either alone or mixed with boric acid. Dyer has found as much as 1.35 per cent. calculated as boric acid, but the usual quantity is much smaller, *i.e.*, about .1 per cent. Boron preservatives are also nearly always added to margarine, which usually contain more than butter; .5 per cent. (calculated as boric acid) or more being quite common.

The Departmental Committee on Food Preservatives, *etc.*, 1901, recommended "(D) that the only preservative permitted to be used in butter and margarine be boric acid or mixtures of boric acid and borax, to be used in proportions not exceeding .5 per cent. expressed as boric acid." All the preservatives mentioned under milk might be looked for in butter, but the only ones likely to be found besides borax and boric acid are sulphites and nitrates. Rideal has found formalin, but it is now rarely, if ever, used. (The paper wrapped round Canadian butter shipped to England is sometimes dipped in a solution of salt and formalin.) J. F. Geisler found paraffin in margarine, and C. A. Crampton 10 per cent. of glucose in a highly coloured French butter intended for exportation to a hot climate (*Jour. Amer. Chem. Soc.*, 1898, 207).

Otto and C. W. Hehner have very recently found a number of Brittany butters containing *sodium fluoride* in quantities varying from about 1 to 4 grains per pound.

The detection of preservatives in butter and margarine may be carried out by means of the methods described under milk (p. 234-240). The most convenient method of testing butter and margarine for borax and boric acid is to melt a small quantity of the sample in a dish, add a few c.c. of water made acid with HCl, mix while warm, and when the water has separated pass it through a wet filter and apply to the filtrate the usual test with turmeric paper.

Otto and C. W. Hehner, having observed that boric preservatives in the presence of fluorides render the usual test unreliable, suggest the following process:—The aqueous liquor is separated from 50 grms. of the butter, calcium chloride is added, the liquid boiled, and a small excess of sodium carbonate added to precipitate the calcium compounds. The precipitate is filtered, burnt to an ash, and treated with hot dilute acetic acid; the residue is collected on a filter, ignited, and treated with strong H_2SO_4 in a platinum crucible. The crucible is covered with a waxed slip of glass, upon which a mark has been scratched. The crucible and its contents are gently heated upon a sand-bath for two hours. Distinct etching of the glass results in the presence of a fluoride.

The estimation of preservatives in butter may be carried out on the general lines already laid down in the article on milk, the only difficulty being to get rid of the large amount of fat. This can usually be accom-

plished by shaking the melted fat with several quantities of hot water, or by obtaining the ash of 25 to 50 grms. of the sample, as in making a proximate analysis.

For the rapid estimation of boric acid in butter, Richmond and Harrison's method¹ may be used:—Weigh out 25 grms. of butter in a beaker, add 25 c.c. of a solution containing 6 grms. of milk-sugar and 4 c.c. N. sulphuric acid to 100 c.c.; place in the water-oven till the fat is just melted and stir well; allow the aqueous portion to settle for a few minutes and draw off 20 c.c.; add a few drops of phenolphthalein, bring to the boil, and titrate with $\frac{N}{2}$ soda till a faint pink colour just appears; add 12 c.c. glycerol, and titrate till a pink colour appears. The difference between the two titrations, less the amount of alkali required by 12 c.c. of glycerol, multiplied by 0.0368, will give the amount of boric acid in 20 c.c., and this multiplied by $\frac{100 + \text{percentage of water}}{20}$ will give the percentage.

§ 203. *Colouring Matter of Butter and Margarine.*—The colouring matter of natural butter is the 'lactochrome,' already described (p. 202).

Many artificial colouring matters are also added, the original object being, as it was with milk, to maintain an uniform colour during both winter and summer. It is now asserted that the public demand a highly coloured article. Margarine is, of course, coloured in imitation of butter.

The following colouring matters are said to have been added to butters:—Annatto, turmeric, saffron, safflower, marigold leaves, yellow wood, and carotin. Also the coal-tar colours—aniline yellow (amido-azo-benzene), butter yellow (dimethyl-amido-azo-benzene), methyl-orange (dimethyl-amido-azo-benzene-sulphonic acid), Victoria yellow (sodium dinitro-ortho- and para-cresols), and Martius yellow (sodium dinitro-naphthol).

The only artificial colouring matters likely to be met with in butter at the present time are annatto and butter yellow (dissolved in cotton seed oil), or mixtures of these substances, turmeric, carotin, and possibly saffron. The most common colouring matter found in margarine is butter yellow.

The Detection of Colouring Matters in Butter and Margarine.—The colouring matters may, as a rule, be extracted from the filtered fat by shaking with dilute potash solution; the potash solution may then be directly tested for annatto and turmeric (see Milk, p. 241). It will sometimes be found of advantage to dissolve the fat in ether and to shake the ethereal solution with the alkali.

Butter yellow is not easily extracted from fat by alkaline solutions, but we have found that it dissolves in strong hydrochloric acid with a pink colour; if a fat containing butter-yellow be warmed and shaken with an equal volume of hydrochloric acid, a strong pink colour will be developed, but will disappear on dilution. Wool heated in the hydrochloric acid will be dyed a magenta colour, turning to light yellow on washing with water, but becoming the original colour when again placed in strong hydrochloric acid.

J. F. Geisler mixes the filtered fat with a little Fuller's earth on a porcelain plate, when in the presence of certain coal-tar dyes a pink mass is obtained; we have not found this test satisfactory.

§ 204. 2. *Examination and Analysis of the Fat.*—By far the most important process in butter analysis is the examination of the fat. The

¹ *The Analyst*, p. 179, June 1902.

data by which the analyst judges whether a butter consists of foreign fats, entirely or partly, are derived from—(a.) Certain simple tests; (b.) the melting-point; (c.) the angle of refraction; (d.) the specific gravity; (e.) viscometry; (f.) acid value; (g.) Koettstorfer's test; (h.) the relative proportion of soluble and insoluble fatty acids; (i.) Reichert-Wollny distillation process. For many of these tests the first requisite is a pure dry fat. This is easily accomplished by melting a sufficient quantity of the butter over the water-bath. In a short time the water, curd, and salt sink to the bottom, and the nearly pure fat can be poured off. Should it not be clear, it must be filtered through filtering paper or glass-wool. This operation will necessitate the filter being kept warm in a suitable steam-jacket.

§ 205. *Certain Simple Tests.*—(For the patterns assumed by solidifying fats, see the 4th edition of this work, pp. 346–348; although of scientific interest, they have been found of little practical value, and are therefore omitted.)

W. G. Crook¹ has experimented upon the solvent action of carbolic acid on butter as compared with other fats. 1 grm. of purified butter-fat is put in a test-tube and liquefied, 2½ c.c. of carbolic acid solution (10 acid, 1 water) are added, after which the mixture is shaken, and then put on one side for a little time. If the sample is pure butter, it wholly dissolves; if beef, mutton, or pork-fat is present, the mixture will resolve itself into two solutions of different densities, with a clear line of demarcation. If beef-fat, the lower layer will occupy about 49.7 per cent. of the total volume; lard, 49.6; and mutton, 44.0. W. Lenz has also tried this process, and generally confirms the results obtained by Mr. Crook.²

P. Casamajor (*Chem. News*, xliv., 309, 310) has proposed a novel method of distinguishing margarine from genuine butter. Pure butter at 15° has the same specific gravity as alcohol of 53.7 per cent., specific gravity = .926, and margarine as alcohol of 59.2 per cent., specific gravity = .918. Any butter, therefore, which is adulterated with margarine will float in alcohol of 53.7 per cent. In alcohol, 56.8 per cent. (mean of 53.7 and 59.2), pure butter when melted sinks, margarine floats. In alcohol, 59.2 per cent., butter sinks, whether solid or fluid. The amount of adulteration is calculated as follows:—Determine the strength of alcohol, the same specific gravity as the sample, let it e.g. = 57 per cent., from it take 53.7 per cent., which is a constant, being the butter expressed, as it were, in equivalent percentage of alcohol, and multiply the remainder by the reciprocal of the difference between the strengths of alcohol of the same specific gravity as margarine and butter = $(57 - 53.7) \cdot 0.18 = 5.94$ per cent. of margarine.

Valenta's Test.—By far the most valuable simple test is the behaviour of butter fat with acetic acid; this is known under the name of Valenta's test. The most accurate method of carrying out this test is that proposed by Chattaway, Pearmain, and Moor:—

The butter-fat, melted and filtered at as low a temperature as possible, is further dried by filtration through a dried filter-paper. 2.75 grms. of this fat are weighed into a stoppered test tube, and 3 c.c. of 99.5 per cent. acetic

¹ *Analyst*, 1879, 1111.

² *Zeitschrift für analyt. Chemie*, 1880, 370. C. Husson (*Compt. Rend.*, lxxxv., 718) has proposed an ancient test depending on the different solvent properties of alcohol for margarine, etc.; and F. Filsinger has a very similar method (*Pharm. Central Halle*, xix., 42). Both these tests, however, are of little practical value among such a number of positive reactions.

acid added. The tube is placed in a beaker of water which is gradually warmed, until, on shaking the tube, a clear solution is obtained; the temperature is carefully noted. Several determinations should be made. They obtained the following figures:—

	Maximum.	Minimum.	Average.
Butter fat,	39·0	29·0	36·0
Margarine,	97·0	94·0	95·0

The acetic acid should, in all cases, be tried on a sample or samples of genuine butter-fat; if this is done, no mistake will be made. Jean¹ relies rather upon the amount of acetic acid dissolved; he places about 8 c.c. of fat in a graduated test-tube, 1 c.c. in diameter, immersed in water of 50° C.; then removes the excess of fat by a pipette until the fat is exactly 3 c.c. at 50° C.; then 3 c.c. of glacial acetic acid, specific gravity 1·0565, are introduced, the acid being measured at 22° C.; the contents are warmed for a few minutes, the tube corked and well shaken. The tube is then placed in the water at 50° C., and the volume of undissolved acetic acid read off. Nine samples of butter averaged 63·33 per cent. of acetic acid dissolved; margarines vary from 27 to 32 per cent. It is obvious that the turbidity test, and the amount of acetic acid dissolved can be done on the same sample at the same time.

M. Crismer² has proposed a turbidity method under the name of "the critical temperature of dissolution," which differs from the Valenta test, but is probably equal to it in value. About half a c.c. of the filtered fat is introduced into a tube of small diameter; to this is added about 0·75 c.c. of alcohol; the tube is hermetically sealed and attached by means of a platinum wire to the bulb of a thermometer; the bulb and tube are immersed in a small sulphuric acid bath, and the temperature slowly raised until the meniscus separating the two layers becomes a horizontal plane. At this point the thermometer and tube are withdrawn from the bath, and the two liquids mixed together by shaking the tube with the thermometer; they are then again placed in the bath and the temperature allowed to fall, the thermometer with attached tube being shaken all the time. The moment in which there is a marked turbidity is noted, and this is considered the temperature of dissolution. Fourteen genuine butters gave from 98° to 102° C.; mean 100° C. Margarines varied from 122° to 126° C.

As margarine, as a rule, contains certain vegetable oils, valuable information may often be obtained by applying to the filtered butter-fat the following tests:—

(1) *Baudouin's Test*, fully described in article on olive oil (*post.*).

The butter-fat should be shaken several times with hydrochloric acid to purify it, and only 1 c.c. of a 1 per cent. alcoholic solution of furfuraldehyde used. Commercial furfuraldehyde should be distilled under reduced pressure before using. The reaction should take place at once.

H. Weigmann³ experimented with cows to which sesamé cake was given in increasing quantities up to 3 kilos per day, but could obtain no reaction for sesamé oil in the butter. C. Annato⁴ made some similar experiments; he, however, got what he considered a sesamé reaction; his results have been

¹ *Corps gras industriels*, 1892, xix., 4.

² *Bull. de l'Assoc. Belge des Chimistes*, ix., 1895; also abstract in *Analyst*, September, 1895.

³ *Milchzeit.*, 1898, 529.

⁴ *Chem. Centr.*, 1901, ii., 836.

contested by other chemists, and there can be no doubt that butters which give a sesamé reaction are almost certain to contain margarine.

(2) *Becchi's Test* for cotton seed oil. Article on 'olive oil' (*post.*).¹

The results obtained by this test must be accepted with reserve, as butter made from the milk of cows fed on cotton seed cake may give a distinct reaction for cotton seed oil.

(3) *Wellman's Test* for vegetable oils (p. 315) may also be applied.

§ 206. (b.) *The Melting-Point.*—Various methods have been proposed for the determination of the melting-points of fats. The one used by most analysts is to take the melting-point in a fine tube. A piece of quill-tubing is drawn out, so as to make a tube about the diameter of a knitting needle, and from 2 to 3 inches in length. The fat is now drawn up to the extent of about an inch, and permitted to solidify. The tube thus charged is placed in some cold water in a small beaker, which is 'nested' in a second beaker, a little water being between the two, the inner beaker carrying also a thermometer. Heat is now applied, and the moment the fat runs up the tube the temperature is noted. A modification of this process¹ is to take a short capillary tube, blow a bulb on it, and while the bulb is still hot, plunge the open end into the melted fat; let it run up a short distance, and then solidify the fat by the application of cold. To take an observation, the tubes are placed in water, so that the bulb is uppermost; on melting, the fat runs up into the semi-vacuous bulb, and this rise is somewhat more easily observed than in the simpler process.

Another method is the employment of a little bulb weighted with mercury, so as to weigh from 3 to 4 grms.; the bulb rests on the surface of the fat in a test-tube, which is immersed in a beaker of water provided with a thermometer, and the moment the bulb sinks is noted. A modification of this is the employment of a light float sunk to the bottom of the fat, and retained there until it is solid; on now applying heat, the float rises at a certain temperature, which is taken as the melting-point. These processes are not entirely satisfactory, and different observers obtain results which do not agree well. Reinhardt² takes the melting-point of fats as follows:—The fat is drawn up when melted into a fine tube, *b*, immersed in a beaker of water, *c*; the tube, *b*, is attached as shown in the diagram (fig. 35) to a simple form of pressure apparatus, consisting of a stoppered cylinder, *g*, the caoutchouc stopper of which carries a graduated thistle-head funnel, *f*, and is connected with the tube carrying the fat by the short right-angled tube, *a*; it is, therefore, possible to put a water pressure on the fat by filling up the graduated stem of the funnel to any desired height. To make comparative observations, the tube, *b*, must be each time filled to the same height, and immersed the same depth in the water; the pressure must also be the same. Heat should be applied very gradually, and the thermometer, *t*, graduated so as to allow of fifths being read. The end of the observation is when the first bubble of fat is forced out and rises to the surface of the water.

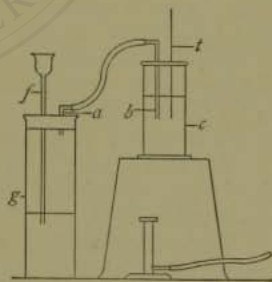


FIG. 35.

Disc Method of taking Melting-Points.—A method of taking melting-

¹ O. Kellner, *Zeitschrift für anal. Chemie*, xx., 1.

² *Repert. anal. Chem.*, 1886.

points, adopted by the American Association of Official Agricultural Chemists at their meeting in Chicago, 1893, is thus described :¹—

The apparatus for determining the melting-point consists of (1) an accurate thermometer for reading easily tenths of a degree; (2) a cathetometer for reading the thermometer (this may be done with an eye-glass if held steadily and properly adjusted); (3) a thermometer; (4) a tall beaker-glass 35 cm. high and 10 cm. in diameter; (5) a test tube 30 cm. long and 3.5 cm. in diameter; (6) a stand for supporting the apparatus; (7) some method of stirring the water in the beaker; for example, a blowing bulb of rubber, and a bent glass tube extending to near the bottom of the beaker; (8) a mixture of alcohol and water of the same specific gravity as the fat to be examined.

The discs of the fat are prepared as follows :—The melted and filtered fat is allowed to fall from a dropping tube from a height of 15 to 20 cm. on a smooth piece of ice floating in water. The discs thus formed are from 1 to 1.5 cm. in diameter and weigh about 200 mgrms. By pressing the ice under the water the discs are made to float on the surface, whence they are easily removed with a steel spatula, which should be cooled in the ice water before using.

The mixture of alcohol and water is prepared by boiling, in two separate vessels, distilled water and 95 per cent. alcohol for ten minutes to remove the gases which they may hold in solution. While still hot the water is poured into the test-tube already described until it is nearly half full. The test-tube is then nearly filled with the hot alcohol. It should be poured in gently down the side of the inclined tube to avoid too much mixing. If the tube is not filled until the water has cooled, the mixture will contain so many air bubbles as to be unfit for use. These bubbles will gather on the disc of fat as the temperature rises and finally force it to the top.

The test-tube containing the alcohol and water is placed in a tall beaker containing water and ice until cold. The disc of fat is then dropped into the tube from the spatula, and at once sinks until it reaches a part of the tube where the density of the alcohol-water is exactly equivalent to its own. Here it remains at rest and free from the action of any force save that inherent in its own molecules.

The delicate thermometer is placed in the test-tube, and lowered until the bulb is just above the disc. In order to secure an even temperature in all parts of the alcohol mixture in the vicinity of the disc, the thermometer is moved from time to time in a circularly pendulous manner.

The disc having been placed in position, the water in the beaker-glass is slowly heated, and kept constantly stirred by means of the blowing apparatus already described.

When the temperature of the alcohol-water mixture rises to about 6° C. below the melting-point, the disc of fat begins to shrivel, and gradually rolls up into an irregular mass.

The thermometer is now lowered until the fat particle is even with the centre of the bulb. The bulb of the thermometer should be small, so as to indicate only the temperature of the mixture near the fat. A gentle rotatory movement should be given to the thermometer bulb. The rise of temperature should be so regulated that the last two degrees of increment require about ten minutes. The mass of fat gradually approaches the form of a sphere, and when it is sensibly so, the reading of the thermo-

Chem. News, June 22, 1894.

meter is to be made. As soon as the temperature is taken, the test-tube is removed from the bath and placed again in the cooler. A second tube, containing alcohol and water, is at once placed in the bath. The test-tube (ice water having been used as the cooler) is of low enough temperature to cool the bath sufficiently. After the first determination, which should be only a trial, the temperature of the bath should be so regulated as to reach a maximum of about 1°·5 C. above the melting-point of the fat under examination.

The distilled water for floating the piece of ice on which the discs are made should be recently boiled, to free it of all air particles.

The edge of the discs should not be allowed to touch the sides of the tube. This accident rarely happens, but in case it should take place, and the disc adhere to the sides of the tube, a new trial should be made.

Triplicate determinations should be made, and the second and third results should show a near agreement.

Example.—Melting-point of sample of butter :—

First trial,	33°·15 C.
Second trial,	33°·05 C.
Third trial,	33°·00 C.

The following melting-points are taken by the old methods, and are somewhat high, but are given as the values usually accepted :—

Margarine,	31°·3 C.
Cocoa butter,	34°·9 C.
Butter (average),	35°·8 C.
Beef-dripping,	43°·8 C.
Veal-dripping,	47°·7 C.
Mixed,	42°·6 C.
Lard, from	42° to 45° C.
Ox-fat, from about	48° to 53°·0 C.
Mutton-fat, from	50° to 51°·6 C.
Tallow,	53°·3 C.

It hence follows that a low melting-point indicates the probable presence of margarine, especially that which is partly manufactured from a concrete oil, obtained from the seeds of *Garcinia Indica*, and is known under the name of Mangosteen oil, or kokum butter. A higher melting-point indicates, as a probable adulterant, dripping, lard, or other animal fat.

The Titer Test.—A melting-point method, which gives useful results, is the 'freezing' point of the fatty acids; unfortunately it requires considerable material. At least 50 grms. of the butter-fat are saponified, the fatty acids separated and allowed to solidify; the acids are melted and poured into a test-tube 16 cm. long and 3·5 cm. wide, filling the tube half full. The tube is put into the neck of a suitable flask, and a Beckman's¹ thermometer inserted; when the mass begins to cloud, the thermometer is given a rotatory movement, and the mercury watched. At first it falls regularly, then stops, and gradually rises one or more tenths of a degree, to again stop, and then fall as before. The last stationary point is called the 'titer' or solidifying point.

It is advisable in all the above processes to allow the melted butter or fatty acids, as the case may be, to 'set' a definite time previous to the

¹ Since Beckman's thermometer only contains five degrees divided into tenths, it requires adjusting to within two degrees of the probable melting-point, and should not be inserted until the fat is about that temperature.

determination. The same fat melted and cooled will give two or more melting-points, if such melting-points are determined at different intervals of time, it is best then, in all cases, to prepare the fats one day and determine the melting-points on the following day.

§ 207 (c) *Application of the Refractometer to the Testing of Butter-fat.*—J. Skalweit¹ has made some determinations of the angle of refraction of various fats, and believes there is a sufficient difference between the angle of margarine and pure butter to enable substitutions and adulterations to be detected; he uses an Abbe's refractometer. The fat, kept for some time at 20° C., is spread out on a watch-glass, and covered by a piece of Swedish filter-paper; the fat is absorbed by the paper, and a clear grease spot forms in the centre. The grease spot is applied to the edge of the Nicol prism to which it readily adheres; the apparatus is closed, and the angle estimated at 20° C. The following results are given:—

Water,	1.333	
Olein, from commercial oleic acid,	1.4635	
Oleic acid, at 17°,	1.4638	
Do. at 20°,	1.4639	
Genuine butter,	1.4652	
Do.	1.4658	
Cocoa butter,	1.4680	
Lard,	1.4690	
Margarine, 1st quality,	1.4692	
Do., 2nd "	1.4720	
Do., 3rd "	1.4796	
Do., Oil,	1.4680	
Do.,	1.4712	
Do.,	1.4693	
Do.,	1.4698	Hanoverian manufacture.
Do.,	1.4698	
Do.,	1.4698	English make.
Do.,	1.4733	
Refined cotton-seed oil,	1.4748	
Crude " "	1.4732	
Cod-liver oil,	1.4801	
Linseed oil,	1.4835	

The authors have made a number of determinations of angles of refraction by Abbe's instrument. The apparatus was enclosed in a copper air-bath, open at the top, but provided elsewhere with double walls, between which were placed glycerin and water; the boxing was completed by a cover of caoutchouc, in which a slit was cut to allow of the eyepiece protruding through; a small window in the copper box admitted light to the mirror. By carefully heating this copper bath, the whole instrument could be maintained at any desired temperature.

It is neither advisable nor necessary to place the drops of fat on filter-paper, it is best to examine a thin film between the prisms themselves, the edge of the shadow is then sharply defined. By means of the long arm, called by the inventor the 'alhidade,' the shadow is adjusted so that its border is exactly coincident with the cross lines, and the field is made colourless by working a milled head which regulates and measures the dispersion. These operations the authors perform in the copper air-bath itself, and if after 15 minutes' interval at any desired temperature there is no further change in the refraction, the instrument is removed and the

¹ Abbe's refractometer is fully described in "Neue Apparate zur Bestimmung des Brechungs- u. Zerstreungsvermögens fester u. flüssiger Körper." Von Dr. E. Abbe. Jena, 1874. A more modern and better instrument is that of Zeiss, described at page 286.

index read. To ensure accuracy a second observation is desirable, and the mean of the two taken.

The best temperature for observation the authors conclude to be the same one at which the specific gravity is taken, viz., 38° C. At that temperature the most uniform results were obtained, and this temperature gives the necessary data for the calculation of what Dr. Gladstone has termed *the specific refractive energy, i.e., the angle of refraction, minus unity, divided by the density.*

In the following tables the angle of refraction, the specific refractive energy, and various other data are given as determined from genuine and adulterated butter-fats.

TABLE XXI.—THE ANGLE OF REFRACTION, SPECIFIC REFRACTION, MELTING-POINT, SPECIFIC GRAVITY, AND THE RESULTS OF THE REICHERT AND KOETTSTORFER TESTS AS REPRESENTED IN 33 GENUINE BUTTERS.

Angle of Refraction at 33°.	Specific Refraction (i.e., Angle of Refraction, -1 divided by density).	Specific Gravity, at 38°.	Melting-Point.	c.c.'s of d. n. Alkali used in Reichert's Test. ¹	Koettstorfer's Test. Mgrms. of KHO used.
1.4543	.49838	.91158	31.6	14.4	226.0
1.4565	.50037	.91233	31.2	14.9	229.0
1.4570	.50145	.91138	30.3	13.9	223.5
1.4556	.51094	.91246	32.0	15.3	224.1
1.4570	.50147	.91134	31.6	14.3	224.1
1.4575	.50155	.91217	31.4	13.6	221.2
1.4562	.50088	.91079	31.6	14.1	220.6
1.4558	.49969	.91217	30.7	14.0	223.7
1.4575	.50174	.91085	33.5	13.9	221.2
1.4565	.50018	.91266	35.0	15.3	224.4
1.4565	.50004	.91293	33.0	14.5	222.9
1.4560	.49944	.91310	32.0	15.0	234.0
1.4570	.50051	.91290	32.8	15.2	231.4
1.4575	.50068	.91378	33.0	15.7	234.4
1.4570	.50097	.91225	33.5	13.8	233.3
1.4555	.51071	.91266	33.5	15.5	229.6
1.4565	.49962	.91369	34.5	15.6	234.9
1.4555	.49918	.91251	30.5	15.0	227.5
1.4555	.49887	.91307	35.0	16.0	233.3
1.4555	.49900	.91281	35.0	14.9	227.2
1.4565	.49937	.91206	33.5	15.4	229.6
1.4555	.49890	.91303	35.0	16.0	231.5
1.4555	.49987	.91124	35.0	14.4	223.1
1.4555	.49910	.91264	31.0	15.8	232.9
1.4560	.49987	.91222	34.8	15.8	234.2
1.4568	.57236	.91235	32.5	15.1	234.4
1.4560	.49570	.91276	32.5	15.5	235.2
1.4560	.50023	.91156	33.0	13.5	233.8
1.4562	.50055	.91133	33.4	13.3	226.7
1.4565	.50012	.91277	32.0	15.0	231.9
1.4560	.51140	.91286	31.5	15.4	232.1
1.4570	.51270	.91214	33.3	15.0	230.9
1.4555	.51061	.91285	33.2	...	233.7
Max. 1.4575	.51270	.91378	35.0	16.0	232.9
Min. 1.4543	.49887	.91079	30.5	13.3	220.6
Mean 1.4562	.50212	.91234	32.8	14.8	232.0

¹ To convert these numbers into approximate Reichert-Wolny values they must be multiplied by 2.

EXAMPLES OF LOW-CLASS AND ADULTERATED BUTTERS.

Angle of Refraction at 38° C.	Specific Refraction.	Specific Gravity, at 38° C.	Melting-Point.	c.c.'s of d. n. Alkali used in Reichert's Test.	Koettstorfer's Test. Mgrms. of KHO used.
¹ 1.4577	.50255	.91073	29.8 C.	12.0	230.7
² 1.4585	.50422	.90932	29.0 "	7.4	213.9
³ 1.4580	.50293	.91066	30.0 "	11.4	226.8
⁴ 1.4624	.51050	.90578	26.0 "	2.6	199.4
⁵ 1.4595	.50550	.90902	26.5 "	7.7	212.8
⁶ 1.4582	.50343	.91016	27.0 "	9.7	218.4

The angle of refraction in milk-fat, as extracted by ether, and genuine butters, varies from a maximum of 1.4575 to a minimum of 1.4543 at 38° C.; the mean is 1.4562, and the average specific refractive energy is .502. The refractive angle of margarine is always above 1.4620, generally 1.4639 or 1.4640, at 38°. A mixture of 50 per cent. by weight of margarine (specific gravity .90578, melting-point 25.5 C., refractive angle 1.4624) and 50 per cent. of butter (specific gravity .91073, melting-point 29.8 C., refractive angle 1.4577) gave an angle of, at 38° C., 1.4595, to which a genuine milk-fat may attain; the same materials, mixed in the proportion of 25 per cent. of margarine and 75 per cent. of butter, gave 1.4582 as the angle at 38° C.

Hence it is certain that to the analyst the refractometer has some utility, although it will not assist much in the case of moderate mixtures; but what may be said is that a butter at 38° C., having an angle at or below 1.4570, is almost certain to be genuine, but above that angle is probably adulterated.

§ 207a. *Zeiss' Butyro-refractometer.*—This is an instrument which is now largely used for the rapid examination of butter, margarine, lard, and other fats. It is constructed upon the same principle as Abbe's refractometer; the prisms are, however, provided with a jacket through which water is passed, the temperature being indicated by a thermometer. (See fig. 35a.) To make a reading the prisms are opened, a drop of the filtered fat is placed on the surface of the lower prism. The prisms are closed, and a reading taken either by ordinary light or better by the sodium light. The temperature is read off, and the reading corrected to any desired temperature, by adding or subtracting .55 scale degrees for every 1° C. Thus, if the scale reading at 35° C. be 47, and it is required to know the scale reading at 40° C., .55 × 5 must be subtracted from the reading at 35° C., or if the reading at 45° C. be 41, the reading at 40° C. will be 41 + .55 × 5, or 43.75 scale divisions.

Genuine butters give, as a rule, readings under 44° at 40° C., with an average reading of about 43°; should higher values than these be obtained foreign fat may be present; should a low value be found cocoa-nut oil may be present. Margarine has at 40° C. a value of about 49°, and cocoa-nut oil about 38°.

¹ A low-class butter.

² An adulterated butter, for which vendor was prosecuted and fined.

³ A low class butter, probably adulterated.

⁴ Margarine.

⁵ 50 per cent. margarine, No. 4, and 50 per cent. of low-class butter, No. 1.

⁶ 25 per cent. margarine, No. 4, and 75 per cent. of low-class butter, No. 1.

The following table by A. L. Winton and A. W. Ogden gives the relationship between the Zeiss reading and the refractive index:—

REFRACTIVE INDICES CORRESPONDING TO READINGS OF THE ZEISS BUTYRO-REFRACTOMETER.

Reading.	Refractive Index.	Reading.	Refractive Index.	Reading.	Refractive Index.
40.0	1.4524	53.5	1.4616	67.0	1.4704
40.5	1.4527	54.0	1.4619	67.5	1.4707
41.0	1.4531	54.5	1.4623	68.0	1.4710
41.5	1.4534	55.0	1.4626	68.5	1.4713
42.0	1.4538	55.5	1.4629	69.0	1.4717
42.5	1.4541	56.0	1.4633	69.5	1.4720
43.0	1.4545	56.5	1.4636	70.0	1.4723
43.5	1.4548	57.0	1.4639	70.5	1.4726
44.0	1.4552	57.5	1.4642	71.0	1.4729
44.5	1.4555	58.0	1.4646	71.5	1.4732
45.0	1.4558	58.5	1.4649	72.0	1.4735
45.5	1.4562	59.0	1.4652	72.5	1.4738
46.0	1.4565	59.5	1.4656	73.0	1.4741
46.5	1.4569	60.0	1.4659	73.5	1.4744
47.0	1.4572	60.5	1.4662	74.0	1.4747
47.5	1.4576	61.0	1.4665	74.5	1.4750
48.0	1.4579	61.5	1.4669	75.0	1.4753
48.5	1.4583	62.0	1.4672	75.5	1.4756
49.0	1.4586	62.5	1.4675	76.0	1.4759
49.5	1.4590	63.0	1.4678	76.5	1.4762
50.0	1.4593	63.5	1.4681	77.0	1.4765
50.5	1.4596	64.0	1.4685	77.5	1.4768
51.0	1.4600	64.5	1.4688	78.0	1.4771
51.5	1.4603	65.0	1.4691	78.5	1.4774
52.0	1.4606	65.5	1.4694	79.0	1.4777
52.5	1.4609	66.0	1.4697	79.5	1.4780
53.0	1.4613	66.5	1.4700		

§ 207b. *The Oleo-Refractometer of MM. Amagat and Jean.*¹—This is a more convenient and better instrument for observations on fats than ordinary refractometers, the differences between butters and margarines being greater. The centre of this instrument (fig. 36) consists of a circular reservoir made of metal, B B, closed by two plates of glass in front of two lenses, E and E', the one lens belonging to the collimator tube, S, and the other to the telescope tube, S'. In the middle of this reservoir is placed a receptacle in the shape of a prism, its glass sides having an angle of 107°. In front of the field glass of the telescope is a photographic scale, H (shown enlarged at K), and there is an adjustable shutter placed vertically, so arranged as to divide the luminous field into two parts, the one bright, the other in shadow. The readings of the apparatus are made from the edge of the shadow. If in both reservoirs is placed the same liquid at the same temperature, then the shadow occupies the zero of the scale; but if a different liquid be placed in the inner prism, there will be, according to its nature, a deviation either to the right or left, the amount of the deviation being appreciated by the numbers on the scale. The makers supply a typical oil having no refraction, by means of which the instrument is set;

¹ Ladan Bockairy in Ch. Girard and A. Dupré's *Analyse des Matières Alimentaires*, Paris, 1894; also Muter in *Analyst*, May 1890.

this oil is put in both receptacles, the temperature adjusted to 22° C., and the shutter moved so as to mark zero. The oil is now run out of the

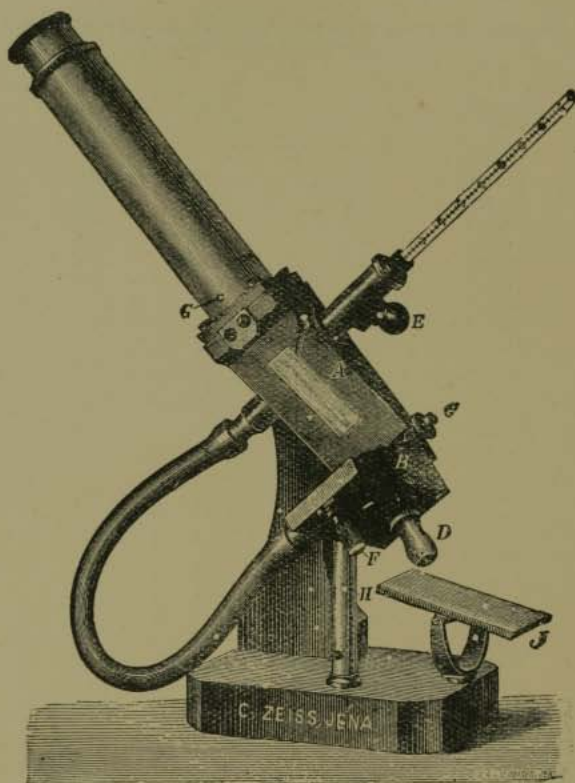


FIG. 35a.

prism, and replaced by the oil to be tested. Solid fats, such as butter or lard, are tested at a temperature of 45° C. In all cases the oil or butter

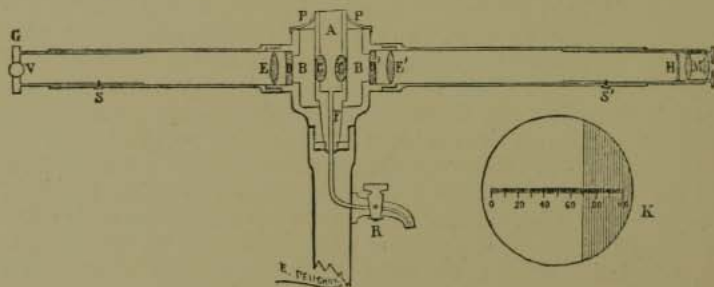


FIG. 36.—Amagat and Jean's Oleo-Refractometer.

is to be freed from free fatty acids. The butter is melted, filtered, and shaken up with a little powdered gypsum, the gypsum is allowed to settle,

and the fat is then filtered again, and poured into the prism. The fat should never be heated above 50° C. The rule is that vegetable oils and fats rotate to the right, animal oils and fats to the left. Thus—

Olive oil,	+ 1·5 to 2	Neatsfoot oil,	- 3
Colza ,,	+16·5 to 17·5	Horsefoot ,,	-12
Ground nut oil,	+ 4·5	Lard, ,,	-12·5
Sesame oil,	+17	Beef tallow,	-16
Cotton ,,	+20	Mutton, ,,	-20
Linseed ,,	+53	Butter-fat, {	-25 to -34
Castor ,,	+40	} average about	-30
Hempseed oil,	+33	Margarine,	-15
Poppy ,,	+30	Cocoa-nut oil deviates to the left like	
Almond ,,	+ 6	an animal fat; it deviates some	59°.
Japonica ,,	+50		

§ 208. (d.) *Specific Gravity*.—One method of obtaining the specific gravity of butter-fat is to fill a counterpoised specific gravity bottle, provided with a thermometer stopper, of 50 to 100 grms. capacity, with water of 35° C. (95° Fahr.), and immerse it in a beaker of water of about 43° C. (109°·4 Fahr.). By thus heating the specific gravity bottle by a liquid which is falling in temperature, the water in it can be brought exactly to 37°·7 C. (100° Fahr.), at which temperature the bottle is taken out, slightly cooled and weighed; and in this manner the weight of that particular bulk of water at 37°·7 C. (100° Fahr.) is obtained, and this value used for the subsequent operations. To take the specific gravity of the fat, the pure filtered fat, at 35° C. (95° Fahr.), is poured into the clean dry bottle, and the exact process just detailed followed. Latterly many chemists have preferred to take the specific gravity at 100° C. Butter, as compared with water at 15° C., has a specific gravity at 100° C. from 0·865 to 0·8685; margarine 0·859 to 0·863.

Skalweit has found that the greatest difference in specific gravity is to be found at the temperature of 35° C., and therefore recommends the specific gravity to be taken at that temperature; thus, butter-fat at 100° C. had a specific gravity of 0·8672, margarine 0·8598—a difference of ·0074; but at 35° C. the butter specific gravity was 0·9121, the butterine 0·9019—a difference of ·0102. Specific gravity at 100° C. may be taken either in a specific gravity bottle or by means of a Westphal's balance, but best of all by a Sprengel tube. In a former edition of this work Mr. Wigner's proposal to use specific gravity bubbles was mentioned; in butters of 0·911 specific gravity (at 38° C.) a bead of specific gravity 0·889 slowly sinks at 63° C.; but the process is not sufficiently accurate to have found favour among analysts.

The specific gravity, as first pointed out by Mr. Bell, of Somerset House, has a direct relation or correspondence to the percentage of insoluble acids, a fact, it must be remembered, only applicable to pure unadulterated butter-fat. Thus—

Specific Gravity at 37°·7 C. (100° F.).	Actual Insoluble Acids Found. Per cent.	Specific Gravity at 37°·7 C. (100° F.).	Actual Insoluble Acids Found. Per cent.
·91382	87·47	·91286	88·52
·91346	87·89	·91276	88·62
·91337	87·98	·91258	88·80
·91290	88·48	·91246	89·00 ¹

¹ Muter, *Analyst*, i., p. 7, 1877.

The fats used for the adulteration of butter are of low density. Vegetable butterine has a specific gravity .90294, dripping .90659, so that a low specific gravity at 37.7° C.—that is, anything below .91101¹—is strongly indicative of foreign fat.

§ 209. (e.) *The Viscometry of Butter.*—Killing² has described an apparatus for taking the viscometry of butter; it essentially consists of a 50 c.c. pipette, provided with a thermometer within the pipette, and a glass cylinder which acts as a jacket to the pipette and enables the contents to be maintained at a suitable temperature. The cylinder is also provided with a thermometer. The butter-fat, properly clarified, melted, and brought to a temperature of 40°·5 C., is sucked up into the pipette above the mark, then water at 42° C. is poured into the outer jacket, and the butter-fat, when both thermometers stand at 40° C., run down to the mark, so as to measure exactly 50 c.c.; then the tap of the pipette (which is at the top, not at the bottom) is opened wide, and the time accurately noted which the butter takes to run into a beaker to a mark below the body of the pipette; this is compared with the same quantity of water at 20° C. run in the same manner from the same cylinder, water being taken as 100; thus, if a butter-fat took 222 seconds and water 80·33 seconds, the calculation is—

$$\frac{222.00 \times 100}{80.33} = 276.3.$$

Various margarines gave a mean time of 4 minutes 12·77 seconds, equal to a viscosity number of 314·7; various butters, a mean time of 3 minutes 43·76 seconds; mixtures gave proportionate numbers. Lard and beef-fat both took a longer time than margarine. By carefully standardising a particular pipette for water, for margarine, and for pure butter, this process may give useful results.

R. Brulle³ distinguishes butter from margarine by treating 5 c.c. of the clarified fat at 130° C. with a little pumice-stone and 8 drops of fuming nitric acid, the whole mixed and heated for twelve minutes; it is then cooled to 21° C., and, after one hour, tested in an 'oleo-grammeter,' which consists of a rod gliding on a bearing, and surmounted by a little table on which weights can be placed. On an average, 250 grms. are required for pure butter, and 5000 grms. for margarine, mixtures giving intermediate figures. Should this process be used, each analyst had better obtain his own standard.

§ 210. (f.) *The Acid Value.*—The acid value of a fat is the number of milligrammes of potassium hydrate required to saturate the free fatty acids in 1 gm. of the fat. This becomes of importance in the examination of rancid butters, but in butters which are not rancid it is seldom estimated. A weighed quantity of the melted, filtered, clear butter-fat is heated on the water-bath with a mixture of 2 parts of ether and 1 of alcohol, until a perfect solution is obtained; phenol-phthalein is added, and then the solution is exactly neutralised by decinormal alkali. Thus, 10 grms. of butter-fat, dissolved as above, required for neutralisation 3 c.c. of decinormal potash; since each c.c. of d. n. potash is equal to 5·61 mgrms. of KOH, it follows that the 3 c.c. are equal to 16·83 mgrms. of KOH; therefore, as 10 grms. were taken, the acid value is one-tenth of this—namely, 1·683 mgrms.

¹ With the exception of cocoa-nut, cotton seed, arachis, and sesamé oils.

² *Zeit. f. angewandte Chemie*, 1894 and 1895.

³ *Comptes Rendus*, 1893, cxvi., 1255.

§ 211. (g.) *Direct Titration of Butter by Alcoholic Potash.*—This elegant test was first suggested by Dr. Koettstorfer, and is a general method more or less applicable to all fats, but especially suited to butter-fat, for most other fats contain only the higher fatty acids; as the lower acids have a smaller molecular weight, butter must contain more molecules of acid than equal weights of another fat—in other words, requires more potash for saponification. Koettstorfer used semi-normal hydrochloric acid and an alcoholic solution of potash, this alkaline solution being about the same strength as the acid;¹ the indicator was a dilute alcoholic solution of phenol-phthalein. From 1 to 2 grms. of the purified filtered fat are weighed in a tall beaker of about 70 c.c. capacity, 25 c.c. of the potash solution are added, and heated on a water-bath.² When the alcohol is nearly boiling, the mixture is stirred with a glass rod till all the fat is dissolved, which does not take more than a minute. The glass rod is washed with a little alcohol, and the beaker covered with a watch-glass, and heated further for fifteen minutes, in such a manner that the alcohol does not boil too violently. At the end of the quarter of an hour, the watch-glass is washed with spirit, and the alcoholic solution is stirred for one minute longer with the glass rod before used, so as to saponify any fat that may still adhere to it. The solution is now taken from the water-bath; 1 c.c. of an alcoholic solution of phenol-phthalein added, and it is titrated back with semi-normal hydrochloric acid. The exact point is very sharply indicated by the phenol-phthalein changing from a crimson to a yellow. (This reagent is very sensitive to CO₂; it is therefore better to use a flask than an open beaker.) Thirteen butters treated in this way by Dr. Koettstorfer used for every gm. of fat from 221.5 to 232.4 mgrms. of KHO. Thirty-three genuine butters examined in one of the authors' laboratory gave as a maximum 232.9 mgrms., as a minimum 220.6 mgrms. KHO (see Table at p. 285).

On the other hand, there is a wide difference between this amount and that required by other fats, the following being about the saturation capacity in mgrms. of potash for 1 gm. of various fats:—

	Potash KHO. Milligrammes.
Oleo-margarine,	195.5
Beef-dripping,	196.5
Tallow,	196.8
Lard from kidneys,	195.8
Lard from unsmoked bacon,	196.7
Commercial lard,	195.0
Dripping,	197.0

Or if the suggestion of Mr. Allen³ be accepted, and the results be translated into equivalents of the fat by dividing 56.1 by the grms. of potash, the results are as follows—

Oleo-margarine,	286.5
Beef-dripping,	285.5
Tallow,	285.1
Lard from kidneys,	286.5
Lard from unsmoked bacon,	286.7
Commercial lard,	287.1
Mutton dripping,	284.8

¹ The relation of the acid to the alkali must be very carefully determined by several blank experiments.

² A flask and inverted condenser may be used while the saponification is proceeding.

³ *Analyst*, 1879, 162.

The chief convenience in expressing the number in equivalents is, that it then becomes a matter of indifference whether potash or soda¹ is used for the saponification. The practical question in the use of this test is: what is the lowest limit above which a butter may be passed as genuine, but below which it will be necessary to examine the butter by other means? The general opinion of analysts as to this point is, that butter-fat, 1 grm. of which uses less than 226 mgrms. of KHO (equivalents 248·2), is probably adulterated. The formula for calculating the amount of admixture which has been proposed is

$$(227 - n) \times 3 \cdot 17 = x,$$

x being the percentage of admixed fat, n the number of mgrms. of potash used.

§ 212. (*h.*) *The Decomposition of the Fat into Fatty Acids and Glycerin.*—This is effected by saponifying with an alcoholic solution of potash, decomposing the soap with sulphuric acid, washing the subsequent fatty acids with water, titrating the soluble, and weighing the insoluble acids. The details of the process have been so simplified by successive improvements, that what was formerly a tedious and even difficult operation is now moderately speedy and simple. The solutions requisite are as follows:—²

(1.) Approximately semi-normal alcoholic potash solution, 28 grms., roughly weighed, of KHO, dissolved to a litre with alcohol (specific gravity ·840).

(2.) Approximately semi-normal sulphuric acid—*i.e.*, 25 grms. of the strong acid to the litre.³

(3.) Deci-normal soda solution of exact strength, most conveniently made by dissolving metallic sodium in water, in the exact proportion of 2·3 grms. to the litre. [1 c.c. equals ·0088 of butyric acid.]

It is necessary to know with the greatest exactitude the relationship between the potash and the sulphuric acid solution; the exact quantity of alcoholic potash that is to be used in the analysis is delivered from a 25 c.c. or 50 c.c. pipette, as the case may be, phenol-phthalein solution added, and then titrated by the acid. It is also necessary to know the relationship between the d. n. soda and the sulphuric acid, which must be found in the usual way. 4 to 5 grms. of the pure dry fat are weighed by difference into a flask, and 50 c.c. of potash solution added; the flask, closed by a glass marble, is now heated on the top of the water oven, and by occasionally giving it a rotatory motion, saponification is complete under the hour at the low temperature of 50° C.

The senior author does not himself follow the above process, but uses the strong small assay flasks recommended by Dr. Dupré. These flasks are of about 70 c.c. capacity, and with rather long narrow necks, the whole capable of bearing considerable pressure. 4 to 5 grms. of the fat are poured into such a flask, 25 c.c. of potash solution added, well corked with a caoutchouc stopper, which must be secured by string and strong linen or canvas, and then the flask suspended in the boiling water of a water-bath.

¹ It is scarcely necessary to add that should soda be used, then 40, the equivalent of soda, must be divided by grms. of the alkali used.

² "Butter-Fat," by E. W. Jones, F.C.S. *Analyst*, May, 1877.

³ An accurate $\frac{N}{2}$ solution of HCl is now usually employed.

At the end of an hour or less it may be taken out completely saponified.¹ When cool the flask is opened, the soap gently melted and poured into a flask of about 500 c.c. capacity, having a long, rather narrow neck (see fig. 37), which carries the tubes *a* and *b*—the tube *a* for the admission of air, the tube *b* furnished with a stopcock. In this flask the soap is decomposed by adding about 1 c.c. more sulphuric acid than is necessary to neutralise the potash; if, for example, the latter is neutralised by 25 c.c. of the sulphuric acid 26 c.c. are added, and after this addition the fatty acids melted so as to form a layer on the surface of the acid water.² At this point the whole may be diluted with warm water up to 200 c.c. or 300 c.c., the cork carrying the tubes inverted, and the flask turned upside down, as represented in the figure. After standing a few hours the cake is more or less solid, and the lower stratum of liquid may be run off almost clear. It will, however, always be safest to pass it through a filter. By adapting an india-rubber tube to *a*, warm water may be sucked up through *b*, and the fat washed in the flask (perfectly closed by pinching the india-rubber), and then the cake allowed to form as before. The fluid is now again run off from the solid, and this time cold water may be sucked up through *a*, and the whole process of alternations of hot and cold water repeated. Lastly, the cork with its tubes is removed, any adherent fat washed off with warm water into the flask, the latter adapted either to an upright Liebig's condenser and boiled, or connected in the usual way with a receiving flask adjusted by a cork tightly to the bent tube of the condenser, and furnished with a mercury valve, the whole forming a closed system. In the latter case, also, the heat is applied to boiling for five or ten minutes, and the distillate added to the filtrates; lastly, the cork with tubes is again connected, the flask inverted, the liquid when cool run off, and the fat finally washed with a little cold water and allowed to drain.

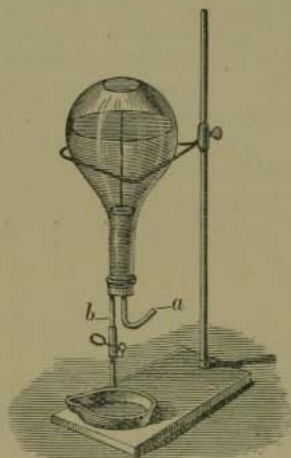


FIG. 37.

The watery liquid contains sulphuric acid, glycerin, sulphate of potash, alcohol, butyric, and the other soluble fatty acids; it will be in bulk from 600 to 700 c.c., and may be made up to any definite quantity. In any case, a portion of it—a quarter, a fifth, or even a tenth—must be taken and titrated with d. n. soda, which, when the quantity required to neutralise the 1 c.c. of sulphuric acid in excess is subtracted, indicates the amount of soluble acid, and is always returned as butyric, which is near enough to the truth.

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Instead of this method it may be useful to distil the acid liquid until all the volatile acid which can be obtained has gone over, and then titrate the distillate. It is also possible to separate the volatile fatty acids from such a solution by shaking up with ether in the tube figured at p. 223, the ether dissolving the acids freely.

¹ The reason for preferring this method is, that less potash is required.

² The American Association of Official Analysts recommend connecting the flask, while the acids are being melted, with a long upright narrow glass tube.

The *insoluble fatty acids* remaining partly in the flask, with a trace on the filter, are now united in a flat porcelain dish. This is done by melting the acids in the flask, pouring off, and extracting by alcohol and ether—the same solvent also dissolving the acids from the filter. On evaporation of the alcohol and ether, one or two large bubbles of water may be formed in the acids, and it is best to add a few drops of absolute alcohol. The dish is now placed on the top of the water-bath (the water in which should only boil gently), and weighed at short intervals; if after twenty minutes only 1 or 2 mgrms. are lost, the weight is considered constant. See also Johnson's method, p. 297.

The following are a few examples of percentages of fatty acids found in genuine butters:—

	(1.)	(2.)	(3.)	(4.)
Soluble,	5·92	5·76	5·37	4·77
Insoluble,	87·86	88·10	87·68	88·44
	<hr/> 93·78	<hr/> 93·86	<hr/> 93·05	<hr/> 93·21

It is generally accepted that 88 per cent. of insoluble acids, if associated with 6·3 of soluble acids, is a fair standard of butter calculation, and that if a butter shows anything less than 89·5 insoluble, with 5 soluble, it may be passed as genuine.

A few examples of adulterated butter-fat are as follows:—

	(1.) A Commercial Butter.	(2.) A Commercial Butter.	(3.) Margarine.
Soluble,	1·98	2·34	·58
Insoluble,	93·30	93·82	95·51
	<hr/> 95·28	<hr/> 96·16	<hr/> 96·09

§ 213. (i.) *Estimation of the Volatile Fatty Acids.*—*Reichert-Wollny Process.*—The original Reichert process and the modification of it known as the Reichert-Meißl process, described in previous editions of this work, are now no longer used, owing to the careful research of Wollny, which proved that the processes were open to error in many ways. The process generally used both for butter and margarine, and which may be almost called the official method in this country, is the Reichert-Wollny process. In order that results may be uniformly comparable, it has been agreed by the Principal Chemist of the Government Laboratory, and a committee appointed by the Society of Public Analysts, that the following method of working shall be adopted. This method is recommended for margarine, but it is equally applicable to all fats. It is essential that the measurements given, the quantity of fat taken, and all other details should be followed closely. Should it be convenient to take 2·5 grms. of fat instead of 5 grms., half the other quantities must be taken and 55 c.c. distilled. The result multiplied by two will be slightly higher than the true Wollny figure. The method is as follows:—

Five grms. of the melted and filtered sample are introduced into a 300 c.c. flask (see fig. 37a), 2 c.c. of aqueous sodium hydroxide (1 : 1) free from carbon dioxide are added, and also 10 c.c. of 92 per cent. alcohol, and the mixture is heated under a reflux condenser, connected with the flask by a T-piece, for 15 minutes in a bath containing boiling water. The alcohol is distilled off by heating the flask on the water-bath for about half an hour, or until the soap is dry. One hundred c.c. of hot water which has been

kept boiling for at least 10 minutes are added, and the flask heated until the soap is dissolved. Forty c.c. of *N*-sulphuric acid and three or four fragments of pumice or broken pipe-stems are added, and the flask is at once connected with the condenser, as indicated in the figure. The flask, which is supported on a circular piece of asbestos 12 cm. in diameter having a hole in the centre 5 cm. in diameter, is first heated with a very small flame, to fuse the insoluble fatty acids without causing the liquid to boil. The heat is then increased, and when fusion is complete 110 c.c.

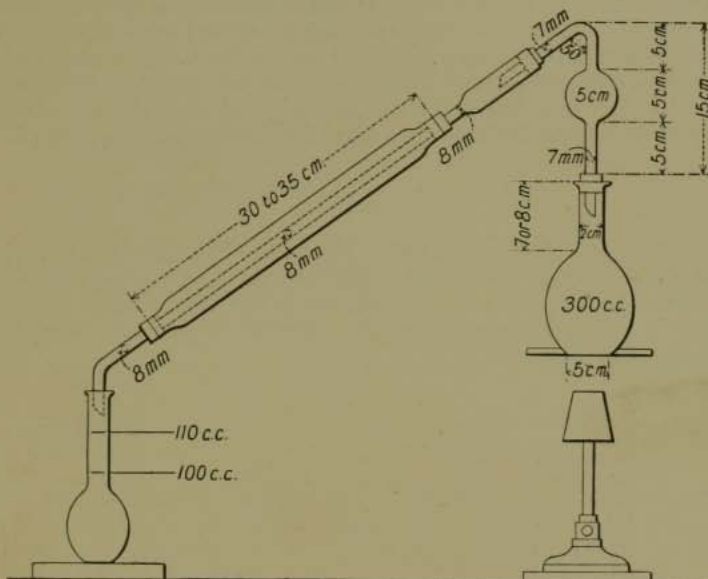


FIG. 37a.—Apparatus for the Reichert-Wollny process.

are distilled off into a graduated flask, the distillation lasting about 30 minutes (say from 28 to 32 minutes). The distillate is shaken, 100 c.c. are filtered off, transferred to a beaker, 0.5 c.c. of phenol-phthalein solution (1 gm. in 100 c.c. of alcohol) added, and the filtrate titrated with *N*/10 sodium or barium hydroxide. As the chemicals used may yield a slightly acid distillate, a blank experiment should be made, and the result allowed for; this should, however, not exceed 0.3 c.c. of *N*/10 alkali. The final result multiplied by 1.1 is the Reichert-Wollny number.

The following table gives, I, the Reichert-Wollny number as determined by this method, and, II, the corresponding amount of butter-fat that is assumed to be present in the margarine:—

I.,	.	.	4.0	4.3	4.6	4.9	5.2	5.5	5.9	6.2	6.5	6.8	7.1
II.,	.	.	10	11	12	13	14	15	16	17	18	19	20

No presumption against the margarine in regard to its content of butter-fat should be raised unless the Reichert-Wollny number thus ascertained exceeds 4.

Leffmann and Beam's Process.—This is in some respects superior to the one described, but chiefly because it gives a sharper end reaction.

In place of alcoholic potash, *glycerol-soda* prepared as follows is used:—

Dissolve 100 grms. of pure sodium hydroxide in 100 c.c. of distilled water and allow to stand until clear. Mix 20 c.c. of this solution with 180 c.c. of pure concentrated glycerol.

Place 5 grms. of the fat in a 300 c.c. flask, add to it 20 c.c. of the glycerol soda, and heat over a Bunsen burner; the mixture will require careful watching, as there is considerable frothing; saponification is complete in a few minutes. The rest of the operation may be carried out as in the Reichert-Wollny process.

English butters made from the mixed cream of several cows have a mean value of 29, and the minimum value adopted in this country is 24, but smaller values may be obtained from the butter of single cows, and under these circumstances the minimum may fall as low as 20.4 (see page 304). Reichert and Meissl have proposed to calculate the amount of real butter-fat by taking 28.78 as a standard by the following formula:—

$$B = \frac{100(n - C)}{28.78 - C}$$

n is the Reichert-Meissl value, and C the value of the admixed fat. Meissl thinks that C may be taken as 3. The formula is, of course, only a rough guide, because the value for the original butter may be higher or lower than 28.78. The following are results obtained by various observers with the Reichert process:—

	D. N. Soda.
	c.c.
Butter-fat,	24 to 32
Lard,	4 to 6
Rape oil,6
Sesamé oil,7
Olive oil,6
Palm oil,	1.0
Kidney-fat,5
Cocoa-nut oil,	7.0 to 8.0
Margarines,	0.8 to 3.0
Butter-fat + 10 per cent. cocoa-nut oil,	26.8
" 50 " " "	18.0
Butter-fat 50, cocoa-nut oil 22.5, margarine 27.5,	17.4

H. Kreis¹ has proposed to saponify by sulphuric acid; his method has been subsequently modified by several chemists, the most important addition to the process being the use of potassic permanganate. Dr. Rideal adds 10 c.c. of sulphuric acid, specific gravity 1.836, to 2.5 grms. of melted butter-fat; the butter dissolves. 100 c.c. of water are now added, and the sulphurous acid developed is destroyed by about 1 c.c. of strong potassium permanganate solution. The mixture is distilled until 80 c.c. of distillate are obtained. In nineteen different butters examined by the Reichert-Meissl process and the sulphuric acid process, the mean number of c.c. of decinormal alkali consumed was 28.8 for the former and 29.4 for the latter.

A process of saponification has been proposed and practised by Mr. West Knight,² which is based on the insolubility of the oleate, stearate, and palmitate of barium, and the ready solubility of the volatile fatty acid combinations with barium. The butter-fat is saponified with alcoholic potash in the ordinary way. The soap solution is diluted to 300 c.c., and a solution of chloride of barium added until a curdy precipitate separates, and

¹ *Chemiker Zeitung*, 1892, 16, 1394.

² *Analyst*, 1880.

the liquid is no longer rendered milky by a fresh addition—the insoluble barium fatty acids are collected on a filter, and ultimately transferred to a tube such as is used by Muter (p. 298), and the fatty acids liberated by sulphuric acid and shaken up with ether; when separation has been effected, a fractional part of the ether is taken, and evaporated in a tared flask.

Wm. Johnstone¹ has proposed a method of dealing with butter-fat which has the advantages of Koettstorfer's process, and gives the amount of soluble and insoluble fatty acids, 2.5 grms. of the butter-fat are saponified with a known quantity of alcoholic potash in a closed flask, the titre of the potash having been carefully ascertained.

After saponification the liquid is exactly neutralised, and thus—

A. The amount of alkali required to saturate all the acids of the butter is ascertained.

The alcohol is now boiled off and an excess of acid added to decompose the soap. The fatty acids are washed with hot water (as before described), the insoluble fatty acids collected on a filter and dried in air;² the filter is transferred to a Soxhlet tube, and its contents exhausted with dry ether, the ethereal solution being received in a weighed flask. When the extraction is complete the ether is driven off, the flask and its contents carefully dried and weighed; this gives—

B. Insoluble acids by weight.

Next the fatty acids remaining in the flask are saponified by a known volume of standard alkali and titrated with standard acid; this gives—

C. Amount of alkali required to saturate the insoluble acids.

By subtracting C from A, the amount of alkali required to saturate D, the soluble fatty acids, is obtained. An example will make the working clear.

2.5 grms. of butter were saponified by 25.00 c.c. of normal alcoholic potash; after saponification the liquid was neutralised by 14.78 c.c. of standard acid, therefore (A) the alkali was equal to 25.00 - 14.78 c.c.; that is, 10.22 c.c. of normal potash.

(B) The insoluble fatty acids weighed 2.2487 grms. = 89.95 per cent. The insoluble fatty acids were saponified by normal potash, the difference of acid taken was equal to 8.13 c.c. of potash—that is to say, the insoluble acids were neutralised by (C) 8.13 c.c. of normal potash; but the total acids consumed 10.22 c.c. of normal potash; therefore, the soluble acids (D) must be equal to 10.22 - 8.13 c.c., or 2.09, equal to 7.35 per cent. of butyric acid.

§ 214. *Further Analysis of the Insoluble Fatty Acids.*—The insoluble fatty acids are, as already stated, oleic, palmitic, and stearic; it is their total weight which is alone valuable, and to separate the three with accuracy is not easily effected. The first can, however, be isolated by the following process, the details of which have been worked out by Dr. Muter. The process depends upon the well-known fact that the oleate of lead, $Pb2C_{18}H_{32}O_2$, can be separated from plumbic palmitate, $Pb2C_{16}H_{31}O_2$, and plumbic stearate, $Pb2C_{18}H_{35}O_2$, by taking advantage of the solubility of the former in ether.

Muter's method of estimating oleic acid is as follows:—3 grms. of the fat are saponified by means of alcoholic potash. The potash is carefully

¹ *The Analyst*, xiv., 1889, No. 158.

² Richmond dissolves the fatty acids in warm alcohol, makes up to 100 c.c. with alcohol; of this he takes 25 c.c., and evaporates and weighs. He takes 50 c.c., and titrates with alcoholic soda solution—this gives both B and C as above.

neutralised by acetic acid, using phenol-phthalein as an indicator. 200 c.c. of water, to which 30 c.c. of a 10 per cent. solution of plumbic acetate have been added, are boiled and, while boiling, the soap solution is slowly poured in with constant stirring. The whole is allowed to cool, the supernatant fluid poured off, and the lead soap washed with hot water by decantation. The precipitate is transferred to a stoppered bottle, 80 c.c. of ether added, and, finally, the ether made up to 120 c.c. The bottle is



Fig. 38.—Muter's olein tube.

allowed to stand, with occasional shaking, for twelve hours, during which time the whole of the oleate will have dissolved. The ethereal solution is now filtered into a special tube (fig. 38), the plumbic stearate, etc., being washed with ether until free from lead; this usually entails the use of about 100 c.c. of ether. Hydrochloric acid, 25 per cent. strength, is poured in up to the first mark on the tube and the contents shaken. The liquids are now allowed to separate and the acid layer drawn off by means of the stop-cock. Water is then poured in and the contents again shaken, the water being removed as before, and the whole process repeated until the washings are free from acidity. The total volume of the ether is noted, a fractional portion run off into an Erlenmeyer's flask, and the ether distilled until a very little ether remains. It is not distilled completely, so as to avoid exposing the oleic acid to the air; 50 c.c. of pure alcohol are added to this residue and the solution titrated by decinormal soda, using phenolphthalein as an indicator; each c.c. of d. n. acid is equal to 0.0282 oleic acid. Having thus ascertained the strength of the remaining ethereal solution in the tube, the next step is to run off as many c.c. as correspond to 0.5 gm. of oleic acid into a stoppered bottle of at least 350 c.c. capacity; this flask is also provided with a doubly perforated cork carrying two tubes, one of which is connected with a carbon dioxide apparatus. The flask is placed in warm water and the gas passed through until all trace of ether has been removed. To the residue 50 c.c. of Hubl's solution (see p. 301) are added, the stopper inserted and allowed to stand the usual time and then titrated.¹

E. Twitchell² has proved that 100 c.c. of ether dissolves 0.015 of the lead salts of purified commercial stearic acid at 0° C., and that this is an objection to Muter's process.

He proposes a modification of Jean's process as follows, of which he gives the following example:—A lard, the liquid fatty acid of which gave when tested by Muter's process an iodine absorption of 94.1 per cent., was treated as follows:—4 grms. of the fatty acids were dissolved in 50 c.c. of 95 per cent. alcohol and 2.5 grms. of lead acetate in 20 c.c. of the same alcohol added, both solutions being hot. The liquid with precipitate was put on one side for two hours, the temperature being strictly 15° for the last hour. A part of the whole was then filtered into a separating funnel, treated with ether and HCl, the acids washed and dried in a current of CO₂, their iodine number obtained, and their percentage in the original solution determined. The precipitate was washed with 95 per cent. alcohol,

¹ *Analyst*, April, 1889.

² *Journ. Amer. Chem. Soc.*, 1895, xviii., 289-295; *Analyst*, July, 1895.

decomposed with HCl, and the solid fatty acids dried and weighed; the results were:—

	Per cent.	Iodine number.
Solid fatty acids,	46·24	4·9
Liquid fatty acids,	51·82	103·37

figures indicating that all the solid and part of the liquid acids had been precipitated.

If the iodine number of the total fatty acids is obtained, the method of calculating results is as in the following example:—4 grms. of fatty acids gave an iodine number of 62·57; the fatty acids from the filtrate at 15° gave an iodine number of 109·35, and were equal to 46·81 per cent.

The percentage of liquid acids multiplied by the iodine number of these acids and deducted from the iodine number of the total acids equals oleic acid. Dividing this by 0·9 gives the percentage of oleic acid precipitated with the solid acids. Thus in the above case $\frac{46\cdot81 \times 109\cdot35}{100} = 51\cdot19 - 62\cdot57 = 11\cdot38$. Dividing 11·38 by 0·9 = 12·64 in the precipitate, which, added to 46·81, represents the total liquid acids, 59·45.

Mr. Wanklyn¹ has proposed estimating butyric acid derived from the formation of butyric ether in saponifying with alcoholic potash as follows:—The butter is clarified in the usual way, and then 5 grms. are weighed and taken for the analysis. The butter is placed in a small retort of about 200 c.c. capacity, and fitted to a condenser. About 100 c.c. of alcohol (specific gravity 0·838) is added to the butter in the retort, and then 0·5 gm. of solid potash is added. The retort is then gently heated, and the contents are distilled, the distillation being continued to dryness. The distillate is received in a bottle fitted with a stopper, and containing 40 c.c. of accurately measured normal caustic potash or soda. When the distillation is complete, the stopper is placed in the bottle and the contents are shaken for a short time, and presently it will be found that the smell of butyric ether has vanished. Phenol-phthalein is now added to serve as an indicator, and the solution is titrated with normal sulphuric acid.

Good butters treated in this way yield from 2·8 to 3·4 per cent. of butyric acid as ether, while common margarine only a trace.

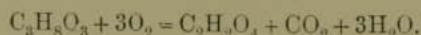
Haberland² has worked out a scheme of quantitatively separating acetic, butyric, propionic, and formic acids. The four acids are made free by phosphoric acid and distilled, the distillate evaporated with lead oxide, the residue dissolved in cold water, and the solution heated to boiling. Basic lead propionate is precipitated; the other lead salts remain in solution and are filtered off. The filtrate is treated with H₂SO₄, evaporated to dryness with excess of zinc oxide, and the mass is then treated with a large volume of absolute alcohol and filtered. Zinc formate and lead sulphate remain on the filter. The alcoholic filtrate is evaporated to dryness, and distilled with phosphoric acid. To the distillate silver carbonate is added, and the solution is evaporated, when, in consequence of the lower solubility of silver butyrate (1 part in 250 water), this salt separates with almost quantitative completeness. Silver acetate (solubility 1 part in 100 water) remains in solution.

§ 215. *Estimation of Glycerin.*—The determination of glycerin in fats, hitherto most unsatisfactory, can now, thanks to the labours of Wanklyn, Fox, Benedikt, and Zsigmondy, be accomplished with fair accuracy.

¹ "The Analysis of Butter," by W. Fox and J. A. Wanklyn. *Analyst*, 1884.

² *Zeit. anal. Chem.*, 1899, xxxviii., 217-226.

The process is based upon the oxidation of the glycerin by alkaline permanganate, the consequent formation of oxalic acid, carbon dioxide, and water; and the estimation of the oxalic acid; whence the glycerin is calculated according to the formula—



100 parts of oxalic acid are equal to 102.2 parts of glycerin, or, since the oxalic acid is usually determined as lime carbonate, 100 parts of lime carbonate equal 92 parts of glycerin.

The details of the process are as follows:—10 grms. of the filtered butter-fat are saponified by a known volume of a solution of KHO in pure methyl alcohol,¹ or, better still, by a strong aqueous solution of potash. Mr. Allen has recommended saponifying the fat by means of 25 c.c. of a 16 per cent. potash solution in a flask well stoppered down by means of an india-rubber stopper, heat being applied by means of a water-bath for several hours, until from the homogeneous appearance of the liquid it is certain that saponification has been effected. The soap is now decomposed in the usual way by means of dilute sulphuric acid; the fatty acids separated; the filtrate made up to a known bulk, and a fractional part of this taken for oxidation. The oxidation is carried out as follows:—The solution is alkalisied by potash solution containing at least 5 per cent. of free alkali, and then a 5 per cent. solution of potassic permanganate is added until the liquid is blackish in colour. The solution is boiled, whereupon manganese oxide is precipitated, but the excess still tinges the liquid red, the red colour is discharged by adding sulphurous acid, the whole is then filtered and the precipitate well washed with boiling water. The filtrate is boiled, and, while boiling, an excess of calcium acetate is added. The precipitate, consisting of an impure calcic oxalate, is collected, washed, dissolved in dilute sulphuric acid, and titrated with permanganate solution, or it may be ignited, and the resulting calcic carbonate dissolved in d. n. HCl, titrating back with a known volume of a solution of d. n. soda, and using methyl orange as an indicator. The best results are obtained when the amount of glycerin to be oxidised is from .3 to .5 grm., and the dilution not greater than .1 per cent.²

§ 216. *The Iodine Value.*—Hubl (*Dingl. polyt. Journ.*, ccliii., 281-295) has proposed a method which is of the greatest value in the examination of oils and fats generally, and can be applied to the analysis of butter. The melted fat is treated with an alcoholic solution of iodine in presence of an alcoholic solution of mercury bichloride; under these circumstances the *unsaturated* fatty acids or their glycerides absorb iodine in a regular and definite manner. With regard to butter, the only unsaturated fatty acid is oleic acid; hence the amount of iodine absorbed has relation to the content of olein or to oleic acid.

¹ Ethyl alcohol cannot be used, for on treatment with boiling alkaline permanganate, some oxalic acid is produced.

² See papers by Messrs. Fox & Wanklyn, *Chem. News*, Jan. 8, 1886; by R. Benedikt & R. Zsigmondy, *Analyst*, x., 205; and by Allen, *op. cit.*, xi., 52.

J. David, *Compt. Rend.*, xciv., 1477-1479, estimates glycerin as follows:—100 grms. of the fat are melted, 65 grms. of barium hydrate are added, with brisk stirring; when most of the water has been expelled the heating is discontinued; 80 c.c. of 95 per cent. alcohol are poured on the mass, and the whole stirred; 1 litre of water is then added, and the mixture boiled for an hour. The barium soap remains insoluble, whilst the glycerol is dissolved by the water, which is freed from the excess of barium, reduced in volume by boiling, and finally evaporated in a vacuum at a low temperature. The glycerin might evidently be estimated by the oxalic method.

The following solutions are required:—

(1) Solution of iodine and mercury bichloride. 25 grms. of iodine are dissolved in half a litre of alcohol of 95 per cent. strength, and 30 grms. of mercury bichloride are dissolved in another half litre of alcohol; the two solutions are now mixed, and allowed to stand, before standardising, for twenty-four hours.

(2) Solution of sodium hyposulphite, 24 grms. to the litre.

(3) 3·8747 grms. of potassium bichromate dissolved in a litre of water.

(4) A solution of potassium iodide, 10 per cent. strength.

The thiosulphate is standardised as follows:—10 c.c. of the potassium iodide solution are placed in a stoppered bottle, and 5 c.c. of HCl added, together with 20 c.c. of the bichromate solution. This will liberate exactly 0·2 gm. of iodine. The thiosulphate solution is run in carefully until a light straw colour only remains, then a little freshly-prepared starch solution is added, and the thiosulphate run in until the blue colour disappears. The number of cubic centimetres added will, of course, be equal to 0·2 gm. of iodine. The next thing is to titrate with the thiosulphate the solution of iodine and mercury bichloride. It is best to take for this purpose 25 c.c. of the iodine solution, and to operate in a similar way—that is, running in the thiosulphate until there is only a pale straw colour, and finishing with starch as an indicator.

To obtain the iodine value of butter-fat, from 0·6 to 0·8 gm. of the clear melted fat is dropped on to the bottom of a tared flask, the flask and its contents weighed. The fat is dissolved in 10 c.c. of chloroform, and 25 c.c. of the iodine solution run into the flask, which should be at least 500 c.c. in capacity; the flask is now stoppered and put in the dark for four hours. Should the iodine solution become decidedly pale at the end of two hours, a second 25 c.c. of iodine is run in, for unless an excess is present, accurate results will not be obtained. At the end of the stated time, 20 c.c. of the potassium iodide solution are added (or, should there be a red precipitate of mercury iodide, even more), the liquid diluted with from 300 to 500 c.c. of water, well shaken and titrated with the thiosulphate, using as an indicator starch solution. The difference in the number of cubic centimetres of thiosulphate used on the original iodine solution, and that on the solution which has acted on the fat, gives the requisite data from which to calculate the amount of iodine the fat has absorbed; this is calculated into per cent. of the fat.

Rowland Williams¹ has examined in this way thirty butters, the mean of which gave 35·34 per cent. of iodine absorbed, the extremes being 23·6 and 40·3. Seven margarines gave from 62·29 to 75·22 per cent.

The same process is applicable to the insoluble fatty acids, a weighed quantity from 0·2 to 0·4 of the fatty acids being dissolved in chloroform and treated as above. The following are a few iodine values for various fats and oils:—

	Iodine number.		Iodine number.
Linseed oil,	158·0	Tallow,	40·0
Castor oil,	84·4	Cocoa butter,	34·0
Olive oil,	82·8	Nutmeg butter,	31·0
Lard,	59·0	Butter-fat (from 26·0 to 35·1),	31·0
Margarine,	55·3	Cocoa-nut oil,	8·9
Palm oil,	51·5	Japanese wax,	4·2

¹ *Analyst*, June, 1894.

In fats, like butter-fat, in which it is believed there are no other unsaturated acids than oleic; olein, and the oleic acid can be estimated in this way.

The theoretical amount of iodine absorbed by oleic acid is 90.07 per cent., and the theoretical iodine value for olein is 86.20. Hence, if the iodine value of a fat equals 1, the percentage of olein will be found from the following formula—

$$\text{Olein} = \frac{100 \times 1}{86.2}$$

Or, simpler still, the iodine value multiplied by the factor 1.1601 gives the olein. Similarly the amount of oleic acid in the fatty acids is found by multiplying the iodine value found by the factor 1.1102.

Hehner¹ has proposed to treat fats with bromine, and weigh the product. 1 to 3 grms. of the fat are introduced into a wide-mouthed flask, and dissolved in a few cubic centimetres of chloroform; then pure bromine is added drop by drop until in excess. The flask is then heated in the water-bath, and the bromine driven off by adding from time to time a little chloroform. Finally the brominised fat is dried at 125° C., and weighed. To compare the results with those obtained with iodine, the gain in weight is multiplied by $\frac{127}{80}$ —that is, 1.587; the results fairly

agree.

§ 217. *The Cryoscopic Method of Examining Butter-Fat.*—Raoult's method of determining molecular weights is applicable to butter-fat, and may be used as a means of distinguishing it from margarine.² F. Garelli and L. Carcano, using benzene as a solvent, and considering the constant for benzene to be 53 (it is usually taken as 49), found that pure butter had a molecular weight of from 696 to 716, while margarines ranged from 780 to 883.³

The apparatus required is (1.) a Beckman's thermometer, D (fig. 39); this thermometer has a mercury reservoir at the top, *e*, so that variable amounts of mercury may be introduced into the lower bulb; it has a range of only 5 or 6 degrees divided into tenths; (2.) a tube with a side limb, A A'; and (3.) a wide test-tube into which the first one fits, so as to (as it were) jacket it with air.

The whole is supported in a wide beaker, in which the freezing mixture is placed, and suitable stirrers, C and *r*, provided. The first thing is to ascertain the freezing point of a known weight of the solvent. The tube with side limb is weighed on the balance, and then about 20 grms. of the solvent are introduced, and the tube with its

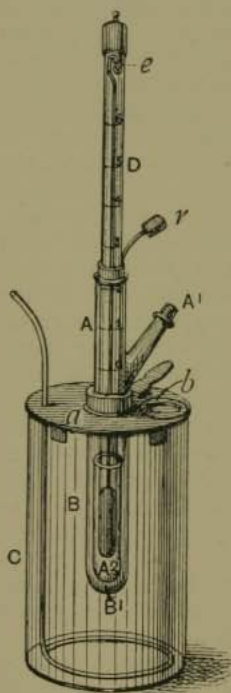


FIG. 39.—Cryoscopic Apparatus.

¹ *Analyst*, xx., p. 50.

² Pouret (*Bull. Soc. Chem.*, 1899, iii., xxi., 738-740), using benzene, found for pure butter values from 631 to 662 and for margarine 840 to 860. He also found 15 per cent. of margarine increased the molecular weight by 30-35 units.

³ *Analyst*, March, 1894.

contents again weighed; the weight subtracted from the tare gives the amount of liquid. Some ice and salt and water are placed in the beaker, and the whole apparatus arranged as in the diagram. The mercury of the thermometer gradually falls, and as it falls both stirrers must be worked; on approaching the freezing point the thermometer becomes for a moment stationary, then suddenly rises, and then becomes stationary to again fall; when it last becomes stationary is considered the freezing point of the liquid; the tube is taken out, the liquid allowed to melt, and the operation repeated once or twice, the mean of the determinations being taken. Next from .5 to .8 grm. of the butter, or other fat, is dissolved in the same liquid, and the operation again repeated; this time there will be a difference of one or more tenths of a degree in the freezing point; it will be lower than when operating on the pure substance; this lowering is the molecular depression. The molecular weight is calculated by means of a constant, which, for the chief solvents, has already been ascertained. The two solvents applicable for butter are benzene, for which the constant is usually taken as 49, and paraxylene,¹ which may be taken as 43. Of the two solvents, paraxylene is much to be preferred, for it has the low freezing point of about 16° C. and the high boiling point of 136° C.; hence, although expensive, it is readily recovered with but little loss.

The senior author has made several determinations of the cryoscopic value of butter-fat; but with paraxylene has obtained much lower values than Garelli and Carano. He is inclined to put the average molecular weight of butter-fat at about 580.

An actual example of a cryoscopic determination with paraxylene will illustrate the above remarks:—

Weight of paraxylene taken,	19.505 grms.
Weight of butter-fat dissolved in the above,	1.0784 „
Depression of freezing point,	0.41°.

If M equals the molecular weight, C the constant, G the per cent. of substance dissolved, and t the depression, then the molecular weight, M , is found from the equation—

$$M = \frac{G \cdot C}{t}$$

In the above example, replacing the letters by the experimental values—

$$\frac{5.528 \times 43}{.41} = 579.8$$

SUMMARY.

§ 218. From what has been said the analyst will have no difficulty in distinguishing normal genuine butter from margarine, nor will he have any difficulty in detecting the presence of large quantities of margarine in butter. Even small quantities of margarine in butter may be detected if the analyst has full information as to the breed of cow, the state of lactation, the season of the year, nature of feeding, etc., or if he is able to obtain a genuine sample of butter from the same source within a reasonably short space of time and under the same conditions. If, however, he has no such information, as, for instance, in the majority of samples taken

¹ Paterno and Montemartini, *Gazetta Chimica Italiana*, xxiv., ii., p. 197.

for analysis under the Sale of Food and Drugs Act, the problem he will have to solve is very difficult, as he will have to distinguish between adulterated samples and samples which are not adulterated, but which yield figures quite as low as do moderately adulterated samples.

Thus Allen found samples of genuine butter which gave a Reichert-Wollny figure of 22.5, and Vieth and others have found Reichert-Wollny figures of only 20.

Van Ryn¹ has recorded some exceedingly low figures obtained from Dutch butters, the Reichert-Wollny figure being in one case as low as 17.0. One of his samples gave the following series of abnormal figures:—

Sp. gr. at 37.8° C.	Reichert- Wollny.	Insol. acids.	Sol. acids.	Koett- storfer.	Iodine value.
.9105	19.2	91	3.8	212.9	46.6

Such figures as these cannot, however, be accepted as representing normal butters, and they are only obtained from butters made in the autumn in the northern countries of Europe, where it is the custom to leave the cows out late in the cold autumn. It has also been observed that a rise in the volatile acids begins shortly after such cows have been stabled. That the occurrence of these abnormal butters could be readily prevented by the proper management of the cows is shown by the fact that in the provinces of Limberg and North Brabant, where the cows are stabled over night practically during the whole year, the lowest Reichert-Wollny figure obtained by van Ryn was 28. An advanced state of lactation also tends to cause abnormal figures, as does disease, as has been observed by Nilson. Putting aside these very abnormal butters, the following table compiled from the results of various authorities may be taken as representing the composition of normal butter, and adulteration may be suspected if the figures obtained are outside the limits mentioned. Where no information as to the origin of the sample is obtainable the analyst will do well not to certify to adulteration even on such a low Reichert-Wollny figure as 20, unless this conclusion is supported by other figures, by a distinct colour reaction for vegetable oil, or by the microscopical examination.

TABLE GIVING AVERAGE VALUES FOR THE STANDARD TESTS.

	Butter.			Margarine.	Cocoa- nut oil.
	Mean.	Max.	Min.	Mean.	
Reichert-Wollny,	28	33.5	24	.8	7.5-8.0
Valenta,	36°	39°	29°	95°	...
Specific gravity at 100° C., 15.5	.8668	.8685	.8650	.861	.874
Refraction (Jean) at 45° C.,	-30	-34	-25	-15	-59
„ (Zeiss) at 35° C.,	46	49	43.7	52	41
Koettstorfer value,	227.0	233.0	221.5	195	268-246
Insoluble acids per cent.,	87.5	90	85.5	95.5	82-90
Soluble acids per cent.,	5.0	7.0	4.0	traces	...
Iodine value,	34	40	24	55	0.9.5
Melting-point,	32.8	35	30	26	20-28

Palm-nut oil like cocoa-nut, but an iodine value of 10-17.

¹ "On the Composition of Dutch Butter," by Dr. J. J. L. van Ryn, 1902.

From the figures given it will be seen that, although cocoa-nut oil has a comparatively high Reichert-Wolny figure, it has a high specific gravity, a high refraction, and a low iodine absorption. Vandam¹ has devised a process for the detection of cocoa-nut oil in butter, based upon the fact that it contains a large proportion of volatile acids which are insoluble in water, but soluble in rectified spirits.

Ceylon oil also has a rather high Reichert-Wolny figure (7.6); it can, however, be detected by its low refraction² (36).

§ 219. *The Analysis of Rancid Butter.*—According to Hanus,³ when butter goes rancid the glycerides of both the volatile and non-volatile acids are hydrolysed, the non-volatile being set free sooner and in larger proportion than the volatile. The glycerides of the saturated and unsaturated acids are equally affected. C. Amthor⁴ states that rancid butter contains alcohol and also free volatile acids, esters of the volatile acids, particularly butyric ethyl-ester. How far the analytical figures are affected by keeping butter is a matter of some doubt; according to C. A. Brown⁵ there is a decided increase in the acid, saponification and Reichert numbers, a slight increase in the ether number, and a marked increase in the iodine value, with a decrease in the soluble acids and glycerol. The experiments of other observers show, however, that the changes are only slight. Besana, analysing a number of butters kept for various times, only got a difference in the Reichert-Wolny figure in the first decimal place, some showing an increase, others a decrease. Richmond⁶ has shown that the changes in the analytical figures depend on whether the butter itself or the separated fat is kept. The volatile and soluble acids in the butter itself being increased, giving an apparent decrease in the filtered fat, he suggests that the volatile acids should be determined on the butter itself without separation of the fat, with subsequent determination of the actual fat taken.

CHEESE.

§ 220. Cheese may be made from the milk of any animal, but the great majority of cheeses in commerce are made from that of the cow. Rennet is added to the milk, which causes the casein to split up into two compounds, one of which is soluble and allied to albumen, the other⁷ in the presence of calcium phosphate being insoluble, this latter carries down with it most of the milk-fat, as well as some of the milk-sugar. The rennet brings about this change by the action of an enzyme (rennin), which acts only in an acid or neutral solution, and has an optimum temperature of about 41° C. The thin whey is allowed to run off, and the precipitated 'curds'⁸ submitted to pressure, which has the effect of not

¹ *Ann. Pharm.*, 1901, 201.

² W. G. Indemans, *Med. Tydschr. Pharm.*, xii., 306-310.

³ *Zeit. Nahr. Genussm.*, 1900, iii., 324-328.

⁴ *Zeit. analyt. Chem.*, 1899, 18.

⁵ *Journ. American Chem. Soc.*, 1899, xxi., 975-994.

⁶ *Dairy Chemistry*, 1899, pp. 293-296.

⁷ Various names have been given this compound, such as casein (*Halliburton*), tyrein (*M. Foster*), dys-chymo-caseose (*Richmond*).

⁸ An analysis of curds by M. Rubner is as follows:—

	Per cent.
Water,	60.27
Casein,	24.84
Fat,	7.33
Ash,	4.02
Milk-sugar,	3.54
Total solids,	39.73
'Solids not fat,'	32.40

only getting rid of the whey, but also of giving to the mass shape and consistency. Cheese may be made from sour milk without the addition of rennet, the lactic acid precipitating the casein; but most of the cheeses in commerce are made from fresh milk. Cheeses may be divided into two varieties—the soft and the hard; the former are manufactured by precipitating with rennet at a low temperature, and using but little pressure; they have mostly an alkaline reaction. The hard cheeses are subjected to a higher temperature and stronger pressure, and have, when first made, an acid reaction.

§ 221. *Soft Cheeses*.—Examples of soft cheeses are cream cheese, Neufchatel (a Swiss cream cheese), *fromage de Brie*, and Camembert.

Hard Cheeses.—Examples of hard cheeses are American, Cheddar, Stilton, Dunlop, Gloucester, and others.

The general composition of the chief cheeses of commerce may be gathered from the following table:—

TABLE XXII.—COMPOSITION OF CHEESE.

	Water.	Ash.	Fat.	Nitrogen.	Analysts.
American cheese, mean of six samples,	27.5	4.1	32.5	4.54	{ Chattaway, Pearmain, and Moor.
American cheese, mean of two samples,	27.2	4.1	32.1	5.80	{ A. Wynter Blyth.
Camembert, mean of two samples,	45.6	4.25	32.2	3.63	{ Chattaway, Pearmain, and Moor.
Cheddar (English), mean of three samples,	34.2	4.10	28.5	4.30	{ Do. Do.
Cheddar (English), mean of two samples,	28.1	3.34	22.5	7.3	{ A. Wynter Blyth.
Cheddar (Canadian),	33.3	3.60	30.6	4.3	{ Chattaway, Pearmain, and Moor.
Cheshire, mean of two samples,	34.7	4.30	33.3	4.1	{ Do. Do.
Cheshire, mean of six samples,	44.6	4.61	30.7	4.6	{ C. M. Blades.
Cream (double),	57.6	3.4	39.3	3.14	{ Chattaway, Pearmain, and Moor.
Cream (York),	63.1	1.4	6.5	2.76	{ Do. Do.
Dunlop,	38.5	3.8	31.8	4.1	{ Johnstone.
Dutch, mean of two samples,	39.6	6.4	11.5	4.8	{ Chattaway, Pearmain, and Moor.
Gloucester (double), mean of two samples,	35.2	4.8	25.8	4.7	{ Do. Do.
Gloucester,	35.8	4.2	21.9	6.2	{ Johnstone.
Gloucester (single),	21.4	4.1	25.4	7.7	{ A. Wynter Blyth.
Gruyère, mean of five samples,	34.7	3.8	28.9	4.9	{ Payen, Lindt, and C. Müller.
Gruyère, mean of two samples,	26.9	4.2	30.2	4.7	{ Chattaway, Pearmain, and Moor.
Fromage de Brie,	51.9	5.0	24.8	2.9	{ A. Wynter Blyth.
Neufchatel,	37.9	3.4	41.3	3.7	{ Do.
Parmesan,	32.5	6.2	17.1	6.9	{ Chattaway, Pearmain, and Moor.
Roquefort,	29.6	6.7	30.3	4.45	{ Do. Do.
Skim cheese,	43.1	6.2	0.9	7.7	{ A. Wynter Blyth.
Wensleydale,	28.3	3.7	38.3	4.3	{ Chattaway, Pearmain, and Moor.

§ 222. *Parmesan Cheese* is a peculiar cheese, never made in this country, but imported from Parma and elsewhere. The essential points in the manufacture are, that the rennet is heated to about 46°·6 C. (116° Fahr.), and an hour afterwards the milk set over a slow fire until heated to about 65°·5 C. (150° Fahr.). These operations cause the curd to separate in hard lumps. It is usually coloured with saffron. The outer crust of the cheese at the end of fourteen days is cut off, the new surface varnished with linseed oil, and one side coloured red. It is a very dry cheese, with a large amount of casein, and only a moderate percentage of fat.

§ 223. *The Ripening of Cheese.*—The complicated changes which take place in cheese during ripening are not fully understood. They appear to be brought about (1) by micro-organisms, (2) by enzymes, (3) by chemical action. During ripening, changes take place both in the proteids and in the fat.

The Cheese Proteid is decomposed into, first, albumoses and peptones, then into amines, and finally into ammonia. Freudenrich, Cohn, and others held the opinion that bacteria, especially the lactic class, were the chief agents in bringing about this decomposition. Chodat and Hofman Bang¹ have, however, shown that the lactic acid producing bacteria have no decomposing action on the cheese proteid even after it had been partly dissolved by *Tyrothrix*. The lactic acid bacteria undoubtedly play a preliminary and important part in the ripening of cheese, but the splitting up of the cheese proteid is chiefly brought about by the action of enzymes. These enzymes are, according to Babcock and Russell, contained in the original milk.

The Cheese Fat is at first affected to a very small extent, but subsequently the change is very great (*Duclaux*), the glycerides of the soluble fatty acids being split up into free fatty acids and glycerol, but subsequently the oleic acid glyceride is attacked. According to Karl Windisch,² these changes in the fat are brought about by micro-organisms, the ammonia produced by the decomposition of the proteids also acting on the glycerides of the fatty acids. The acids produced are chiefly non-volatile fatty acids.

The following example illustrates the changes taking place in the cheese proteid:—

		Per cent.
Cheddar, six months old, A. Vivian, ³	Insoluble nitrogen,	3·18
	Nitrogen as albumoses and peptones,	·22
	„ amides,	·86
	„ ammonia,	·09

§ 224. *The Analysis of Cheese.*—The chief difficulty in the analysis of cheese is in the extraction of the fat; one method, recommended and used by the American chemists, is to take a weighed quantity of cheese, mix it intimately with anhydrous copper sulphate, and then exhaust it in a Soxhlet tube with petroleum, ether, or other suitable fat solvent. A second method is to rub up 25 to 50 grms. of the cheese with sand or gypsum, and similarly exhaust with ether or petroleum. The fat in cheese may also be estimated by 'whirling.' Chattaway, Pearmain, and Moor operate as follows, using a Leffmann and Beam apparatus:—2 grms. of cheese are reduced to as fine a division as possible, transferred to a small dish, and heated in the water-bath with 30 c.c. of concentrated hydrochloric acid

¹ *Ann. Inst. Pasteur*, 1901, xv., 36-48.

² *Chem. Centr.*, 1901, i., 128-130.

³ *Wisconsin State Rep.*, 1899, 179-186

until dissolved. The mixture is poured into a Leffmann-Beam bottle, the dish rinsed with the hydrochloric acid and fusel-oil mixture into the bottle, and, finally, enough strong hot acid added to fill the bottle up to the mark. It is then 'whirled.' The Leffmann-Beam bottles are graduated, so that ten divisions equal 1 per cent. by weight of fat in the 15.55 grms. of milk taken; hence, the factor to be used is $\frac{15.55}{2} = 7.7$.

The Werner-Schmid method will also be found satisfactory.

The water in cheese is best estimated by extracting the powdered cheese by alcohol and ether, and drying the alcohol ether extract and the fat-free solids separately. This is a better method than drying the solids in the ordinary way, which will be found in many cases to be extremely tedious and inaccurate. With alkaline cheeses there appears no other way of obtaining the amount of water than careful neutralisation of the free alkali by an acid before drying. The sugar must be determined on the principles laid down at p. 224; the lactic acid and the ash on the principles laid down at pp. 230 and 228.

The proteids are best determined by subtracting all the other constituents from 100. The nitrogen may be determined by Kjeldahl's process, but the percentage of nitrogen contained in the organic nitrogen compounds of cheese varies from 15.8 in the original cheese proteid to 7.73 in tyrosine. Nitrates¹ are also said to be sometimes present.

If a further examination of the nitrogenous compounds contained in the cheese is required in order to judge of its ripeness, the following process may be employed:—

Total Nitrogen.—The nitrogen in 2 grms. of the cheese is estimated by Kjeldahl's method.

Nitrogen as Ammonia.—5 grms. of the cheese, well ground up with clean sand, are mixed with 200 c.c. of water, barium carbonate is added and the whole distilled into standard acid.

Nitrogen as insoluble proteids, Albumoses, Peptones, and Amides.—50 grms. of the cheese are well ground up with sand, mixed with dilute acetic acid, boiled and filtered; the filtrate is divided into two parts; the total nitrogen is determined in an aliquot portion of one part; the percentage of nitrogen found deducted from the total nitrogen gives the nitrogen as *insoluble proteids*. To the other portion rendered acid with dilute sulphuric acid (1:4), phosphotungstic acid is added until no more precipitate is formed; the liquid is filtered, and the nitrogen determined in an aliquot portion of the filtrate; the nitrogen found represents that due to *amides* and *ammonia*. This deducted from the nitrogen in the first portion gives the nitrogen as *albumoses* and *peptones*.

§ 225. *Adulterations.*—The chief adulteration is the incorporation of animal fat—this is effected as follows:²—“An emulsion of lard is made by bringing together in a disintegrator lard and skimmed milk, both previously heated to 140° Fahr. in steam-jacketed tanks; the disintegrator consists of a cylinder revolving within a cylindrical shell; the surface of the cylinder is covered with fine serrated projections, each one of which is a tooth with a sharp point; as this cylinder revolves rapidly within its shell the mixture of melted lard and hot skimmed milk is forced up in the

¹ Leffmann and Beam, “Select Methods of Food Analysis,” 1901, p. 240.

² U.S. Dep. of Agriculture, *Bull.*, No. 13, 1887, “Foods and Food Adulteration,” H. W. Wiley.

narrow interspace and the lard becomes very finely divided and most intimately mixed or emulsionised with the milk. This emulsion consists of from two to three parts of milk to one of lard; it can be made at one factory and taken to another to be used for cheese, but it is usually run at once into the cheese vat.

"In making the cheese a quantity of this emulsion, containing about 80 lbs. of lard, is added to 6000 lbs. of skimmed milk and about 600 lbs. of butter milk in the cheese vat, and the lard that does not remain incorporated with the milk or curd (usually about 10 lbs.) is carefully skimmed off. These quantities of materials yield from 500 to 600 lbs. of cheese, containing about 70 lbs. of lard, or about 14 per cent. About half of the fat removed in the skimming of milk is replaced by lard."

Hence, the fat extracted from cheese should always be tested by the Valenta test and by the Reichert-Wollny process, and if a suspicious result is obtained, it must then be analysed in the same way as butter. In such a case it is best to extract the fat from 100 to 150 grms. of the cheese, so as to have sufficient for the various methods described under 'Butter.'

According to G. Pescetti and F. Ghigi¹ in genuine cheeses the Reichert-Wollny number exceeds 18. Margarine cheeses give values below 15 and generally below 6. The refractometer value (Zeiss) of genuine cheese lies below 47.5 at 35° C., and for margarine cheese above 48°, but these fall during ripening, as does the Reichert-Wollny figure and to a less extent the saponification number. The iodine value² first decreases and then increases during ripening.

Skim Milk Cheese.—Other adulterations of cheese which have actually been found are not numerous. All mineral adulterations, save those of volatile metals, must be looked for in the ash, which consists normally of common salt, alkaline, and earthy phosphates. Cheese has from the earliest to the present time been coloured by vegetable matters, and so long as the latter are not injurious, such addition cannot be considered as adulteration.

Thin slices of cheese should be examined microscopically after dissolving out the fat, etc., by ether; in this way starches and vegetable substances may be detected. Arsenical washes and lead pastes have often been applied to the rind to prevent the attacks of the fly. As this part is habitually eaten by a few people, it is necessary to examine it, especially for these metals, and, in a complete investigation, to make two separate analyses, one of the rind, and the other of the substance of the cheese.

In past times, a few isolated instances have occurred in which it was found that the manufacturers of cheese had mixed preparations of arsenic with the cheese itself as a preservative:—e.g., such was the case in the year 1841, when several of the inhabitants of Chatillon were poisoned by this means. In 1854, the same thing occurred, and a Parisian family suffered, but not fatally (*Chevallier*). It is to be hoped that such ignorance is a thing entirely of the past.

Tyrotrotoxin, diazobenzol $C_6H_5N_2(OH)$, has been discovered by Vaughan ("Report of Michigan State Board of Health," 1886) in a cheese which caused the illness of some 300 people in Michigan. The crystalline substance was isolated from a watery extract of the cheese by the process detailed on pp. 504–505 of *Poisons*, by A. Wynter Blyth. 3rd edition.

Whey.—After the removal of the curd from milk, a 'whey' remains, containing

¹ *Chem. Centr.*, 1901, i., 573, 574.

² Karl Windisch (*Chem. Centr.*, 1901, i., 128–130).

proteids, milk-sugar, lactic acid, and salts. The mean of thirty-two analyses collected by König gives the following as the average composition of 'whey':—

Water,	93.31
Nitrogenous substances,82
Fat,24
Milk-sugar,	4.65
Lactic acid,33
Salts,65

Whey, on account of its high content of milk-sugar, is used for the preparation of the latter, and there is also a 'whey-vinegar.' In very many places, however, whey is used merely as a food for pigs.

LARD.

§ 226. Lard is, from an analyst's point of view, the fat of the pig. Lard used to be defined as the fat from particular parts of the pig—that is, the solid fat from around the kidneys, and from the peritoneum; but the enormous American industry in lard has thrown on the market pig-fat from all portions of the animal, and the term 'lard' cannot properly be restricted to fat from particular parts. On the other hand, 'leaf-lard' should always be fat from what the butchers call the 'leaf'—that is, the peritoneum.

The American packing trade, according to Wiley,¹ 'render' the following varieties:—

1. *Neutral Lard*.—Fat derived from the leaf of the pig rendered in a fresh state at a temperature of from 40° to 50° C.; this lard is not much exported, but is mainly used up in the manufacture of oleo-margarine. It is almost neutral.

2. *Choice Lard, Choice Kettle-rendered Lard*.—This is lard made from leaf and trimmings only. It is rendered in steam-jacketed open kettles, hence its name.

3. *Prime Steam Lard*.—This is apparently pig-fat rendered by steam from all or any portions of the animal.

4. *A low quality lard* made from the 'guts,' by which term the hog packers mean the whole of the abdominal viscera.

It is obvious, therefore, that different lards will show analytical variations. Spaeth has made some useful observations on the chief analytical and physical distinctions of lard derived from various portions of eight animals. His results are as follows:—

	European Lards.			American Lards.		
	Fat from the Back.	Fat from the Kidney.	Fat from the Leaf.	Fat from the Head.	Fat from the Back.	Fat from the Leaf.
Specific gravity at 100° (water at 15°=1),	0.8607	0.8590	0.8588	.8632	.8616	.8626
Melting-point of fat,	33° 8'	43° 2' C.	44° 5' C.
" of fatty acids,	40° 0' "	43° 2' "	42° 9' "
Iodine value of fat,	60.58	52.60	53.10	65.9	63.8	61.4
" of fatty acids,	61.90	54.20	54.40
Free fatty acids, KOH per 100 grms.	0.54	0.58	1.28
" " calc. as oleic acid,	0.152	0.163	0.360

¹ U.S. Dep. of Agriculture, "Foods and Food Adulteration," by H. W. Wiley.

§ 227. *The Physical Characteristics of Lard.*—Lard possesses a pure white colour and a granular texture. The specific gravity of lard at 40° C. is about 0.898; at 100° C. about 0.861—that is, in each case, water at 15° C. being taken as 1. The increase in density for each degree C. is .00062. The melting-point of lard is from 34° to 48° C.; Wiley, for American lards, considers a wide deviation from 40° C. as a sign of impurity. The solidifying point of butcher's lard is 28° to 29° C. American steam lard from 25° to 27° C.

A better test is the solidifying point of the fatty acids; this, in the prime steam lards, varies, according to Wiley, from 41° to 43° C.; in other lards from 36° to 46° C.; these numbers agree fairly well with those found by Gladding.

The refraction of lard has been studied by Wiley, using an Abbe's refractometer. The rate of variation for each degree of temperature was found to be 0.000288, and the refractive index varies inversely as the temperature. The refractive indices were taken at various temperatures between 30° and 40° C., and then reduced to 25° C.; at that temperature water had a refractive index of 1.3300, and the mean number for pure lard was 1.4620. Zeiss' butyro-refractometer or Jean's oleo-refractometer will be found preferable to Abbe's instrument. Cotton-seed oil may be detected with certainty by this means; it may also be used as a corroborative test in the case of beef tallow. The following table gives some of the figures obtained:—

	Degrees of Oleo-Refractometer. Jean.	Butyro-Refractometer at 40° C. Mansfeld.
European lard, . . .	- 12.5	48.6 to 51.2
„ stearin, . . .	- 10 to - 11	...
American lard, . . .	- 4 to - 11.5	51.4
Beef tallow, . . .	- 16 to - 17	49.0
„ stearin, . . .	- 34	...
Cotton-seed oil, . . .	+ 12 to + 23	61.0
„ stearin, . . .	+ 25	...
Cocoa-nut oil, . . .	- 54	35.5

Microscopical Appearances of Lard.—If forty drops of melted lard are dissolved in 10 c.c. of ether and allowed to cool, crystals are soon deposited, which have the appearance of oblong plates with oblique terminals; these occasionally occur in radiated groups.

Beef-fat, on the other hand, always forms more or less radiated groups of crystals, and the individual crystals are in the form of needles, not plates (see *Frontispiece*). Some of the crystals are of a wavy *f* shape.

This test must not be taken as absolute proof of the presence of beef stearin, as Hehner and Mitchell have shown¹ that the form of the crystals depends entirely upon the proportion of stearic acid contained in the sample. Hehner has more recently² shown that in pure lards containing a high percentage of stearic acid the crystals often cannot be distinguished from those obtained from lard largely admixed with beef-fat.

§ 228. *Chemical Characteristics of Lard—Moisture in Lard.*—Lard consists of the glycerides of palmitic, stearic, and oleic acids, and also a

¹ *The Analyst*, vol. xxi., p. 328.

² *The Analyst*, vol. xxvii., p. 248.

small amount of linolin. Hehner has obtained stearic acid in quantities varying from 6 to no less than 24.91 per cent. The quantity of water in lard is fractional; it should never exceed 0.7 per cent. An amount of water equal to 1 per cent. would be an adulteration.

Insoluble Fatty Acids.—These vary from 93 to 95 per cent.

Volatile Acids.—The volatile acids are always small in quantity. If a Reichert-Wollny test be applied and 5 grms. be saponified, decomposed, and distilled, the distillate does not neutralise more than 0.5 c.c. of d. n. soda. Should the amount of volatile acid be more than this, it is a suspicious sign.

Saponification Value.—This has been stated by Koettstorfer to be equal to 195.6 mgrms. of KHO, by Valenta as from 195.3 to 196.6, and by Wiley as varying from 193.4 up to 203.1 with a mean value of 198.

Iodine Number.—This is a value depending on the amount of olein in the lard; it rises above the normal when lard is adulterated with oils of larger olein content and sinks below the normal when lard is mixed with stearins, having a small iodine absorption. Mixtures can be made, however, which will pass the iodine test.

Unfortunately the normal iodine number varies much in the fat of the pig derived from different parts of the animal; compare, for instance, the iodine values given at p. 310. Wiley, for prime steam American lards, gives the value as from 60.34 to 66.47 per cent. of iodine absorbed, with a mean value of 62.5; Hubl gives a mean number of 59; Dieterich from 49.9 to 63.8. So that all that the analyst can say is that a number below about 49 or higher than 64 is a suspicious sign. Hehner has obtained from a lard, melted out by himself, a value of only 45.6.

Another method is to separate the insoluble fatty acids, and as in Muter and de Koningh's process (already described, p. 297), determine the iodine number of the liquid fatty acids. Pure lard, according to Muter, yields a liquid fatty acid having an iodine number of 94; cotton-seed oil, on the other hand, has an absorption under the same conditions of 136; so that if the iodine number is above 94 adulteration with vegetable oil is indicated. Muter's iodine number of 94 must not be considered final, for Wallenstein and Finck¹ have made some researches on the liquid fatty acids of European lards, and give the iodine number as from 93 to 96, and the iodine number of the liquid fatty acids of American lards as high as from 103 to 106.

The Maumené Test.—This is the rise of temperature when a definite quantity of an oil is mixed with a definite quantity of strong sulphuric acid. The drying oils examined in this way give a much higher temperature than the non-drying oils. Comparable results are obtained by operating on strictly the same conditions. These conditions are to use the same volumes or weights of oil and acid, to bring the two to the same initial temperature before mixing, and to have the same strength of acid. This latter may, however, not be essential if Thomson and Ballantyne's suggestion be adopted of referring the rise of temperature obtained with 50 grms. of oil and 10 c.c. of sulphuric acid, to the rise of temperature which 50 grms. of water give under the same conditions; the rise of temperature with the oil, divided by the rise of temperature with the water, gives a quotient which they have named "specific temperature reaction." This in some degree meets the difficulty of testing with different strengths of acids. The best method of bringing about a temperature reaction after Maumené's method is as follows:—

¹ *Chem. Zeit.*, 1894, 1189.

Sulphuric acid of 97 per cent. strength is poured into dry stoppered bottles of small capacity, and all save one securely protected by a lute from the air. The analyst can now get his standard from the small bottle in use, and this standard will hold good for all the rest.

To apply the principle to lard, 50 grms. of the lard are melted in a small flask and kept at a temperature some 2° or 3° C. above the melting-point, by standing in a water-bath kept at a constant temperature. The small bottle of sulphuric acid is also placed in the same water-bath, and when both acid and melted lard are equal in temperature, 10 c.c. of sulphuric acid are transferred to the oil at the same moment that the flask is immersed in a beaker and packed with cotton wool; the mixture is then well stirred with the thermometer and the rise of temperature noted. Such various degrees of heat have been stated that it would be unwise to rely upon any published statement, but a standard should be made by operating on pure lard; under these circumstances good comparative results will be obtained. For example, Engler and Rupp found a lard giving a rise of temperature of 31° to 32° C.; the same lard with 10 per cent. of cotton-seed oil gave a reaction of 34° C.; with 20 per cent. cotton seed oil, 40° to 42° C.; and with 50 per cent. seed oil, 58° C. In all cases, indeed, the same operator will find a difference between the temperature reactions of pure lard and lard mixed with oils of high temperature reactions.

Hehner and Mitchell¹ have proposed to treat the fat with bromine and ascertain the rise of temperature. 1 grm. of the oil or fatty acid is dissolved in 10 c.c. of chloroform in a test tube enclosed in a vacuum jacket; 1 c.c. of bromine is added, and the rise in temperature noted by a delicate thermometer. The bromine, oil, and solvent must all be at the same temperature before mixing.

The rise of temperature of 10 samples of lard gave a mean number of 10°·7 C.; a sample of lard with 10 per cent. cotton oil, 11°·6 C.; cotton oil, 19°·4 C.; cod-liver oil, 28° C.; olive oil, 15° C.; butter, 7° C.; by multiplying the number of degrees by the factor 5·5 a close approximation to the iodine number is obtained. Thus the process checks a Hübl determination.

§ 229. *Adulteration of Lard.*—The chief sophistication of lard is the admixture of beef stearin, either alone or with vegetable oils, especially cotton seed oil, cocoa-nut oil, sesamé oil, or maize oil. Cotton-seed stearin has also been used, either with beef stearin or alone. Water is generally enumerated as an adulterant, but the wilful addition of water at the present time is rare.

The adulteration of lard is on a large scale, and great ingenuity has been shown in devising mixtures which will pass the analytical tests. Hence the analyst, when he examines a suspicious sample, must never condemn on one test, but should ascertain what are the chief lard constants before he pronounces an opinion.

The chief analytical values of the different adulterants of lard are tabulated in Table XXIII., from which the following facts will be apparent:—

(a.) *Specific Gravity.*—All the adulterants save beef stearin raise the specific gravity of lard; beef stearin is, on the other hand, about the specific gravity of lard.

(b.) *The Solidifying- or Freezing-Point of the Fat.*—Cotton-seed oil, arachis oil, sesamé oil, maize oil, and even cocoa-nut oil, have all lower

¹ *Analyst*, July, 1895.

figures for the freezing-point than lard; on the other hand, beef stearin is somewhat higher, so that a 10 per cent. admixture of beef stearin will have but little effect on the freezing-point of the mixed fat. Cotton-seed stearin also may have the same freezing-point as lard.

(c.) *The Freezing- or Solidifying-Point of the Fatty Acids.*—The same remarks apply to the solidifying-point of the fatty acids, but here another valuable distinction comes in—viz., the very great difference between the freezing-point of some of the oils and fats and the freezing-point of their fatty acids; for example, pure lard giving a solidifying-point of 25° to 29° C., the fatty acids are on an average 10° to 15° C. higher; on the other hand, cotton-seed oil as compared with the fatty acids will show 30° of difference, while maize oil and cocoa-nut oil will differ but to a slight, often inappreciable, extent.

TABLE XXIII.—CHEMICAL AND PHYSICAL CONSTANTS OF LARDS AND SOME OILS USED FOR THE ADULTERATION OF LARD.

	Specific Gravity. Water=1.		Solidifying Point.		Insoluble Acids. Helmert Val.	Saponifi- cation. Mgrms. KOH.	Iodine Value.	Maumené Thermal Test.
	At 15°.	At 100°.	Fat.	Fatty Acids.				
LARD,861	25°-29°	37°-47°	94.0	196	59.0	24°-35
Cotton-seed oil,922-.930	.867-.868	0° to -1°	31°-36°	96.0	191-196	100-117	75°-90°
Arachis oil,916-.922	.864-.869	-3° to -7°	24°-30°	96.0	190-196	87-100	44°-67°
Sesamé oil,921-.924	.868-.871	-4° to -6°	18°-22°	96.0	188-191	103-112	63°-64°
Maize oil,921-.922	.868-.869	-10° to -15°	14°-16°	96.0	189-190	111-119	79°-86°
Cocoa-nut oil,923	.873	16°-20°	16°-20°	88.0	246-268	8.9	(Cocoa-nut olein 26°-27° (Allen))
Beef stearin (tallow),943-.952	.860	33° to 48°	43°-0	96.0	193-200	35-40	...
Cotton-seed stearin,923	.865-.867	16°-32°	35°-0	96.0	195	89-93	48

N.B.—None of the above (save cocoa-nut oil) give more than sufficient volatile acid to neutralise 0.5 c.c. of d. n. soda when submitted to the Reichert-Wollny test. Cocoa-nut oil, however, gives a definite Reichert-Wollny value of about 7 c.c. of d. n. soda for 5 grms. of the fat.

(d.) *The Insoluble Acids.*—These acids, save perhaps in the case of cocoa-nut oil, are of little value to determine, all the oils and fats in the table (save cocoa-nut oil) yielding approximately the same amount of fatty acids.

(e.) *Volatile Acids.*—A Reichert-Wollny determination of the volatile acids becomes of great importance when cocoa-nut oil is suspected. A somewhat low content of insoluble acid and an appreciable amount of volatile acid, such as sufficient to neutralise over 2 c.c. of decinormal soda, points strongly to some fat like cocoa-nut oil, containing soluble and volatile fatty acids.

(f.) *The Saponification Value.*—The only oil in the table that is likely to raise materially the saponification value is cocoa-nut oil.

(g.) *Iodine Value.*—All the oils, with the exception of cocoa-nut oil, raise the iodine value; on the other hand, the stearins generally depress it.

(h.) *Maumené Test.*—Mixtures of either cotton-seed oil, arachis oil, sesamé or maize oil, raise the thermal indications.

Having obtained some indication of adulteration from a comparison

with the constants above enumerated, the results will have to be confirmed, or otherwise, by special tests, as follows:—

Special Tests for Vegetable Oils.—As lard is wholly animal, and as there are some analytical differences between animal and vegetable fats or oils, the sample may be tested as follows:—

Process for Detection of Phytosterol.—Salkowski¹ was the first to show that animal fats contained cholesterol only, and vegetable fats phytosterol. His original process has been modified and improved by A. Bömer² and others.

50 grms. of the fat or oil are saponified by means of alcoholic potash. The soap is heated in a beaker for about an hour on the water bath, then transferred to a large separating vessel provided with stopcock and stopper. 200 c.c. of water are added, and the liquid, when cool, shaken up with 500 c.c. of ether. The ether is separated from the soap, which is again treated with 200 to 250 c.c. of ether. The ether is recovered by distillation. The ether extract is dried and obtained alcohol free by heating on the water bath and again saponified by alcoholic potash, and the process repeated on a small scale, as above. The ultimate product is dissolved in hot alcohol (absolute) and allowed to crystallise. Cholesterol usually crystallises in rhombic plates, phytosterol in fine needles. In cases of adulteration, however, both cholesterol and phytosterol will be present. Cholesterol melts at 148°·4 to 150°·8, phytosterol from 138°·0 to 143°·8 C. A mixture of cholesterol and phytosterol (1:3) melts at 135°–136°, while one part of phytosterol to three parts of cholesterol melts at 144° to 144°·5; other mixtures follow more closely the calculated mean melting points of mixtures.

Besides the microscopical appearances and the melting point there are a few special tests for cholesterol. The crystals, dissolved in acetic acid, and after the addition of a small lump of zinc chloride, warmed with acetyl chloride, give a red colour with green fluorescence (L. Tschugaern).³

A. Bömer now prefers to change the ultimate crystals into their acetic esters. The dry crystals are heated with acetic anhydride in a covered watch glass for a few seconds to boiling, the unused anhydride got rid of by evaporating over the water bath, and crystallisation effected from absolute alcohol. The acetic ester of cholesterol melts at from 114°·3 to 114°·8 C., phytosterol at 125°·6 to 137°·0 C.

Bömer has worked out the melting points of the esters of artificial mixtures in per cent. of the plant oil or fat added as follows:—

117–118°,	1 to 2 per cent.	vegetable oil added.
120–121°,	2 to 3	„
123–125°,	3 to 4	„

Welman's Colour Test.—1 grm. of the melted lard is dissolved in 5 c.c. of chloroform in a test tube, and agitated with 2 c.c. of a freshly prepared solution of phospho-molybdic acid, or of sodium phospho-molybdate and a few drops of nitric acid. After standing for a short time the chloroform layer remains colourless; the upper layer, in the case of animal fats or oils generally (cod-liver oil excepted) also remains colourless; but if the lard is mixed with a vegetable oil, or if it is rancid, a green colour appears in the upper layer. On alkalisng with ammonia, the green colour changes into blue. This colour test must not be too much relied upon, but it is useful as a preliminary test.

¹ *Zeitschrift f. anal. Chemie*, xxvi. 557.

² *Zeitschrift f. Untersuchung d. Nahrungs- u. Genussmittel*, i. 21, 81, 532, 559.

³ *Ibid.*, iv. 974.

Linoleic Acid.—This acid is stated by Benedikt and Hazura not to be contained in lard. It is a constant constituent of vegetable oils. The method of detection is based upon oxidising it into sativic acid, a crystalline acid insoluble in cold water, ether, benzene, and carbon disulphide. On the other hand, Fahrion states that he has obtained sativic acid from pure lard; therefore there is at present some doubt in the matter. Hence, until the point is definitely settled, attempts to separate sativic acid will not be of great utility.

Special Tests for Cotton-seed Oil, Sesamé Oil, Arachis Oil, and Maize Oil.—These are fully detailed in the article on Olive Oil, to which the reader is referred. It must be remembered that lard made from animals fed on cotton-seed cake, etc., may give faint indications of cotton-seed oil.

Detection of Beef Stearin.—By far the best method is one worked out by Stock, who prepares two sets of mixtures. The one mixture consists of lard melting at from 34° to 35° C., mixed with 5, 10, 15, and 20 per cent. of beef-fat—melting-point, 56° C. A second mixture is prepared, also of pure lard, but with a higher melting-point—viz., 39° to 40° C.—and mixed with similar proportions of beef-fat, the beef-fat having a lower melting-point—viz., 50° C. The melting-point of the sample under examination is taken by the capillary tube method, and, according as to whether it has a high or a low melting-point, the first or the second series of comparison mixtures is used. 3 c.c. of the melted fat are run into a graduated stoppered tube of 25 c.c. capacity; 21 c.c. of ether are added, and the fat dissolved at a temperature of 20° to 25° C.; 3 c.c. of each of one of the sets of mixtures are treated in precisely the same way. The five cylinders are cooled down to 13° C., and kept approximately at that temperature or a little lower over-night. The volume of crystals in each case is noted, and an approximate estimate in this way obtained. The ether is poured off as completely as possible, and 10 c.c. of ether added at 13° C. The cylinders are again shaken, and allowed, as before, to deposit. Finally the content of crystals is weighed, and this weight compared with the weight of the crystals of known composition. Stock found that with regard to lards melting below 39° C., no sample yielded (under the above conditions) more than 11 mgrms. Samples of higher melting-points, such as 45·8 C., might, however, yield as much as 146 mgrms. The crystals are carefully examined by the microscope. In the case of mixtures, intermediate characters between the crystals of pure lard (*Frontispiece*) and beef fat (*Frontispiece*) will be obtained, and in a good many cases the chief crystalline form will be that of beef crystals. The process, when combined with chemical data supporting the suspicion of admixture, is without doubt of great value, but requires experience.

Lardérine.—There are some mixtures in the market sold honestly as such under various fancy names; one examined by Mr. Bodmer gave the following figures:—

Melting-point,	38°·0 C.
Specific gravity at 38°·5,	0·9105 (= 0·8724 at 100°).
Reichert test 2·5 grms.,	1·00 c.c. d. n. baryta.
Temperature reaction (Maumené),	43° C.
Iodine absorption,	65·6 per cent.
Saponification figure,	201·8 mgrms. KHO.
Saponification equivalent,	277·7.
Nitrate of silver,	Much reduction.
Crystallisation from ether,	Beef crystals.
Ether deposit (Stock's process),	37 mgrms.
Oleo-refractometer,	6°.

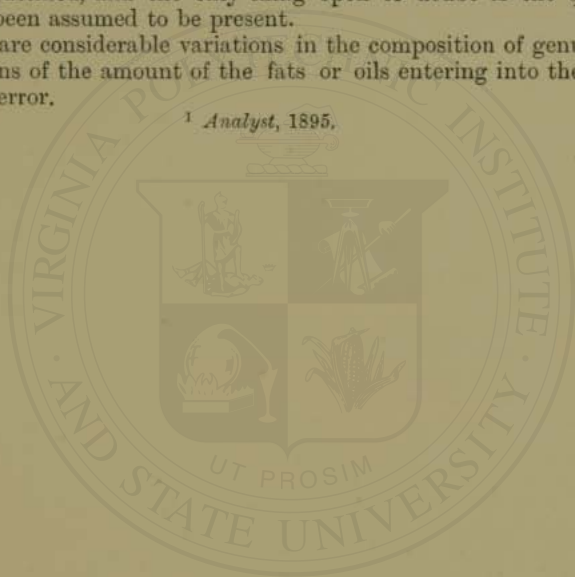
Mr Bodmer considered the sample to probably contain 10 per cent. beef stearin, 25 cocoa-fat, 45 cotton-seed oil, 20 per cent. lard.¹

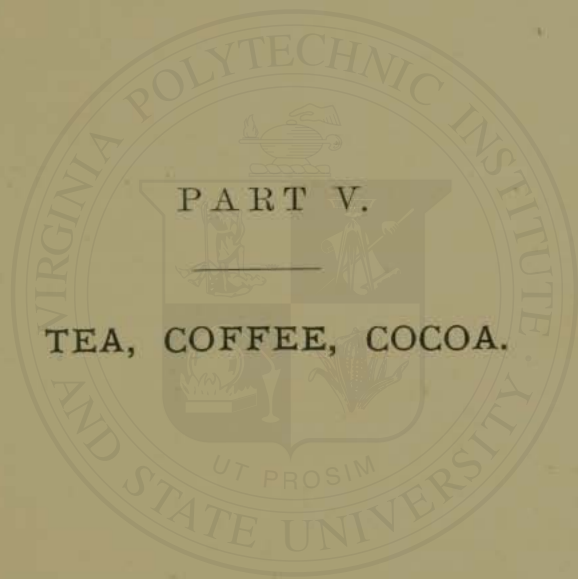
Since the above is a typical case of adulterated lard, it may be useful to examine the figures in detail. First, the melting-point affords no indication of impurity. The specific gravity is higher than usual, and is suspicious. The Reichert test gives decided indications of more volatile acid than in lard, and in itself points clearly to cocoa oil; the saponification figure is fairly high, but not beyond genuine lards.

The iodine number, although not very high, is yet higher than in the majority of genuine lards; Bechi's nitrate of silver gave a decided reaction indicative of cotton-seed oil; lastly, beef stearin crystals were discovered, and the ether deposit weighed 37 mgrms., which for a lard of 38° melting-point, according to Stock, is never found in genuine samples. Hence the diagnosis of a mixture of beef stearin, cotton-seed oil, cocoa oil, and lard is perfectly justified, and the only thing open to doubt is the quantities which have been assumed to be present.

As there are considerable variations in the composition of genuine lard, all calculations of the amount of the fats or oils entering into the mixture are liable to error.

¹ *Analyst*, 1895.





PART V.

TEA, COFFEE, COCOA.

PART V.—TEA, COFFEE, COCOA.

TEA.

§ 230. *Varieties of Tea.*—Tea is the dried leaf of different species of *Thea*, a section of the genus *Camellia*. The botanical varieties do not appear to be numerous. *Thea Bohea*, *T. viridis*, and *T. sinensis*, all Chinese plants, *Thea Assamica*, indigenous to Assam, and one or two hybrids, are the principal plants from which the numerous teas of commerce are derived. The difference depends on the selection of young or old leaves, and special treatments in drying and otherwise preparing the leaf, rather than on essential botanical variation.

The varieties of tea imported into this country are extremely numerous; but seldom does any one of them reach the consumer unmixed, for the wholesale tea merchants carefully improve their teas by 'blending.' The most common sorts are—Gunpowder, Hyson, Congou, Capers, and Indian tea. Of these, the Gunpowder and Hyson are dried at a higher temperature than the others, and contain less hygroscopic moisture. The Capers may be generally told by the leaves being rolled up into little lumps with starch or gum; as a class, they are much adulterated, and, in fact, can hardly be called genuine tea.

Besides these, there are a number of special teas, some of a very high price, and imported in a state of great purity; but such teas are used almost entirely for mixing or blending.¹ They are known under the names of Moyone, Moyone gunpowder, Oolong, Mannuna Kaisow, scented Pekoes,

¹ In Cooper's travels ("Travels of a Pioneer of Commerce," by T. T. Cooper, London, 1871), there is an interesting account of the brick tea used in Thibet, some of which may find its way to Russia, but none, probably, to this country. It is the staple produce of the city and district of Yatow, and the tree from which it is prepared attains not unfrequently the height of 15 feet. It has a large coarse leaf, and is cultivated with little care, growing along the borders of fields and homesteads. There are three qualities of the tea: the first is gathered in June and July, when the leaf is about an inch long. It is spread out in the sun to dry slightly, then rolled in the hand until the sap begins to exude, and when in this state made up into balls, and laid on one side to ferment. After fermentation, it is pressed into wooden moulds, and dried by charcoal fires. The bricks on their removal from the moulds are enveloped in yellow paper covers, bearing a Government stamp and the trade mark of the exporter, and then they are packed in baskets four feet long, made of the thin strips of the bamboo. The bricks thus packed form a 'packet of tea,' and weigh about 20 lbs.; at Lhassa, this quality of tea sells for 4s. 8d. per lb. The second variety consists of the older and yellower leaves which, when exported to Lethang and Bithang, sell for 1s. 6d. per lb. The third variety is merely chopped twigs stuck together by rice water, and is only used in the neighbourhood of Ta-tsian-too, where it is sold at 9d. per lb.

Indian Souchong, Assam, Java, etc. The names by which the teas of commerce are most familiar to the public are simply 'green' and 'black,' which differ merely in accordance with the method of preparation followed. Green tea is prepared from young leaves, which are roasted over a wood fire within an hour or two after being gathered. The black tea leaves, on the other hand, are allowed to lie in heaps for ten or twelve hours after they have been plucked, during which time they undergo a sort of fermentation; the leaves then pass through certain processes, and are slowly dried over charcoal fires.

§ 231. *Structure of the Tea Leaf.*—The border is serrated nearly, though not quite, up to the stalk (see fig. 40). The primary veins run out from the midrib almost to the border, and then turn in, so that a distinct space is left between them and the border.



TEA PLANT

FIG. 40.

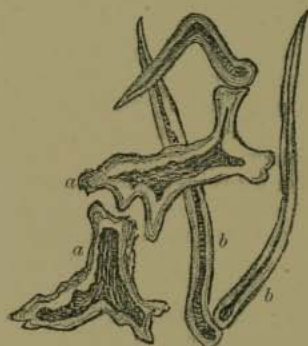


FIG. 41.

A section through a tea-leaf is difficult to make successfully; but when this is accomplished, it will be seen that the upper and lower epidermis are very similar in thickness and structure, both being formed of a single row of oblong thick-walled cells, the upper epidermal layer being also covered with a cuticle.

The mesophyll, or parenchyma of the leaf, contains two kinds of cells, the one being a very regular single or double row, filled with chlorophyll, just beneath the upper epidermal layer, whilst a spongy parenchyma, containing large spaces, occupies the rest of the leaf-thickness. There are also cells containing crystals, and, lastly, there are the peculiar structures called 'idioblasts' (see *a*, fig. 41).

Idioblasts are long, tough, tenacious, branched cells, which seem to act as pillars, or beams, keeping the two layers of the leaf apart; they do not occur in any other leaf with which the tea-leaf is likely to be confused, so that their presence would indicate tea, their absence would point to foreign leaves. A very convenient method of detecting idioblasts is given by

Moeller¹:—Small fragments of the leaf are warmed in a very strong solution of caustic potash, and then placed under the thin covering glass and pressed firmly (see fig. 41).

The surface-view of the leaf is different on the two sides; the upper surface consists of small (.05 mm.) cells, with wavy crumpled contours; the under surface has larger cells (.07 mm.) of irregular wavy outline, and studded over with numerous stomata, formed of two reniform cells, of an average length of .00075 inch and breadth of .000588 inch.

The hairs of tea are very characteristic, but not constantly found; they disappear from old leaves, but the under-surface of young leaves is always thickly beset with them. The hairs are 1 mm. long and about .015 mm. broad (see *b*, fig. 41).

§ 232. *Chemical Composition of Tea.*—The constituents of tea are—*Essential oil, theine, boheic acid, quercetin, tannin, querci-tannic acid, gallic acid, oxalic acid, gum, chlorophyll, resin, wax, albuminous, woody, and colouring matters, and ash.* The essential oil of tea varies from 0.6 to 1 per cent. It is citron-yellow, lighter than water, has a strong odour of the tea plant, solidifies easily by cold, and resinifies on exposure to air.²

Theine, Caffeine, $C_8H_{10}N_4O_2$ $\left(\begin{array}{c} CH_2N - CO \\ CO \quad C - N(CH_3) \backslash \\ CH_3N - C - N = CH \end{array} \right)$.—A crystalline substance identical with 1, 3, 7—trimethylxanthin—and therefore belonging to the uric acid group of organic compounds; it can be synthetically

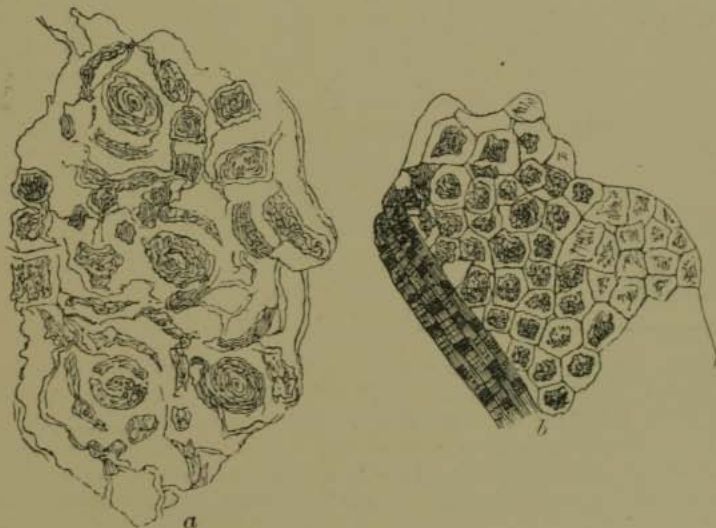


FIG. 42.—Epidermis of Tea Leaf, $\times 300$. *a*, under, *b*, upper surface.

formed by replacing in a methyl group the H of the NH of theophyllin. It was first separated in an impure condition by Runge, from coffee berries,

¹ *Mikroskopie der Nahrungs- u. Genussmittel.* Berlin, 1886.

² A fixed oil, serving many purposes in China, is extracted from the seeds of the tea plant. It is citron-yellow, specific gravity 0.927, and is composed of one part of stearin and one of olein.

in the year 1820. It was found by Corput and Stenhouse also to be a constituent of the leaves of the coffee tree. Oudry in 1827, finding it in various species of tea, named it 'Theine,' and Oudry's 'Theine' Mulder and Jobst showed, in 1838, to be identical with caffeine. The alkaloid has also been discovered in guarana, maté, and the kola nut, by Martius, Stenhouse, and Attfield respectively.

Theine crystallises from an aqueous solution with 1 molecule of water; from ether, in an anhydrous state. It sublimes in minute dots at $78^{\circ}8$ C., in crystals at $79^{\circ}4$ C. (175° Fahr.), and above.¹ The sublimate consists of microscopic needles: that which is first produced is of extremely fine, light elements; after a little time, at such temperatures as 120° , the crystals become longer and larger. The melting-point of theine is somewhere between 177° and 228° C. Theine possesses a slightly bitter taste, but is without odour. According to a recent research, the solubility of theine in different solvents is as shown in Table XXIV.

TABLE XXIV.—SOLUBILITY OF THEINE.

	100 grms. of Solvent at 15° to 17° dissolved of Theine.		Coefficient of Solubility at 15° to 17°.		100 grms. of Solvent at the boiling point dissolved of Theine.		Coefficient of Solubility at the boiling point.	
	Hydrated.	Anhydrous.	Hydrated.	Anhydrous.	Hydrated.	Anhydrous.	Hydrated.	Anhydrous.
Chloroform.	...	12.97	...	1/7.72	...	19.02	...	1/5.25
Alcohol of 85 per cent.,	2.51	2.30	1/40	1/44.4
Water,	1.47	1.35	1/68	1/74.2	49.73	45.55	1/2.01	1/2.19
Absolute alcohol,	...	0.61	...	1/164.7	...	3.12	...	1/32
Commercial ether,	0.21	0.19	1/476	1/526	...	0.454	...	1/220
Carbon bisulphide,	...	0.0585	...	1/1709
Purified anhydrous ether,	...	0.0437	...	1/2288	...	0.36	...	1/277
Light petroleum,	...	0.025	...	1/4000

Theine forms numerous salts of definite composition—the hydrochlorates, $C_8H_{10}N_4O_2$, HCl and $C_8H_{10}N_4O_2$, 2HCl; a platinum compound, $C_8H_{10}N_4O_2$, HCl, $PtCl_2$; a chloride of gold compound, $C_8H_{10}N_4O_2$, HCl, $AuCl_3$; a chloride of mercury compound, $C_8H_{10}N_4O_2$, 2HgCl; an argentic nitrate compound, $C_8H_{10}N_4O_2$, $AgNO_3$, and many others.² Some of these, such as the silver compound, separated from a concentrated watery

¹ The subliming point given by Pelouze, 178° C., and by Mulder, 184.7° C., must have been obtained by extremely faulty methods. If a little theine is placed between two watch-glasses on the water-bath, the almost instantaneous rise of crystals to the upper glass will at all events show that the subliming point is below 100° C. See "Temperature at which the Alkaloids Sublime," by A. Wynter Blyth, *Journal of Chemical Society*, 1878.

² The salts with hydrochloric, hydrobromic, and sulphuric acids are decomposed by water. The sulphate crystallises with difficulty, the hydrobromide well. Theine forms compounds with sodium acetate, citrate, sulphate, chloride, benzoate, cinnamate, and salicylate. Theine gives no precipitate with potassium mercuric iodide, and it has a neutral reaction. See Tanret., *J. Pharm. Chim.* [5], v., 591-595.

solution, and the mercury compound, almost insoluble in ether, and capable of being dried at 100° C., might possibly be of use in the estimation of theine.

Theine is, in large doses, a poison. Frerichs, C. J. Lehmann, Husemann, and others, have made themselves the subject of experiment. Lehmann, after taking .5 grm., suffered from frequency of the pulse, irritation of the bladder, cerebral excitement, slight hallucinations, and, lastly, desire to sleep. Husemann took .25 grm. with somewhat similar symptoms. Pratt, with subcutaneous injections of from .12 to .8 grm., suffered from symptoms rather different from the foregoing; .3 grm. lessened the pulse and caused sleeplessness; .4 to .5 grm. quickened the pulse, and caused a desire for frequent micturition, but no dilation of the pupil; .8 grm. caused great uneasiness and anxiety, trembling of the hands and arms, so that he was unable to write, and later a restless sleep, with continual dreaming. In opposition again to all these statements, is that of the late Mr. Cooley,¹ who is said to have taken 20 grains (1.29 grms.) of pure theine every day for a month without experiencing any other symptom than some slight elevation of spirits. According to Strauch, the least fatal dose for cats is .25 grm., a quantity which killed a cat in thirty-five minutes. In all experiments on animals there has been increased frequency of the heart's action, and repeated emptying of the bladder and intestine. No case of poisoning in the human subject appears to be on record. When given to animals it has been chemically separated from the blood, urine, and bile.

Tests for Theine.—Concentrated sulphuric and nitric acids dissolve theine in the cold without the production of colour. If the alkaloid is treated with fuming nitric acid, and evaporated to dryness, the reddish-yellow residue becomes, when moistened with ammonia, of a splendid purple-red colour. If a solution of theine be evaporated with chlorine water in a watch-glass, a red-brown residue is obtained, which on cooling, and exposure to the vapour of strong ammonia, becomes purple-violet. The chief precipitants of theine are—phospho-molybdic acid, yellow precipitate; iodine with potassic iodide, dirty brown precipitate; chloride of platinum, yellow hair-like crystals, insoluble in cold hydrochloric acid, slowly separating; chlorides of gold, mercury, and nitrate of silver also give precipitates.

Boheic Acid, $C_7H_{10}O_6$, was first separated by Rochleder in 1847,² from the leaves of *Thea sinensis*. The hot watery decoction of tea is precipitated whilst boiling by sugar of lead, filtered, the filtrate neutralised by ammonia, the resulting precipitate collected, suspended in absolute alcohol, and freed from lead by SH_2 ; the filtrate from the lead precipitate is evaporated to dryness in a vacuum, and purified by re-solution in water, etc. It is a pale yellow amorphous powder, melting at 100° C. into a tenacious mass, and decomposing at common temperatures if exposed to the air. It is soluble in all proportions in water and alcohol, is coloured brown (but not precipitated) by chloride of iron, and forms for the most part amorphous salts insoluble in water.

Quercitannic Acid, $C_{33}H_{30}O_{17}$, first discovered by Chevreul and Brandt in the *Quercus tinctoria*, and stated by Hlasiwetz to be in tea leaves, can be crystallised from an aqueous solution. It forms sulphur or chrome-yellow microscopic tables, containing 3 molecules of water, part of which is

¹ Vide Cooley's "Dictionary of Practical Recipes," Art. Caffeine.

² Rochleder: *Ann. Chem. Pharm.*, lxxiii., 202.

expelled at 100°, the rest at from 165° to 200° C. Its reaction is neutral, and it is without odour, but has a marked bitter taste when in solution. It melts at from 160° to 200° C. to a resinous, amorphous mass. Its solubility is as follows:—Cold water 2485, boiling 143; cold absolute alcohol 23·3, boiling 3·9; ether dissolves it slightly, warm acetic acid copiously. Sugar of lead precipitates almost completely; the precipitate is soluble in acetic acid.

Quercetin, first obtained by Rigaud, 1854, from the splitting up of quercitanic acid, is, according to Filhol, to be found in the green leaves and flowers of all plants. Its formula is given as $C_{15}H_{10}O_7$; it forms fine yellow needles, or a citron-yellow powder, which gives up at a temperature of 120° C., 7 to 10 per cent. of water of crystallisation. It melts, according to Zwenger and Dronke, above 250° C. without decomposition, solidifying again in a crystalline mass, and it may be also sublimed with only partial carbonisation. It is very little soluble in water. Warm acetic acid dissolves it copiously, but it separates on cooling. It is soluble in 229·2 parts of cold, and 18·2 parts of hot absolute alcohol. A solution of quercetin colours linen bright yellow; sugar of lead precipitates the alcoholic solution cherry red, and chloride of iron dark red. A combination with sodium or potassium can be obtained, $Na_2O, C_{15}H_{10}O_7$.

The other constituents of tea, such as gallic and tannic acids, gum, etc., are too well known to need description.

COMPOSITION OF TEA.

§ 233. The mean of sixteen analyses quoted by König gives the following:—

	Per cent.
Water,	11·49
Nitrogenous substances,	21·22
Theine,	1·35
Ethereal oil,	·67
Fat, chlorophyll, wax,	3·62
Gum and dextrin,	7·13
Tannin,	12·36
Other nitrogen free matters,	16·75
Woody fibre,	20·30
Ash,	5·11
	100·00

And this may be taken to represent average tea. The annexed table gives determinations of several constituents of teas in Russian commerce by Dragendorff—the chief features of which may be summarised as follows:—

Dragendorff's Analyses of Twenty-three Teas in Russian Commerce.

	Mean. Per cent.	Maximum. Per cent.	Minimum. Per cent.
Water,	10·00	12·66	7·10
Extract,	32·67	44·50	24·80
Theine,	1·90	3·09	1·36
Tannin (4 determinations only),	11·42	9·42	12·70
Ash,	6·23	6·82	5·23

An interesting research on the changes taking place in the tea-plant through age has been made by O. Kellner.¹ Tea leaves from the same plants were collected twice a month from May to November, and a sample also obtained at the end of the twelve months. The main results may be briefly summarised thus:—The water regularly diminished from 76 to 60 per cent.; the theine in the young leaves amounting to 2.85 per cent. of the *dried* substance diminished to .84 per cent. of the old dried leaves, and the total nitrogen from 4.91 to 2.67 per cent. The nitrogen from the amido-acids, equal in the first month to .66 per cent. of the dried substance, rapidly decreased, so that in September the amido-nitrogen only attained .08 per cent., and in the twelfth month was still less—viz., .01 per cent.; since the theine did not diminish in anything like the same proportion, the inference is that the amido-nitrogen became theine-nitrogen. On the other hand, the tannin increased from 8 to 11 or 12 per cent., and the ash from 4.69 to 5.14 per cent. It is, therefore, clear that in young tea leaves there is more water, more theine, and more amido-acid than in old leaves. Whereas in old leaves, with the decrease of the constituents mentioned, there is more ash and tannin.

MICROSCOPICAL METHODS OF DETECTING ADULTERATIONS IN TEA.

Preliminary Examination of Tea.—The tea leaves should be soaked in hot water, carefully unrolled, and their shape and structure examined. Sections of leaves can be made by placing them between two pieces of cork, and cutting fine slices off both the cork and the enclosed leaf; on floating the sections in water, the film of cork may be readily separated from the leaf. The epidermis of the lower or upper surface can, with a little practice, be detached in small portions by the aid of a sharp razor, and examined in water, glycerin, or dammar balsam under the microscope. Its structure has been already detailed.

§ 234. *Process for the Examination of Leaves and Vegetable Tissues generally under the Microscope.*—One of the authors has suggested an easy process for examining vegetable leaves. A portion of a leaf is enclosed between two of the thin circles of glass used by all microscopists, and a weight having been placed upon the upper glass, the portion of leaf thus enclosed is heated with a strongly alkaline solution of permanganate of potash. The action begins at once, and the substance under examination must be examined from time to time to see that the oxidation does not proceed too far. Alkaline permanganate attacks the colouring matters and the contents of the cells first, and afterwards the cell membranes; the object of this treatment is to make the leaf transparent, and yet to preserve its structure. Tea leaves are very opaque, and it is impossible without some mechanical or chemical treatment to render them transparent.² When from the appearance of the leaf-fragment the oxidation is considered sufficient, it is removed, washed in water, and treated with a little strong hydrochloric acid, which at once dissolves the manganese oxide that has been precipitated on the leaf, and leaves the latter as a translucent white membrane, in which the details of structure can be readily made out—tea-leaf in this way being quite different in appearance from other leaves. A second method of very great value is to place a fragment of a leaf between

¹ *Land. Versuchs. Stat.*, 1886, 370-380. *Journ. Chem. Soc.*, Jan. 1887, 73.

² See also method to discover 'idioblasts,' described at page 323.

two circles¹ of glass, weight the upper one with a silver coin, and burn on a bit of sheet platinum the leaf thus prepared. Since it is impossible for the ash to curl up and become disarranged, a complete skeleton of siliceous ash remains, which may be called *the skeleton ash*.

These skeleton ashes of leaves (so far as the authors have hitherto been able to investigate the subject) show such decided differences the one from the other, that a great number of leaves may with a little practice be recognised by this method alone. [It is particularly useful in detecting the adulterations of tobacco, the skeleton-ash of the tobacco-leaf being special and peculiar.] It is sometimes well not to burn to an absolute ash, but to leave little bits of partially-consumed carbon, forming objects for the microscope of great beauty. To preserve the 'skeleton-ash' the two circles may be cemented together, or the edges may be fused by the flame.

Figs. 43, 44, 45 and 46 are examples of *skeleton ashes*, as drawn on the block to scale.



FIG. 43.—Ash of Tea Leaf, $\times 170$.



FIG. 44.—Ash of Sloe Leaf, $\times 29$.



FIG. 45.—Ash of Lime Leaf, $\times 29$.



FIG. 46.—Ash of Tobacco Leaf (Cigar), $\times 29$.

§ 235. *Chemical Method for the Detection of Foreign Leaves in Tea.*—A chemical method for the detection of foreign leaves (adulterants) was first described by one of us in June 1877.² It is based upon two facts—first, that every part of a *theine*-producing plant—wood, stem, leaf, flowers, and even hairs—contains theine; and, secondly, that this can be readily sublimed. The leaf, or fragment of a leaf, is boiled for a minute in a watch-glass with a very little water, a portion of burnt magnesia equal in bulk is added, and the whole heated to boiling, and

¹ That is, the covers of thin glass used by the microscopist.

² "Micro-Chemistry as applied to the Identification of Tea Leaves." By A. Wynter Blyth. *Analyst*, 1877.

rapidly evaporated down to a large-sized drop. This drop is transferred to the 'subliming cell,' described fully in 'Poisons,'¹ and if no crystalline sublimate be obtained, when heated up to 110° C. (a temperature far above the subliming point of theine), the fragment cannot be that of a tea-plant. On the other hand, if a sublimate of theine be obtained, it is not conclusive evidence of the presence of a tea-leaf, since other plants of the *camellia* tribe also contain theine.

Finally, there is a negative test which may occasionally be valuable. All fragments of tea hitherto examined contain manganese, and there are a few foreign leaves in which manganese is constantly absent. Hence, if a leaf be burnt to an ash, and a fragment of the ash be taken up on a soda-bead to which a little potassic nitrate has been added, the absence of the green manganate of soda would be sufficient evidence that the leaf had not been derived from the tea-plant, while conversely, as in the case of theine, the presence of manganese is not conclusive of tea.

Another portion of the tea leaves should be thoroughly bruised, spread on a glass plate, and carefully searched with a magnet for ferruginous particles—the so-called iron-filings, which are occasionally found, especially in *Capers* and certain species of *Congou*. It is almost unnecessary to state that the black, irregular masses found in tea, and attracted by a magnet, are not metallic iron.² Their chemical composition is somewhat variable; they all contain magnetic oxide of iron, and many of them in addition phosphate of iron, titanate of iron, quartz, and mica, with a little sand. They are, without doubt, sometimes an adulteration (the senior author has himself found over 1 per cent.), and sometimes an impurity, for in a few teas mere traces only of this ferruginous sand may be discovered. Any particles of the kind extracted by the magnet should be collected and treated with hot water, which soon disintegrates them; the adherent tea-dust is separated, and the sand dried and weighed.

To detect facing, the tea in its dried state should be mounted as an opaque object.³ If it has the appearance of being heavily faced, soaking in warm water will soon detach the film; and indigo, Prussian blue, or similar substances will sink to the bottom, and may be collected and examined. Indigo may be identified by the microscope. Prussian blue may be tested for by warming the deposit with caustic alkali, filtering acidifying the filtrate with hydrochloric acid, filtering again if necessary, and testing the filtrate with ferric chloride. The residue left after treatment with caustic alkali may be tested for magnesium silicate, by first extracting with HCl, and then collecting the insoluble residue, and fusing it with an alkaline carbonate. The silica is now separated in the usual way by evaporation with HCl to dryness, subsequent solution of the remaining ash in weak acid, and filtration; any lime is removed by

¹ *Poisons: their Effects and Detection*. By A. Wynter Blyth. 3rd ed., 1895, p. 258.

² Mr. Allen appears to have found metallic iron in tea. The test for metallic iron is, that nitric acid, 1·2 specific gravity, dissolves it with the production of red fumes; it also precipitates metallic copper, if added to an acidulated solution of cupric sulphate.

³ The facing of tea is thus described by M. S. Julien: "The leaves are mixed either with powdered indigo, with powdered plaster, or with slaked lime, sometimes even all three substances being put together in small proportion to tea leaves. These matters are introduced into the basins at the commencement of the operation, when the leaves begin to be covered with a light dew under the influence of heat. These matters attach themselves to the leaves, and communicate to them the bluish-green characteristic of green tea. . . . In certain manufactories Prussian blue is used instead of indigo." *Industries Anciennes et Modernes de l'Empire Chinois*, par MM. Stanislaus Julien et O. Champion. Paris, 1869.

ammonia and ammoniac oxalate; and lastly, magnesia is precipitated as ammon. mag. phosphate. Magnesia found under these circumstances must have been present as steatite or other magnesian silicate.

LEAVES USED, OR SUPPOSED TO BE USED, AS ADULTERANTS.

§ 236. The following is a brief description of the principal leaves supposed to be used as adulterants :—

Beech (*Fagus sylvatica*).—The leaves of the beech are ovate, glabrous, obscurely dentate, ciliate at the edges, the veins running parallel to one another right to the edge. The leaf, slightly magnified, is seen to be divided into quadrilateral spaces by a network of transparent cells. On section, the parenchyma of the leaf is found to consist of an upper layer of longitudinal cells, and a lower of loose cellular tissue, enclosed between the epidermis of the upper and under surface. The whole section is thus divided into oblong spaces by transparent cells connecting the cuticle of the upper and lower surfaces. The epidermis of both the upper and lower surfaces is composed of cells with an extremely sinuous outline (see fig. 47).

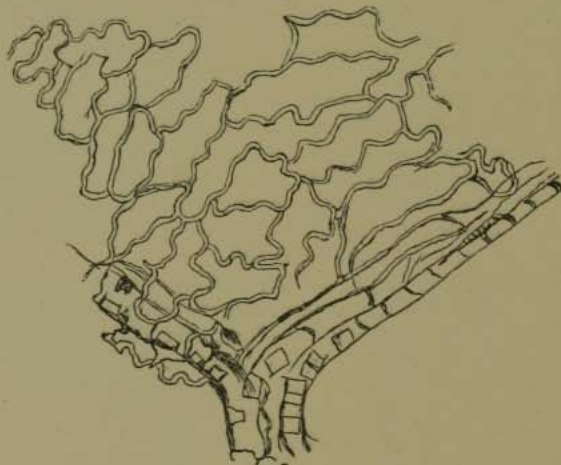


FIG. 47.—Epidermis of Beech Leaf, $\times 300$.

The stomata are small, not numerous, and almost round. Beech leaves contain manganese.

Hawthorn (*Crataegus oxyacantha*).—At least two varieties, the more common of which is the *C. monogyna*, with obovate three- to four-deeply lobed leaves, with the lobes acute. The leaf is divided into quadrilateral spaces, like the beech and many other leaves, by a transparent network. The epidermis of the upper surface is composed of a layer of thin-walled cells, generally quadrilateral, outline seldom sinuous. The epidermis of the lower surface has a layer of thin-walled cells, with a very sinuous outline. Stomata large, distinct, and numerous, in many instances nearly round, but the shape mostly oval. (See fig. 48.)

Camellia Sasanqua.—The leaves of *Camellia sasanqua* are oval, obscurely serrate (the younger leaves entire), dark green, glabrous, of somewhat leathery consistence; the lateral veins of the leaf are inconspicuous.

Micro-structure.—The parenchyma of the leaf is placed between two thickened epidermal layers; the epidermis of the upper surface, as seen upon a section, forms a wrinkled, continuous, thick membrane, in which a cellular structure is not very evident. Below this there are two or three layers of large cells, more or less oblong, with their long diameter at right

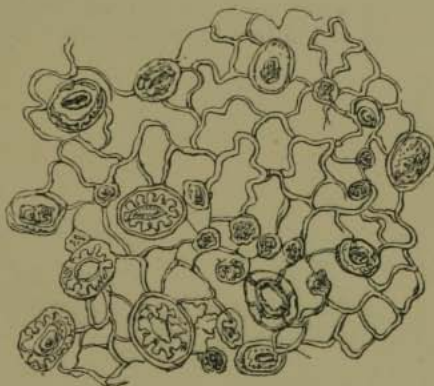


FIG. 48.—Epidermis from the under surface of the Hawthorn Leaf, $\times 300$.

angles to the surface of the leaf; and underneath this again is a loose network of cells, resting upon an epidermis in every respect similar to that of the upper surface, but only half as thick. A thin layer of either the upper or lower epidermis shows a peculiar dotted or reticulated appearance,

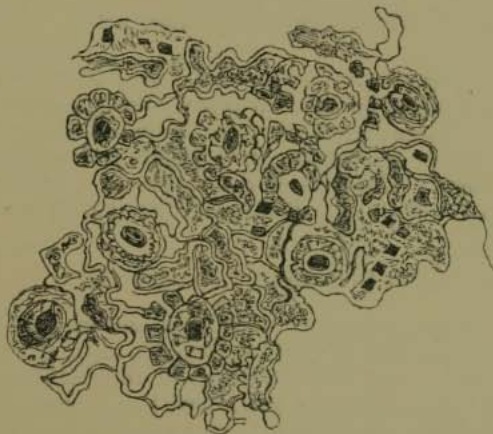


FIG. 49.—Epidermis of under surface of the leaf of the *Camellia Sassanqua*, $\times 300$.

not unlike the rugæ of a stomach. The lower epidermis is studded with frequent stomata, small, and of an oblong shape (see fig. 49).

Sloe (*Prunus communis*).—The leaves of the common sloe are rather small, elliptic or ovate-lanceolate in shape, and slightly downy beneath. The sectional thickness of the leaf is the same as that of tea. The stomata

on the lower surface are scanty. The microscopical appearances are wholly different from those of tea leaves, more especially as seen in section. (See fig. 50.)

Chloranthus Inconspicuus.—The leaves of the *Chloranthus inconspicuus* are long, oval, serrate, wrinkled, the veins running nearly to the edge, and there forming a network in such a manner, that at the point of intersection little knots are formed, which give the margin of the leaf a very rough feeling. The structure of the leaf is very simple. The epidermis of the



FIG. 50.—Section of Sloe Leaf, $\times 300$.

upper surface is formed of one or two layers of thin-walled cells, the epidermis of the lower of one or two layers also of cells, and between the two there is a parenchyma of loose cellular tissue. The stomata are oval and rather numerous; their length is from $\cdot 0010$ inch, their breadth $\cdot 00073$ inch. The cells of the epidermis are large, some of them $\cdot 005$ inch or more in their long diameter.¹ (See fig. 51.)

THE CHEMICAL ANALYSIS OF TEA.

The sample is next submitted to chemical analysis. If the question to be decided is simply that of adulteration, the taste of the infusion, the percentage of extract, and a determination of the chief constituents of the ash are in most cases all that is necessary; but a more or less complete examination embraces a quantitative estimation of hygroscopic moisture, theine, total nitrogen, tannin, extract, gum, and ash.

§ 237. *Hygroscopic Moisture*.—The ordinary method of taking the hygroscopic moisture of tea is to powder as finely as possible an indeter-

¹ The leaves of *Epilobium angustifolium* (common willow herb) are said to be extensively used in Russia for the adulteration of tea. The dried leaves are sold for from four to six roubles a pound, and are used by the poorer classes in the place of tea. Alcohol produces in infusions of epilobium a precipitate of mucilage.—*Pharm. Zeitsch. für Russland* and *Year-Book of Pharmacy*, 1876.

minate quantity of from 1 to 2 grms., and to heat it in a watch-glass over the water-bath until it ceases to lose weight. It should be finally weighed between two watch-glasses, since it rapidly absorbs moisture from the air.

The method given is in its results incorrect, since some volatile oil and a small proportion of theine are always volatilised. That theine is actually lost is capable of rigid demonstration; it is only necessary to heat a few leaves of tea between two watch-glasses over the water-bath, and theine crystals can be readily discovered on the upper glass. To devise a process

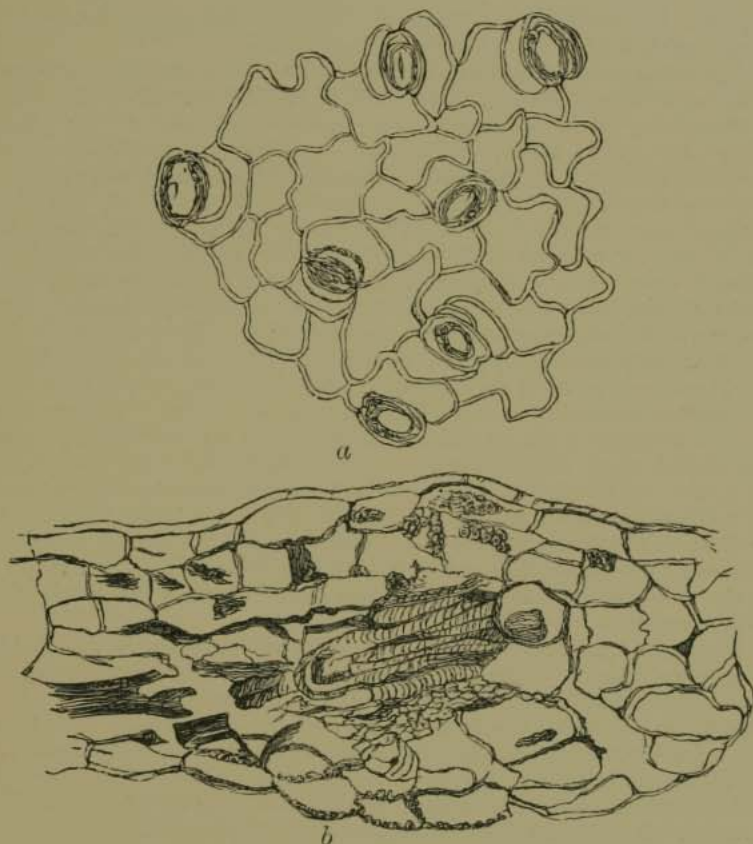


FIG. 51.—(a) Epidermis of under surface of the leaf of *Chloranthus Inconspicuus*, $\times 300$.
(b) Section near edge.

of drying tea which will represent water only is not easy; but since the loss both of volatile principles and theine does not materially affect the results, it is scarcely worth while to complicate the analysis by the use either of a lower temperature or of processes of absorption. The highest amounts of moisture in a genuine tea which are on record are two specimens from Cachar, analysed by Professor Hodges—the one (indigenous) gave 16.06 per cent., the other, a hybrid, 16.2 per cent. These were, however, not commercial teas, and appear to have been simply dried in heated rooms. The average hygroscopic moisture found by Mr. Wigner in thirty-five teas,

consisting of Hysons, Capers, Souchongs, Gunpowders, and others, was 7.67 per cent., the driest teas being the Hysons and Gunpowders, the moistest the Congous :—

	Per cent.
The maximum amount of moisture found in Hyson, .	5.68
The minimum	4.84
The maximum	6.55
The minimum	4.94
The maximum	10.33
The minimum	6.36

§ 238. *The Estimation of Theine or Caffeine.*—The modern processes for extracting theine fall chiefly under three heads :—

(1.) *Extraction by treating a decoction of the theine-containing substances with lime or burnt magnesia, evaporation to dryness, and subsequent solution by chloroform, ether, or benzine.*—The fundamental idea of this process, perhaps, belongs to Müller; it has also, with various modifications, been recommended by Clous, Commaille, Dragendorff, and many other chemists.

Commaille adopts the following method :—5 grms. of the finely powdered and carefully sifted substance are made into a hard paste with 1 gm. of calcined magnesia. This, after standing for twenty-four hours, is dried upon a water-bath and powdered. The resulting green powder is exhausted three successive times in a flask with boiling chloroform, the flask being connected with an inverted Liebig's condenser, so that the action may be continued for a long time. The cool solution is filtered, the chloroform recovered by distillation, and the residue in the flask dried. This residue consists of resinous fatty matters and theine; the former are removed by treating the contents of the flask with hot water and 10 grains of powdered glass, which have been previously washed with dilute hydrochloric acid. The water is boiled and the contents shaken up with the glass; the resinous matters attach themselves to the latter in the form of little globules. The solution is poured on a wet filter, and the residue completely exhausted by repeated boiling with fresh quantities of water. On evaporating the united filtrates in a tared capsule, pure theine is left in the form of white crystals.

Dragendorff takes 5 grms. of the substance, exhausts it with boiling water, evaporates to dryness, adding 2 grms. of burnt magnesia and 5 of ground glass; the finely powdered residue is soaked in 60 c.c. of ether for twenty-four hours, and finally thoroughly exhausted by ether. The latter, when separated and evaporated, leaves the theine in a tolerably pure state. He also states that ether may be replaced by chloroform. Cazeneuve and Caillot recommend a very similar process, but magnesia is replaced by recently slaked lime, ether by chloroform. Markownikoff uses benzine instead of the solvents mentioned.

In all the above processes there is one source of error which does not appear sufficiently guarded against—viz., loss of theine during the evaporation to dryness, since it is absolutely impossible to evaporate a decoction of tea and magnesia to dryness at 100° C. without loss of the alkaloid—a loss which, so far as our experiments go, does not take place until the mixture is quite dry. The following modification may therefore be proposed: 4 to 5 grms. of the tea are boiled in a flask provided with an inverted Liebig's condenser for a couple of hours, the liquid and leaves are transferred to an evaporating dish, some magnesia added, and the whole concentrated to a pasty condition. This paste is treated and thoroughly exhausted by chloroform; the latter is separated and evaporated, and the

chloroformic extract redissolved in a little boiling water, the solution filtered, evaporated to dryness at a very gentle heat, and weighed.

(2.) *Simple Treatment of the Powdered Leaves by Solvents.*—Legrif and Petit soften the leaves first with boiling water, and then extract the moist mass by the aid of chloroform. Other chemists simply exhaust the powdered substance by chloroform or ether; subsequent purification may, of course, be necessary.

(3.) *Sublimation.*—A method of utilising tea dust by making it a source of theine, was recommended by Heijnsius (*Journ. Prak. Chem.*, xlix., 317). The tea dust was simply treated in a Mohr's benzoic acid subliming apparatus. Stenhouse improved this process by precipitating either a spirituous extract, or a decoction of tea by acetate of lead, evaporating the filtrate to dryness, mixing the residue with sand, and subliming. These processes of sublimation, however, were proposed simply for the extraction, not the estimation, of theine.

One of us, in 1877,¹ proposed the following quantitative method of sublimation:—A convenient quantity of the tea was boiled in the way mentioned, magnesia added, and the whole evaporated to a paste, which was spread on a thin iron plate, and covered with a tared glass funnel. The heat at first was very gentle, but was ultimately raised at the later stages of the process to 200° C. The theine sublimes perfectly pure and anhydrous, and forms a coherent white coating on the sides of the funnel; the increase of weight is simply anhydrous theine. To ensure success it is absolutely necessary—

- (1.) That the layer be as thin as possible.
- (2.) That the heat be only gradually increased.
- (3.) That the mixture be occasionally cooled, and then thoroughly stirred.
- (4.) That the sublimation be prolonged for a sufficient time.

The sublimation is finished when a funnel, inverted over the substance, heated to about 150° C., and left for half an hour, shows no crystals

An improvement on this process is to place the paste on a ground glass plate, to which a flanged funnel has been ground so as to fit air-tight. The apparatus is then connected with a mercury-pump, and an absolute vacuum produced. By the aid of a shallow sand-bath, the theine may be sublimed at a very gentle heat.

§ 239. *Determination of Total Nitrogen.*—Peligot and Wanklyn have laid particular stress on the large amount of nitrogen contained in tea leaves. This nitrogen is, of course, largely dependent on the theine, and it is questionable whether, with the improved methods for the extraction of the latter, it is worth while to make a combustion, more especially as the exhausted leaves are highly nitrogenous, from the presence of an albuminous body. The N may be determined by Kjeldahl's method or by combustion with copper oxide. The following are a few determinations of total nitrogen:—

	Per cent.	Analysed by
A sample of genuine tea from Cachar, .	4.74	Hodges.
A hybrid variety, do., .	2.81	"
Another sample from Cachar, .	4.42	"
Sample taken from 60 green teas slightly faced, .	3.76	"
60 Black teas,	3.26	Wigner.
6 Assam teas,	3.64	"
6 Caper teas,	3.32	"
Assam tea, from Dr. M'Namara's garden, .	3.88	"
Sample of exhausted leaves,	3.80	"

¹ *Op. cit.*

§ 240. *Determination of Tannin.*—The methods proposed for the determination of tannin are very numerous. Four only, however, require any notice here—viz., the gelatin process, the copper process, Mr. Allen's acetate of lead process, and Löwenthal's process.

(1.) *By Gelatin.*—The best process by gelatin is decidedly that which dispenses with the drying and weighing of the precipitate. A solution of gelatin is carefully made by first soaking the gelatin in cold water for twelve hours, then raising the heat to 100° C., by placing the bottle on the water-bath (the strength should be about 3 per cent.), and finally about 8 per cent. of alum should be added. A portion of the solution thus prepared is put into an alkalimeter flask (*e.g.*, Schuster's), and carefully weighed. A solution containing a known quantity of tannin is now titrated with the gelatin until a precipitate no longer occurs; the flask is reweighed, and the loss shows approximately the strength of the solution. One or two more exact determinations will be required to get the correct value. It is necessary to allow the precipitate now and then to settle, and a few drops of the supernatant fluid should be placed on a watch-glass, to which a drop of gelatin may be added, and thus the point of saturation ascertained. The tannin in a decoction of tea is, of course, estimated on precisely similar principles.

(2.) *Copper Process.*—When a single determination of tannin is required it is best to precipitate by copper-acetate. 2 grms. of tea are boiled for an hour in 100 c.c. of water, the solution filtered, the filtrate boiled, and while boiling 20 to 30 c.c. of solution of copper acetate [1:20] added. The precipitate is collected, dried, burnt to an ash, oxidised with nitric acid, and again ignited and weighed. 1 gm. of $\text{CuO} = 1.3061$ of tannin, if Eeler's¹ figures be accepted; if Woolf's,² then 1 gm. $\text{CuO} = 1.304$ tannin.

(3.) *Mr. Allen's Lead Process.*—A filtered solution of lead acetate 5 per cent., a solution of 5 mgrms. of pot. ferridecyanide, 5 c.c. of strong ammonia water, and 5 c.c. of pure water, and lastly, solution of pure tannin (1 per cent.) are required. The process essentially depends upon the precipitation of tannin by lead acetate, and using ammoniacal pot. ferridecyanide as an indicator. The latter agent strikes a pink colour with tannin. The solution is standardised by taking a known volume of the lead solution, and dropping in the tannin liquid until a small portion filtered gives a pink colour with the indicator.

Tea is tested in a precisely similar manner. Mr. Allen's method is tolerably speedy and accurate. The final reaction is, however, somewhat difficult to observe.

(4.) *Löwenthal's Process.*—Up to the present time this method (originally worked out for barks) is the best we possess; it depends on the oxidation by permanganate, and indigo is used as an indicator. It not only gives us the tannin, but the amount of other astringent matters as well. The following solutions are required:—

- (1.) A solution of potass. permanganate, 1.333 grms. per litre.
- (2.) Precipitated indigo, 5 grms. per litre.
- (3.) Dilute sulphuric acid (1:3).
- (4.) A solution of gelatin, 25 grms. to litre, saturated with sodium chloride.³

¹ *Dingler's Poly. Journ.*, ccxxix., 81.

² *Zeitschrift f. an. Chem.*, 1, 104.

³ Löwenthal prepares the solution by steeping 25 grms. of the finest Cologne glue in cold water over night; it is then melted on the water-bath, saturated with NaCl , and made up to 1 litre with saturated NaCl solution, filtered, and kept well corked.

(5.) A saturated solution of pure salt, containing 25 c.c. of sulphuric, or 50 c.c. of hydrochloric acid per litre.

The analysis as applied to the determination of tannin in barks is performed thus:—10 grms., say, of sumach are taken and exhausted by boiling with water, and the solution made up to 1 litre; of this infusion, 10 c.c. are mixed with 75 c.c. of water, 25 c.c. of the indigo solution added, and 10 c.c. of the dilute sulphuric acid. The permanganate solution is run drop by drop from the burette with constant stirring, till the blue colour changes to yellow, when the amount of permanganate used is noted (x). The same process is repeated with indigo and sulphuric acid, and the amount read off (y); subtracting y from x = total astringent matters. The permanganate oxidises both tannin and indigo; but the tannin being the easier to oxidise, is consumed first. In order to obtain accurate results, the proportion of indigo should be such as to require about twice the quantity of permanganate which would be consumed by the tannin alone. Thus, if indigo alone requires 10 c.c. of permanganate to decolorise it, the indigo and tannin together must not take more than about 15 c.c.; if it does so, the tannin must be diluted accordingly. The total astringent matters being known, the next step is to throw the tannin out, and estimate the gallic acid and impurities. 100 c.c. of the infusion are mixed with 50 c.c. of the salted gelatin infusion; after stirring, 100 c.c. of the salt acid solution are added, and the mixture allowed to stand for twelve hours. It is then filtered, and an aliquot part of the filtrate is oxidised by permanganate and indigo, as before.

Löwenthal gives the following example: 10 grms. of sumach were boiled in 750 c.c. of water and after cooling made up to one litre:—

		Permanganate.
(1.)	10 c.c. of sumach infusion,	} consumed, 16·6
	25 c.c. of indigo solution,	
	Do.,	repeated, 16·5
		33·1
	50 c.c. of indigo solution alone,	13·2
	Total permanganate for 20 c.c. of sumach,	19·9
(2.)	50 c.c. filtrate from the gelatin,	} consumed, 11·2
	25 c.c. indigo solution,	
	Do.,	repeated, 11·1
		22·3
	50 c.c. indigo alone,	13·2
	Gallic acid and impurities,	9·1

Deducting 9·1 c.c. from 19·9 c.c. equals 10·8 c.c. as permanganate, equivalent to the tannin of 20 c.c. of sumach infusion, or 0·2 gm. of dry sumach. It is well to ascertain the value of the permanganate solution by oxalic acid, adopting the numbers given by Neubauer and Oser—viz., that 0·063 oxalic acid is equal to 0·04157 gallo-tannic and 0·062355 quercitanic acids. Should it be preferred to use tannin, the purest commercial tannin must be precipitated by lead, the precipitate freed from lead in the usual way, and the solution of pure tannin then evaporated to complete dryness, and a solution of convenient strength made. The process requires but little modification to be applicable to tea.¹

¹ F. Becker has proposed (*Chem. News*, xli., 229) to estimate tannin as follows:—50 c.c. of a solution of methyl violet—5 per cent. strength—is made up to half a litre and heated to 50°, it is then standardised by a 1 per cent. solution of tannin; which is run in until all the colouring-matter is precipitated and the filtrate is colourless. When operating on solutions of unknown strength, guided by the first rough essay, they should

The amount of tannin in genuine teas seems to be variable. S. Janke, using the acetate of copper process, has determined the tannin in eighteen samples of black tea, and found as a maximum 9.142 per cent., as a minimum 6.922 per cent., and as a mean 8.1 per cent. Three samples of green tea gave 9.94, 8.56, and 9.57 per cent. Mr. Wigner, as a sample of very astringent teas, gave the following:—

	Per cent.
Moyone young Hyson,	39.0
Very choice Assam,	33.0
Indian young Hyson,	39.0
Assam tea from Dr. M'Namara's garden,	27.7
Caper, mixed,	42.3

Exhausted tea leaves yield from 2 to 4 per cent. of tannin. A tea giving only 6 per cent. of tannin is to be regarded as suspicious, but care must be taken not to rely upon any single indication.

§ 241. *The Extract.*—The extract is a measure of the soluble matter in tea. Peligot exhausted the leaves and then redried them, and thus estimated the soluble matter by difference. Wanklyn, however, has proposed a more rapid and convenient method. It consists in taking 10 grms. of tea, and boiling with 500 c.c. of water, the flask being adapted to a Liebig's condenser. When 50 c.c. are distilled over, the process is stopped, and the 50 c.c. returned to the flask; 50.3 grms. of the hot strained liquid are then weighed out and evaporated to dryness. Wigner boiled with a vertical condenser for an hour, and found that 1 per cent. strength yielded the most constant results. —Perhaps, on the whole, the best process is the following:—Place one part of tea in 100 of water, boil for one hour with a vertical condenser, and then take an aliquot part of the filtered liquid for evaporation. In every case the time occupied in boiling, and the strength, should be mentioned in reporting, for two analysts operating by different methods may differ as much as 6 or 8 per cent.—the soluble matter not being entirely removed for a very long time. Since the substances that are at once dissolved are really those upon which its commercial value depends, it is a question whether it would not be better simply to pour boiling water on the leaves, let the infusion stand for one hour, and then estimate the extract, calling it *extract of infusion*.

Any addition of exhausted leaves lowers the percentage of extract. The following are some determinations of extract:—

	Per cent.	Analysed by
Java tea, dried,	35.2	Peligot.
„ not dried,	32.7	„
Pekoe, ordinary, dry,	41.5	„
„ undried,	38.0	„
Gunpowder, dry,	51.9	„
„ undried,	48.5	„
„ dry,	50.2	„
„ undried,	46.9	„
Moyone Gunpowder,	40.7	Wigner.
„	39.3	„
„	38.5	„
„	37.9	„
„	33.3	„
Imperial, dry,	43.1	Peligot.
„ not dried,	39.6	„
„ dry,	47.9	„
„ not dried,	44.0	„

be diluted or concentrated to about 1 per cent. If this method be applied to tea, it would be well to prepare some pure gallo-tannic acid from tea, and to use this substance for the purpose of standardising.

	Per cent.	Analysed by
Hyson, dry,	47.7	Peligot.
" not dried,	43.8	"
Hyson skin, dry,	43.5	"
" not dried,	39.8	"
Congou,	36.8	"
" dried,	40.9	"
" bon,	40.7	"
" " dried,	45.0	"
"	33.0	Wigner.
"	29.8	"
"	29.8	"
"	26.2	"
"	26.1	"
Caper, dried,	39.3	Peligot.
" not dried,	35.8	"
"	37.9	Wigner.
"	37.7	"
"	32.4	"
"	30.0	"
Assam, dried,	45.4	Peligot.
" not dried,	41.7	"
"	33.3	Wigner.
Hyson,	36.8	"
Moyone Young Hyson,	44.8	"
Tea direct from China, dry,	41.7	Wanklyn.
" "	40.2	"
" "	41.2	"
Indian tea, dry,	33.9	A. Wynter Blyth.
"	43.8	Wigner.
Broken Indian,	43.4	"
Indian Souchong,	32.5	"
Scented Orange Pekoe,	34.2	"
Manuna, fine,	37.0	"
Himalayan Tea,	38.6	Wanklyn.
"	35.4	"

TABLE XXV.

	ZOLLER.	HODGES.		ZOLLER.	WIGNER.	
	Ash of fine young Himalaya Tea.	Tea from Cachar (indigenous).	Tea from Cachar (hybrid).	Exhausted Tea Leaves.	Ash of a number of Mixed Black Teas.	Ash of a number of Mixed Green Teas.
Potash,	39.22	35.200	37.010	7.34	30.92	28.42
Soda,	0.65	4.328	14.435	0.59	1.88	2.08
Magnesia,	6.47	4.396	5.910	11.45
Lime,	4.24	8.986	5.530	10.76
Oxide of iron,	4.38	2.493	2.463	9.63
Manganous oxide,	1.03	1.024	0.800	1.97
Phosphoric acid,	14.55	18.030	9.180	25.41
Sulphuric acid,	trace.	5.040	6.322	trace.	4.88	5.66
Chlorine,	0.81	3.513	2.620	trace.
Silica and sand,	4.35	0.500	1.300	7.57	1.70	7.50
Charcoal,	2.900	1.830
Carbonic acid,	24.30	13.590	12.600	25.28	11.60	6.43
Percentage of total ash soluble in water, }	100.00	100.00	100.00	100.00	57.00	52.85

Since the extract of genuine tea appears to vary from 26 per cent. up to more than 50 per cent., it is unfortunately of no very great value for purposes of valuation. The extract, after being weighed, is burnt up to an ash, which will always be found to be heavy, rich in alkaline salts, and varying usually from 4 to 7 per cent.

§ 242. *The Ash*.—The percentage of total ash is taken by burning up 1 to 5 grms. of the tea in a platinum dish. The leaves readily ignite, and the operation may take place at a very low temperature, so that there is, with care, very little volatilisation of chlorides. The comparative composition of the ash of fresh and of exhausted tea leaves is shown in Table XXV.

The ash, on being cooled and weighed, is next boiled up with a little water, the soluble portion filtered from the insoluble, and washed in the

TABLE XXVII.

	Total Ash. Per cent.	Ash soluble in Water. Per cent.	Ash soluble in Acid. Per cent.	Silica,	Potash.	Authority.
Average of 17 ordinary Teas from original chest, consisting of 2 Indian, 12 Congous, 2 Gun-powders, and 1 Hyson,	5·75	3·07	2·25	0·43	1·38	G. W. Wigner.
Maximum,	6·03	3·35	2·87	0·76	1·88	„
Minimum	5·53	2·75	1·99	0·15	1·17	„
Average of 25 special Teas,	5·95	3·33	2·09	0·53	1·38	„
Maximum	7·02	3·88	2·68	1·67	1·96	„
Minimum	5·17	2·64	1·33	0·04	1·08	„
Genuine Indian Tea,	5·61	2·90	A. Wynter Blyth.
Common Tea,	5·92	3·55	Wanklyn.
Paraguay Tea,	6·28	4·22	„
Average of 7 Teas,	5·75	A. S. Wilson.
„ 9 „ „	5·66	3·00	A. H. Allen.
Horniman's p. black,	5·30	3·50	„
„ green,	5·60	3·80	„
Ambrosial black,	5·60	3·40	„
Genuine black, 2s. 6d. lb.,	5·60	3·09	„
„ „	5·70	3·28	„
„ „	6·02	3·26	„
„ „	6·34	3·20	„
„ „	6·10	3·96	„
„ „	5·75	3·06	„
„ 3s. lb.,	5·50	3·55	„
Broken leaf, with stalks,	5·40	2·80	„
Caper (4·8 silica),	11·40	1·50	„
Mixed dryexhausted leaves from various Teas,	4·30	0·52	„
Coffee leaves,	10·32	3·77	„
Beech,	4·52	2·00	Wanklyn.
Bramble,	4·53	1·84	„
Raspberry,	7·84	1·72	„
Hawthorn,	8·05	3·78	„
Willow,	9·34	4·16	„
Plum,	9·90	5·66	„
Elder,	10·67	3·19	„
Gooseberry,	13·50	7·83	„

ordinary way. The filtrate is evaporated to dryness, very gently ignited, and returned in percentage as soluble ash. The insoluble portion is next treated with acid, and the remaining sand dried, ignited, and weighed. The alkalinity of the soluble portion should also be taken, and may be returned as potash. This simple examination of the ash, consuming very little time, gives tolerably well all the information afforded by a complete and exhaustive analysis. The table (XXVII.) shows a few percentages of ash, and may be compared with the percentages of the ashes of beech, bramble, and others.

All the analyses hitherto published show that the percentage of ash in genuine tea never reaches 8 per cent. An ash beyond 8 per cent., calculated on the dried tea, is certainly adulterated. In the same manner, all genuine tea possesses a soluble ash not less than 3 per cent.

Mr. John White¹ has given some analyses of the ash of adulterated Caper teas as follows :—

TABLE XXVIII.

	Total Ash.	Ash soluble in Water.	Ash insoluble in Water.	Ash insoluble in Mineral Acid.
(1)	12·20	2·76	9·44	5·86
(2)	11·34	2·79	8·55	4·66
(3)	11·31	2·84	8·47	5·10
(4)	9·90	3·24	6·66	4·40
(5)	9·88	2·98	6·90	3·84
(6)	13·47	2·80	10·67	6·26

He suggests on these analyses that the limit for sand in the ash of tea might be fixed at 3 per cent.

§ 243. *Determination of Gum.*—If it is necessary to determine the gum in tea, as sometimes happens, the aqueous decoction should be evaporated nearly to an extract, and the residue treated with methylated spirit, filtered, and washed with the spirit. The gum is dissolved off the filter by the aid of hot water, and the solution evaporated to dryness, and weighed; it is then ignited to an ash, and the mineral deducted from the total weight.²

§ 244. *General Review of the Adulterations of Tea.*—The most frequent are, certainly, the addition of sand, generally strongly impregnated with iron, the addition of foreign and exhausted leaves, and the addition of astringent principles, such as catechu, etc. All these adulterations must take place abroad, there being no evidence that a single hundredweight of tea has been tampered with in England,—the blame *may* lie with the home-traders, but proof is wanting. On the other hand, it not unfrequently happens that cargoes of tea recovered from sunk vessels, or teas damaged in some other way, are sold and blended by wholesale manufacturers with those that are genuine. Such samples contain usually an excess of salt, and show more or less evidence of the addition of exhausted leaves.

¹ *Analyst*, May 1899.

² H. Hager is (*Pharm. Central. Halle*, 1879, 258) the author of a general process of analysis, which possesses some good points; 10 grms. of tea are infused in 100 c.c. of warm water, and allowed to stand for two days; the solution is poured off, and another

The facing of tea is rapidly decreasing. There has been much dispute as to whether this is to be considered an adulteration or not; a thin film of graphite, or any other harmless substance, in such quantity as to add no appreciable weight, can hardly be called adulteration. Each case, however, must be judged of by its merits. A small addition of such a substance as catechu, to impart astringency, is probably frequent, and difficult of detection. Any amount present, to the extent of 3 per cent. or over, is shown by precipitating an infusion of the tea with a slight excess of neutral lead acetate, filtering, and adding a little dilute ferric chloride solution. If catechu be present there is a bright-green colour, and ultimately a precipitate of a greyish-green colour. [See also Hager's method.²] The same infusion filtered from the lead precipitate gives a copious precipitate with argentic nitrate. Mr. Allen has pointed out one advantage of the lead process in cases of adulteration with catechu, catechuic acids possessing a precipitating power so widely different from that of tannin, that, if reckoned as tannin, there are always anomalous results.

Soluble iron salts, alkaline carbonates, and other substances, are stated to be occasionally added, but no conviction relative to these appears to be on record. The soluble iron salts may, of course, be dissolved from the tea leaves by a little cold dilute acetic acid, and the liquid tested in the usual way; there is then no confusion between the iron naturally present and that added.

§ 245. *Bohemian Tea*.¹—It would seem that for some time there has been cultivated in Bohemia the *Lithospermum officinale*, the common 'Gromwell' of our country, and the leaves have been dried and sold as *Thea Chinensis*, under the name of 'Bohemian Tea.' They have also been used for the purpose of adulterating Chinese tea. The 'Gromwell' is a plant belonging to the borage order, growing in dry and stony places, from a foot to a foot and a half high. The flower is greenish-yellow, the stem erect and branching, and the leaves are lanceolate, hairy beneath, with bulbous adpressed bristles above. They are totally unlike tea leaves, and the hairiness itself would be diagnostic of a leaf other than that of tea. The chemical composition is also entirely different. The mineral constituents are excessive, and there is neither any alkaloid nor any essential oil.

The average composition of 'Bohemian Tea' is as follows:—

Cellulose,	5.96
Tannin,	8.25
Fat,	9.29
Other nitrogen-free organic substances,	26.49
Albuminous matters,	24.54
Ash, -	20.60
Water,	4.86

100 c.c. of water added to the partially-exhausted leaves, which are then unrolled and botanically examined. 50 c.c. of the solution are evaporated to dryness; 10 c.c. of the solution should give no turbidity in the cold when an equal volume of alcohol is added. For the estimation of theine, 10 grms. of tea, 3 grms. of sodic carbonate, and 3 grms. of litharge are made into a paste with 10 grms. of water dried up and extracted with chloroform. For the special detection of catechu, 1 gm. of tea is extracted by 100 c.c. of boiling water. This solution is boiled with excess of lead oxide, and the filtrate (which must be clear) mixed with a solution of silver nitrate. Pure tea gives only a slight grey-black deposit of metallic silver, but tea adulterated with catechu a strong yellow flocculent precipitate.

¹ A. Belohouben: *Chem. Centrabl.*, 1880, p. 152.

MATÉ.

§ 246. "The *Maté*, or Paraguay Tea Tree (*Ilex paraguayensis*), is a small tree, belonging not to the family of the *Ilicieæ*, as stated by some, but to the *Celastrineæ*; it reaches in height ordinarily 4 or 5 metres, sometimes 7. Its trunk is about 20 cm. in circumference, and is covered by a whitish bark. The leaves are oblong, cuneiform, obtuse, and finely dentate. It has ancillary, multipartite peduncles, calyx tetrasepalous, the corolla with four petals in the form of a crown, style none, stigma 4-fid, fruit a four-celled berry. The plant grows very abundantly in Paraguay, North Corrientes, Chaco, and South Brazil, where it forms woods called 'Yerboles.'

In the Spanish Republic three different sorts of maté are sold:—
1. Caá-cuy or caá-cuip, which consists of the new leaves and scarcely developed shoots. 2. Caá-mirim, the leaves separated from the twigs and stalks (the midrib of the leaf is also removed). 3. Caá-guacu or Jerva de Palos, large old leaves with twigs and fragments of wood.

The microscopical structure of the leaves is fairly characteristic, the upper surface of a maté leaf is seen to be built up of thin-walled wavy cells, .05 mm. (.002 inch) in diameter, the cuticle is thick and wrinkled, the under surface has similar cells, but the contour is still more wavy, while it is only the cuticle over individual cells which shows fine wrinkles. The number of stomata is extremely great, being about 12 to the square millimetre, the stomata are nearly circular, and but .03 mm. (.001 inch) in diameter. Here and there may be found glands with reddish contents. The leaf on section may present, a little below the upper epidermis, cells filled with oxalate of lime crystals, but has no other special peculiarity.

Maté is prepared in Paraguay thus:—The entire trees are cut down, and the small branches and shoots are taken with the leaves, and placed in the *tatacúa*, a plot of earth about 6 feet square, surrounded by a fire, where the plant undergoes its first roasting. From thence it is taken to the *barbacua*, or grating supported by a strong arch, underneath which burns a large fire. Here it is submitted to a particular torrefaction, determined by experience, which develops the aromatic principle. Then it is reduced to a coarse powder in mortars formed of pits dug in the earth, and well rammed. It is next put into fresh bullock skins, well pressed, and placed in the sun to dry. The packages (*tercois*) thus obtained weigh from 90 to 100 kilograms, and have an average commercial value of 1 to 2 dollars the kilogram.

Senor Araté gives an analysis of maté—

	In 100 parts.
Organic combustible substances,	91.685
Ash,	8.315
The ash contains—	
Lime (CaO),	12.344
Magnesia,	11.395
Soda,	7.281
Potash,	2.984
Manganese dioxide,	2.500
Ferric oxide,	3.410
Sulphuric acid,	0.926
Hydrochloric acid,	0.716
Phosphoric acid,	5.540
Carbonic acid,	8.150
Sand, silica, carbon, and loss,	44.754

The enormous relative quantity of sand is a result of the mode of preparation in excavations made in the soil. The plant contains—

Principles soluble in ether,	9·820
„ „ alcohol,	8·432
„ „ water,	26·208
„ „ water acidulated with HCl,	7·260
„ „ in solution of caustic soda,	16·880
Cellulose,	13·280
Water,	9·000
Sand,	9·120
	<hr/>
	100·00

T. Peckolt (*Pharm. Journ. Trans.* [3], xiv., 121-124) has analysed the fresh leaves of *Ilex paraguayensis* from the Orgu mountains in Neufreeburg with the following results:—

	Per 1000 (Leaves simply air dried).
Stearoptene,	·021
Volatile oil extracted by ether,	·099
Fat and wax,	19·800
Green colouring-matter,	10·900
Chlorophyll and soft resin,	20·966
Brown acid resin,	48·500
Theine,	6·398
Bitter extractive matter,	2·033
Sugar,	39·266
Extractive matter and organic acids,	8·815
Maté, tannic acid (pure),	27·472
Maté, viridic acid (crystalline),	·024
Albumen, dextrin, salts,	47·660
Cellulose,	166·600
Moisture,	601·356

He has also analysed Parana leaves and maté as follows:—

	Air-dried leaves from which maté is prepared. Per 1000.	In commercial maté. Per 1000.
Volatile oil extracted by ether,	·179	·026
Theine,	16·750	5·550
Chlorophyll and soft resin,	51·200	16·755
Brown acid resin,	84·500	25·500
Maté, tannic acid (pure),	44·975	16·755
Maté, viridic acid (crystalline),	·025	·024
Extractive matter,	65·130	16·610
Albumen, salts, dextrin,	36·102	18·159
Cellulose and moisture,	700·643	908·379

Theine averages 1·3 per cent. The tannin of maté is peculiar; it does not tan hides, and requires a special method for its estimation; it amounts to about 1·6 per cent. Maté also contains a large quantity of a peculiar fatty matter. Maté does not exalt the peripheric nerves like tea, nor the cerebriac like coffee, but appears to have, in some degree, a narcotic action. The usual way of taking it is by sucking it up through a reed called a 'bombilla.'

MM. d'Arsenal and Couty have administered maté to dogs by injection into the stomach. They found it diminish the carbon dioxide and oxygen of the arterial and venous blood to a considerable extent, sometimes to a third or even half the normal quantity. This action, which is less

intense during digestion, and has no necessary relation to phenomena of excitation of the sympathetic nerve-system, is somewhat obscure as to its 'mechanism'; but its existence proves directly the importance and nutritive value of the aliment in question.

A species of *Ilex*—viz., *Ilex cassiva*, employed as a tea in Virginia, has been analysed by M. Ryland and T. Brown, who found

Volatile oil,	0·011
Wax and fatty matter,	0·466
Resin,	3·404
Chlorophyll,	2·491
Theine,	0·122
Tannin,	2·409
Colouring-matter soluble in alcohol,	4·844
Extractive matter soluble in alcohol,	10·149
Extractive matter insoluble in alcohol,	4·844
Amidon and pectine,	15·277
Fibre,	33·827
Ash,	3·995

COFFEE.

§ 247. The coffee berry is the seed dried, and deprived of its fleshy coverings, of the *Coffea arabica*, Nat. Ord. *Cinchonaceæ*. Before use the berries are roasted to a chocolate brown, and then ground in a mill.

Microscopical Structure.—The main portion of the coffee berry is composed of strong angular, thick-walled cells, the walls of which, cut in radial



Fig. 52.—Endosperm of the coffee berry, $\times 160$.

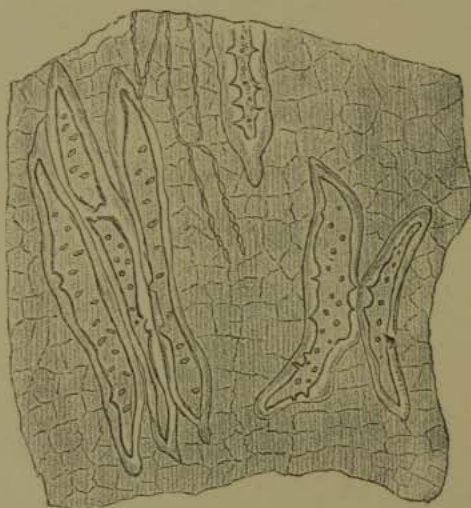


Fig. 53.—Membrane from the coffee berry, showing the peculiar pitted and thickened cells, $\times 160$ (after MOELLER).

sections, are seen to possess a peculiar knotty appearance (fig. 52), but a tangential section shows merely a network of thickened cells (fig. 54), and the knotty appearance is then not to be seen. The thickened cell-walls

are mainly composed of cellulose, and are nearly entirely dissolved by ammoniacal solutions of copper oxide, and strike a blue colour when treated with the chloriodide of zinc, or with sulphuric acid and iodine. The cells all contain dark-coloured contents, and sometimes crystals of theine. When roasted, these characteristics are never entirely obliterated, but the cell walls are often coloured, and the whole structure more difficult to define. A thin tough Japanese-paper-like membrane (fig. 53) may be detached from the berry; most of this in grinding is separated from its connections, but it may always be found; it is composed of a series of spindle-shaped fibres attached to a thin membrane; the fibres in the undeveloped seed form a continuous sheet, but in older berries they are separated.

Besides these elements there is a small embryo composed of the usual regular, oval, tender-walled embryonic cells. These are so different from the usual coffee tissues, that they might be mistaken for foreign substances, but of course the quantity in ground coffee of embryonic tissue is very small indeed, and is detected but rarely.

The microscopical structure described above separates and distinguishes coffee at once from all other known berries or seeds, while the tissues of roots, such as chicory (consisting of loose, thin-walled, vegetable cells, with a greater or less admixture of large spiral vessels), are entirely different, and may be said, indeed, to be built on a different plan.

To investigate the ground coffee microscopically, it is best to examine with a low power a little in a watch-glass, and then to remove those portions which are most transparent; if necessary, still further clearing them up by soaking in a strong solution of potash, and teasing them out by the aid of a needle. It is also quite practicable to cut sections of fragments of coffee held firmly between two pieces of cork, or embedded in sealing-wax.

§ 248. *Chemical Changes during Roasting.*—The effect of roasting is to drive off a large quantity of water, to volatilise a small quantity of theine, to change a portion of the sugar into caramel, to rupture the cell-layers containing fat and albumen, and to swell the berry by the extrication of gases, consisting mainly of carbon dioxide. There is also developed a fragrant aromatic substance, a single drop of which is sufficient to scent a large room with the peculiar coffee odour; the best temperature for the production of this aroma is 210° C. That during roasting there is an actual loss of theine, is easily proved by holding a glass plate over the heated berries; in a very little time crystals of theine condense. Tenneck found in unroasted coffee .75 per cent., and in the same roasted, .42 per cent. theine. It would appear that roasted coffee gives up more to water than does raw coffee; for Cadet found that beans roasted to a light brown yielded to water 12.3 per cent.; to a nut-brown, 15.5 per cent.; and to a dark-brown, 21.7 per cent. Vogel also gives the soluble matters in raw coffee as 28 per cent.; and in roasted, 39 per cent. The amount of sugar changed is always considerable; thus Graham and Stenhouse found the

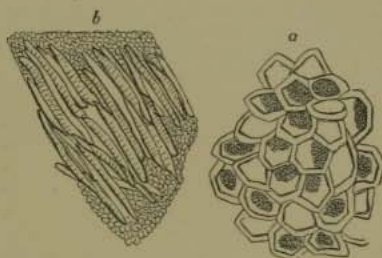


FIG. 54.—Coffee tissue, $\times 170$.

following differences in the percentage of sugar between the raw and roasted coffees:—

	Raw. Per cent.	Roasted. Per cent.
Highest amount,	7.78	1.14
Lowest amount,	5.70	...
Average of twelve specimens grown in different places,	6.97	0.26

König has studied the changes taking place in roasting, and his views and experiments are as follows:—300 grms. of coffee-berries containing 11.29 per cent. water gave, on roasting to a light brown colour, 246.7 grms. of roasted coffee, containing 3.19 per cent. water. We have, therefore—

1. Taken 300 grms. coffee-berries,	= 266.15 grms. dry substance.
2. Obtained 246.7 grms. roasted coffee,	= 238.83 " "
Then loss, 53.3 grms.,	= 27.32 " "
Or in per cent., 17.77,	= 9.11 per cent. organic substance.

Hence, in roasting, 8.66 per cent. of water and 9.11 per cent. of organic substances have been lost. This is divided among the constituents of coffee as follows. (See Table XXIX.)

The products obtained in roasting coffee, according to Bernheimer, are palmitic acid, theine, caffel (an oil, boiling-point 195° to 197°), acetic acid, carbonic acid, hydrochinon, methylamine, pyrrol, and acetone.

§ 249. *Constituents of Coffee.*—The main properties of coffee are apparently due to:—(1.) An essential oil, not yet completely studied; (2.) caffee-tannic acid; (3.) theine or caffeine; (4.) an alkaloid 'caffearine'; (5.) a concrete oil or fatty substance; (6.) manno-arabinose.

Caffeo-tannic acid, $C_{14}H_8O_7$, was first observed by Pfaff in the seeds and leaves of the coffee plant; it also occurs in the root of the *Chiococca racemosa*, and in the leaves of the *Ilex paraguayensis*, S. Hil. It may be separated from coffee by fractional precipitation of the infusion with acetate of lead. The precipitate, at first falling, consists of citrate with caffee-tannate of lead; but later on, the latter occurs alone, and can be washed with water and decomposed by SH_2 in the usual way. Caffeo-tannic acid thus obtained is a brittle, yellowish mass, easily powdered, and of feeble acid reaction. It is supposed to exist in the plant in combination with potash and theine. It is scarcely soluble in ether, but dissolves easily in water or in alcohol. The solution gives a dark-green colour with chloride of iron, or if dissolved in aqueous solution of ammonia, the alkaline earths, or the alkalies, a reddish-yellow or yellow colour. Decomposed with 3 parts of KOH, the end product is protocatechuic acid. Caffaic acid can be obtained in a crystalline form by boiling with 5 parts of potash lye and neutralising by sulphuric acid. It colours chloride of iron grass-green, is oxidised by nitric acid into oxalic acid, and is decomposed by potash into protocatechuic and acetic acids.

Caffearine, $C_{14}H_{16}N_2O_4$, is an alkaloid discovered in coffee by Pietro Paladino;³ it possesses narcotic properties, but is only in small quantity. This alkaloid requires farther investigation.

Coffee Fat.—The coffee fat can be obtained from an alcoholic extract of coffee; part separates on cooling the fluid to 0° C., the rest on dilution with water. It is white, without odour, of a buttery consistence, melting at

¹ *Nahrungs-und Genussmittel*, ii., 479.

² Theine is described at p. 323.

³ *Gazetta Chimica Italiana*, xxv., 1895; abstract in *Analyst*, June, 1895.

TABLE XXIX.

	Total Matters soluble in Water.	Nitrogen = { Nitrogenous Substances } ¹	Theine.	Fat.	Sugar.	Other N-free Substances.	Cellulose.	Ash.	Water.
1. In 300 grms. unroasted coffee,	82.32	5.610 = 28.69	3.540	39.09	9.75	90.88	83.16	10.44	33.87
2. In 246.7 grms. roasted coffee,	73.29	5.698 = 29.43	3.403	38.56	3.23	91.49	59.87	9.85	7.87
Then in the last more + { or less -,	- 9.03	+(0.078) = (0.74)	- .137	- 1.13	- 6.52	+ 3.61	- 23.29	- 0.59	- 26.00
Or in per cents. of the { original quantity,	- 10.9%	+	- 3.1%	- 2.1%	- 66.9%	+ 3.9%	- 28.0%	- 5.7%	- 76.7%
The percentage composition of the dry substance was as follows:—									
1. Unroasted coffee,	30.93	2.21 = 11.43	1.33	14.91	3.66	34.55	31.24	3.92	...
2. Roasted,	28.36	2.38 = 12.31	1.42	16.14	1.35	39.84	25.07	3.87	...
Or reckoned on the natural substance,									
1. Unroasted coffee,	27.44	1.87 = 8.43	1.18	13.23	3.25	31.52	27.72	3.48	11.19
2. Roasted,	27.45	2.31 = 12.05	1.38	15.63	1.32	38.41	24.27	3.75	3.19

¹ Theine subtracted.

37°5 C, and becomes rancid on exposure to the air. According to Rochleder (*Wien Akad. Ber.*, xxiv., 40), it contains the glyceride of palmitic acid and of an acid of the composition $C_{12}H_{24}O_2$.

C. O. Cech¹ exhausted 50 lbs. with alcohol and ether, and obtained 1200 grms. of a thick green fluid oil, and after a time fine crystals of theine separated. After six months the oil, although in closed flasks, began to be turbid, and gradually little groups of crystals separated and sank to the bottom. After the lapse of three years, the flask was found to be about two-thirds filled with crystals of the more solid fats, but the upper layer was beautifully green.

There are also citrates, and probably other organic acids and nitrates in coffee. J. Buig found in raw dry coffee .054 per cent. of nitrate of potash, in roast .041 per cent.

The following table gives the general composition of various commercial varieties of coffee:—

	Gummy Matter.	Theine.	Fat.	Tannic and Caffeotannic Acids.	Cellulose.	Ash.	Potash.	Phosphoric Acid.
Finest Jamaica Plantation,	25.3	1.43	14.76	22.7	33.8	3.8	1.87	0.31
Finest Green Mocha,	22.6	0.64	21.79	23.1	29.9	4.1	2.13	0.42
Ceylon Plantation,	23.8	1.53	14.87	20.9	36.0	4.0	...	0.27
Washed Rio,	27.4	1.14	15.95	20.9	32.5	4.5	...	0.51
Costa Rica,	20.6	1.18	21.12	21.1	33.0	4.9	...	0.46
Malabar,	25.8	0.88	18.80	20.7	31.9	4.3	...	0.60
East Indian,	24.4	1.01	17.00	19.5	36.4

Some analyses by Dragendorff of Mocha, Preanger, Ceylon, Malabar, Mysore, Java, and other coffees (twenty-five in all) give the following results:—

	Maximum per cent.	Minimum per cent.	Mean per cent.
Theine,	2.21	0.64	1.16
Ash,	4.87	3.83	4.41
Phosphoric Acid,	0.72	0.28	0.42

The carbo-hydrates of coffee have been partially investigated in the chemical laboratories of the U.S. Department of Agriculture. Cane sugar is the chief soluble hydrocarbon. By hydrolysis the insoluble hydrocarbons yield galactose; and by distillation with hydrochloric acid, an abundance of furfuraldehyde was obtained, indicating the presence of a pentose-yielding substance. By treating the substances insoluble in water with 5 per cent. NaOH solution, and precipitating with alcohol, a gummy substance was obtained. The gum yielded, by suitable treatment, galactose and furfuraldehyde.

One of the chief constituents of the coffee berry is stated by Mr. Baker² to consist of a manno-arabinose or manno-xylose, which, on hydrolysis, yields mannose in quantities of from 80 to 90 per cent.

§ 250. *Analysis of Coffee—Specific gravity.*—The chemists of the Municipal Laboratory, Paris, lay stress upon a determination of the density

¹ *Journ. für prak. Chemie*, xxii., 398.

² *Analyst*, xxvii., April 1902, 116.

of the powdered coffee, considering that it is of special value in showing artificial moistening of the berries. The density is determined in Regnault's volumemeter modified by M. Dupré. This apparatus consists of a vertical cylinder, V (fig. 55), closed by means of discs of ground glass made tight by screws, E and E; this cylinder communicates by means of a narrow tube with a glass syphon having a bulb of known capacity, U; the long limb of the syphon, T, is open to the air, graduated; at the bend of the syphon a third tube is let in, communicating with a mercury reservoir, B, and having an air trap, *r*; the whole is fixed on a wooden stand (see fig. 55); the tube has a mark below the bulb, U, and also a mark just where it joins the narrow tube leading to the cylinder. 50 or 100 grms. of the coffee are introduced into V. The cylinder, by means of the screws and a little vaseline, is made air tight. The stop-cock, R, is opened so as to put V in communication with the atmosphere. The reservoir is raised so as to bring the mercury on a level with the lower mark, and the barometric pressure noted (*h*) on a barometer. The stop-cock, R, is then closed, and the reservoir raised until the mercury coincides with the higher division, and the difference in height of the mercury in the two tubes noted (*H*). The volumes of V and U are first and once for all determined.

To make a measurement proceed as above, and take the height of the mercury in a barometer placed near the instrument; this may be called *h*. If *V* = the volume of the vertical cylinder, *v* = the volume of the smaller bulb, and *H* the difference in height of the

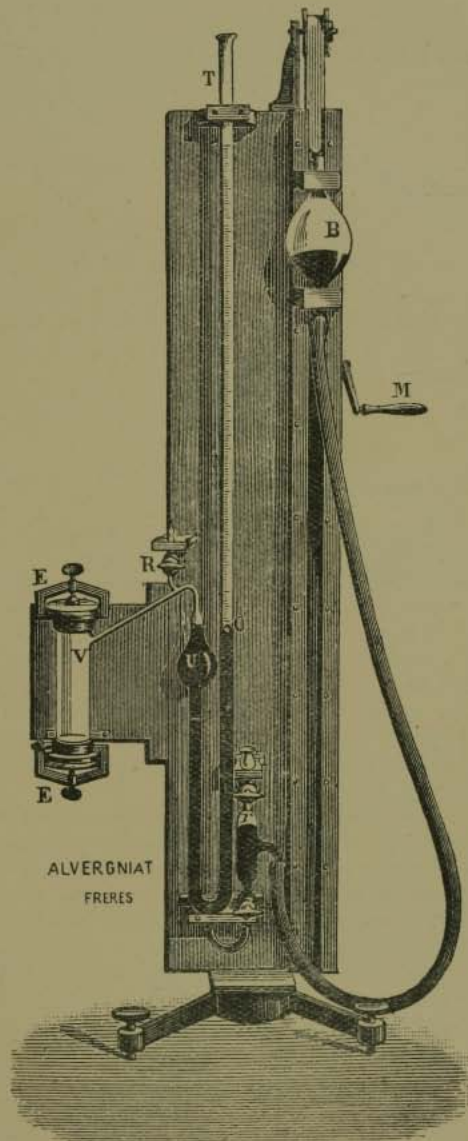


FIG. 55.—Regnault's volumemeter (Dupré's form).

mercury in the manometer tube; then the volume u is obtained by the following formula:—

$$u = V - v \frac{h}{H}$$

and if w = weight of coffee and d = density, then (u being the volume of coffee)

$$d = \frac{w}{u}$$

or by replacing the value of u from the first equation, the complete formula becomes

$$d = \frac{w}{V - v \frac{h}{H}}$$

To give an actual example:—100 grms. of unroasted green coffee are taken:
 $h = 75.5$ cm.; $H = 27.5$ cm.; $V = 230.1$ c.c.; $v = 54.5$ c.c.

then

$$u = 230.1 - \frac{54.5 \times 75.5}{27.5} = 230.1 - 149.63 = 80.47.$$

The density therefore equals $\frac{100}{80.47} = 1.2427$.

An accurate estimate of volume of a weighed portion of coffee or other powdered substance, and therefore of density, may be made with ordinary laboratory appliances as follows:—A suitable small flask provided with ground stopper and stop-cock is clamped on to the mercury pump, and the air it contains pumped out, measured, and reduced to 760 mm. and 0° C. A sample of the substance is next weighed and introduced into the flask. The flask now, of course, contains less air. The air is again pumped out and measured, the measurement being reduced as before to normal temperature and pressure. By dividing the weight of the substance by the difference in the air volume of the empty and the charged flask the density is obtained.

The great advantage of determinations of density by such means as the foregoing, as compared with ordinary determinations of density by means of the displacement of various liquids, is that there is neither waste nor alteration of the substance.

The experiments of the French chemists have given as a minimum for green coffee a density of 1.041, a maximum of 1.368; for roasted coffee, a minimum of 0.500, and a maximum of 0.635; a very high roasting will, however, give less than 0.500. The density taken in this way is also stated to indicate factitious coffees.

The hygroscopic moisture, theine, gum, astringent principles, and ash are all determined precisely as in the case of tea. According to the chemists of the Paris Municipal Laboratory the hygroscopic moisture in green coffee should not exceed 12 per cent., in roasted 3 per cent.

The coffee fat may be conveniently estimated by putting a known quantity in the fat-extraction apparatus figured at page 50; the best solvent to use will be petroleum ether, since it has less solvent action on the theine than ether. When the process is finished, the petroleum is evaporated to dryness in a tared dish. As thus obtained, the fat is almost, but not quite, pure.

ADULTERATIONS OF COFFEE AND THEIR DETECTION.

§ 251. The sophistications of coffee are numerous; chicory, roasted cereals of all kinds, wheat, rye, buck-wheat, potato flour, mangel-wurzel, acorns, lupine seeds, ground date-stones, caramel, and various leathery seeds have all been detected. Coffee damaged by sea-water is also commonly washed, first with water, then with lime water, dried and roasted, or sometimes coloured with an azo dye to give it a bright appearance.

Imitation coffees or coffee substitutes are manufactured on a large scale in America, Hamburg, and also in this country. Wiley has given a useful list of imitation coffees which have been examined in the laboratory of the U.S. Department of Agriculture; the list comprises the following mixtures which were pressed and moulded into berries or pellets:—¹

Coffee, bran, molasses.
 Wheat flour, coffee and chicory.
 Wheat flour, bran and rye.
 Chicory, peas (or beans), barley.
 Wheat, oats and buckwheat.
 Wheat flour and sawdust.
 Hulls of leguminous seeds formed into granules with molasses and roasted.
 Pea hulls and bran.

Dr. Bernard Dyer² has made some careful analyses of chicory; he finds the moisture varies from 1 to 4 per cent., and gives the following percentage data of 11 samples of chicory carefully dried:—

TABLE XXX.—CHICORY.

	Total insoluble in Water.	Ether Extract.	N.	Total Ash.	Soluble Ash.	Sand.
Chicory nibs and medium roast,	22.40	2.57	1.53	4.63	2.50	0.70
Chicory nibs and dark roast,	50.30	1.43	1.67	4.70	2.99	0.30
Ground chicory	22.27	2.17	1.33	5.53	2.43	1.43
"	21.50	1.90	1.34	5.23	2.07	1.43
"	35.50	3.43	1.50	5.13	2.57	0.77
"	37.80	3.87	1.52	8.23	1.60	3.97
"	22.77	3.17	1.25	5.13	3.30	1.60
"	22.50	3.67	1.23	5.73	3.23	1.63
"	23.50	2.60	1.29	5.63	2.97	1.47
"	22.50	2.60	1.29	5.33	3.22	1.47
"	22.63	2.57	1.29	5.70	2.80	1.47

It may be stated generally that such adulterations are best detected by a microscopical examination, but that they usually also present marked chemical differences, being for the most part deficient in theine, high in sugar, low in fat, and having high aqueous extracts. The general percent-

¹ U.S. Dep. of Agriculture, Div. of Chem., *Bull.*, No. 13.

² *Analyst*, Sept., 1898.

age composition of various imitation berries, as given by various chemists, is summarised in the following table:—

TABLE XXXA.—IMITATION COFFEE BEANS.

	W. Kisch.	E. Fricke.	K. Portele.	C. Kornauth.	
				Barley.	Coffee.
Water,	5.14	?	1.46	3.45	6.41
Albuminoids,	10.75	17.90	13.93	9.38	10.56
Fat,	2.19	2.03	3.80	3.25	1.04
Cellulose,	3.96	10.83	15.83	4.25	10.56
Sugar,	1.99	0.71	6.18	...
Extractive matter,	76.66	64.04	63.30	70.13	68.36
Aqueous extract,	29.88	24.85	21.53	31.20	34.37
Theine,	0.94	0.07
Glucose formed on boiling with H ₂ SO ₄ ,	50.02	69.28	67.19

Microscopical Detection of Adulterations in Coffee.—A practical, though not a scientific, distinction of substances used for the adulteration of coffee, is to divide them, for microscopical purposes, into two classes, the hard and the soft. If ground coffee is sprinkled on water, and allowed to soak for a little time, there is scarcely a particle which does not feel resistant or hard when rubbed between the fingers, the only soft portions of the coffee berry being the embryo, and this is so insignificant in quantity that it is not likely to cause embarrassment. The soft substances which have been used to adulterate coffee are chicory, figs, dandelion roots, and portions of the fruit of the ceratonia or carob-bean; while the hard substances are vegetable ivory, date-stones, acorns, and other hard seeds.

The soft tissues of the roots of the chicory¹ and dandelion abound with vascular bundles, and it is never difficult to see the spiral and other vessels; similarly, the laticiferous vessels in the tissues of the fig are extremely distinctive. In the softer tissues of the ceratonia or carob-tree fruit are to be found large cells containing bodies which, at first sight, look like starch granules, the surface being in wrinkles (see fig. 56). They are insoluble in water, alcohol, and dilute sulphuric acid. The characteristic test is, however, that they strike a blue or violet colour with a warm solution of

¹ Chicory is so readily detected that we scarcely require a direct chemical test. A. Franz has, however, pointed out that an infusion of coffee, when treated with copper acetate and filtered, yields a greenish-yellow filtrate; an infusion of coffee containing chicory yields, when similarly treated, a dark red-brown filtrate.—*Arch. Pharm.* [5], ix., 298-302.

A method of detecting chicory has been described by C. Husson. According to the latter, the chicory is often prepared by roasting with rancid fat. He discovers this by putting in a flask 10 grms. of chicory with 50 grms. of glycerin and 20 drops of hydrochloric acid, and the mixture is boiled and filtered. The filtrate is added to an equal volume of ether, and placed in a flask which, again, is put in a bath with boiling water. When pure ether-vapour rushes out of the bottle, the vapour is lit, and under the combined heat, the fatty matter rises to the surface of the glycerin, and is dissolved in the ether. When the flame diminishes in intensity, it is extinguished, and the ether allowed to evaporate spontaneously. On exposure to cold, fatty drops form gradually; these are examined by the microscope, and are drops of crystalline fats, such as are not yielded by pure coffee.

potash. And as these bodies are widely distributed throughout the fruit, the test reacts with the smallest fragment.

Finely ground date-stones and vegetable ivory are not so easy to detect, mainly because the particles are, for the most part, opaque, and it entails considerable trouble and some skill to prepare sections sufficiently clear to make out the details of structure.

Vegetable ivory is derived from the *Phytelephas macrocarpa*, and is much used for making buttons and various other purposes. In the working of the ivory there is considerable waste, hence it may be obtained at a very low rate, being, indeed, so much refuse. The ivory consists of elongated, equally thickened cells (see fig. 57 B), perfectly colourless, and dotted all over with pores; the contents of the cells are granular, and seem to be composed of a vegetable albumen, with here and there a few drops of fat. In sections of the ivory nut itself there is also to be seen a thin shell or epidermal layer (see fig. 57, A) consisting of peculiar fibres. The ends of the fibres are often thickened or clubbed; they are about .02 mm. broad and .2 long, are thickened by deposit, and contain a reddish-brown substance. The largest fibres are on the outside of the layer, while the shorter fibres occupy the inner part of the seed coating; the fibres are interlaced or felted in every direction. The seed-coating bears a very small proportion to the vegetable ivory, and is not so likely to be found as fragments of the ivory itself.

Date-stones¹ possess a very distinctive structure. The epidermis consists of elongated cells, forming, when looked at from above, irregular oblongs, the cell-walls are thickened irregularly, and they are frequently pitted (see fig. 58). In the parenchyma of the stone are curious irregularly-shaped long cells, which contain a tannin, and are coloured dark-green by ferric chloride solution. The endosperm is the hardest part of the date-stone, and is very similar in structure to vegetable ivory, but the cells are smaller and rounder, and the cell membranes are not so thickened by deposit.

The leguminous seeds are all built on a similar plan; they have at least two characteristic structures—viz., a layer of regular lengthened cells with their long axis set radially (see fig. 59), the palisade layer of the Germans; seen under a low power, they look so much like the dentine tubes of a tooth that they might be called “the dentine layer”; they

¹ *Date Coffee*.—There was established, some years ago, a company for the manufacture of what is termed ‘Date coffee,’ a preparation made from torrifed dates, and mixed with coffee, in the proportion of one-fourth coffee and three-fourth dates. A sample presented the appearance of a dark-brown, rather sticky powder, having a sweetish smell, but no coffee odour. On being thrown into water the water was immediately coloured, and the powder sank to the bottom. The specific gravity of the infusion was nearly that of pure chicory, viz., 1019.6. The microscope showed some fragments of coffee, as well as large loose cells and structures, quite different from those of coffee, and there was scarcely a trace of theine. The general analysis gave:—

	Per cent.
Water,	5.25
Sugar,	15.29
Extract,	46.50
Total ash, { Soluble, 1.78 {	2.85
{ Insoluble, 1.07 {	

The ash contained .262 P₂O₅ and .13 silica; .628 per cent. of an oily and resinous matter was also separated. The large amount of sugar would alone be sufficient to distinguish it from coffee, and there will not be the slightest difficulty in the identification of the substance should it be ever used in such a manner as to come under the Sale of Food and Drugs Act.

consist of narrow prismatic long cells, the base not unfrequently swollen out flask-like—and are covered with a thin cuticle. If looked at from above, of course only the hexagonal ends are seen presenting a tessellated pavement appearance—there is no 'lumen.' The most remarkable thing about these cells is that they are all the same size, save, indeed, in *Cicer arietinum*, where the dentine layer is in regular waves, and the individual cells range in length from 100 to 300 micro-millimetres.

The length and breadth of the palisade layers in various leguminosæ have been carefully determined by Moeller,¹ as follows:—

	Length. Micro-millimetres.	Breadth. Micro-millimetres.
Canavalia,	240	20
Parkia,	150	15
Lupine,	120	15
Pea,	100	12
Vetch,	75	6
Astragalus,	150	20
Cassia,	60	6
Cicer,	100-300	25
Bean,	45	15
Lentils,	40	6
Soya,	60	15

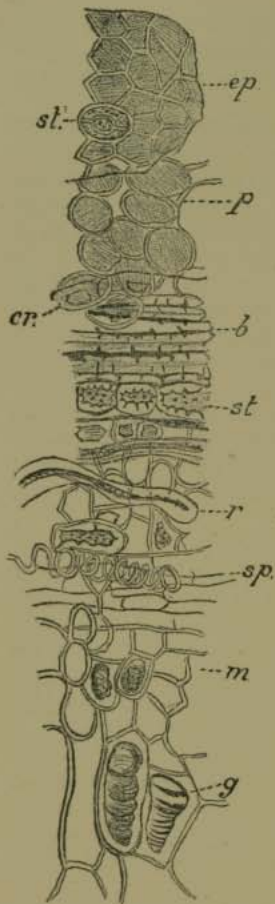


FIG. 56.—A section of the carob-tree fruit, $\times 160$ —*ep*, epidermis superficial view, showing a stoma, *st'*; *p*, brown parenchyma; *cr*, cells containing crystals; *b*, bent fibres; *st*, hard cells, thickened by deposit; *r*, a layer of thin walled cells, but scattered throughout are fibres and thickened hard cells; *m*, the middle layer containing spirals, *sp*, and the peculiar bodies, *g*, described in the text.

The second characteristic is a single layer of cells which support the palisade layer; these cells in form suggest various similes: one observer sees in them 'pillars'; another, 'hour-glasses'; a third, 'reels of cotton' (see fig. 59 s), and, indeed, they are more or less like all these. Their peculiar shape is only to be fully made out in section; looked at from above, they are merely polygonal or many-sided cells. These little pillars have very different shapes in different species, and afford a valuable means of diagnosis. In the bean, they are prismatic, contain one or two large monoclinic crystals, and form no intercellular substance. In soya, lupine seed, and canavalia, the pillar-cells are as high as broad, and very much like cotton reels; canavalia is peculiar in possessing several layers of them. In the lentil, again, they are broader than high, cone-like, and when looked at from above have a regular five- to six-sided outline with an inscribed circle; the lumen is filled with a granular dark or greenish mass.

§ 252. The seeds of *Cassia occidentalis*² have been, to some extent, used as an adulterant, and as a substitute for coffee.

¹ Moeller, *Mikroskopie der Nahrungs- u. Genussmittel*.

² Holler: *Dingl. Pol. Journ.*, cccxxvii., p. 61; cccxxviii., p. 164.

In Germany the ground and roasted seeds have been sold under the name of 'Mogdad' coffee, and it is said that neither by the taste nor by the general appearance can the addition of cassia seeds be detected, if such addition does not exceed one-fifth of the weight of the coffee. The seeds are small, flattened, oval, smooth, marked on each side of the two flattened

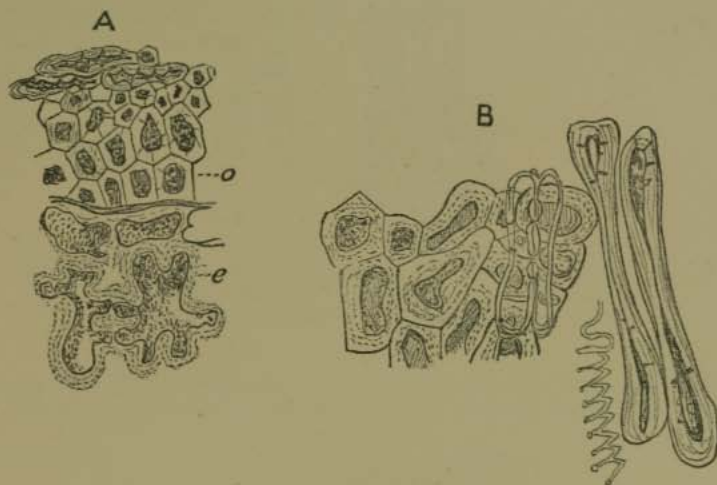


FIG. 57.—A, Section through the vegetable ivory nut—*o*, outer coating of the seed; *e*, endosperm, composed of thickened porous cells.
B, Individual fibres separated from *o*, the outer coating of the seed, $\times 160$ (after MOELLER).

surfaces with a slight circular groove or depression; when magnified the surface of the seed is somewhat tuberculated.

The integuments are wonderfully hard and leathery, and in the fresh state most difficult to grind or cut; they are, indeed, about the consistence

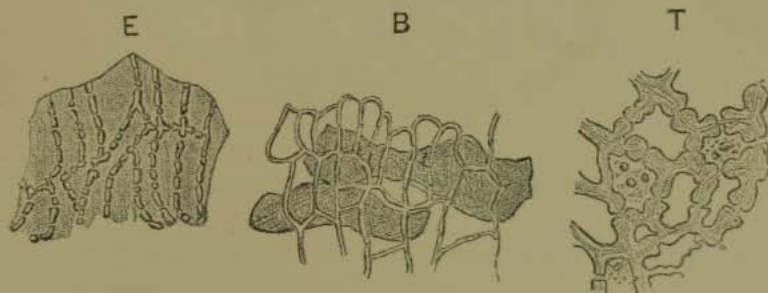


FIG. 58.—Tissues of the date-stone, $\times 160$ —E, endosperm; B, epidermis; T, irregularly shaped tubular cells filled with tannin-like substances.

of the leathery seeds of *Nux vomica*. The microscopical structure is very distinctive; the covering of the seed has first a layer of hard tissue, with fine striæ-like, perpendicular tubes radiating from the centre towards the circumference (palisade layer). Beneath this palisade or dentine layer

are 'the pillar-cells,' spool-shaped, as broad as high, and very thick-walled, and these pass into some thick-walled four- to five-sided oblong cells, filled with an orange-red colouring-matter. Within the coloured cells are oval,

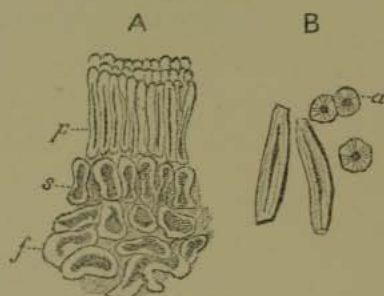


FIG. 59.—A, A section of a seed of *Parkia*—*p*, the 'palisade' layer; *s*, the pillar-like cells; *f*, spongy parenchyma, $\times 160$ (after MOELLER).
B, Isolated 'palisade' cells—*a*, the apex alone showing.

round, or angular cells (according to the pressure), filled with granular matter, and making up most of the substance of the seed (see fig. 60).

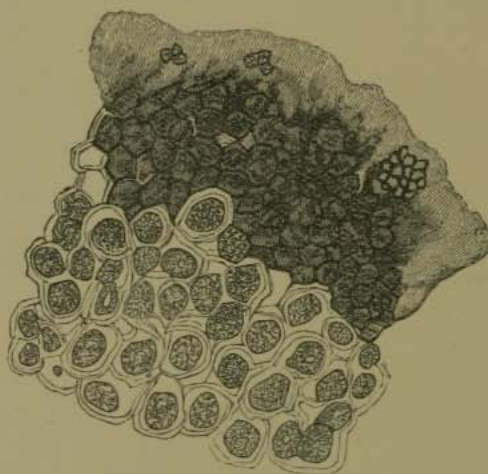


FIG. 60.—Section of seed of *Cassia occidentalis*, $\times 170$.

The composition of Mogdad coffee is as follows :—

Cellulose,	21.21
Fatty oil,	2.55
Plant mucus,	36.60
Astringent substance, gallo-tannic acid,	5.23
Inorganic salts,	4.33 ¹
Nitrogenous organic matters and loss,	15.13
Nitrogen-free organic matter,	3.86
Water,	11.09

¹ The seeds of *Cassia occidentalis* give 10 per cent. of ash.

CHEMICAL METHODS OF DETECTING ADULTERATIONS.

§ 253. When chicory is mixed with coffee, the chemical composition of the mixture shows, in some particulars, a marked deviation from that of pure coffee.

Letheby's analysis of chicory is as follows:—

	Raw root.	Kiln dried.
Hygroscopic moisture,	77.0	15.0
Gummy matter,	7.5	20.8
Glucose or grape sugar,	1.1	10.5
Bitter extractive,	4.0	19.3
Fatty matters,	0.6	1.9
Cellulose, inuline, and woody matter,	9.0	29.5
Ash,	0.8	3.0

Composition of the roasted root:—

	(1.)	(2.)
Hygroscopic moisture,	14.5	12.8
Gummy matter,	9.5	14.9
Glucose,	12.2	10.4
Matter like burnt sugar,	29.1	24.4
Fatty matter,	2.0	2.2
Brown or burnt woody matter,	28.4	28.5
Ash,	4.3	6.8

The ash of these had the following composition:—

Chloride of potassium,	0.22	0.45
Sulphate of "	0.97	0.98
Phosphate of "	1.41	1.37
" " magnesia,	0.90	0.53
" " lime,	0.40	0.81
Carbonate of "	0.10	0.26
Alumina and oxide of iron,	0.20	0.20
Sand,	0.70	2.20

Chicory influences the composition of coffee as follows:—

(1.) It decreases the gum, the latter seldom rising in chicory to more than 15 per cent., whilst in coffee it has not been found less than from 21 up to 28 per cent.

(2.) It increases in sugar, roasted coffee having seldom so much as 2 per cent. of sugar; whilst chicory, when roasted, usually has at least 8 or 9 per cent.

(3.) It decreases the fatty matter, the fat of chicory ranging from 1 to over 2 per cent., that of coffee from about 14 per cent. up to over 20 per cent.

(4.) It decreases the tannin and caffen-tannic acids, chicory being destitute of tannin.

(5.) It decreases the theine, chicory possessing none.

(6.) It profoundly modifies the constitution of the ash, especially by introducing silica, which is not a component of coffee ash. The main differences are thus as follow:—

	Coffee Ash. Per cent.	Chicory Ash. Per cent.
Silica and sand,		10.69 to 35.88
Carbonic acid,	14.92	1.78 to 3.19
Sesquioxide of iron,	0.44 to 0.98	3.13 to 5.32
Chlorine,	0.26 to 1.11	3.28 to 4.93
Phosphoric acid,	10.00 to 11.00	5.00 to 6.00
Total soluble ash,	75.00 to 85.00	21.00 to 35.00

Some approximate idea (although no accurate results) may be obtained of the amount of chicory present, if a careful determination of the soluble ash is made. If we take the soluble ash of chicory as 1.74, and that of coffee as 3 per cent., then Table XXXI. may be used.

TABLE XXXI.—GIVING THE THEORETICAL QUANTITY OF SOLUBLE ASH, CORRESPONDING TO VARIOUS ADMIXTURES OF CHICORY AND COFFEE.

Percentage of Chicory.	Percentage of Soluble Ash.	Percentage of Chicory.	Percentage of Soluble Ash.
5	2.94	50	2.58
10	2.88	55	2.56
15	2.82	60	2.52
20	2.79	65	2.50
25	2.74	70	2.48
30	2.70	75	2.46
35	2.67	80	2.43
40	2.64	85	2.41
45	2.60	90	2.40

By making standard infusions of pure coffee and chicory, and then taking the same weight of the suspected coffee, and comparing the colour, it is possible to obtain some idea of the quantity of chicory added.

Leebody recommends this to be done as follows:—Take 1 grm. of the unknown mixture, and 1 grm. of a standard mixture of equal parts of chicory and coffee; remove all the colouring-matter from each sample, and make the extract of each up to the same bulk. Put 50 c.c. of the filtered extract from the *unknown mixture* in the Nessler cylinder, and determine by trial how many c.c. of the extract from the *standard mixture*, together with sufficient distilled water to make up the 50 c.c., will give the same colour. In calculating the percentage of chicory present, closely accurate results are obtained in practice by assuming the tinctorial power of chicory to be three times that of coffee.—(J. R. LEEBODY, *Chemical News*, xxx., 243.)

Messrs. Graham, Stenhouse, and Campbell proposed to take the density of different infusions of coffee, etc., as a guide to its adulteration; and this is found in practice to work tolerably well, and to give approximative results. The following solutions were made by them by first treating the powder of the roasted substance with ten times its weight of cold water, boiling, and filtering, and determining the density at 60° Fahr.:—

Spent tan,	1002.14	Carrots,	1017.10
Lupine seed,	1005.70	Bouka,	1018.50
Acorns,	1007.30	Black malt,	1021.20
Peas,	1007.30	Turnips,	1021.40
Mocha coffee,	1008.00	Rye-meal,	1021.60
Beans,	1008.40	Dandelion root,	1021.90
Neilgherry coffee,	1008.40	Red-beet,	1022.10
Plantation Ceylon coffee,	1008.70	English chicory,	1021.70
Java coffee,	1008.70	Yorkshire chicory,	1019.10
Jamaica coffee,	1008.70	Foreign chicory,	1022.60
Costa Rica coffee,	1009.00	Guernsey chicory,	1023.26
"	1009.05	Mangel-Wurzel,	1023.50
"	1009.00	Maize,	1025.30
Native Ceylon coffee,	1010.90	Bread raspings,	1026.30
Brown malt,	1010.90		
Parsnips,	1014.30		

It has been shown by Hehner and Sketchly¹ that a determination of furfural in chicory and coffee may be of use. If the furfural be calculated into pentosans, then about double the quantity of pentosan is yielded by coffee as by chicory.

The method to be pursued is as follows:—

From 3 to 4 grms. of the substance are placed in a suitable distilling flask provided with a tap funnel, and 100 c.c. of dilute hydrochloric acid (strength 12 per cent.) added. The liquid is distilled, and when 30 c.c. of the distillate are collected, 30 more c.c. of the acid are added through the tap funnel, and so on until the distillate measures about 400 c.c. The progress of the reaction is traced by testing for furfural by taking a drop of the distillate on filter paper and touching the drop with a solution of aniline acetate containing sodium acetate. The presence of furfural is denoted by a pink colour. On adding a solution of pure phloroglucinol to the distillate and allowing to stand overnight, a black precipitate forms of furfural-phloroglucoside. This is collected on a weighed filter paper, washed and dried. The weight divided by 1.82 = furfural. This furfural may be calculated into pentosan by subtracting 0.0804 and multiplying the remainder by 1.88.

Genuine roasted coffee gave 2.5 per cent. of pentosan; coffee containing 32 per cent. chicory gave 2.71 per cent. The same sample of chicory roasted for different lengths of time gave from 4.8 per cent. to 5.56 per cent. of pentosan.

When the microscope has detected chicory, and it is certain that nothing but chicory is present, the most reliable guide is the density of the infusion. Of the strength given, if the specific gravity of coffee infusion be taken as 1008.5 and that of chicory as 1023.0, then the following table, from the authors' experiments, will be a fair guide to the amount of adulteration. The error usually lies within 3 per cent., while with calculations based on soluble ash the possible error is higher. It hence follows that the analyst must be very careful in his statements as to the percentage of chicory. The certificate should say "*about . . . per cent.*;" or supposing that from the soluble ash and from the specific gravity, as well as from the microscopical examination, a coffee seems to contain 40 per cent. of chicory, it will be safe to subtract 5 per cent. for error, and give the amount as "*at least 35 per cent.*"

§ 254. W. L. Hiepe, taking into consideration the fact that pure coffee has .03 per cent. of chlorine, while chicory has .28 per cent. of chlorine, has proposed to calculate the percentage of mixtures on these data. If this method should be accepted, it will necessitate a most careful incineration; for in the majority of ordinary cases involving ash-taking, two-thirds of the chlorine is volatilised.

Prunier, again, has attempted to determine the coffee directly by weight: 2 grms. of the mixture of coffee and chicory are weighed out, and the finer powder is separated by sifting through fine silk. This is composed entirely of coffee, as may be proved by microscopic examination. That which remains on the silk sieve is moistened with water in a test glass; after some hours it is thrown upon a piece of stretched cloth, and crushed with the fingers. The grains of coffee resist the pressure, whilst those of chicory penetrate under these circumstances into the cloth, and adhere to it. The cloth is dried, and it is then easy to detach the coffee, which is added to

¹ *Analyst*, vol. xxiv., 178.

the fine powder from the first operation, and weighed after complete drying; the chicory is calculated from the loss.

TABLE XXXII.—GIVING THE APPROXIMATE PERCENTAGE OF CHICORY IN A DECOCTION OF COFFEE AND CHICORY.

Specific Gravity.	Percentage of Coffee in Mixture.	Specific Gravity.	Percentage of Coffee in Mixture.	Specific Gravity.	Percentage of Coffee in Mixture.	Specific Gravity.	Percentage of Coffee in Mixture.
1009.0	96.55	1011.6	78.6	1014.2	60.7	1016.8	42.8
1009.1	95.8	1011.7	77.9	1014.3	60.0	1016.9	42.1
1009.2	95.1	1011.8	77.2	1014.4	59.4	1017.0	41.4
1009.3	94.4	1011.9	76.5	1014.5	58.7	1017.1	40.7
1009.4	93.8	1012.0	75.7	1014.6	58.0	1017.2	40.0
1009.5	93.1	1012.1	75.1	1014.7	57.3	1017.3	39.3
1009.6	92.4	1012.2	74.5	1014.8	56.6	1017.4	38.6
1009.7	91.8	1012.3	73.8	1014.9	55.9	1017.5	37.9
1009.8	91.0	1012.4	73.1	1015.0	55.2	1017.6	37.3
1009.9	90.3	1012.5	72.4	1015.1	54.5	1017.7	36.6
1010.0	89.6	1012.6	71.7	1015.2	53.8	1017.8	35.9
1010.1	88.9	1012.7	71.0	1015.3	53.1	1017.9	35.2
1010.2	88.2	1012.8	70.3	1015.4	52.4	1018.0	34.5
1010.3	87.5	1012.9	69.6	1015.5	51.8	1018.1	33.8
1010.4	86.8	1013.0	68.9	1015.6	51.1	1018.2	33.1
1010.5	86.1	1013.1	68.3	1015.7	50.4	1018.3	32.4
1010.6	85.5	1013.2	67.6	1015.8	49.7	1018.4	31.7
1010.7	84.8	1013.3	66.9	1015.9	49.0	1018.5	31.0
1010.8	84.1	1013.4	66.3	1016.0	48.3	1018.6	30.4
1010.9	83.5	1013.5	65.6	1016.1	47.6	1018.7	29.7
1011.0	82.8	1013.6	64.9	1016.2	46.9	1018.8	29.0
1011.1	82.1	1013.7	64.2	1016.3	46.2	1018.9	28.3
1011.2	81.4	1013.8	63.5	1016.4	45.5	1019.0	27.6
1011.3	80.7	1013.9	62.8	1016.5	44.8		
1011.4	80.0	1014.0	62.1	1016.6	44.2		
1011.5	79.3	1014.1	61.4	1016.7	43.5		

H. Hager's investigations into coffee adulteration may be detailed as follows:—To examine the unroasted coffee for artificial colouring-matters, he treats the berries with cold water; when, if the berries are in their natural state, the water is scarcely coloured. 50 grms. are next macerated with water, to which 1 per cent. of nitric acid has been added, and then hydric sulphide is passed through the filtrate; from this solution chloroform will extract indigo if present. Berlin-blue and alkanet pigment may be dissolved out from the berries by carbonate of potash solution, and then precipitated by hydrochloric acid. He finds also that when thrown into water, imitation or artificial berries will sink to the bottom, while good berries swim. On treating 3 grms. of powdered coffee with 20 grms. of cold water, and filtering, after the lapse of half an hour the filtrate should be feebly yellow, and not taste in the least degree bitter; in presence of lupine-seeds the taste is markedly bitter.

With regard to the 'swimming test,' he recommends a saturated solution of rock salt. 2 grms. of the coffee are placed in a narrow test cylinder with 15 to 20 c.c. of the cold saline solution; the coffee is shaken up with this, and then allowed to stand for an hour; after this time the coffee swims to the surface, and the water remains uncoloured. Lupine-seeds generally colour the salt solution yellow, and give a strong deposit. The

filtrate from pure coffee gives no precipitate with picric acid, tannin, iodine, or alkaline solution of copper. Ferric chloride strikes a green colour with false coffee; with starches iodine strikes a blue colour; with astringent matters, ferric chloride a black colour; if sugar from chicory, dates, etc., is present, alkaline copper solution is reduced.

Lupine-seeds give, when extracted by weak sulphuric acid water, only a slight turbidity with mercury potassic iodide. Coffee, on the contrary, under the same circumstances, gives a strong turbidity; but if this is doubtful, the theine can be extracted from the solution by shaking it with chloroform or benzole, which dissolves the theine, but leaves the lupine.

Hager has also a different method of taking the 'extract': 10 grms. of coffee, 1 gm. of oxalic acid, and 80 c.c. of water are mixed by shaking, and digested at 100° C. for 3 hours, filtered, and washed with water until the filtrate is no longer coloured. The filtrate is evaporated to dryness. Pure coffee at the most yields in this manner 2.5 to 3 grms. of extract (including the oxalic acid), while chicory gives 5 to 7 grms., and other substances similarly much increase the extract. His reason for using oxalic acid is because of its changing starch into dextrin, and quickening the filtration.

In regard to other adulterations, a great variety of starch-holding substances, with the cereals, may be entirely excluded, as certainly not present, if no dirty-blue or violet colouration is produced by iodine in an infusion of coffee. In order to apply this test properly, the infusion should be decolorised, which is most rapidly done by a solution of permanganate of potash. Coffee itself, as before stated, contains no starch.

Burnt sugar, or caramel, is usually detected by observing the rapid darkening of water on which a little coffee is sprinkled, and the particles (on examination in water by the microscope) reveal themselves by the absence of organised structure, and the coloured ring, arising from partial solution, round each.

COCOA AND CHOCOLATE.

§ 255. The cocoa of commerce is made from the roasted seeds of the *Theobroma cacao*, a tree belonging to the natural order *Byttneriaceae*, whole forests of which exist in Demerara. It is also more or less extensively grown in Central America, Brazil, Peru, Caraccas, Venezuela, Ecuador, Grenada, Essequibo, Guayaquil, Surinam, and some of the West Indian Islands; and its cultivation has also been attempted (in most cases successfully) in the East Indies, Australia, the Philippine Islands, the Mauritius, Madagascar, and Bourbon.

The principal kinds of cocoa in commerce are known under the names of Caraccas, Surinam, Trinidad, Grenada, Jamaica, Dominica, Guayaquil, Venezuela, Bahia, Brazil, and St. Lucia. The seeds are officinal in the French and Norwegian pharmacopœias. They are ovate, flattened, 2 to 2½ cm. [.7 to .9 inch] long, and 1 to 1½ cm. [.39 to .58 inch] broad, and covered with a thin red or grey-brown friable shell. The taste of the fresh seed is oily, bitter, and rather unpleasant.

The seeds, on being submitted to a kind of fermentation (technically called the *sweating* process), lose in a great measure this disagreeable flavour, and develop an aromatic smell. Seeds which have been subjected to this treatment are best suited for the manufacture of chocolate, while those which have been simply roasted are richer in cocoa-butter.

The cocoas of commerce are deprived of the husk, and consist of the

ground or entire cotyledons. The cotyledons are made up of a parenchyma of small, polygonal, thin-walled cells (fig. 61). Within these cells are the very small starch granules, measuring about 5 micro-millimetres [$\cdot 002$ inch]. In some of the cells the starch is in little lumps, mixed with fat and albuminous matters. The starch is distinguished from rice-starch by the granules being round and not angular. It possesses considerable resistance to hot water, and the reaction with iodine is slow—a quick-blue colour being only brought out by the addition of the chlor-iodide of zinc. Careful measurements made by Éwell give the maximum diameter as $0\cdot 012$ mm., minimum $0\cdot 002$ mm., average about $0\cdot 006$ mm. Some of the cells contain 'cocoa red,' which is coloured blue by a solution of ferric chloride, and dissolved by strong potash to a green, by dilute sulphuric acid to a red, and by acetic acid and alcohol to a violet solution; but the fresh seeds are colourless.

The cotyledons are covered with a thin glass-clear epidermis, consisting of at least two layers of cells, and where it folds between the cotyledons of several layers.¹ These are very transparent, they often contain crystals of theobromin, and other crystals supposed to be those of a fatty acid. Attached to the epidermis are to be found curious club-shaped hairs (fig. 61).

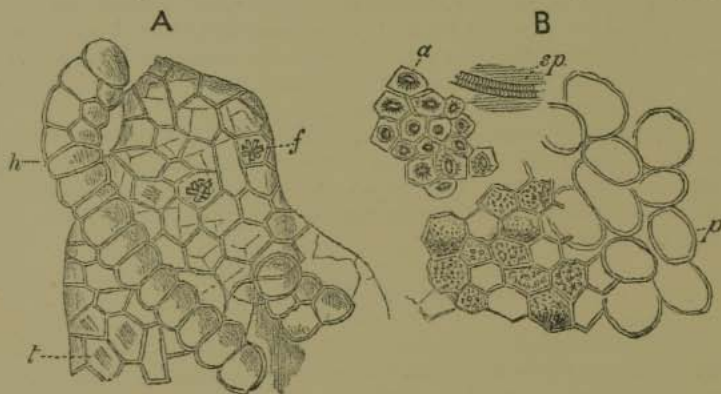


FIG. 61.—Tissues of cocoa, $\times 160$. A, Epidermis, beneath which are seen cells containing two different kinds of crystalline cell contents; *f*, crystals of fatty acids; *t*, crystals of theobromin; *h*, a hair. B, The tissues of the seed proper; *sp*, spiral vessels; *a*, thickened cells; *p*, parenchyma, some of the cells of which are filled with various cell contents.

To examine ground cocoa microscopically, if not already fat free, it must be exhausted with ether—the finer particles may then be examined in glycerin and water for foreign starches, and lastly, the dark opaque portions cleared up by treating with a strong solution of potash. Unless methods of the above kind are adopted, but little can be made out from a microscopical examination.

§ 256. The commercial varieties of cocoa are very numerous:—*Cocoa nibs* are simply the bruised, roasted seeds deprived of their coverings; and *flake cocoa* is composed of the nibs ground in a particular form of mill. The cocoa powders may be divided into those mixed with starches and

¹ The chief distinguishing marks of the husk are a large-celled epidermis, and small thick-walled cells; but, unless as an adulteration, the husk will not occur in ordinary cocoa.

sugar and partly deprived of fat, and those which, like the Dutch cocoas, have been treated with salts of soda, potash, or ammonia, with the idea of softening the cocoa fibre. These latter contain no added starch. Some of them have not been deprived of fat. Another method of manufacture is to soften the fibre by heating in closed vessels with water to 150° C.

Epps's cocoa, according to an analysis advanced as evidence in the case of *Gibson v. Leifer*, is composed of cocoa 40 per cent., sugar 44 per cent., and starch 16 per cent.

Granulated cocoa is mostly a mixture of nibs, arrow-root, and sugar; *Homœopathic cocoa*, a preparation of the same kind without the sugar; *Maravilla cocoa* contains sugar and much sago flour; and *cocoa essence*, *cocoatine*, etc., consist of pure cocoa deprived of 60 to 70 per cent. of its fat.

§ 257. *Chocolate*.—In the manufacture of chocolate the cocoa-nibs are ground in a mill, the rollers of which are usually heated by steam, so as to soften the cocoa-butter; and in this way a paste is formed which is mixed with refined sugar, and very often other substances, and pressed into moulds. Some of the recipes for chocolate are as follows:—

(1.) *French Chocolate*.—Two beans of Vanilla rubbed into a powder with sugar, and 1 lb. of best sugar to every 3 lbs. of cocoa-nibs.

(2.) *Spanish Chocolate*.—(a.) Curaçoa cocoa 11, sugar 3, Vanilla $\frac{1}{16}$, cinnamon $\frac{1}{16}$, cloves $\frac{1}{128}$. (b.) Caraccas cocoa 10, sweet almonds 1, sugar 3, Vanilla $\frac{1}{84}$.

(3.) *Vanilla Chocolate*.—A chocolate paste highly flavoured with Vanilla, and generally with other spices as well. (a.) Caraccas cocoa 7, Mexican Vanilla $\frac{1}{16}$, cinnamon $\frac{1}{32}$, and sufficient cloves to flavour. (b.) Best chocolate paste 21, Vanilla 4, cinnamon $\frac{1}{8}$, cloves and musk in small quantities.

The chocolates of English commerce yield but little cocoa-butter, since they are mostly prepared from the cake left after expression of the oil.

§ 258. The average chemical composition of cocoa, according to some English analyses, is as follows:—

	J. A. Wanklyn.	J. Muter.
Cocoa butter,	50·00	42·94
Theobromin,	1·50	·90
Starch,	10·00	19·03
Albumen, fibrin, and gluten,	18·00	12·21
Gum,	8·00	6·40
Colouring-matter,	2·60	3·69
Water,	6·00	5·98
Ash,	3·60	2·90
Loss, etc.,	0·30	...
Cellulose,	5·95
	100·00	100·00

The peculiar constituents of cocoa are three—viz., cocoa-butter, theobromin, and cocoa red.

Cocoa or Cacao-butter, Oleum theobromæ.—This fat is a yellowish-white concrete vegetable fat of about the consistency of tallow. Its specific gravity at 15° C. is from 0·95 to 0·96; and at 100° C. about 0·856. The melting-point is from 28° to 32° C., the solidifying-point of the fat from 21° to 23° C., while the mixed fatty acids melt at from 48° to 50° C., and solidify at from 45° to 47° C. It has a definite Reichert value, the distillate being sufficiently acid to use from 1 to 2 c.c. of decinormal alkali. The insoluble fatty acids are somewhat low, about 94 to 95 per cent.; the saponification

value is from 193 to 195, and the iodine value from 32 to 38. The iodine value of the fatty acids, according to de Negri and Fabris, is 39.1.

The acid value of commercial samples, according to Dieterich, is from 1.0 to 2.3; samples slowly increase through age in acidity, the absolutely fresh butter only requiring from 0.06 to 0.22 c.c. of decinormal alkali.

The above constants are explained by the composition of cocoa-butter; it consists chiefly of the glycerides of stearic, palmitic, and lauric acids, with small quantities of linoleic, arachidic, formic, acetic, and butyric acids. Cocoa-butter is soluble in five parts of absolute alcohol, but insoluble in 90 per cent. alcohol.

It is often adulterated, particularly with tallow, almond, arachis, sesamé, and cocoa-nut oils, beeswax, stearic acid, and paraffin wax.

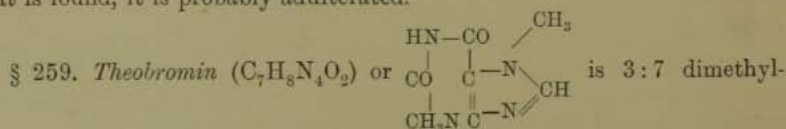
Indications of such adulterations will be afforded by a determination of the constants given above. Thus a low saponification value suggests beeswax or paraffin wax. A high iodine value suggests some of the vegetable oils, such as sesamé, arachis, or almond. As regards tallow there are two special tests, Björklund's test as modified by Filsinger and Hager's aniline test.

Björklund's Test.—The first test is performed as follows:—2 grms. of the sample are melted in a test-tube and agitated with 6 c.c. of a mixture of four parts of ether (specific gravity 0.725) and one part of alcohol (specific gravity 0.810); should tallow be absent the solution remains clear; otherwise it becomes turbid, especially if the solution is cooled to 0°.

Hager's Aniline Test.—This is a test for not only tallow, but several other fats. A gramme of the sample is warmed with 2.8 grms. of aniline until it dissolves, and then allowed to stand for an hour at 15°, or, at higher temperatures, longer. Pure cocoa-butter forms a layer on the top which does not solidify for hours, but if it has been adulterated with tallow, stearic acid, or small quantities of paraffin wax, granules appear in the oily layer; if large quantities of stearic acid or paraffin wax are present, the whole may become a solid crystalline mass. A rough quantitative determination may be made by comparing standard mixtures of the adulterant found and in any case the test should be confirmed by getting out the constants. Cocoa-butter is stated to be replaced in some cocoas by mixtures of fats; but this is rare; at all events, no prosecution for such has been instituted in England.

Cocoa-fat is readily soluble in either ether or petroleum ether. It may be determined by exhausting a weighed portion in a Soxhlet tube with either solvent, and since cocoa or cocoa mixtures contain a good percentage of fat, small quantities may be taken, such as 5 grms., for this determination.

Cocoa should contain at least 20 per cent. of cocoa fat; if less than that is found, it is probably adulterated.¹



xanthin; it may be transformed into theine by dissolving in ammonia, precipitating the ammoniacal solution by silver nitrate, and heating the crystalline precipitate with methyl-iodide. Theobromin was discovered in 1841 by Woskresensky, in the seeds of the *Theobroma cacao*; it principally

¹ 20 per cent. is the standard of the Society of Analysts; but this, according to published analyses, is much too low.

resides in the cotyledons, and in smaller quantities in the seed coverings. The average yield of theobromin appears to be $1\frac{1}{2}$ per cent.

The original method of separation pursued by Woskresensky was—extraction on the water-bath with distilled water, filtering through linen, precipitating with sugar of lead, re-filtering, freeing the filtrate from excess of lead by SH_2 , evaporating to dryness, and subsequent purifying of the residue by solution in spirit, and treatment with animal charcoal. Mitscherlich, again, boils the cocoa with a weak solution of sulphuric acid in order to change the starch into sugar, saturates the fluid with carbonate of lead, and ferments it with yeast to destroy the sugar. On the conclusion of the fermentation, the fluid is boiled, neutralised with soda, filtered, concentrated by evaporation, and the impure brown theobromin which separates boiled in hot nitric acid. This nitric acid solution is precipitated by ammonia, again dissolved in nitric acid, and the nitrate obtained by evaporation. According to Mitscherlich, the quantity obtained in this way is much greater than by other processes.

Wm. E. Kunze¹ has made an exhaustive study of the published processes for the separation and estimation of theobromin and theine in cocoa of Weigmann, Mulder, Wolfram, Legler, Trojanowsky, Zipperer, Süß, Diesing, and James Bell, and finds them all imperfect or inaccurate; and after much research, he has adopted the following, which, from test analyses, appears to give results of great accuracy:—

10 grms. of cocoa are boiled with 150 c.c. of 5 per cent. sulphuric acid for 20 minutes, the fluid is filtered, and the residue well washed with boiling water. While warm the mixture is precipitated by phosphomolybdic acid,² and after standing 24 hours the precipitate is filtered and washed with much 5 per cent. sulphuric acid. The precipitate while still moist is transferred to a flask, and decomposed with baryta water. The alkaline fluid is then saturated with CO_2 and evaporated to dryness. The dry mass is transferred to a flask, and exhausted by boiling chloroform, the flask for this purpose being connected with an inverted condenser. The chloroform extract is placed in a tared flask, the chloroform distilled off, and the residue dried and weighed. The weight equals theobromin + theine, with a little mineral matter, so small that, in technical analyses, it may be neglected. If it is desired to estimate these crystalline substances separately, the residue after weighing is dissolved in a little water, ammonia added, and to the ammoniacal solution silver nitrate added. On prolonged boiling the crystalline silver theobromin falls down ($\text{C}_7\text{H}_7\text{AgN}_4\text{O}_2$) containing 37.488 per cent. Ag. This may be weighed, or dissolved in dilute nitric acid, and the silver precipitated as chloride. Kunze, however, prefers a volumetric process of separation. He adds a measured quantity of silver nitrate solution of known strength (say 5 per cent.), and after filtration, titrates the unused silver by ammonium sulphocyanide—by multiplying the silver thus found by difference in the precipitate by the factor 1.66 (for 108 silver = 180 theobromin) the weight of the theobromin is obtained, and this weight subtracted from the combined weight of theobromin and theine gives, of course, the theine.

Theobromin forms microscopic rhombic needles. It is generally thought

¹ *Zeit. f. analyt. Chem.*, 1894, 1. See also Brunner and Heinrich Leins, *Schweiz. Wochen. f. Chem. u. Pharm.*, March, 1893.

² Prepared by precipitating ammonium molybdate, by sodic phosphate, suspending the well-washed precipitate in water, and adding sodium carbonate, and warming until dissolved. The solution is evaporated to dryness and ignited. The ignited residue is powdered, mixed with nitric acid into a paste, and again ignited. The residue is warmed with water, nitric acid added to strong acid reaction, and diluted to 10 per cent.; the solution is warmed for some time and ultimately filtered; the result should be a clear yellow liquid.

to sublime between 296° and 295° C. without decomposition, but this temperature is many degrees too high. The senior writer finds that a minute fragment, placed in the subliming cell elsewhere described, begins to give fine nebulae at 134° C., and on examining the mists by a high power, they are resolved into extremely minute dots; distinct crystals are obtained at temperatures of 170° C. and above. Theobromin is insoluble in petroleum ether, and not very soluble in ether, one part requiring 600 parts of boiling and 1700 parts of cold ether. It is soluble in alcohol, 1 part requiring 47 parts of boiling and 1460 of cold alcohol. Its solubility in water is stated to be 1 in 55 parts at 100° C., 1 in 600 parts at 20° C., and 1 in 1600 at 0° C. It is somewhat soluble in chloroform and warm amyl-alcohol, but with difficulty soluble in benzole.

Theobromin¹ forms easily crystallisable salts. The simple neutral salts are decomposed by water, with the formation of basic salts, and lose their acid, if it is volatile, at 100° C. A hydrochloride of theobromin, $C_7H_8N_4O_2, HCl$; a nitrate, $C_7H_8N_4O_2, NHO_3$; a platinum salt, $C_7H_8N_4O_2, HClPtCl_2 + 2H_2O$; are all very definite crystalline compounds. A noteworthy salt is that with nitrate of silver, which is formed by adding a solution of argentic nitrate to a solution of nitrate of theobromin; in a short time there separate silver-white needles, very insoluble in water, of the composition $C_7H_8N_4O_2, NHO_3 + AgNO_3$.

The other precipitants of theobromin are—phospho-molybdic acid (yellow) and chloride of gold (long needles). Tannic and picric acids only produce turbidity, while potass. mercuric iodide and potass. cadmium iodide do not precipitate. A characteristic reaction of theobromin is that produced by peroxide of lead and sulphuric acid. If peroxide of lead and diluted sulphuric acid are heated with theobromin, avoiding an excess of the oxidising agent, CO_2 is developed, and if filtered from lead sulphate the filtrate gives off ammonia with potash, separates sulphur on treatment with SH_2 , stains the skin purple-red, and colours magnesia indigo-blue.

Theobromin is poisonous to kittens (and other animals of similar size) in such large doses as a gramme. It appears to be separated by the kidneys, and could probably be discovered in the urine of any person taking large quantities of cocoa. The method of research successfully used by Mitscherlich is as follows:—The urine is acidified with HCl, filtered, and to the filtrate, acidified with nitric acid, a solution of phospho-molybdate of soda is added. The precipitate is collected, and treated with baryta water until it is strongly alkaline, warmed, filtered, and the filtrate evaporated; the residue extracted with alcohol, refiltered, and the filtrate again evaporated. This last residue is dissolved in a drop of hydrochloric acid, and precipitated by ammonia. The alkaloid may now be collected, and, if necessary, purified.

§ 260. Cocoa red is an astringent colouring-matter found in cocoa. On saponification it breaks up into glucose, tannin, resin, and the ill-defined brown amorphous substance to which has been given the name of phlobaphene.

When cocoa is freed from fat the red colouring-matter is only partially extracted by solvents, unless a mineral acid has been added. By adding a few cubic centimetres of hydrochloric acid and gently warming for a few seconds, the red colouring-matter is dissolved with great ease by amyl alcohol, as well as by ethyl alcohol. It can be in part shaken out of an aqueous solution by amyl alcohol. It is insoluble in ether or petroleum ether, but dissolves to a slight extent in carbon bisulphide. The best way

¹ E. Fischer (*Ber. d. d. chem. Gesells.*, xv., 453-456) has transformed xanthine, $C_5H_4N_4O_2$, into theobromin by heating lead xanthinate, dried at 130° in a sealed tube, at 100° with $\frac{1}{2}$ its weight of methyl iodide.

to extract cocoa red is, apparently, as follows:—From 2 to 3 grms. of the fat-free cocoa are made into a paste with hydrochloric acid, the acid paste put into a Soxhlet tube and exhausted by 100 c.c. of absolute alcohol, the alcohol being heated in a flask standing in a beaker of boiling water. Before beginning the extraction, sufficient silver oxide is added to the alcohol in the flask to fix the hydrochloric acid. The alcoholic liquid is cooled, filtered, and then precipitated by an alcoholic solution of lead acetate. The precipitate is of a purple black. It is collected on a filter, well washed with boiling water, and then transferred to a small flask, some 70 per cent. alcohol added and the lead salt decomposed by SH_2 ; on getting rid of the SH_2 , filtering and evaporating to dryness, the red colouring-matter is obtained in a solid form, and by repetition of the process purification may be effected.

A solution of cocoa red obtained in this way gives a diffuse band in the green, allowing the red, blue, and most of the yellow rays to be transmitted. The solution in alcohol is capable of being estimated on colorimetric principles, but low results are obtained because the process always entails some decomposition.

If estimated by quantitative spectroscopy, the wave lengths between 590 and 578, between 578 and 565, and between 539 and 529 are the best adapted.

The absorption factors for these wave lengths are as follows:—

$\lambda 590$ to 578 ,	0.001332
$\lambda 578$,, 565 ,	0.00148
$\lambda 539$,, 529 ,	0.000995

Cocoa red is a sensitive reagent to acids and alkalis, alkalis generally striking a dirty green, mineral acids a red with a violet shimmer. The solution in water is bitter, and gives precipitates with the salts of iron, copper, and silver.

Zipperer's Method of Determining Cocoa Red and the Products of its Decomposition.—100 grms. of cocoa are exhausted of fat by petroleum ether. The residue is dried and then macerated for 8 days in a litre of absolute alcohol. The alcohol is filtered off, and most of the alcohol is recovered by distillation; in any case the alcohol is evaporated to an extract and weighed. This mainly consists of cocoa red with resin and phlobaphene, products of its decomposition; the weight may be designated by p . The cocoa which has been exhausted by petroleum ether and treated with alcohol is now macerated in a litre of water for two days, at the end of which period it is filtered. The filtered liquid is precipitated by four times its volume of absolute alcohol and again filtered. The filtrate is evaporated to an extract. The extract is treated with a definite quantity of water, and a fractional portion of this is precipitated with neutral acetate of copper; the tannate of copper is collected, dried, weighed, and incinerated, the loss of weight on incineration being returned as tannin; let this weight be denoted as p^1 ; then $p + p^1$ equals 'cocoa red.' The phlobaphene and resin may also be determined. To do this the alcoholic extract, p , after being weighed, is treated on a filter with acidulated water (which dissolves the tannin) and washed with water. The residue thus freed from tannin is washed with 200 c.c. of ammoniacal water (50 per cent.) which dissolves the phlobaphene; and the solution is filtered, evaporated to dryness, and weighed as phlobaphene. The residue freed from tannin and phlobaphene is dried and weighed and returned as resin. Zipperer obtained from a sample treated in this way, $p = 2.64$ per cent., $p^1 = 2.85$, phlobaphene 2.0,

resin 0.07; in other words, the total cocoa red was 5.49, the tannin glucoside being equal to the difference between the weight of the total cocoa red and the united weight of the phlobaphene and resin—that is to say, $5.49 - 2.07 = 3.42$ per cent.

Zipperer's method with its large quantities of absolute alcohol is expensive and time-consuming, and without a doubt could be simplified. With careful work certainly a tenth of the quantities may be used—that is to say, 10 grms. of cocoa instead of 100, and the solvent diminished in the same proportion.

§ 261. *Determination of Crude Fibre.*—A gramme of the substance is freed from fat by exhaustion in a Soxhlet tube with ether or petroleum ether. It is then boiled under an inverted condenser with 1.25 per cent. of dilute sulphuric acid; for this purpose from 100 to 150 c.c. of acid will be required; it is then filtered and thoroughly washed with hot water. The matter on the filter is next washed into a flask and boiled with 100 c.c. of 1.25 per cent. soda solution. This is filtered and the residue thoroughly washed with hot water and, lastly, with alcohol, collected on to a platinum dish, dried, weighed, and ignited.

§ 262. *The Ash.*—The composition of the ash of cocoa seeds is stated by Mr. Wanklyn to be as follows:—

COMPOSITION OF ASH OF COCOA SEEDS.		Per cent.
Potash,		29.81
Chloride of sodium,		6.10
Ferrous oxide,		1.60
Alumina,		2.40
Lime,		7.72
Magnesia,		7.90
Phosphoric acid,		24.28
Sulphuric acid,		1.92
Carbonic acid,		0.98
Silica,		5.00
Sand,		12.15

The percentages of ash found in cocoa are given as follows:—

	Percentage of Ash.
Common Trinidad,	3.27
Very fine Trinidad,	3.62
Fair, good, fine Trinidad,	3.64
Fine Grenada,	3.06
Caraccas,	4.58
Bahia (Brazil),	3.31
Fine Surinam (small),	3.06
Mexican,	4.27
Dominican,	2.82
African,	2.68
Mean of the ten being	3.43

Thus the lowest determination is 2.68, the highest 4.58 per cent. The nibs show a lower ash than the shell. The nibs of the Caraccas give 3.95 per cent. of ash, 2.00 being soluble and 1.95 insoluble in water. The nibs of Mexican seeds give 2.59 per cent. of ash, .89 part being soluble and 1.70 insoluble in water. The ash of the shell is rich in, but the nib almost devoid of, carbonates. The ash should in all cases be dissolved in a measured quantity of d. n. sulphuric acid, boiled to expel carbonic acid gas, and then, after adding litmus, titrated back with d. n. soda; the difference between the two results is a measure of the alkalinity of the ash, and is

called by the American chemists the "acid equivalent." Dutch cocoas and others which have been treated with alkalis show a high alkalinity. Mr. Heisch has examined the cocoas of commerce, with the results embodied in the following tables:—

TABLE XXXIII.—EXAMINATION OF ROASTED BEANS AFTER REMOVAL OF THE HUSK.

	Husk.	Fat.	Nitrogen.	Albuminoids.	Ash.	Ash soluble in Water.	Ash soluble in HCl.	H ₃ PO ₄ in Ash.	Moisture.	Starch, Gum, Cellulose, etc.
Caraccas, . . .	13·8	48·4	1·76	11·14	3·95	2·15	1·80	1·54	4·32	32·19
Trinidad, . . .	15·5	49·4	1·76	11·14	2·80	0·90	1·90	0·93	3·84	32·82
Surinam, . . .	15·5	54·4	1·76	11·14	2·35	0·80	1·85	1·23	3·76	28·35
Guayaquil, . . .	11·5	49·8	2·06	13·03	2·50	1·75	1·75	1·87	4·14	30·47
Grenada, . . .	14·6	45·6	1·96	12·40	2·40	0·60	1·80	1·35	3·90	35·70
Bahia, . . .	9·6	50·3	1·17	7·40	2·60	0·90	1·70	1·26	4·40	35·30
Cuba, . . .	12·0	45·3	1·37	8·67	2·90	0·95	1·95	1·13	3·72	39·41
Para, . . .	8·5	54·0	2·00	12·66	3·05	1·40	1·65	1·00	3·96	26·33

There are also some analyses by König as follows:—

TABLE XXXIV.—I. COCOA BEANS DEPRIVED OF THE SHELL.

	Water.	Nitrogenous Substances.	Fat.	Starch.	Other Nitrogen Free Matters.	Cellulose.	Ash.
1. Caraccas I., . . .	4·04	14·68	46·18	12·74	(18·50)	...	3·86
2. " II., . . .	4·72	14·06	49·36	13·99	9·46	4·20	4·21
3. Guayaquil I., . . .	3·63	14·68	49·64	11·56	12·64	4·13	3·72
4. " II., . . .	2·61	16·25	46·99	10·82	16·12	3·53	3·68
5. Trinidad I., . . .	2·81	15·06	48·32	14·91	12·06	3·62	3·22
6. " II., . . .	2·28	15·12	52·14	14·38	8·82	3·87	3·39
7. Puerto Cabello, . . .	2·96	15·03	50·57	12·94	11·49	3·07	3·94
8. Socosusco, . . .	2·95	13·19	48·38	15·13	13·20	3·34	3·21
Mean, . . .	3·25	14·76	49·00	13·31	12·35	3·68	3·65

II. COCOA HUSKS.

	Amount of Shell.	Water.	Nitrogenous Substances.	Fat.	Nitrogen Free Constituent.	Cellulose.	Ash.	Sand.
1. Caraccas I., . . .	15·03	7·41	13·93	4·94	41·78	12·91	7·41	12·62
2. " II., . . .	20·09	7·74	11·68	5·99	35·29	12·79	8·32	18·19
3. Guayaquil,	8·93	13·44	8·12	48·01	13·87	6·81	0·82
4. "	9·11	12·94	10·75	47·08	13·12	6·79	0·21
5. Trinidad, . . .	15·35	9·04	14·94	6·18	44·80	16·36	6·39	2·29
6. " . . .	14·04	8·30	15·44	4·23	46·05	18·00	7·06	0·92
7. Puerto Cabello, . . .	14·92	6·40	13·75	4·38	47·12	14·83	6·06	7·46
8. Socosusco, . . .	18·58	6·48	19·12	6·48	39·39	15·67	8·15	4·71
Mean, . . .	16·33	7·83	14·29	6·38	43·79	14·69	7·12	5·90

Sixty-four analyses of commercial samples of cocoa and chocolate have been made in the U.S. Agricultural Laboratory, and the general results may be classified as follows :—

TABLE XXXV.

	Moisture.	Fat.	Fibre.	Total Ash.	Acid Equivalent.	Acid Equivalent by Ash.	Sugar.
<i>A. Samples of cocoa or chocolate in commerce without any admixture, but most of them evidently showing treatment by steam or alkalis.</i>							
Maximum, . . .	3.5	50.85	5.02	8.64	16.6	4.9	...
Minimum, . . .	2.5	27.56	2.87	3.17	4.9	1.35	...
Mean, . . .	3.0	35.0	3.90	5.28	12.0	2.2	...
<i>B. Cocos and chocolates mixed only with sugar.</i>							
Maximum, . . .	1.75	25.74	1.99	2.20	2.90	1.20	63.0
Minimum, . . .	0.55	18.47	0.94	1.28	1.50	.72	51.0
Mean, . . .	1.13	22.3	1.31	1.58	2.31	1.40	57.3
<i>C. Cocos and chocolates mixed with wheat, flour, and sugar.</i>							
Maximum, . . .	2.46	28.72	1.85	3.29	2.70	1.58	60.0
Minimum, . . .	0.85	16.92	0.81	0.97	1.25	0.46	51.0
Mean, . . .	1.71	20.79	1.14	1.55	1.86	1.28	56.7
<i>D. Cocos and chocolates mixed with arrowroot and sugar.</i>							
Maximum, . . .	2.5	28.79	1.20	3.15	4.60	1.63	51.00
Minimum, . . .	0.6	11.13	2.62	1.40	2.25	0.83	25.00
Mean, . . .	1.4	22.30	1.85	2.30	3.08	1.37	36.4

R. Bensemann lays stress on the relations between the fat, starch, and matters insoluble in water. Designating the starch by S, the fat by F, the insoluble matters in water by U, the starch divided by the difference between the fat and matters insoluble in water gives what Bensemann calls "the starch coefficient," and is an aid to estimating the admixture of cocoa with flour or starch. He also gives other coefficients. This method and its results can be gathered from his analysis as follows :—

TABLE XXXVI.—BENSEMANN'S ANALYSES OF COCOA, COCOA HUSKS, AND CHOCOLATE.

	Total Organic Matter Insoluble in Water=U.						Ash.	Ash of Water-insoluble Bodies.	Husks in Air-dried Beans.
	Total Organic Matter Insoluble in Water=U.								
	Moisture at 100° C.	Fat = F.	Starch = S.	Other Organic Bodies Insoluble in Water.	Organic Bodies soluble in Water.				
%	%	%	%	%	%	%	%	%	
Air-dried husked beans—									
Maracabo,	6.87	49.18	13.91	17.32	9.20	4.42	0.84	...	
Caracas,	7.03	49.43	12.74	18.53	8.26	4.01	0.90	...	
Trinidad,	6.45	51.97	10.15	19.25	8.80	3.38	0.63	...	
Machala (Guayaquil),	5.81	53.21	10.82	19.38	6.94	3.84	1.00	...	
Porto Plata,	5.87	53.57	12.04	15.69	9.52	3.31	1.09	...	
Means,	6.41	51.47	11.75	18.03	8.54	3.80	0.89	...	
Air-dried husks—									
Maracabo,	13.98	2.34	8.79	54.43	14.45	6.91	2.63	12.00	
Caracas,	13.62	1.81	8.81	48.96	9.74	17.06	13.60	16.00	
Trinidad,	13.80	2.37	8.63	48.32	18.91	7.97	3.80	14.00	
Machala (Guayaquil),	14.56	2.03	7.07	48.51	14.73	13.11	1.20	13.00	
Porto Plata,	11.55	3.95	10.35	45.58	15.53	13.04	10.21	12.00	
Means,	13.32	2.50	8.73	49.16	14.67	11.62	7.83	13.00	
Chocolate in cakes mixed with sugar only—									
4.8 marks per kilo,	1.92	22.61	5.20	8.35	59.60	2.32	0.27	...	
4.0 " "	3.25	22.50	4.70	8.82	59.56	2.42	0.24	...	
3.2 " "	1.10	22.48	4.27	8.63	61.81	1.71	0.34	...	
2.4 " "	1.53	21.40	3.92	9.02	62.43	1.70	0.25	...	
2.0 " "	1.43	21.14	4.81	8.09	59.73	1.80	0.40	...	
Means,	1.65	22.57	4.58	8.58	60.63	1.99	0.30	...	

TABLE XXXVII.—NUMERICAL RELATIONS BETWEEN THE VARIOUS CONSTITUENTS OF THE COCOAS IN THE PRECEDING TABLE AS CALCULATED BY BENSEMANN.

S = starch ; F = fat ; U = total organic matter insoluble in water.

	$\frac{S}{U-F}$	$\frac{F}{U-S}$	$\frac{S}{U}$	$\frac{F}{U}$	$\frac{S}{F}$
Air-dried husked beans—					
Maracaibo,	0·4289	0·7395	0·1636	0·6185	0·2645
Caracas,	0·4074	0·7273	0·1578	0·6125	0·2577
Trinidad,	0·3452	0·7297	0·1247	0·6337	0·1953
Machala (Guayaquil),	0·3583	0·7330	0·1297	0·6379	0·2033
Portoplata,	0·3660	0·7734	0·1481	0·6589	0·2247
Means,	0·3946	0·7406	0·1446	0·6335	0·2283
Air-dried husks—					
Maracaibo,	0·1390	0·0412	0·1341	0·0357	3·7564
Caracas,	0·1525	0·0356	0·1479	0·0304	4·8674
Trinidad,	0·1515	0·0467	0·1455	0·0399	3·6413
Machala (Guayaquil),	0·1272	0·0402	0·1227	0·0352	3·4827
Portoplata,	0·1850	0·0797	0·1728	0·0660	2·6202
Means,	0·1508	0·0484	0·1446	0·0414	3·4920
Chocolate in cakes mixed with sugar only—					
4·8 marks per kilo,	0·3838	0·7303	0·1438	0·6253	0·2300
4·0 " "	0·3476	0·7161	0·1314	0·6220	0·2112
3·2 " "	0·3310	0·7226	0·1207	0·6354	0·1899
2·4 " "	0·3029	0·7035	0·1141	0·6232	0·1831
2·0 " "	0·3729	0·7490	0·1298	0·6517	0·1992
Means,	0·3480	0·7245	0·1282	0·6317	0·2029

§ 263. Stutzer classifies the nitrogenous constituents of cocoa as follows :—

1. Non-proteids, substances soluble in neutral water solution in presence of copper hydrate (theobromin, ammonia, and amido compounds).

2. Digestible albumen.

3. Insoluble and indigestible nitrogenous substance. In three samples he found the mean numbers in per cent. of total nitrogen to be—soluble non-proteid nitrogen 29·4 per cent., albumen 32·2 per cent., and not digestible 38·4 per cent. Mr. Wigner's examination of the nitrogenous constituents of cocoa, based on 84 commercial samples, is given in Table XXXVIII.

§ 264. *Adulterations of Cocoa.*—The list of adulterations usually given is as follows :—Sugar, starches, Venetian red, brick-dust, and peroxide of iron. Some of these sophistications, such as the starches, may be detected by a preliminary microscopical examination, which in no instance should be neglected. The ordinary chemical examination consists in the extraction of the fat as before described, the estimation of the percentage of ash in the ordinary way, its division into soluble and insoluble, and its content of phosphoric acid. By a simple estimation of the fat and the chief constituents of the ash, supplemented by the use of the microscope, all known adulterations can be detected. The amount of phosphoric acid in the ash

of soluble cocoas has been taken as a basis of calculation of the amount of cocoa, and in the absence of foreign seeds, or other phosphate-producing substance, the calculation will be a fair approximation to the truth. The ash itself and the amount of phosphoric acid will, of course, be very

TABLE XXXVIII.—NITROGENOUS CONSTITUENTS OF COCOA.

	Total Nitrogen.	Coagulable Nitrogen.	Total Albuminoids = N × 6.33.	Coagulable Albuminoids = N × 6.33.	Difference = Non-Coagulable Nitrogen Matter.	Per cent. Total Non-Coagulable.	
1.	1.095	.600	9.92	3.80	3.12	54.9	
2.	1.162	.760	7.35	4.81	2.54	65.5	
3.	2.978	2.335	18.84	14.79	4.05	78.5	
4.965	.375	6.11	2.37	3.74	38.8	
5.699	.330	4.42	2.09	2.33	47.3	
6.	1.201	.770	7.61	4.88	2.73	34.1	
Roasted beans,	Socunza, .	2.040	1.175	12.92	7.44	4.48	57.6
	Para, .	2.000	1.045	12.67	6.62	6.05	52.2
	Trinidad, .	1.490	1.050	9.46	6.65	2.81	70.3
	Grenada, .	2.370	1.335	14.99	7.56	7.43	50.4

notably diminished in the case of the soluble cocoas, and the percentage of the phosphoric acid will in such instances be a fair guide to the amount of foreign admixture. For example, suppose a soluble cocoa to yield an ash of 1.5 per cent., .6 of which is due to phosphoric acid, taking as a basis of calculation .9 per cent.¹ of phosphoric acid in cocoa nibs:—

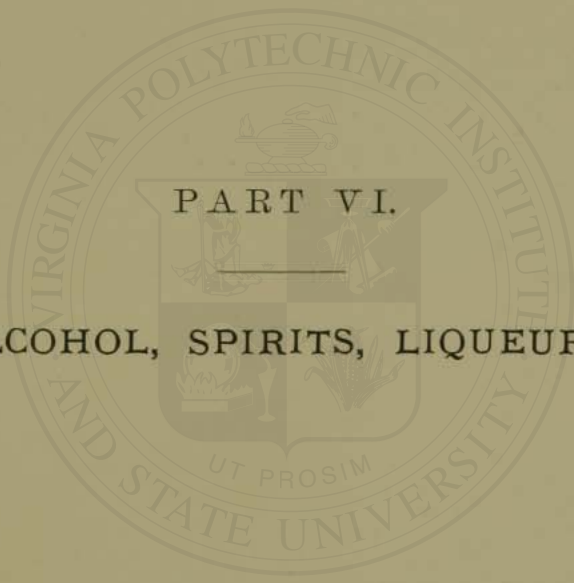
$$\frac{.6 \times 100}{.9} = 66.6.$$

That is, the mixture contains about 66.6 per cent. of cocoa. The amount of starch in cocoa may be determined in the ordinary way, as described at p. 136 *et seq.*, but the process is somewhat tedious, and may be dispensed with, since the extract in cold water is always a guide to the adulteration by starchy substances. Cocoa nibs treated in this way give to water about 6.76 per cent. of organic matter and 2.16 of ash. The determination of theobromin and of cocoa red is also of use.

§ 265. *Adulterations of Chocolate.*—Oil of almonds, cocoa-oil, beef and mutton fat, starches, cinnabar, chalk, and various other substances are usually enumerated; a few of these are, however, apocryphal.

The analysis of chocolate is conducted on exactly the same principles as that of soluble cocoa. If it is desired to separate the different constituents, the method recommended by A. Porrier may be used:—Extract the fat with ether, and the sugar with alcohol of 20 per cent., and dissolve the starch out by boiling water. The liquid holding the starch is then decolorised by animal charcoal, and the starch precipitated by alcohol of 50 per cent., dried, and weighed. But as regards adulteration, the procedure recommended in the case of cocoa will be found quite efficient, and less cumbersome and tedious.

¹ The lowest percentage given by Mr. Heisch, and but little different from Mr. Wanklyn's.



PART VI.

ALCOHOL, SPIRITS, LIQUEURS.

PART VI.—ALCOHOL, SPIRITS, LIQUEURS.

ALCOHOL.

§ 266. The term *alcohol*, in its strict chemical sense, applies only to the neutral compounds of oxygen, carbon, and hydrogen, which, by the action of acids, form ethers. The principal alcohols are enumerated in the following table:—

TABLE XXXIX.—EXHIBITING THE PROPERTIES OF THE PRINCIPAL ALCOHOLS.

Alcohols.	Formula.	Specific Gravity, 15.5°.		Vapour. Rel. Wt. H = 1.	Boiling Point.	
		Liquid.	Vapour.		Fahr.	Cent.
1. Wood spirit, or } methylic alcohol, }	CH ₄ O	0.798	1.12	16	148.6	64.7
2. Spirit of wine, or } ethylic alcohol, }	C ₂ H ₆ O	0.7936	1.6133	23	173.0	78.3
3. Tertiary or propylic, .	C ₃ H ₈ O	0.817	2.02	30	207.3	97.4
4. Tertiary or butylic, .	C ₄ H ₁₀ O	0.8032	2.589	37	242.0	117.0
5. Fousel oil, or amylic, .	C ₅ H ₁₂ O	0.8184	3.147	44	280.0	138.0
6. Hexylic or caproic, .	C ₆ H ₁₄ O	0.833	3.53	51	314.0	157.0
7. Heptylic,	C ₇ H ₁₆ O	0.819	...	58	348.8	176.0
8. Octylic or caprylic, .	C ₈ H ₁₈ O	0.823	4.50	65	384.8	196.0
12. Laurylic,	C ₁₂ H ₂₆ O
16. Ethal or cetylic, . .	C ₁₆ H ₃₄ O
27. Cerotin or cerylic, .	C ₂₇ H ₅₆ O
30. Melissin or mellisyllic	C ₃₀ H ₆₂ O

Of these ethylic alcohol, wood spirit, and fousel oil are the three of most importance to the analyst.

Ethylic Alcohol, C₂H₆O, specific gravity, 0.8059 at 0° C., 0.7936 at 15.5° C.; boiling point, 78.3° C. Absolute alcohol is usually obtained by digesting strong alcohol with freshly ignited lime, and distilling. Young¹ has, however, shown that a mixture of alcohol, benzene, and water distilled with the aid of an efficient still head, and fractionated, will yield at one operation a large fraction of alcohol containing a little over 1 per cent. of water.² This fraction on repeating the operation may be made similarly practically absolute by the use of benzene, the benzene acting as

¹ *Journ. Chem. Soc. Trans.*, 1902, 707.

² Dilute methyl alcohol can be freed from water by simple fractional distillation (Young and Fortey, *op. cit.*, 719).

TABLE XL.—ESTIMATION OF ALCOHOL.

Specific gravity, 15°5'.	Absolute Alcohol by weight. Per cent.	Absolute Alcohol by volume. Per cent.	Proof Spirit. Per cent.	Specific gravity, 15°5'.	Absolute Alcohol by weight. Per cent.	Absolute Alcohol by volume. Per cent.	Proof Spirit. Per cent.
1·0000	0·00	0·00	0·00	·9499	34·57	41·37	72·50
·9999	0·05	0·07	0·12	·9489	35·05	41·90	73·43
·9989	0·58	0·73	1·28	·9479	35·55	42·45	74·39
·9979	1·12	1·42	2·48	·9469	36·06	43·01	75·37
·9969	1·75	2·20	3·85	·9459	36·61	43·63	76·45
·9959	2·33	2·93	5·13	·9449	37·17	44·24	77·53
·9949	2·89	3·62	6·34	·9439	37·72	44·86	78·61
·9939	3·47	4·34	7·61	·9429	38·28	45·47	79·68
·9929	4·06	5·08	8·90	·9419	38·83	46·08	80·75
·9919	4·69	5·86	10·26	·9409	39·35	46·64	81·74
·9909	5·31	6·63	11·62	·9399	39·85	47·18	82·69
·9899	5·94	7·40	12·97	·9389	40·35	47·72	83·64
·9889	6·64	8·27	14·50	·9379	40·85	48·26	84·58
·9879	7·33	9·13	15·99	·9369	41·35	48·80	85·53
·9869	8·00	9·95	17·43	·9359	41·85	49·34	86·47
·9859	8·71	10·82	18·96	·9349	42·33	49·86	87·37
·9849	9·43	11·70	20·50	·9339	42·81	50·37	88·26
·9839	10·15	12·58	22·06	·9329	43·29	50·87	89·15
·9829	10·92	13·52	23·70	·9319	43·76	51·38	90·03
·9819	11·69	14·46	25·34	·9309	44·23	51·87	90·89
·9809	12·46	15·40	26·99	·9299	44·68	52·34	91·73
·9799	13·23	16·33	28·62	·9289	45·14	52·82	92·56
·9789	14·00	17·26	30·26	·9279	45·59	53·29	93·39
·9779	14·91	18·36	32·19	·9269	46·05	53·77	94·22
·9769	15·75	19·39	33·96	·9259	46·50	54·24	95·05
·9759	16·54	20·33	35·63	·9249	46·96	54·71	95·88
·9749	17·33	21·29	37·30	·9239	47·41	55·18	96·70
·9739	18·15	22·27	39·03	·9229	47·86	55·65	97·52
·9729	18·92	23·19	40·64	·9219	48·32	56·11	98·34
·9719	19·75	24·18	42·38	·9209	48·77	56·58	99·16
·9709	20·58	25·17	44·12	·9199	49·20	57·02	99·93
·9699	21·38	26·13	45·79	·9198	49·24	57·06	100·00PS
·9689	22·15	27·04	47·39	·9189	49·68	57·49	100·76
·9679	22·92	27·95	48·98	·9179	50·13	57·97	101·59
·9669	23·69	28·86	50·57	·9169	50·57	58·41	102·35
·9659	24·46	29·76	52·16	·9159	51·00	58·85	103·12
·9649	25·21	30·65	53·71	·9149	51·42	59·26	103·85
·9639	25·93	31·48	55·18	·9139	51·83	59·68	104·58
·9629	26·60	32·27	56·55	·9129	52·27	60·12	105·35
·9619	27·29	33·06	57·94	·9119	52·73	60·56	106·15
·9609	28·00	33·89	59·40	·9109	53·17	61·02	106·93
·9599	28·62	34·61	60·66	·9099	53·61	61·45	107·69
·9589	29·27	35·35	61·95	·9089	54·05	61·88	108·45
·9579	29·93	36·12	63·30	·9079	54·52	62·36	109·28
·9569	30·50	36·76	64·43	·9069	55·00	62·84	110·12
·9559	31·06	37·41	65·55	·9059	55·45	63·28	110·92
·9549	31·69	38·11	66·80	·9049	55·91	63·73	111·71
·9539	32·31	38·82	68·04	·9039	56·36	64·18	112·49
·9529	32·94	39·54	69·29	·9029	56·82	64·63	113·26
·9519	33·53	40·20	70·46	·9019	57·25	65·05	113·99
·9509	34·10	40·84	71·58				

TABLE XL.—*continued.*

Specific gravity, 15°.	Absolute Alcohol by weight. Per cent.	Absolute Alcohol by volume. Per cent.	Proof Spirit. Per cent.	Specific gravity, 15°.	Absolute Alcohol by weight. Per cent.	Absolute Alcohol by volume. Per cent.	Proof Spirit. Per cent.
.9009	57.67	65.45	114.69	.8449	81.40	86.64	151.83
.8999	58.09	65.85	115.41	.8439	81.80	86.96	152.40
.8989	58.55	66.29	116.18	.8429	82.19	87.27	152.95
.8979	59.00	66.74	116.96	.8419	82.58	87.58	153.48
.8969	59.43	67.15	117.68	.8409	82.96	87.88	154.01
.8959	59.87	67.57	118.41	.8399	83.35	88.19	154.54
.8949	60.29	67.97	119.12	.8389	83.73	88.49	155.07
.8939	60.71	68.36	119.80	.8379	84.12	88.79	155.61
.8929	61.13	68.76	120.49	.8369	84.52	89.11	156.16
.8919	61.54	69.15	121.18	.8359	84.92	89.42	156.71
.8909	61.96	69.54	121.86	.8349	85.31	89.72	157.24
.8899	62.41	69.96	122.61	.8339	85.69	90.02	157.76
.8889	62.86	70.40	123.36	.8329	86.08	90.32	158.28
.8879	63.30	70.81	124.09	.8319	86.46	90.61	158.79
.8869	63.74	71.22	124.80	.8309	86.85	90.90	159.31
.8859	64.17	71.62	125.51	.8299	87.23	91.20	159.82
.8849	64.61	72.02	126.22	.8289	87.62	91.49	160.33
.8839	65.04	72.42	126.92	.8279	88.00	91.78	160.84
.8829	65.46	72.80	127.59	.8269	88.40	92.08	161.37
.8819	65.88	73.19	128.25	.8259	88.80	92.39	161.91
.8809	66.30	73.57	128.94	.8249	89.19	92.68	162.43
.8799	66.74	73.97	129.64	.8239	89.58	92.97	162.93
.8789	67.17	74.37	130.33	.8229	89.96	93.26	163.43
.8779	67.58	74.74	130.98	.8219	90.32	93.52	163.88
.8769	68.00	75.12	131.64	.8209	90.68	93.77	164.33
.8759	68.42	75.49	132.30	.8199	91.04	94.03	164.78
.8749	68.83	75.87	132.95	.8189	91.39	94.28	165.23
.8739	69.25	76.24	133.60	.8179	91.75	94.53	165.67
.8729	69.67	76.61	134.25	.8169	92.11	94.79	166.12
.8719	70.08	76.98	134.90	.8159	92.48	95.06	166.58
.8709	70.48	77.32	135.51	.8149	92.85	95.32	167.04
.8699	70.88	77.67	136.13	.8139	93.22	95.58	167.50
.8689	71.29	78.04	136.76	.8129	93.59	95.84	167.96
.8679	71.71	78.40	137.40	.8119	93.96	96.11	168.24
.8669	72.13	78.77	138.05	.8109	94.31	96.34	168.84
.8659	72.57	79.16	138.72	.8099	94.66	96.57	169.24
.8649	73.00	79.54	139.39	.8089	95.00	96.80	169.65
.8639	73.42	79.90	140.02	.8079	95.36	97.05	170.07
.8629	73.83	80.26	140.65	.8069	95.71	97.29	170.50
.8619	74.27	80.64	141.33	.8059	96.07	97.53	170.99
.8609	74.73	81.04	142.03	.8049	96.40	97.75	171.30
.8599	75.18	81.44	142.73	.8039	96.73	97.96	171.68
.8589	75.64	81.84	143.42	.8029	97.07	98.18	172.05
.8579	76.08	82.23	144.10	.8019	97.40	98.39	172.43
.8569	76.50	82.58	144.72	.8009	97.73	98.61	172.80
.8559	76.92	82.93	145.34	.7999	98.06	98.82	173.17
.8549	77.33	83.28	145.96	.7989	98.37	99.00	173.50
.8539	77.75	83.64	146.57	.7979	98.69	99.18	173.84
.8529	78.16	83.98	147.17	.7969	99.00	99.37	174.17
.8519	78.56	84.31	147.75	.7959	99.32	99.57	174.52
.8509	78.96	84.64	148.32	.7949	99.65	99.77	174.87
.8499	79.36	84.97	148.90	.7939	99.97	99.98	175.22
.8489	79.76	85.29	149.44				
.8479	80.17	85.63	150.06				
.8469	80.58	85.97	150.67				
.8459	81.00	86.32	151.27				
				Absolute Alcohol.			
				.7938	100.00	100.00	175.25

a dehydrating agent. Absolute alcohol does not dissolve common salt, nor does it give a blue colour when digested with anhydrous sulphate of copper, if perfectly water-free. Filter paper saturated in the following solution—viz., two parts of citric and one of molybdic acids heated to incipient fusion, then dissolved in 30 to 40 pints of water and dried at 100° C., is not bleached when soaked in absolute alcohol; but should water be present, the blue of the paper is entirely discharged. Pure absolute alcohol burns with a white flame, but if water is present the flame is blue.

There is no cloudy appearance when mixed with water, showing the absence of oily matters. It should be also perfectly neutral to test paper, and leave no residue on evaporation. It must be remembered that, in a commercial sense, 'absolute alcohol' is any stronger spirit than can be obtained by ordinary distillation; and, since this is the case, it would be most unwise for any action to be taken under the "Sale of Food and Drugs Act," unless a distinctly fraudulent statement has been made. "Absolute alcohol," as bought over the counters of the chemist, is seldom above from 93 to 95 per cent. of real alcohol.

§ 267. *Rectified Spirit*, as defined by our own pharmacopœia, should be of specific gravity 0·838; by that of the Netherlands 0·830 to 0·834; of Germany, Switzerland, and Norway, 0·8336; of Austria, 0·838; of France, 0·835 to 0·841. It should be neutral, colourless, volatilising without residue, and free from other alcohols.

Proof Spirit,—a term in constant use for purposes of excise,—is a diluted spirit, which was defined by Act of Parliament (58 George III.) to be "such as shall, at the temperature of 10°·0 C. [51° Fahr.] weigh exactly twelve-thirteenth parts of an equal measure of distilled water." According to Drinkwater it consists of—

Alcohol by weight,	49·24
Water by weight,	50·76
	100·00

and its specific gravity at 15°·5 C. is 0·91984.

In the analysis of all spirits (seeing that the terms 'proof' and 'under proof' are used and known in the trade), the statements of results should always include the percentage of proof spirit.

Spirits weaker than proof are described as U.P., under proof; stronger than proof as O.P., over proof; thus a spirit of 50 U.P. means 50 water and 50 proof spirit, 60 U.P., 60 water and 40 proof spirit. On the other hand, 50 O.P. means that the alcohol is of such a strength, that, to every 100 of the spirit, 50 of water would have to be added to reduce it to proof strength. In all the above the strengths are only good for the normal temperature of 15°·5.

§ 268. *Tests for Alcohol*.—The principal tests for alcohol are the following:—

(1.) *Production of Acetic Ether*.—To a distillate or aqueous solution supposed to contain alcohol, some acetate of soda is added and sulphuric acid in amount more than sufficient to decompose the acetate. The flask containing the mixture is connected with a Liebig's condenser, placed vertically, and boiled for a few minutes; any volatile vapour is condensed, and falls back again into the flask. On removing the cork, if acetic ether has been produced, it can readily be detected by its odour.

(2.) *Reduction of Chromic Acid or Bichromate of Potash to Oxide of*

Chromium.—A crystal of chromic acid, placed in a test-tube, with a fluid containing alcohol warmed to a boiling temperature, is decomposed into the green oxide of chromium. Instead of chromic acid, a test-solution of one part of bichromate of potash dissolved in 300 parts of sulphuric acid may be used. A portion of the liquid to be tried is mixed with twice its volume of concentrated sulphuric acid. On pouring a small quantity of this mixture into a quantity of the test-solution, a deep green is produced where one fluid touches the other. This is a very good test in the absence of other reducing agents, such as formic acid, ether, etc.

(3.) *Dr. Edmund Davy's Test.*—Dr. Davy has proposed a test for alcohol founded on a colour reaction, and produced also by methyl, propyl, butyl, and amyl alcohols, ether and aldehyde. A solution of one part of molybdic acid in ten of strong sulphuric acid, is warmed in a porcelain capsule, and the liquid to be tested allowed to fall gently on it. If alcohol is present, a blue coloration appears either immediately, or in a few moments; the liquid gradually absorbs moisture from the air, and the colour disappears, but it may be reproduced by evaporation.¹

(4.) *The production of Iodoform.*—According to Lieben, one part of alcohol in 2000 of water can be detected by adding to some of the warmed liquid a few drops of a 10 per cent. solution of soda, and dropping in a solution of potassium iodide, fully saturated with free iodine, until the liquid is yellowish-brown; then the alkali solution must be added until the whole is colourless, and the mixture allowed to stand for many hours, when a yellowish crystalline deposit of iodoform is obtained. Under the microscope the latter presents the appearance of hexagonal plates, six-rayed, or other varieties of, stellar crystals: $C_2H_5O + 4I_2 + 6NaHO = CHI_3 + NaCHO_2 + 5NaI + 5H_2O$. The objection to this test is, however, that other alcohols, aldehyde, gum, turpentine, sugar, etc., give a similar reaction.²

(5.) *The production first of Acetic Acid, then of Kakodyl.*—A very delicate test for alcohol, and one specially suited for its detection in the blood, etc., is recommended by Bucheim.

The finely-divided substances are put in a tubulated retort, and, if acid, carefully neutralised. In the neck of the retort is placed a little porcelain or platinum boat, containing platinum black, and at each end there is a moistened piece of strongly-blued litmus-paper. On warming the retort, if alcohol be present, it is oxidised by the platinum black to aldehyde and acetic acid; hence, the hinder piece of litmus-paper will be reddened, the front one unchanged. If only a drop of acetic acid be present, it is possible to detect it in the following way:—The platinum black is washed, the washing water neutralised with potash, and dried after the addition of a few grains of arsenious acid. On warming the dry residue in a small glass tube, if even a very small admixture of acetic acid be present, the smell of kakodyl will be perceptible.

(6.) *The Action of Alcohol on Benzoyl Chloride.*—This test, proposed by Berthelot, is based on the fact that very small quantities of alcohol decompose benzoyl chloride with the formation of ethyl benzoate. The sample is shaken up in the cold with a few drops of benzoyl chloride; any

¹ *Proceedings of the Royal Irish Academy* [2] ii., 579-582; *Journ. Chem. Soc.*, vii., 1877, p. 108. Gladstone and Tribe (*Journ. Chem. Soc.*, No. cexlix.) have shown that the test is unreliable, for the same reaction takes place with ammonium sulphide, sodium sulphate, formic acid, sugar, and, in fact, with most reducing agents.

² Rajewsky has found that the brain of a rabbit, which had been starved for two days before death, gave a marked reaction for alcohol with the iodoform test, and the same result was obtained from the muscles and tissues of rabbits. He therefore considers that alcohol always exists in the animal organism, or that it is produced during distillation.—*Pflüger's Archiv für Physiologie*, xi., 122, 127.

ethyl benzoate formed sinks to the bottom with the excess of benzoyl chloride. This heavy layer is removed by a pipette, and heated with a little caustic soda or potash, which dissolves at once the benzoyl chloride, but not the ethyl benzoate, and the latter may be recognised by this insolubility, by its general properties, and by its boiling point. It is nearly insoluble in water, burns with a smoky flame, has a characteristic pleasant odour, and boils at 213° C.

Mr. J. Hardy has proposed a very simple test for the detection of alcohol, which may be performed as follows:—Two common 'Nesslerising' glass cylinders are taken, and a little guaiacum resin, which has been removed from the interior of a freshly broken lump, is shaken up with the sample to be tested. The liquid is filtered, and a few drops of hydrocyanic acid as also a drop of very weak solution of sulphate of copper added. Exactly the same process is adopted with an equal bulk of distilled water. On now placing the two liquids in the glass cylinders side by side, over a porcelain plate, the liquid to be tested, if it contains alcohol, will be found of a blue colour decidedly darker than that of the distilled water.

§ 269. *Separation of Alcohol from Animal Matters.*—In order to obtain alcohol from organic matters (*e.g.*, the contents of the stomach, or the tissues), the following process will be found convenient:—Solid matters, such as the tissues, are cut up as finely as possible, and placed with water in a retort attached to a suitable condenser. Most liquids require no previous preparation, and are merely poured into the retort or flask, as before described; but it is desirable in the treatment of *urine* to add a little tannic acid. About one-third to one-half of the liquid is distilled over into a flask closed by a mercury valve. The product is now exactly neutralised with decinormal alkali to fix any volatile acid, and again slowly distilled, about one-third being drawn over. The liquid is next neutralised with sulphuric acid, to fix volatile alkalies, and redistilled. This final distillate contains all the alcohol, but neither volatile acids nor alkalies. The liquid thus obtained may even now be too dilute to respond conveniently to tests, and it may therefore be digested for some hours with a little caustic lime, and then very slowly distilled. The distillate should finally be carefully measured or weighed, and divided into two parts, one of which serves for the application of the usual tests, the other (if alcohol be found) can be oxidised in the manner described at p. 389 and estimated volumetrically as acetic acid.

J. Bechamp (*Compt. Rend.*, lxxxix., 573, 574) has succeeded in obtaining a sufficient quantity of alcohol from the fresh brain of an ox to estimate it by the specific gravity, and has also separated it from putrefying animal matter. In fact, it is capable of proof, that all putrefaction is accompanied by the production of minute quantities of alcohol, and that the living cells of the body also produce it. Hence, in questions of poisoning, it is not enough to obtain qualitative reactions for alcohol, but the quantity also must be accurately estimated.

ESTIMATION OF ALCOHOL AND ANALYSIS OF LIQUIDS CONTAINING ALCOHOL.

§ 270. Excellent tables for the use of analysts have been published both by Mr. Hehner and by Dr. Stevenson. The table on p. 380 *et seq.* will be found, in the absence of the tables mentioned, sufficient for ordinary use; any specific gravity not given can be intercalated by the ordinary rules of arithmetic.

Another method, sometimes called Gröning's, of arriving at the strength of dilute spirits, is based on the fact that the temperature of the vapour is an exact measure of the strength of the alcohol. The bulb of a thermometer is put (on the small scale) into a flask with a bilateral tube, and the temperature of the vapour carefully noted. The following table (XLI.) may be used:—

TABLE XLI.—SHOWING THE CONTENT OF ETHYL ALCOHOL BY VOLUME OF BOILING SPIRITS AND OF THEIR VAPOUR, FROM THE TEMPERATURE OF THE LATTER, AS OBSERVED BY A THERMOMETER. BY GRÖNING.

Temperature of the Vapour (F.).	Ethyl Alcohol in the Distillate. Per cent.	Ethyl Alcohol in the Boiling Liquid. Per cent.	Temperature of the Vapour (F.).	Ethyl Alcohol in the Distillate. Per cent.	Ethyl Alcohol in the Boiling Liquid. Per cent.
170·0	93	92	189·8	71	20
171·8	92	90	192·0	68	18
172·0	91	85	194·0	66	15
172·8	90½	80	196·4	61	12
174·0	90	75	198·6	55	10
174·6	89	70	201·0	50	7
176·0	87	65	203·0	42	5
178·3	85	50	205·4	36	3
180·8	82	40	207·7	28	2
183·0	80	35	210·0	13	1
185·0	78	30	212·0	0	0
187·4	76	25			

The boiling-point is also a useful guide; for within certain limits the boiling-point of liquids containing alcohol is not materially altered by admixture with saline and organic matter. A thermometer with a movable scale is employed. Before using it, the thermometer is immersed in boiling distilled water, and the 100° [212° Fahr.] of the scale accurately adjusted to the level of the mercury; it is then ready for an operation of several hours, or even an entire day, if no considerable variations of atmospheric pressure are experienced. The minimum boiling-point 78°·174 C. [172°·7 F.] of an aqueous ethyl alcohol corresponds to 96 per cent. by weight of alcohol, absolute alcohol being 78°·3 C. [172°·9 F.].

TABLE XLII.—EXHIBITING THE BOILING-POINTS OF ALCOHOL AND WATER OF THE GIVEN STRENGTHS. BY GRÖNING.

Boiling-point (F.).	Alcohol per cent. per Volume.	Boiling-point (F.).	Alcohol per cent. per Volume.
205·34	5	179·96	55
199·22	10	179·42	60
195·89	15	178·70	65
192·38	20	177·62	70
189·50	25	176·54	75
187·16	30	175·46	80
185·00	35	174·92	85
183·38	40	174·20	90
182·12	45	173·14	95
181·58	50	172·90	100

The methods used in the Municipal Laboratory, Paris, for the analysis of alcohols are the most practical and the easiest to apply to the small quantities of alcohols submitted to an analyst under the Sale of Food and Drugs Act, and have been made considerable use of in the following pages.

The estimations of the higher alcohols, of aldehydes, of furfural, and so forth, are to a great extent determined by colour reactions, the alcohol to be tested being always brought to the definite strength of 50°; by this device variations in colour, owing to varying strengths of alcohol, are avoided.

The French chemists estimate the following:—(1.) Alcohol; (2.) acidity, expressing it as acetic acid; (3.) aldehydes; (4.) ethers, returning the ethers as ethyl acetate; (5.) higher alcohols, returning them as isobutylic alcohol; and (6.) bases.

The scope and character of such an analysis may be gathered from the following example of the analysis of a sample of brandy:—

BRANDY MADE FROM WINE.

Sp. gr. at 15° C.,	0.9414
Alcohol, per cent. (volume),	48.2
Extract, per litre,	12.64 grms.
Sugars { Saccharose, per litre,	8.2 "
{ Inverted sugar, per litre,	3.7 "
Colour,	Tannin and caramel.

	Grammes.		Proportion of each Impurity, per cent. of Total Impurity.
	Per Litre of Brandy.	Per cent. of Absolute Alcohol.	
Acidity, as acetic acid,	0.3360	0.0697	32.23
Aldehydes, as acetic aldehyde,	0.0620	0.0130	6.01
Furfural,	0.0027	0.0005	0.26
Ethers, expressed as ethyl acetate,	0.2024	0.0419	19.38
Higher alcohols, expressed as isobutylic alcohol,	0.3990	0.0828	38.29
Bases { Ammonia, amides,	0.0350	0.0072	3.33
{ Alkaloids, pyridine bases, etc.,	0.0056	0.0011	0.50
Coefficient of impurities, per cent. of absolute alcohol,	0.2162	100.00

§ 271. In the examination of alcoholic liquors, one of the analyst's first steps is to determine the percentage of alcohol, and the methods by which this is done are equally applicable (with slight modifications) to all liquids containing alcohol. The percentage is ascertained—

- (1.) By distillation, and taking the specific gravity of the distillate.
- (2.) By Tabarie's method, applied especially to wines and beers.
- (3.) By Geissler's vaporimeter.
- (4.) By determination of the refractive index.
- (5.) By oxidation into acetic acid, and by several other methods, which are, however, not much in use by the analyst.

(1.) *Distillation.*—A convenient quantity (*e.g.*, 100 c.c. of beer or wine, 50 c.c. of spirits, measured at $15^{\circ}5$ C.) is placed in a flask (*a*, fig. 62), having an angle tube connected by means of a cork to a Liebig's condenser. The distillate is received in a flask (*b*) provided with a doubly perforated cork, into which the bent tube of the condenser, as well as a tube provided with a mercury valve, to prevent loss, is adjusted; the latter may be readily made by putting a very small quantity of mercury into the bend of an ordinary thistle-head funnel (*c*). This precaution is only necessary when very small quantities are operated upon. Experiments with 50 c.c. of spirit distilled into a flask unprovided with a valve, have shown that there is no appreciable loss; but distillation into an open vessel will always give results far too low. Beer and wine yield the whole of their alcohol when half is drawn over; spirits should be distilled nearly to dryness. In any case, the distillate should be made up to exactly the same bulk as the original liquid at the same temperature, its specific gravity taken in a proper specific gravity bottle, and the percentage of

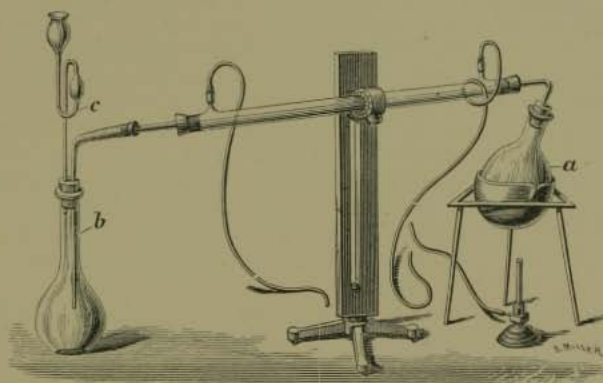


FIG. 62.

alcohol obtained by reference to the tables given at p. 380 *et seq.* Spirits are best returned as containing so much *proof spirit*, by volume; wines and beers, so much alcohol per cent., by weight and by volume.

(2.) *Tabarie's method*, when properly performed, is sufficiently accurate for all practical purposes in the case of beers, wines, and similar liquids. The specific gravity is first accurately taken at $15^{\circ}5$; a measured quantity—say 100 c.c.—is then boiled long enough to evaporate away the whole of the alcohol, made up to the original bulk *at the same temperature*, and its specific gravity again determined. From these data the specific gravity of the liquid, which, if it had been condensed, would have collected in the flask before-mentioned, is determined. Thus, specific gravity of the liquid before boiling, divided by the specific gravity of the de-alcoholised liquid = specific gravity of the dilute alcohol which has been boiled away. An actual example will suffice:—A beer, before boiling, had a specific gravity of 1.014; after boiling, and on making it up to the original bulk, its specific gravity was 1.0172; now $\frac{1.014}{1.0172} = .9968$, and on reference to the table at p. 380, .9968 is found to correspond to 1.7 per cent. of alcohol.

(3.) *Geissler's Vaporimeter* is capable of giving sufficiently accurate results for technical purposes, and as it has the advantage of great expedi-

tion, it may always be used to supplement and check other methods which take more time. It depends on the measurement of the tension or elastic force of the vapour of the liquid, as indicated by the height to which it raises a column of mercury. The apparatus (see fig. 63) consists of four

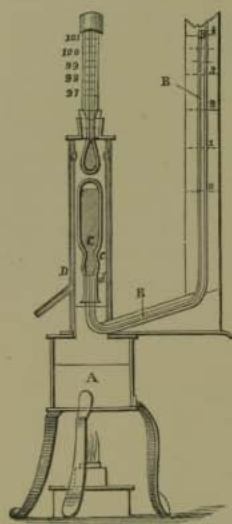


FIG. 63.

parts—viz., (1.) A brass vessel A, containing water; (2.) a doubly bent tube, BB, fastened to a scale; (3.) a cylindrical vessel C, which is filled with mercury and the fluid to be tested; (4.) a brass vessel D, in the upper part of which there is a thermometer. By removing the bent tube and its connections from A, it may be turned upside down, and C detached; the alcoholic fluid is then poured in so as to fill the space between *c* and *d*, which, when the instrument is inverted, is empty. It is now connected with the bent tube, and adjusted exactly as in the figure, the water made to boil in A, when the mercury runs up to a certain height in the tube, and the percentage is directly read from the scale. Should the thermometer not register 100°, certain corrections must be made, which is most conveniently done by a table, sold with the instrument. Care must always be taken to exclude air from the bulb. The senior author has found it always necessary to test various points of the scale with known mixtures of alcohol and water, and to draw up a table of corrections. With this precaution, it will be found a most useful instrument.

(4.) *Estimation of Alcohol by Valuation of the Refractive Index.*—This is an excellent and rapid method of valuation of ethyl alcohol, and can be applied to a distillate or to spirit or to wine.

The method of applying the refraction method to spirit or wine is based on the following principles:—

The refractive index of a liquid containing water, alcohol, and extract is derived from the indices of refraction of the water, alcohol, and extract. Let these be denoted respectively by *a*, *b*, *c*. Taking *N* as the index of refraction of the mixture, then obviously $N = a + b + c$.

If the liquid is freed from alcohol by distillation or evaporation, then the refractive index $N - (a + c) = b$, the index of refraction of the alcohol.

If, again, the index of refraction of distilled water be subtracted from the index found by experiment of the alcohol-free dilute extract and the amount of increase of refraction has been ascertained that 1 grm. per cent. of extract produces, the extract can also be estimated. In other words, by two observations on, first, the index of refraction of an alcoholic liquid, and, secondly, a determination of the same liquid freed from water, it is practical and easy to determine alcohol, water, extract.

E. Riegler¹ by a number of experiments has proved that with regard to wine 1 grm. of extract in 100 c.c. of wine increases the refractive index by 0.00145.

Similarly 1 grm. of alcohol per 100 c.c. of wine increases the index above that of the extract solution by 0.00068.

¹ "Die Bestimmung des Alcohols u. Extractes im Weine auf optischen Wege," by Prof. Dr. E. Riegler, *Zeitschrift f. analytische Chemie*, 1896, 27.

For example—

The refraction index of a wine was found to be 1·34105
 ,, alcohol-free extract, . . . 1·33550
 Difference = $\frac{00555}{}$
 then $\frac{00068}{00555} = 8\cdot1617$ alcohol per cent.

Similarly

the index of refraction of the alcohol-free extract was 1·33550
 Subtract index for distilled water, . . . 1·33263
 Difference = $\frac{00287}{}$
 then $\frac{00145}{00287} = 1\cdot9793$ grms. extract per cent.

The amount of extract and alcohol in wines determined by the ordinary processes do not differ materially from those determined by the optical methods.

The details of the operation itself are simple. 25 c.c. of an alcoholic liquid are placed in a Pulfrich's refractometer or in a hollow prism and observed through a spectrometer, the temperature taken and the index calculated in the usual way; next the alcohol is evaporated away by reducing the liquid to a third of its volume and again made up to exactly the same bulk and brought to the same temperature as the original liquid and the index of refraction again determined in the above calculation.

(5.) *Oxidation into Acetic Acid* is a method of determining vinic alcohol, specially applicable to small quantities. Dr. Dupré recommends the following process¹:—A small quantity of the distillate, representing about a gramme of alcohol, is put in a small strong assay-flask, and mixed with 10 c.c. of an oxidising solution, composed of 147 grms. of bichromate of potash and 220 grms. of sulphuric acid, made up to 1400 c.c. by water. The flask is well stoppered by caoutchouc, and firmly tied down by canvas and string. It is then suspended upright in a water-bath (the neck being above the water), and heated for two hours between 80° and 90° C. The flask is next removed, and the excess of bichromate reduced by zinc and sulphuric acid; the solution is transferred to a small retort (adding some sulphuric acid and bits of tobacco-pipe), and distilled over from a spermaceti-bath (see fig. 62). It will be found necessary to distil at least thrice nearly to dryness, each time adding water to the contents. The united distillates contain acetic acid, the result of the oxidation of the alcohol. This acetic acid may be determined by a volumetric solution of soda, and the amount of alcohol to which it is equivalent calculated by the following short table:—

Acetic Acid.	Alcohol.	Acetic Acid.	Alcohol.
1 . . . = . . .	7666	6 . . . = . . .	4·5996
2 . . . = . . .	1·5332	7 . . . = . . .	5·3662
3 . . . = . . .	2·2998	8 . . . = . . .	6·1328
4 . . . = . . .	3·0664	9 . . . = . . .	6·8994
5 . . . = . . .	3·8330	10 . . . = . . .	7·6666

¹ It has been shown by Wanklyn that alcohol may also be oxidised into acetic acid very readily by an alkaline solution of permanganate of potash; it would appear that in this case there is no previous formation of aldehyde. A. Letteltür also finds that an ammoniacal solution of copper-oxide at 180° has the same effect. *Compt. Rend.*, lxxxix., 1105.

There are several other methods of estimating alcohol, but the above are the most practical and efficient; and whenever the amount of alcohol is important (as, for example, in the case of spirits) the analyst should determine it in at least two different ways. The specific-gravity methods presuppose the presence of ethylic alcohol only; but it is often necessary to test fluids also for methylic and amylic alcohols. For these the following processes are available:—

§ 272. *Methylic Alcohol*.—100 c.c. of the suspected spirit are distilled twice, having been rendered alkaline during the first process and acid during the second, about two-thirds being distilled over each time. The distillate is now shaken up with dry potassium carbonate, and, after standing over night, the upper layer is taken off by a syphon or pipette, and again twice distilled, about 15 c.c. being driven over. This will contain any methylic alcohol present in the original 100 c.c. A portion of the distillate is now diluted with water to a strength from 10 to 15 per cent., and in this diluted spirit the alcohol determined—(1.) By specific gravity; (2.) by Geissler's vaporimeter¹; and, (3.) by oxidation into acetic acid. If ethylic alcohol alone is present, all three methods fairly agree. The specific gravity will give the total amount of both alcohols, the specific gravity of aqueous methylic and ethylic alcohols being almost identical; but since methylic alcohol has a higher vapour tension than ethylic, Geissler's vaporimeter will give a higher result. The oxidation process will, on the other hand, give a lower result, for methylic alcohol yields water and CO₂, so that the acetic acid found is derived wholly from the ethylic alcohol, and the difference between the strength thus found and that derived from the specific gravity gives a rough indication of the proportion of methylic alcohol present. If the methylic alcohol is in sufficient quantity, instead of the usual slight vacuum on opening the flask, there is an escape of carbonic anhydride, and there is no reason why this gas should not be either absorbed or collected and estimated.

Dr. Dupré gives the following example. A pure whisky showed—

Strength by specific gravity,	9·83 per cent.
„ Vaporimeter,	9·75 „
„ Oxidation,	9·75 „

The same whisky, adulterated with 10 per cent. of ordinary methylated spirit, and tested, gave—

Strength by specific gravity,	10·08 per cent.
„ Vaporimeter,	10·45 „
„ Oxidation,	9·50 ² „

The remainder of the distillate may be used in producing methyl-aniline violet or oxalate of methyl.

The general process for the production of methyl-aniline violet is as follows:—10 c.c. of alcohol, with 15 grms. of iodine and 2 grms. of red phosphorus, are put into a small flask, and distilled into 30 or 40 c.c. of water. The alcoholic iodide which settles to the bottom is separated by a pipette, and collected in a flask containing 5 c.c. of aniline. If the action be

¹ Hehner has raised a doubt as to the usefulness of the vaporimeter for the quantitative estimation of methylic alcohol; in the oxidation process he prefers to estimate the quantity of chromate reduced rather than the acetic acid.—*Analyst*, Feb. 7, 1887.

² Note on the Examination of Whisky and other Spirits for Methylated Spirit and Fousel Oil, by Dr. Dupré.—*Analyst*, i., 1876, p. 4.

too violent, the flask can be cooled with cold water; if too slow, a little heating may be necessary. At the end of an hour the crystals are dissolved in hot water and boiled, an alkaline solution is afterwards added, and the bases rise to the top in the form of an oily stratum, which may be separated by bringing the oil, by the addition of water, on a level with the neck of the flask. The oxidation of the bases may be effected in various ways, but best by pouring a cubic centimetre of the oily liquid on 10 grms. of a mixture formed of 100 grms. of quartz sand, 2 of chloride of sodium, and 3 of nitrate of copper. After incorporation it is introduced into a glass tube, and kept at 90° C. in a water-bath for eight or ten hours; it is ultimately exhausted by warm alcohol, thrown on a filter, and made up to 100 c.c. If the alcohol was pure the tint is red; if it contained 1 per cent. of methyl alcohol the colour, by the side of the preceding, is manifestly violet; with 2.5 per cent. the shade is a very distinct violet; and with 5 per cent. it is considerably darker. The process may be made quantitative by having volumetric solutions of methyl and ordinary alcohol. Very minute quantities of methyl alcohol may be detected by adding 5 c.c. of the liquid to 95 c.c. of water, and then again diluting 5 c.c. of this liquid with 400 c.c. of water, and heating it in a porcelain capsule. Fragments of white merino (free from sulphur), immersed in the liquid for half an hour, will remain white, if the alcohol was pure; if methyl was present, they will be of a violet tint.¹

Oxalate of Methyl may be obtained by mixing the distillate with half its bulk of sulphuric acid and double the quantity of hydropotassic oxalate. The whole should stand in the retort for twenty-four hours, and then be distilled. Crystals appear after a time in the cooler parts of the flask or retort; their composition is $(\text{CH}_3)_2\text{C}_2\text{O}_4$. It would also be quite possible to produce such compounds as the salicylate of methyl, etc.²

Formic Acid.—It has been already stated that methyl alcohol, when fully oxidised, is resolved into CO_2 and water. This takes place in two stages; first, formic acid is produced, and then this formic acid breaks up. Thus:—



It hence follows that if a spirit be distilled in the manner recommended by Dr. Dupré, and only partially oxidised, it is possible to get formic acid, which has some very characteristic properties. To obtain this result a small portion of the distillate, 2 to 4 c.c., is taken, and 3 grms. of potassic bichromate are added, with an equal quantity of pure sulphuric acid, and four or five times as much water. This is allowed to act for twenty minutes, and then distilled; the liquid is alkalisied with sodium carbonate, evaporated to half its bulk, acidulated with acetic acid, transferred to a test-tube, and then heated gently with a 5 per cent. solution of nitrate of silver. If

¹ A. Riche and Brady, *Comptes Rendus*, vol. xxx., p. 1096.

² For the detection of methyl alcohol in ethyl alcohol, Van de Vivere (*Chem. Centrbl.*, 1884, 69) employs the property possessed by methyl alcohol of forming a compound with calcium chloride, which is not decomposed at 100° C., but is decomposed by the addition of water; the alcohol is distilled from anhydrous potassic carbonate, and the volume of the distillate determined. A portion is then allowed to remain over an equal weight of anhydrous calcium chloride for 24 hours. The ethyl alcohol is then distilled off. The residue when treated with water and distilled yields a mixture of methyl alcohol and water.

formic acid has been produced, there is a distinct precipitate of metallic silver.

Trillat¹ identifies methyl alcohol as follows:—A quantity of a distillate corresponding to 10 c.c. of absolute alcohol is diluted to 150 c.c. and mixed with 70 c.c. of diluted H_2SO_4 (1 : 5) and 15 grms. of potassic bichromate; this is shaken for twenty minutes, distilled, the first 25 c.c. of the distillate being rejected, then 100 c.c. of the distillate collected. 50 c.c. of this are mixed with 1 c.c. of dimethyl aniline in a flask with ground stopper, and this is placed in a water-bath at 70–80° C. for three hours. The liquid is now alkalisied with NaOH and 25 c.c. distilled over; the distillate contains all the free dimethyl aniline. The residue in the flask is acidified with acetic acid and a few c.c. tested in a test-tube for methyl alcohol by adding a few drops of water with lead dioxide in suspension; if present, a blue colour is produced. The test can be made quantitative by imitating the colour by mixing the reagent with known quantities of diluted methyl alcohol.

Lams' Method of Estimating Methyl Alcohol in Ethylic Alcohol.—The alcohols are converted into iodides by treating 25 c.c. of the alcohol with 5 grms. of amorphous phosphorus and 40 grms. of iodine; when the reaction is over the liquid is distilled; the distillate is first washed with weak NaOH solution, then with water, and finally dried by calcic chloride (the specific gravity of MeI = 1.944, that of EtI = 2.2677). The specific gravity is now taken, and the amount of methyl deduced by the aid of tables given in the original paper (*Zeit. angew. Chem.*, 1898, 125–130).

Acidity of Alcohols.—A definite quantity of alcohol is coloured by phenol-phthalein, and decinormal soda dropped in until the liquid is neutral; the result may be expressed as acetic acid.

Aldehydes.—Aldehydes are detected by a solution of fuchsine made as follows:—Bisulphite of soda solution (sp. gr. 1.3082), 100 c.c.; aqueous solution of fuchsine (1 per 1000), 150 c.c.; pure sulphuric acid (66 per cent.), 15 c.c.; the whole made up to a litre with distilled water. 4 c.c. of this test solution are added to 10 c.c. of a distillate; should aldehyde be present, a red colour is almost immediately produced, attaining its maximum in twenty minutes. To estimate the quantity, a distillate is taken and diluted until it contains 50 per cent. of alcohol, or the distillate may be fortified with pure alcohol until it is of the strength mentioned. The colour is now produced by the test, and the colour imitated by adding the same test to alcohol of 50 per cent. containing 0.050 gm. per litre of acetic anhydride.

Furfurol.—Furfurol may be detected by a number of reactions; one of the most sensitive is the red colour produced by the addition of an alcoholic solution of *a*-naphthol and a very little strong sulphuric acid. The French chemists estimate furfurol by the tint produced by acetate of aniline in a furfurol-holding liquid, as compared with the tint produced by adding to pure alcohol of 50 per cent. a solution of furfurol in alcohol of the same strength, each litre of which contains 5 mgrms. of furfurol. It may also be estimated by quantitative spectroscopy. In both of these methods, since the crimson colour caused by aniline acetate darkens by time, the observations must be made always exactly the same time after the addition of the reagent.

Acetone.—Acetone may be found in spirits. Its boiling-point is 56°·5 C., so that it will be found, if present, in the first fraction of a distillation. It gives the usual reactions of ketones. Special tests are a red colour with

¹ *Ann. de Chim. Anal.*, 1899, ix. 42–44.

nitro-prusside of sodium *plus* ammonia; or the production of indigo blue by the addition of ortho-nitro-benzaldehyde and soda lye until alkaline.

Acetone may also be detected by the Reynolds-Gunning reaction. To an alcoholic solution of mercuric chloride, alcoholic potash is added to alkaline reaction and mixed with the solution supposed to contain acetone, shaken, and filtered; if acetone be present, the filtrate will contain mercury, as shown by a black precipitate with ammonium sulphide. Acetone¹ can be titrated by means of potassium iodide and sodic hypochlorite. The process depends on the transformation of acetone into iodoform, and the fact that until all the acetone is converted, no free iodine is liberated. The hypochlorite solution is thus standardised:—A solution of acetone in water of 0.4 per cent. is prepared; to 100 c.c. of this, 10 grms. of potassium iodide and 20 c.c. of pure soda lye of 28° Béaume are added; when the iodide is dissolved, the hypochlorite solution is added drop by drop from a burette, until a drop of starch solution to which sodium bicarbonate has been added strikes a blue colour. Solutions or distillates of acetone are dealt with similarly.

Ethers.—Ethers are detected and estimated as follows:—A known volume of alcohol is distilled; the distillate is exactly neutralised by decinormal soda or potash; then to every 100 c.c. of the distillate, 20 to 30 c.c. of decinormal alkali are added, and the flask with its contents connected with an inverted condenser and submitted to the heat of the water bath for half an hour; at the end of that time the liquid is cooled, and the amount of alkali remaining unsaturated estimated by titration with decinormal acid. The diminution in acidity is due to the saponification of the ethers, the acid uniting with the alkali, and the alcohol being regenerated. It may be expressed as acetic ether; every cubic centimetre of decinormal alkali equals 0.0088 acetic ether.

§ 273. *The Higher Alcohols.*²—*Fousel Oil* is the name given by most chemists to amylic alcohol; it may, however, be conveniently applied to the mixture of the higher homologues of ethylic alcohol. Dr. Dupré³ detects fousel oil by oxidising one or two grms. of alcohol, previously distilled (if necessary) in the manner already described. When cool the flask is opened, the excess of bichromate reduced by zinc, and the acids are distilled off, neutralised by soda, concentrated to a small bulk, the soda salt decomposed by sulphuric acid, the acids distilled off, and ultimately converted by barium carbonate into the barium salt. The barium salt of the

¹ Robineau and G. Rollin, *Moniteur scientifique*, vii., 272.

² The higher alcohols found in spirits are usually mixtures of amylic, butylic, and propylic alcohols. M. E. Claudon and Ch. Morin have obtained the following results:—

	Sugar Fermented by Elliptic Yeast.	Cognac. Faulty Fermen- tation.	Surgeres Brand.
Normal propylic alcohol,	3.7	11.8	12.1
Isobutylic alcohol,	2.7	4.5	2.9
Normal butylic alcohol,	0.0	4.3	0.0
Amylic alcohol,	93.6	34.4	85.0
	100.0	100.0	100.0

³ *Op. cit.*

acid produced by the oxidation of fousel oil (barium valerianate, $\text{Ba}(\text{C}_5\text{H}_9\text{O}_2)_2$) contains 40.41 per cent. of barium. The barium salt of the acid derived from the oxidation of pure alcohol (barium acetate, $\text{Ba}(\text{C}_2\text{H}_3\text{O}_2)_2$) contains 53.72 per cent. of barium. If the alcohol is free from fousel oil, the barium found will closely approximate to the percentage mentioned; but if the higher alcohols are present, the percentage will be lower, and a fair approximation to the amount of fousel oil will be arrived at by estimating the salt as a mixture of baric acetate and valerianate, and calculating out the percentages on the usual principles.

L. Marquardt (*Ber.*, xv., 1370–1373) utilises the fact that amyl alcohol is dissolved out of dilute alcohols by means of chloroform. The chloroform solution is washed and oxidised, and the barium salt obtained in a similar manner to Dupré's process.

Rose's process is similar, save that the amount of amyl alcohol is estimated by the increase in bulk of the chloroform. This has been much improved by the Swiss chemists. As carried out in the laboratory at Berne, the method is as follows:—First, the alcohol is brought to 30 per cent. at 15°C . An error per cent. *plus* or *minus* in the strength causes a difference, according to M. Sell, of .0199 per cent. in the volume of fousel oil.

A special apparatus is absolutely necessary for the process. This is simply a cylindrical vessel (see fig. 64) surmounted by a globular reservoir. The lower bulb is of a capacity of 19.5 c.c.; the tube is 18.5 c.c. long, with graduations from 19 c.c. to 22.5 c.c., divided into $\frac{1}{50}$ c.c., and the divisions are 1.3 mm. apart, which permits the appreciation of $\frac{1}{100}$ c.c.; the reservoir is of from 200 to 250 c.c. capacity, and altogether the whole apparatus measures 39 cm.



FIG. 64.

20 c.c. of chloroform are placed in the apparatus, all the measurements being made at 15°C .; the meniscus of the chloroform ought, if 20 c.c. be used, to reach at this temperature the lower division of the scale; 100 c.c. of alcohol are then added, and 1 c.c. of sulphuric acid (sp. gr. 1.2857). The mixture is shaken in the perfectly corked apparatus a definite number of times; according to M. Sell it is necessary to shake 150 times. In the Berne laboratory this is done by machinery, so that each sample is submitted rigorously to the same amount of shaking. The volume of chloroform is read after

the apparatus has been allowed to rest for some time. Pure alcohol of the same strength is submitted to the same process, and the difference between the two is reckoned as fousel oil or amylic alcohol; an augmentation of volume of 0.01 is equal to a proportion of 0.006631 per cent. of amylic alcohol.

Savalle's Method of Ascertaining and Estimating the Impurities in Alcohol.—This process depends on the varying tints alcohols take when treated with pure sulphuric acid. 10 c.c. of alcohol are put in a flask with 10 c.c. of pure sulphuric acid; the mixture is heated just to ebullition; when the liquid commences to boil—in fact, immediately the first bubble is seen—the process is stopped and the mixture transferred to a glass prism (the faces of which are at an angle of $2^\circ.5$), and allowed to cool; the colour produced is compared with a series of coloured glass slips which by superposition form a chromatic scale from 1° to 15° . The process is stated to be useful, but as the colour produced with various alcohols is different, and

since the colour is also dependent on the alcoholic strength, it has only a comparative value. To meet the last objection to the process it is recommended to always bring the alcohol operated upon to 50 per cent.

With a uniform strength of 50 per cent. the degrees of colour expressed in Savalle's scale for 1 c.c. per litre of the following alcohols are thus:—

Alcohols.	Aldehydes.	Ethers.
Caprylic, 7	Furfural, Intense black.	Amyl acetate, . . . 3.0
Isobutylic, 6	Isobutylic, 9	Ethyl acetate, . . . 0
Genanthylic, 4	Paraldehyde, 8	Ethyl propionate and other ethers, . . . 0
Amylic, 2	Propionic, 9	
Propylic, 0	Genanthylic, 5	
Isopropylic, 0	Valerianic, 5	
Normal butylic, 0	Ethyl, 3.5	
Methylic, 0	Methylal, 2.5	
Glycerin, 0	Acetal, 1.5	
	Butylic, 0	

The method used by Saglier is essentially a modification of Savalle's process. Aldehydes are first fixed as follows:—50 c.c. of alcohol of 50^o and 1 gram. of metaphenylenediamine hydrochloride are boiled for an hour under an inverted condenser. The result is that a non-volatile combination of 2 molecules of metaphenylenediamine to 1 of aldehyde is produced. The same purpose could be effected by aniline phosphate. The mixture is now distilled as far as possible, and the distillate brought up to the original bulk with distilled water.

Comparison liquids are made as follows:—For comparing with liquids of high alcoholic content, a solution of isobutylic alcohol in 50 per cent. alcohol, equal to 0.5 gram. per litre. For alcohols of lower content, since the reaction succeeds better with strong alcohol, the comparison solution alcohol is of 90 per cent., containing isobutylic alcohol in the proportion of 0.1 gram. per litre. The distillate in the latter case is also brought to 90 per cent. strength.

The operation itself is done as follows:—In two different flasks are placed 10 c.c. of the sample and 10 c.c. of the comparison liquid; to each is added 10 c.c. of monohydrated sulphuric acid; the flasks are shaken, then the two liquids are heated to the boiling-point, allowed to cool, the colours compared, and the quantity estimated on colorimetric principles. In commercial spirits the colour differences of the higher alcohols are so small that it is necessary to exalt the reaction; this is effected by adding to each of the liquids before boiling 20 drops of furfural solution (1 : 1000). This mixture will give a rose colour with the higher alcohols. If the colour produced by isobutyl alcohol equals 10, then the following numbers denote the intensity of the colour with other alcohols:—Crude amylic alcohol, 6; pure iso-amylic, 4.5; normal butylic, 1; iso-propylic, 0.5; propylic, 0.

Bardy's Method.—Bardy first tests the alcohol in order to see whether it is rich or poor in fousel oil. This is done by shaking up 5 c.c. of the sample with 30 to 35 c.c. of brine coloured with aniline violet. If much fousel oil is present, oily drops strongly coloured with the aniline blue float on the surface; if, as is commonly the case, only small quantities are present, there is no oily layer. In the latter case the process is as follows:—100 c.c. of alcohol are introduced into a flask of 750 c.c. capacity

and 450 c.c. of brine; should any of the salt on addition of the alcohol precipitate, 50 c.c. of water are added. Then from 60 to 70 c.c. of carbon bisulphide are added, and the mixture shaken in a separating tube; after shaking for some little time, the carbon bisulphide is run off and a fresh quantity of the latter taken, and this is done three times. The whole of the isobutylic and amylic alcohols will be in the carbon bisulphide.

The next process is to recover the alcohols from the bisulphide and convert them into ethers. For this purpose the carbon bisulphide is shaken up with 2 to 3 c.c. of monohydrated sulphuric acid, and the acid separated, the last traces being recovered by shaking and separating with a single c.c. of the acid three times. The sulphuric acid is freed from the carbon bisulphide by a current of air. 15 grms. of sodic acetate are added, and the acetic ethers formed by heating in a water bath under an inverted condenser. After cooling, brine is used to separate the ethers, the brine with its contained ethers being conveyed to a graduated vessel, and the volume of the ethers floating on the top read; the volume multiplied by 0·8 equals the proportion of isobutylic and amylic alcohols.

Bardy's method for considerable quantities of the higher alcohols is similar, only in that case on treating the alcohol with brine the higher alcohols may rise in the form of an oily layer; in such case the volume is carefully read and the alcohols separated. The brine is then treated by bisulphide of carbon, and small quantities dissolved in the brine recovered, as before stated. Propylic alcohol is insoluble in CS_2 . In order to estimate it Bardy recommends the brine to be freed from CS_2 by filtering it through wet filter paper, then to distil until the distillate contains alcohol of 50 per cent. The alcohol is titrated with permanganate as follows:— 1 c.c. of a 1 per cent. solution of permanganate and 50 c.c. of water are put in a beaker, and the alcohol dropped from a graduated burette until a tint is obtained equal to the tint of a liquid made as follows: 20 c.c. of a solution of fuchsine, 0·01 per cent., is added to 30 c.c. of neutral chromate of potassium of 0·05 per cent., and completed up to 100 c.c. with distilled water. It is necessary to add about 2·5 c.c. of alcohol of 50 per cent., containing 1 per cent. of propylic alcohol, to obtain a red tint. If the number of cubic centimetres employed is equal to n , the amount of propylic alcohol, x , is found by the following equation—

$$x = \frac{2.5}{n}$$

J. Traube's Process.—J. Traube¹ has published an entirely new process for the determination and detection of fousel oil, based upon the variations in height of capillary ascent between pure alcohol and mixtures of amyl and ethyl alcohol. A capillarmeter, consisting of a thin-sided capillary tube, about ·8 mm. in diameter, is used; this is fastened to a very fine scale divided into half-millimetres. The scale terminates at its zero in two points, which are set exactly at the surface of the liquid by means of a stand, movable upon screws. The liquid to be examined is worked up in the capillary tube two or three times, and the bottom of the meniscus is read off a few seconds after the meniscus has come to rest.

The senior author has examined in this way a number of solutions containing ·1 to 1 per cent. by volume both of crude fousel and of pure

¹ See *Journal für prakt. Chemie*, new ser., xxx., 1-177, 178. *Zeitschrift für Spiritus Industrie*, 1886, No. 36. *Chem. News*, June 25, 1886.

isoamylic alcohol dissolved in an alcohol, the specific gravity of which always corresponded to that of a 20 per cent. volume of alcohol.

The following are the main results reduced from the original paper:—

Percentage of the Fousel or Amyl Alcohol in a 20 per cent. vol. of Alcohol.	Pure Iso-amylic Alcohol. Height in mm.	Mean of Results of various samples of Fousel Oil. Height in mm.	Percentage of the Fousel or Amyl Alcohol in a 20 per cent. vol. of Alcohol.	Pure Iso-amylic Alcohol. Height in mm.	Mean of Results of various samples of Fousel Oil. Height in mm.
0	50.0	50.0	0.6	45.2	45.9
0.1	48.6	49.1	0.7	44.3	45.1
0.2	47.5	48.1	0.8	43.4	44.5
0.3	46.9	47.5	0.9	42.2	43.7
0.4	46.3	46.7	1.0	41.7	43.3
0.5	45.9	46.4			

Traube dilutes spirits down to 20 per cent. strength, and then uses the capillarmeter; it would, however, seem most accurate rather to test the distillate. In other words, distil a known volume of the spirit almost to dryness; take the gravity of the distillate in the usual way; and having thus determined the amount of alcohol present, dilute to 20 per cent., and then use the capillarmeter; an error in the determination of the specific gravity, even if corresponding to $\frac{1}{2}$ per cent. of alcohol, makes no essential difference, and some ethylic alcohols of 18 to 22 per cent. depress the rise equally to alcohols of 20 per cent.; an inaccuracy in the dilution is not likely to cause much error. Since the rise in a capillary tube depends on its diameter, and is influenced somewhat by temperature, analysts who try this method should standardise their own capillarmeter by means of known dilutions of ethylic and amylic alcohols.

Bases.—These in wines and spirits consist of ammonia, amides, alkaloids, and pyridine bases. The French school apparently use what is essentially Wanklyn's process; the free ammonia they return as ammonia and amides, the albuminoid ammonia they return as alkaloids and pyridine bases; of course, the ammonia from the last does not represent all the nitrogen the alkaloids, etc., contain, and it would have been better in the table on p. 386 to have expressed them as ammonia and albuminoid ammonia. The process used is shortly as follows:—100 c.c. of the alcoholic liquid are distilled nearly to dryness, first adding 2 c.c. of syrupy ammonia-free phosphoric acid. The residue is now transferred to a retort of 2 litres capacity, containing a litre of water alkalisied by 20 grms. of sodic carbonate and previously distilled until no ammonia comes over. The residue being added, the distillation is recommenced and the ammonia estimated in the way described in the article on 'Water' (*post*); after distillation of the free ammonia, alkaline permanganate is added and the albuminoid ammonia obtained.

BRANDY.

§ 274. Brandy (Cognac), is a spirit derived from the distillation of wine; inferior varieties are made from the refuse of grapes, with admixture of other materials.¹ The constituents of brandy are—alcohol, water,

¹ The brandy made in England is for the most part artificial. A very usual process is to add to every 100 parts of proof spirit from half-a-pound to a pound of argol, some bruised French plums, and a quart of good Cognac; the mixture is then distilled, and a little acetic ether, tannin, and burnt sugar added afterwards.

Artificial Cognac is sometimes made by oxidising palm-oil by potassic bichromate and sulphuric acid, and then distilling it with 70 per cent. of alcohol and concentrated sulphuric acid. Cœnanthic ether is among the products.

acetic, cœnanthic (ethyl pelargonate), butyric, and valerianic ethers, small quantities of grape sugar, minute quantities of a volatile oil, colouring-matter, a trace of tannin, acetic acid, and a little fixed acid. The specific gravity of genuine brandies varies from .929 to .934; the solids from 1 to 1.5; ash, .04 to .2; acidity, .01 to .05 (reckoned as tartaric acid); and the sugar from 0 to .4 per cent.¹

The adulterants to be looked for are—methyl and amylic alcohols in excess, tannin in excess, sulphuric acid, lead, copper, zinc, and hot principles, such as cayenne. The estimation of alcohols and the examination of the distillate have been already described, and an analysis of brandy, according to the French method, is tabulated on p. 386. The standard fixed by statute for brandy, whisky, or rum being 25° under proof, some analysts state in their certificates the excess of water added over that amount. This is easily obtained by the use of the following formula:—Let x = percentage of water by volume added to spirit of 25° U.P. to produce a sample N° U.P., then

$$x = 4 \frac{(N - 25)}{3}$$

For example, a brandy of 30° U.P. = $\frac{20}{3}$ or 6.6 per cent. water added.

Fixed free acids will remain in the residue, and (if no free mineral acid is detected) may be estimated volumetrically, and returned as tartaric acid (1 c.c. of d. n. soda = .0075 tartaric acid).

If the residue taste hot and pungent, some sophistication, such as capsicum, must be present; an attempt should be made to separate such hot substances in a comparative state of purity by benzole, ether, etc.

If the brandy is coloured by caramel alone, on the addition of a persalt of iron there will be no deepening of tint; if, on the contrary, the colouring be derived from the cask, and the brandy contain tannin, it will deepen very decidedly in colour.

Since it appears that in some of the recipes of the trade tincture of oak bark is used,² should the precipitate by a persalt of iron be very evident, the amount of tannin must be estimated. The direct addition of hydric sulphide to acidified spirits produces a colour sufficiently dark to be noticed should lead or copper be present, especially if an equal bulk of the unsulphuretted spirit be compared side by side. If zinc be sought, it will be best to add carbonate of soda, evaporate to dryness, and proceed as in the article on 'Zinc' in the work on 'Poisons,' by A. Wynter Blyth, to which the reader is also referred for special processes for the detection of lead and copper.

¹ Brandy 25 years old has been examined by C. Ordonneau (*Compt. Rend.*, cii., 217-219); he separated by fractional distillation the following substances, and estimated them in grammes per hectolitre:—Aldehyde, 3; ethyl acetate, 35; acetal, traces; normal propyl alcohol, 40; normal butyl alcohol, 218.6; amyl alcohol, 83.8; hexyl alcohol, 0.6; heptyl alcohol, 1.5; propionic, butyric, and caproic ethers, 3; cœnanthyl ether, about 4; amines, traces. Ordonneau considers the peculiar fragrant odour of brandy (as well as that of wines) to be due to a very small quantity of a terpene boiling at 178°. The oxidation of this terpene gives to old brandy its characteristic properties.

² Mr. Griffin found a large quantity of tannin in a sample of brandy. He explains the circumstance by the following recipe for the manufacture of Cognac:—Take of acetic ether three quarters of a pound, French wine eight quarts, oak-bark tincture (made with a quarter pound of oak bark and half a quart of spirit) half a quart, purified spirits so much as to bring the whole to 150 quarts of 54 per cent. by Tralles.—"Chemical Testing of Wines and Spirits." By John J. Griffin, F.C.S., London, 1872.

RUM.

§ 275. The best rum is distilled from fermented molasses, inferior kinds from the *débris* of the sugar-cane. In France a considerable amount of spirit is also derived from the molasses of the beetroot-sugar factories. The specific gravity of rum varies from $\cdot 874$ to $\cdot 926$; it is usually a strong spirit, genuine rum never falling below 28 per cent. of alcohol, and often reaching as high as 70 per cent. It is always slightly acid (about $\cdot 5$ per cent.). The solid residue varies from $\cdot 7$ to $1\cdot 5$ per cent. It may contain sugar; the proportion of ash is very small, seldom more than $\cdot 1$ per cent. The analysis is carried out strictly on the principles before described,¹ and the calculation for added water is the same as for brandy (see previous page).

A. Scala has made some careful analyses of rum, the results of which are as follows:—

	Seven Jamaica Rums.			Sample of Martinique Rum.	Artificial Rum.	Five Samples of Rum-Essence. ²		
	Mean.	Max.	Min.			Mean.	Max.	Min.
Formic ether, . . .	0·0052	·009	·004	0·012	0·026	0·86	1·36	0·46
Acetic ether, . . .	0·079	·141	·024	0·297	0·169	12·81	26·90	6·11
Alcohol (weight), . .	39·38	53·00	28·69	44·36	48·05	54·61	78·40	44·50
Amylic alcohol, . . .	0·109	0·206	0·033	0·266	0·169
Schiff's test for aldehydes, . . .	Two of the 7 samples no colour, the others contained aldehyde.			Dis-tinct colour.	Very weak colour.
Extract, . . .	1·066	2·21	0·194	0·364	...	3·90	7·94	1·98
Ash, . . .	0·016	0·028	0·011	0·062	...	1·31	5·02	0·06
Colour produced by sulphuric acid, . . .	Three samples no colour.			Weak reaction.

All the rum-essences had a cinnamon odour, and were coloured with caramel.

WHISKY.

§ 276. *Whisky* is one of the corn spirits, and is usually manufactured from malted grain. The Scotch distillers for the most part make it entirely from a malt mash, the Irish use malt and raw grain; but in both countries there are considerable differences in its manufacture. The new whisky, impregnated with fousel oil, is called 'pot-still whisky.' This liquor is often blended with so-called improvers—'Hambro' sherry, prime wine, etc. The specific gravities of whiskies are usually from $\cdot 915$ to $\cdot 920$; alcoholic strength from 50 to 60 per cent.; total extract under

¹ Jamaica rum was found by H. Beckurus (*Arch. Pharm.* [3], xvij., 342-346) to vary in alcoholic strength from 63 to 90 per cent. A rum-essence is said to be made by distilling 75 kilos. of alcohol (90 per cent. vol.), 50 kilos. of wood-vinegar, 30 kilos. of sulphuric acid, 20 kilos. of manganese dioxide, and 10 kilos. of potato starch—the distillate is browned by means of burnt sugar. The essence is added in suitable proportions to diluted rectified spirit.

² *Gazzetta Chimica Italiana*, 1891.

1 per cent. ; it has a minute quantity of volatile acid (seldom, perhaps never, so high as .1 per cent.), reckoned as acetic, and a trace of ash. If sugar is found it must have been added.

The chief adulterant popularly supposed to be mixed with whisky is potato spirit, or fousel oil ; but besides this, methyl alcohol, creosote, fixed acids, and, generally speaking, the sophistications used for the other spirits, have either been proved, or their presence suspected, as fraudulent additions. With regard to fousel oil, small quantities in ordinary whisky are incidental to its manufacture, and not an adulteration.

Dr. Dupré found in a sample of Scotch whisky—

For 100 of ethylic alcohol,	0.19 per cent. amylic.
Cape smoke,	0.24 " "
Common samshoe,	0.18 " "
Fine "	0.13 " "

§ 277. The question whether the higher alcohols are injurious depends mainly on the quantity—for in small amounts they may be found in most spirits and fermented liquids. Men engaged in the manufacture of potato-spirit suffer from headache and general nervous indisposition, unless the vapours are conducted away from them. An experiment by Cross on himself showed that the vapour caused intense aching and heaviness of the head, and other unpleasant symptoms. It has also been shown that 20 cgrms. (3.0 grains) of amyl alcohol taken internally produce slight symptoms, the action being first stimulating, then depressing.

From the foregoing we are now in some degree able to judge what amount of amylic alcohol would probably have immediate injurious effects ; as to the chronic effects of small doses nothing definite is known. Turning to Dr. Dupré's first sample (a Scotch whisky containing .19 of amylic to every 100 of ethylic alcohol, the whisky itself containing 54.5 per cent. of alcohol), it would appear that such a whisky has about .49 grain of amylic alcohol to the ounce. A person taking 2 ounces of such a liquid would thus swallow nearly a grain of amylic alcohol, and if it contained three times the quantity, the active properties of the amylic would be added to that of the ethylic ; for it would then be equal to about 3 grains, the lowest amount at which distinct effects can be obtained. Anything, therefore, like a grain and a half of amylic alcohol per ounce (irrespective of the question whether such a whisky may be pronounced *adulterated* or not), may certainly be considered injurious to health. The analysis of whisky is carried out precisely upon the principles already detailed. Sir Lauder Brunton, however, refers the bad effects of spirits mainly to furfural.

§ 278. The prosecutions for mere dilution of whisky¹ have been frequent ; but few *convictions* for other adulterations appear to have been obtained. The statutory standard for the alcoholic strength of whisky is the same as for brandy, and the added water is calculated in the same way (see page 398).

GIN.

§ 279. Gin is a spirit flavoured with various substances. The statutory limit of dilution for gin is 35 degrees under proof. To calculate

¹ The legal limit of dilution of brandy, whisky, or rum is 25 degrees under proof. (See p. 35.)

the amount of water added the excess of degrees U.P. above 35 is multiplied by the factor 1.54. Thus a gin 40° U.P. would have been diluted 5×1.54 , or 7.7 per cent. The different receipts used in the trade include—

Juniper berries.	Cardamom seeds.
Coriander seeds.	Liquorice powder.
Orris root.	Grains of Paradise.
Angelica root.	Cassia buds.
Calamus root.	

These generally impart their essential oils to the spirit. A few of the more important only will be described here.

§ 280. *Oil of Calamus*.—The oil distilled from the root of the calamus is a somewhat thick, yellow, or brownish-yellow oil; neutral, specific gravity 0.950 to 0.952; after rectification, 0.950; boiling point, 196°. It contains oxygen, is but slowly changed by sodium, and does not fulminate with iodine. It dissolves in all proportions in alcohol and bisulphide of carbon. The spirituous solution of the oil takes a brown colour on the addition of a little chloride of iron.

§ 281. *Oil of Cardamoms* is a pale yellow oil, with a strong smell of cardamoms, of neutral reaction, of specific gravity 0.92 to 0.94. It contains a stearoptene of the formula $C_{10}H_{16}3H_2O$.

§ 282. *Angelica root* contains a volatile acid—*Angelic acid*—a bitter principle, a crystalline substance—*Angelicine*—a resinous substance, an essential oil, and other constituents.

Angelic Acid, $\begin{array}{c} \text{H} \\ \diagdown \\ \text{C} = \text{C} \\ \diagup \\ \text{CH}_3 \end{array} \begin{array}{c} \text{CO}_2\text{H} \\ \diagdown \\ \text{C} \\ \diagup \\ \text{CH}_3 \end{array}$, forms transparent glittering prisms and

needles, melting at 45° into an oil, which may be solidified at 0° into a crystalline solid. If the heat be raised up to 185° it boils and distils unchanged; it is inflammable, burning with a luminous flame. The acid reddens litmus, and has the odour of the root. It scarcely dissolves in cold, but is soluble in hot water, in alcohol, ether, turpentine, and the fatty oils. It forms salts with bases, which lose a part of the acid on evaporation. It precipitates lead and silver salts white; iron salts, dark yellow; and copper, bluish. By the aid of hydric iodide and red phosphorus, acting at 180° to 200°, angelic acid is changed into valerianic. Melting the acid with KHO decomposes it into propionate and acetate of potash.

Angelicine is, according to Brunner, probably identical with *hydrocarotin*, a principle described by Husemann, found in the *Daucus carota*, L., and to which the following formula is ascribed, $C_{18}H_{30}O$. Hydrocarotin, or angelicine, forms colourless, large, thin plates, without smell or taste, swimming in water, and becoming at 100° hard and brittle. At higher temperatures (120°.5) it melts without loss of weight to a yellow fluid, which solidifies as a resinous mass, and cannot be again crystallised. It is readily soluble in ether, chloroform, carbon bisulphide, benzene, oil of turpentine, and warm olive oil. It is not changed in colour by concentrated hydrochloric acid; fuming nitric acid dissolves it with the evolution of gas. Concentrated sulphuric acid dissolves it to a red fluid, depositing brownish-white flakes on dilution with water.

Angelica Oil is colourless, and lighter than water; it has a penetrating odour and camphor-like taste, and resinifies on exposure to the air.

§ 283. *Oil of Coriander* is a pale yellow oil, smelling like the fruit; of specific gravity .871 at 14°, and a portion distilling over at 150°. The

volatile part corresponds to the formula $C_{10}H_{18}O_2 = (CH_3)_2C : CHCH_2CH_2C(CH_3)OH.CHCH_3$.

§ 284. *Oil of Juniper* is contained in the unripe berries of the common juniper, in the proportion of from '4 to '75 per cent. It is colourless, or of a pale yellow, dissolving with turbidity in twelve parts of alcohol of 83 per cent. Miscible in all proportions with ether and bisulphide of carbon. Smell and taste mildly aromatic. Specific gravity 0·862 to 0·874, but the poorer commercial samples often have a specific gravity of 0·860. The perfectly colourless oil does not fulminate with iodine, but the commoner kinds explode powerfully. If from 5 to 6 drops of the oil be placed in a test-tube, and five times its bulk of sulphuric acid be added, much heat is developed with the evolution of vapour, and the fluid becomes dark yellow-red and turbid; on now diluting with 10 c.c. of 90 per cent. alcohol, the colour changes to a somewhat dirty rose tint. The pure oil boils between 140° and 150°. On exposure to the air, oxygen is absorbed; and on long standing, colourless tables of juniper camphor are separated. This camphor melts and sublimes without decomposition, is easily soluble in ether and alcohol, and may be obtained in feathery crystals.

The action of warm water on juniper oil, if kept up for some considerable time, results in the formation of a crystalline hydrate. Oil of juniper is official in all the Continental pharmacopœias, as well as our own. In such large doses as from 15 to 30 grms., it is fatal to kittens, apparently acting in the same way as turpentine.

§ 285. *Analysis of Gin*.—The analyst should find in good gin at least 80 per cent. of proof spirit, and a variable amount of sugar and flavouring matters, seldom much over 5 or 6 per cent. Sulphuric acid, sulphate of zinc, alum, and lead should always be looked for. Many writers seem to imagine that grains of paradise is an *adulterant*. It is, however, in its properties merely a pepper, and much nonsense has been talked about it. It is very doubtful whether any just conviction would be obtained for the addition of any harmless flavouring to the spirit; nearly all prosecutions hitherto have been for dilution, and for dilution only.

The alcohol is determined by distillation, as before described (p. 387), and the percentage in the distillate estimated by specific gravity, and, if necessary, in other ways. Neither methyl-alcohol nor fousel-oil appears to have been found in gin.

The residue after the distillation may be treated with petroleum ether, benzine, etc., as in Dragendorff's process for the testing of beers. The essential oils will be taken up by the petroleum ether, and may be identified by their odour and taste, and (if enough is obtained) by their physical properties. Sulphuric acid, if in a free state, may be separated by cinchonine, as recommended under the article 'Vinegar.' The detection of alum, lead, and zinc is elsewhere described.

ARRACK.

§ 286. The best qualities of arrack are manufactured by distillation of the fermented juice of the cocoa-nut tree, palmyra tree, and other palms; the coarser kinds are made from the distillation of fermented rice liquor. Arrack is nearly colourless, a slight tinge of yellow or brown being only

observed in samples kept in casks for a length of time. The average strength and composition of arrack are as follows:—

	Per cent.
Specific gravity,	·9158
Alcohol,	52·700
Extract,	·082
Ash,	·024
Water,	47·194

The Hindoos and Malays consume large quantities of arrack. It would appear in the East to be occasionally drugged, and Indian hemp and the juice from solanaceous plants are said to be employed for this purpose.

LIQUEURS OR CORDIALS.

§ 287. The term cordial, liqueur, etc., is applied to a number of liquids which essentially consist of very strong spirit, flavoured with essences, and often very brightly coloured by vegetable colouring-agents, such as turmeric, cochineal, etc. Occasionally injurious colours are used, and salts of copper, picric acid, and impure aniline dyes have been detected. Of the liqueurs, absinthe is the most important in relation to health, and will be considered separately. The alcoholic strength and general composition of a few others are as follows:—

	Specific Gravity.	Alcohol by Weight.	Extract.	Cane Sugar.	Other Extractive Matters.	Ash.
Bonekamp of Maagbitter,	·9426	42·5	2·05	0·106
Benedictine-bitter,	1·0709	44·4	36·00	32·57	3·43	0·043
Ingwer,	1·0481	40·2	27·79	25·92	1·87	0·141
Crème de Menthe,	1·0447	40·7	28·28	27·63	0·65	0·068
Anisette de Bordeaux,	1·0847	35·2	34·82	34·44	0·38	0·040
Curaçoa,	1·0300	47·3	28·60	28·50	0·10	0·040
Mûmmel-liqueur,	1·0830	28·0	32·02	31·18	0·84	0·058
Pfeffer-munz liqueur,	1·1429	28·6	48·25	47·35	0·90	0·068
Swedish Punch,	1·1030	21·6	36·61

ABSINTHE.

§ 288. Absinthe is a yellowish-green liqueur, which contains, as a peculiar and distinctive ingredient, a poisonous oil having a deleterious action on the nervous system. This, *wormwood oil*, is the produce of the *Artemisia absinthium*. Other flavouring oils are always present, such as peppermint, angelica, cloves, cinnamon, and aniseed. The green colour is produced by the juice of spinach, nettles, or parsley, or, in other words, it is due to chlorophyll. The absorption-spectrum of properly made absinthe is the same as that of chlorophyll. Most samples of absinthe

contain sugar. The average composition of the liqueur as consumed in London (where its use is on the increase) is as follows :—

	Per cent.
Absolute alcohol,	50·00
Oil of wormwood,	·33
Other essential oils,	2·52
Sugar,	1·50
Chlorophyll,	traces
Water,	45·65

On diluting absinthe the essential oils are thrown out of solution, and the liquid becomes turbid. The reaction is always slightly acid, due to a trace of acetic acid.

Adulterations of Absinthe.—The composition of absinthe appears to be fixed by no definite standard of strength ; therefore, practically, the analyst has to look only for such substances as injurious colouring-matters and metallic impurities. Sulphate of indigo with turmeric is not unfrequently employed as a colouring agent, and similarly picric acid has been detected, and salts of copper. The latter is readily discovered by diluting the liqueur and adding ferrocyanide of potassium, which, if copper be present, will give a brown coloration ; picric acid and indigo are detected in the way elsewhere described. (See *Index*.)

Analysis of Absinthe.—The alcohol may be determined by distillation, after diluting the liqueur to cause the oils to separate, and getting rid of some portion by filtration. To make an estimation of the essential oils, a measured quantity of the liqueur is diluted to twice its volume by the addition of water ; carbon bisulphide is added, and the mixture shaken up in the tube described at p. 223. The carbon bisulphide dissolves all the essential oils, and on evaporation leaves them in a state pure enough to admit of their being weighed. Absinthe, when taken habitually and for a lengthened time, produces a peculiar train of nervous symptoms which the French physicians affect to distinguish from the similar symptoms produced in inebriates by alcohol. In epilepsy caused by indulgence in absinthe, M. Voisin states, as the results of clinical observation, that the number of fits is far greater than in alcoholic epilepsy.

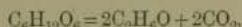
FERMENTATION : FERMENTED LIQUORS.

“The chemical Act of fermentation is essentially a correlative phenomenon of a vital act beginning and ending with it. I think that there is never any alcoholic fermentation without there being, at the same time, organisation, development, and multiplication of globules, or the continued consecutive life of globules already formed.”—*Pasteur*.

§ 289. Fermented liquors are those manufactured by fermentation—*i.e.*, a peculiar, low vegetable growth has been allowed to grow and multiply, assimilating material in one form, and excreting it in another, and this process has evolved, with other by-products, carbon dioxide and alcohol. Fermentation used to be considered as a sort of special action ; but modern research recognises a great variety of ferments, and it is a question whether all animal and vegetable cells, in the exercise of their normal functions, do

not act in a similar manner to the environing fluids, as do the organisms more distinctively called 'ferments.'

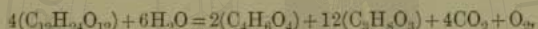
In normal alcoholic or spirituous fermentation, as in the manufacture of beer, a minute unicellular plant, 'yeast,' grows and multiplies, and splits up the sugar, as in the following equation:—



that is, one molecule of sugar furnishes two of alcohol and two of carbon dioxide. In ordinary practice this complete reaction never occurs, but if by the constant removal of carbon dioxide and alcohol, by means of a mercury pump, atmospheric air be excluded, the equation is nearly realised. M. Pasteur, in his quantitative research on the products of ordinary fermentation,¹ found that 100 parts of cane-sugar, corresponding to 105.26 parts of grape-sugar, gave nearly

Alcohol,	51.11	
Carbon dioxide,	48.89 according to Gay Lussac's equation	
	0.53 excess over	" "
Succinic acid,	0.67	
Glycerin,	3.16	
Matter united with ferments,	1.00	
	<hr/>	
	105.36	

M. Monoyer² has represented Pasteur's results in the form of the following equation:—



the free oxygen being supposed to serve for the respiration of the yeast cells. Pasteur's equation is far more complex, and represents no free oxygen as produced; yet the fact of fermentation going on briskly in a vacuum, as well as other considerations, points to the probable correctness of Monoyer's supposition.

As a secondary product, there is also constantly acetic acid. This is generally considered to originate from the ferment itself. With ethyl-alcohol there is also produced, when complex matters are fermented, several other homologous alcohols. For example, if potatoes are fermented, on distilling off the more volatile portions, and collecting separately the final

¹ The method adopted by M. Pasteur to detect and measure quantitatively the glycerin and succinic acid contained in a fermented liquid, was as follows:—"The liquid, when the fermentation was over, and all the sugar had disappeared (which required from fifteen to twenty days under good conditions), was passed through a filter, accurately weighed against another made of the same paper. After having been dried at 100°, the dried deposit of the ferment collected at the bottom of the vessel was also weighed. The filtered liquid was subjected to a very slow evaporation (at the rate of from twelve to twenty hours for each half litre); when reduced to 10 or 20 c.c. the evaporation was completed in a dry vacuum. The syrup obtained was next exhausted with a mixture of alcohol and ether (1 part alcohol of 30° and 1.5 parts ether), which extracted completely all the succinic acid and glycerin. The ether-alcohol was distilled in a retort, then evaporated in a water-bath, and afterwards in a dry vacuum. To this was added a little lime water, to fix the succinic acid, and again the mixture was evaporated, and then the glycerin was separated from the succinate of lime by ether-alcohol, which only dissolved out the glycerin. This ether-alcohol solution on evaporation, which was finished as before in a dry vacuum, left the glycerin in a state fit to be weighed, and the calcium succinate was purified by treatment with alcohol of 80 per cent., which only dissolved out the foreign matters."—Schützenberger on "Fermentation." Lond., 1876.

² "Thèse de la Faculté de Médecine de Strasbourg."

distillate, this is found to consist of propyl, butyl, amyl, caproic, cœnanthyl, and caprylic alcohols.¹

§ 290. *Yeast*.—There are no less than three methods of causing the wort of beer to ferment, but none of the three presents any very distinct variety of the yeast ferment. The one generally employed in England is what is called top fermentation, in which the starch of the malt is changed into sugar by successive infusions, and the fermentation takes place at from 15° to 18°. This in breweries is done in large open vats; the yeast floats on the surface, and can be removed by skimming.

The second method, more in use in Germany, is fermentation by sedimentary yeast (*Unterhefe*). The starch is transformed by decoction, and the temperature (12° to 14°) is much lower than the former. The yeast forms a sediment on the bottom; and when the first and most active fermentation is over, the clear liquid is run off into proper vessels; but the yeast not having been all deposited, a slow fermentation still goes on for a long time.

A third method, employed in Belgium, is to leave the wort to itself, having first placed it under proper conditions.

The microscopical appearance of yeast is that of a number of round or oval cells, from .00031 to .00035 in. (.008 to .009 mm.) in their greatest diameter. They are transparent, with one or two vacuoles; contain often a somewhat granular protoplasm, and occur together, united two by two; or, if in active growth, in groups, of which seven is a very common number. These groups are derived from offshoots or buds from a single cell, which will be found somewhere near the middle, and can be identified by its greater size. The usual teaching as to the mode of propagation has been hitherto that this growth is effected by a true budding; but according to Dr. de Vaureal, the supposed budding is an optical delusion. He considers the utricle of yeast as allied to the spermatogones of Tulasne. The nucleolar elements of the cell are spermatogones; and being set free by the rupture of the cell-wall, produce new cells. This explanation would at all events account for the ready manner in which yeast germs are carried about by the air.

Schlossberger and many other chemists have determined the chemical composition of yeast. Schlossberger gives the mean of two analyses as follows [the elementary composition after removal of ash]:—

	Surface Yeast.	Sedimentary Yeast.
Carbon,	49.9	48.0
Hydrogen,	6.6	6.5
Nitrogen,	12.1	9.8
Oxygen,	31.4	35.7
[Ash,]	2.5	3.5]

The ultimate principles contained in yeast appear to be, certain albuminoid substances, tyrosine, leucine, [nuclein?], cellulose, and some other hydro-carbons, but the matter is not yet fully worked out.

The ash of yeast has been analysed by Mitscherlich, and Schutzenberger arranges his analyses as follows:—

	Surface Yeast.	Sediment Yeast.
Phosphoric acid,	41.8	39.5
Potash,	39.8	28.3
Soda,
Magnesium Phosphate (Mg_2PO_4),	16.8	22.6
Calcium Phosphate (Ca_2PO_4),	2.3	9.7

¹ M. Jeanjean has separated camphyl-alcohol ($C_{10}H_{16}O$) from the distillate of madder.

The researches of Ergol, Rees, Pasteur, and others have shown that there are a great variety of alcoholic ferments: the wine ferment is distinct in size and shape from the beer-yeast ferment; while the ferment of fruit-juice, again, differs in the figure of its cells from either; and to the various alcoholic ferments names have been given—*e.g.*, *Saccharomyces ellipsoideus*, *S. eciquius*, *S. conglomeratus*, *S. apiculatus*, etc. All of them, however, are of quite the same general type as yeast ferment.

§ 291. When liquids become sour, ropy, or putrid, in each case the change has been produced by a particular ferment. The *lactic acid ferment*, *e.g.*, decomposes sugar into lactic acid; the *butyric acid ferment* attacks fatty matters, and separates in a free state butyric acid; and *putrid ferments*, by their avidity for oxygen, split up complex organic matters into a variety of substances. Of these ferments, the most important in reference to beer is the lactic acid ferment. In normal alcoholic fermentation, M. Pasteur has proved that there is not the smallest production of lactic acid, and when this does appear, it is certain that it has originated in and contaminated the yeast used. These ferments can be recognised by the microscope, and they should be looked for generally in the sediment. For the purpose of collecting the sediment of beers, wines, water, etc., the author (A. W. B.) has devised the following tube (fig. 65). The tube is of



FIG. 65.

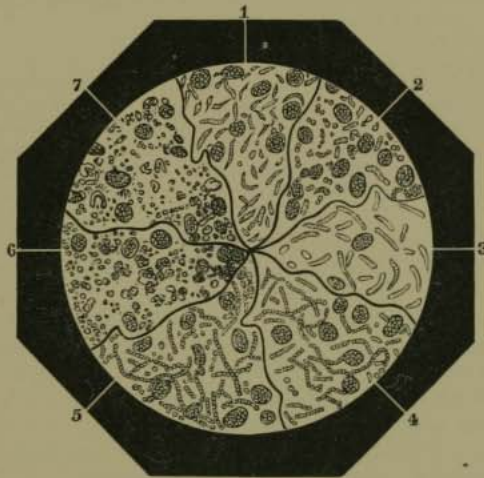


FIG. 66.

the capacity of a litre, and at the lower end is conical and open; on to this conical end is ground a glass cap, C, which is in point of fact a shallow cell about an inch in depth; P is a glass rod, the end of which fits easily into the narrow part of the tube, and is ground so as to make a perfect joint. The use of the tube is as follows:—The liquid under examination is placed in the tube closed by the cap, and the plunger P is removed; when the sediment has all collected in C, the plunger is very slowly and carefully inserted, so as to stopper the lower end of the tube; the cap may then be removed for microscopical examination.

Fig. 66 is a representation, after Pasteur, of the ferments which characterise sour or turned beer.

1. Turned beer—filaments simple or articulated into chains of different size; diameter $\frac{1}{1000}$ inch.
2. Lactic ferments of beer and wort—small, fine, and contracted in the middle; diameter a little greater than No. 1.
3. Ferments of putrid wort and beer—mobile filaments generally, which appear at the commencement of fermentation when it is slow; invariably the result of defective working.
4. Ferments of *viscous* wort and ropy beer, rare.
5. Pungent sour beer with acetic odour—chaplets of *Mycoderma acti*.
6. A wort deposit.
7. Beer of a peculiar acidity—having a detestable taste and smell, generally found with No. 1, but more to be feared than No. 1.

BEER.

§ 292. The most accurate definition of beer,¹ as brewed in the present day, is that of a fermented saccharine infusion, to which has been added a wholesome bitter. The chief constituents of beer, stouts, and porters, are—

- (1.) Water containing in solution (according to its origin) various salts. Distilled water is never used in brewing.
- (2.) Alcohol.
- (3.) Carbonic and acetic acids.
- (4.) Malt extract—malt, sugar, dextrin, albuminous constituents, and ash.
- (5.) Bitter principles, occasionally derived solely from the hop, but very commonly supplemented by so-called 'hop substitutes.' Samples of the latter examined by the author all contained 'quassia,' and portions of the following plants were identified—calumba, chiretta, gentian, and wormwood.
- (6.) The ash derived from the water, the malt, and the bitters.

To these must be added volatile and essential oils, vegetable gelatine, glucinic acid derived from the sugars, lactic acid, and, in porters, caramel and assamar.

The general composition of the chief ingredients of beer may be gathered from Table XLIII., taken from Mr. Watts' Dictionary.

The composition of beers, as a whole, varies in some degree according to the kind of ale or beer, according to the method of manufacture, and according to its age and preservation.

Pale Ale should be made from the finest and highest dried malt and the choicest hops, the bitter being in excess.

Mild Ale is a sweet, rather strong beer. *Table beer* is rarely sold—it is a weak watery ale.

Porter, as drunk in the metropolis, is a rather weak malt-liquor, coloured and flavoured with roasted malt. *Stout* is a richer and stronger description of porter.

¹ From the decisions in *Pashler v. Stevenitt* (35 L.T., 862), and *Webb v. Knight* (L.R. 2, Q.B.D. 530), it is laid down that beer must be "the article ordinarily sold under that name," and that it "would be to the prejudice of the purchaser to sell him or her as beer an article not of the nature, substance, and quality of that ordinarily sold as such, whether containing ingredients injurious to health or not." (The Sale of Food and Drugs Acts, by W. J. Bell, Esq. London, 1886.)

The *German Beers* generally are fermented by sedimentary yeast, and are always, by reason of the after fermentation (*Nachgärung*), well charged with carbon dioxide. The lager, summer, bock, or export beers are separated from the winter beers only by the former being brewed from a richer wort, and containing more alcohol, as well as a greater percentage of malt extract.

Bavarian Beers in some degree derive their peculiar qualities from fermentation at a low temperature. They seldom contain more than two per cent. of alcohol, are only slightly bittered, have a fine aroma, and a peculiar flavour, said to be due to the solution of a minute fraction of the resinous matters used to caulk the casks.

Lambick and Faro Beers are made with unmalted wheat and barley malt. In fermentation the wort is self-impregnated, the process sometimes taking months, and being mostly of a bottom character. The beer contains a large quantity of lactic acid, and is very hard in consequence.

Of the constituents of beer, it will be necessary to notice fully the water, the malt extract, the bitters, and the ash.

TABLE XLIII.—SPECIAL EXAMINATION OF CERTAIN BEERS.

Name of Beer.	Malt Extract. Per cent.	Alcohol. Per cent.	Carbonic Acid. Per cent.	Water. Per cent.	Analysed by
London Porter, Barclay & Perkins,	6.0	5.4	0.16	88.44	Kaiser.
London Porter,	6.8	6.9	...	86.3	Balling.
" " Berlin,	5.9	4.7	0.37	89.0	Ziurek.
Burton Ale,	14.5	5.9	...	79.6	Hoffmann.
Scotch Ale, Edinburgh,	10.9	8.5	0.15	80.45	Kaiser.
Ale, Berlin,	6.3	7.6	0.17	85.93	Ziurek.
Brussels Lambick,	3.4	5.5	0.2	90.90	Kaiser.
" Faro,	2.9	4.9	0.2	92.00	"
Salvator Bier, Munich,	9.4	4.6	0.18	85.85	"
Bock Bier,	9.2	4.2	0.17	86.49	"
Bavarian Draught Beer (Schenk Bier, Munich),	5.8	3.8	0.14	90.26	"
Bavarian Store Beer (Lager Bier, Munich, 16 months old),	5.0	5.1	0.15	89.75	"
Bavarian Store Beer, Munich,	3.9	4.3	0.16	91.64	"
" Draught Beer, Brunswick,	5.4	3.5	...	91.1	Otto.
Bavarian Beer (Waldschlösschen),	4.8	3.6	...	91.5	Fischer.
Prague Draught Beer,	6.9	2.4	...	90.7	Balling.
" Town Beer (Stadt Bier),	10.9	3.9	...	85.2	"
Sweet Beer, Brunswick,	14.0	1.36	...	84.7	Otto.
Josty's Beer, Berlin,	2.6	2.6	0.5	94.3	Ziurek.
Werder's Brown Beer, Berlin,	3.1	2.3	0.3	94.2	"
White Beer, Berlin,	5.7	1.9	0.6	91.8	"
Bière Blanche de Louvain,	3.0	4.0	...	93.0	Le Cambre.
Peterman (Louvain),	4.0	6.5	...	89.5	"

§ 293. The *water used by the brewer* is mainly interesting to the analyst on account of the common salt held in solution, since in prosecutions for the addition of salt the defence generally is, that the latter is a natural

component of the beer. Thus, Dr. Bottinger's analysis of the constituents of the water used at Messrs. Allsopp's brewery is as follows:—

	Per Gallon.
Chloride of sodium,	10·12
Sulphate of potash,	7·65
„ lime,	18·96
„ magnesia,	9·95
Carbonate of lime,	15·51
„ magnesia,	1·70
„ iron,	0·60
Silica,	0·79
	65·28

Messrs. Bass & Co.'s water (according to an old analysis of Cooper) contains chlorine equal to a little over 10 grains of common salt per gallon, and all published analyses of water used in breweries give quantities of salt under 14 grains per gallon. However, since breweries, as a rule, use hard spring water, it is quite possible for the water in particular localities to contain a much larger percentage of salt than the quantity mentioned above.

§ 294. *Malt Extract*.—The constituents of barley and also of malt are given in the following table; but of these it is the ash alone which will remain, comparatively speaking, unchanged; for by the action of mashing a very large portion of the dextrin and starch becomes changed into sugar.

TABLE XLIV.—COMPOSITION OF BARLEY AND MALT.

	Barley.		Malt.	
	Air-dried.	Air-dried.	Kiln-dried, pale.	Kiln-dried, amber.
Produce of torrefaction,	0·0	0·0	7·8	14·0
Dextrin,	5·6	8·0	6·6	10·2
Starch,	67·0	58·1	58·6	47·6
Sugar,	0·0	0·5	0·7	0·9
Cellulose,	9·6	14·4	10·8	11·5
Albuminous substances,	12·1	13·6	10·4	10·5
Fatty substances,	2·6	2·2	2·4	2·6
Ash, etc.,	3·1	3·2	2·7	2·7
	100·0	100·0	100·0	100·0

Barley contains dextrin, starch, albuminous substances, and a small quantity of fat, together with cellulose, and the ordinary saline constituents of seeds. Malt, varying a little in composition according to the heat of the final operations of the maltster, differs from barley in containing a small quantity of sugar (derived from transformation of a portion of the starch), rather more dextrin, and altogether less organic matter, the loss on malting being usually represented as—

Water,	6·00
Saline matter,	0·48
Organic matter,	12·52
	19·00

in 100 per cent. barley.¹

¹ C. O'Sullivan (*Journ. Chem. Soc.*, Jan. 7, 1886) has extracted from barley from '07

Valentine and O'Sullivan give the following as the composition of pale malt. Each substance has been directly estimated.

COMPOSITION OF PALE MALT
(According to Valentine and O'Sullivan).

	(1.)	(2.)
Starch,	44.15	45.13
Other carbo-hydrates (of which 60 to 70 per cent. consist of fermentable sugars), Inulin (1), and a small quantity of other bodies soluble in cold water,	21.23	19.39
Cellular matter,	11.57	10.09
Fat,	1.65	1.96
Albuminoids—		
(a.) Soluble in alcohol of specific gravity .820, and in cold water,63	.46
(b.) Soluble in cold water and at 68°,	3.23	3.12
(c.) Insoluble in cold water, but soluble at 65° to 70°,	2.37	1.36
(d.) Insoluble at 68° to 70°, but soluble in cold water (albumen proper),48	.37
(e.) Insoluble in cold water and at 70°,	6.38	8.49
Ash,	13.09	13.80
Water,	2.60	1.92
	5.83	7.47
	100.12	99.76

§ 295. *The Colouring-Matters of Malt.*—The colouring-matter of malt has been investigated by Sorby, and examined spectroscopically. The colour is of an orange yellow, and may be obtained from the hot water extract of malt, after having got rid of as much sugar and gum as possible. Ammonia colours it a deeper yellow, so does 50 per cent. sulphuric acid. There is no distinctive spectrum. A special test is to add citric acid to a watery or alcoholic solution, and then sodic hypochlorite. The watery solution under these circumstances becomes flesh-coloured, turbid, and after a time precipitates. The spectrum, according to Sorby, is moderately dark from D $\frac{1}{4}$ E to F $\frac{1}{2}$ G, clearer from there to F $\frac{3}{4}$ G, then moderately dark without estimable narrow bands; but in alcoholic solution, the solution remains clear, and there is a well-marked band at the yellow end of the green, which is at $4\frac{3}{4}$ of Sorby's scale (that is, between D $\frac{1}{4}$ E to D $\frac{5}{8}$ E); if the colour is deeper the band is more evident, and goes to D $\frac{2}{3}$ E. These appearances are so characteristic that the colouring of malt in complex mixtures can be discovered by them. In barley the colour does not exist.

The carbonised or high-dried malt, used to colour porters and dark beers, contains at least two colouring-matters different from that just described. The one is orange-yellow, soluble in water and strong alcohol; it gives no flesh-coloured precipitate with sodic hypochlorite and citric acid, nor does it (like the hop colouring-matters) darken by oxidising agents; it is therefore probably a caramel. The other substance may be separated as follows:—The beer is evaporated to a small bulk, and precipitated by to .09 per cent. of raffinose. Raffinose is a sugar $C_{18}H_{32}O_{16} \cdot 5H_2O$; it is capable of fermentation, does not reduce copper solution, and has an optical activity $[\alpha]_D = +135^\circ 3$.

alcohol; the colour is thrown down, mixed with gum, and by redissolving and reprecipitating it can ultimately be obtained nearly pure. In aqueous solution it gives an orange-brown spectrum, which is very slightly shaded from D $\frac{1}{2}$ E, moderately dark to E and $\frac{1}{2}$ C, after that very dark.

A sugar, 'maltose,' is obtained by the action of malt extract on starch; it has a specific rotatory power of + 150°. The behaviour of maltose to Fehling's solution, and its method of estimation, is described at pp. 129, 130. It forms a hard, white, crystalline mass, consisting of needles, which lose their water in a current of air at 100°.

§ 296. *Beer Bitters*.—Beer, bittered by the hop alone, when precipitated by acetate of lead, filtered, and the excess of lead removed by hydric sulphide, gives a filtrate destitute of bitter taste; while, on the contrary, if it has a bitter taste, some bitter other than hop has been used. Whether such bitter is an adulteration or not, will depend altogether on the nature of the substance added; for since the repeal of the hop duty in 1862, and the consequent return of the trade to other bitters, anything harmless in the way of a bitter is perfectly legal. If, however, such poisons as picric acid, picrotoxin, or colchicine, should be found, there can be no difference of opinion as to the course the analyst should pursue.

With regard to these, picric acid has certainly been discovered, and picrotoxin is strongly suspected, but as yet the latter has not been proved to be a common adulterant. The so-called discovery of colchicine in beer appears to have been throughout a mistake. A sample of beer of unknown origin was found to contain a bitter amorphous substance, soluble in water and alcohol, and capable of separation from its acid watery solution by either chloroform or ether. Nitric acid of 1·48 dissolved it with a fine reddish colour, and a mixture of nitric and sulphuric acids turned it rose-red. H. Van Geldern¹ finds that the body which gives these reactions originates in the hop, and that the reactions are only produced in the presence of gelatin; and further, that a mixture of unadulterated hops and gelatin gives all the reactions of colchicine. This observation is quite as important to the toxicologist as to the food analyst.²

§ 297. *Hops*.—Hops are the catkins of the *Humulus lupulus*, and consist of imbricated scales enclosing the so-called nut. The scales themselves are covered with aromatic superficial glands, which are designated 'yellow powder,' or lupulin. Freed as far as possible from the lupulinic grains, the scales consist of astringent matter, chlorophyll, gum, colouring-matter, and ash. The lupulin itself, according to Dr. Yves, contains the following matters:—

Volatile oil,	?
Tannin,	4·16
Extractive,	8·33
Bitter principle,	9·16
Wax,	10·00
Resin,	30·00
Lignin,	38·33
Loss,	·02

100·00

This analysis does not include the oil of hops, which is a very important

¹ *Arch. Pharm.*, 3, ix., 32.

² Phospho-molybdic acid gives a *thick yellow* precipitate with lupulin. On adding ammonia, and placing in a watch-glass, the precipitate becomes surrounded by a blue zone. Colchicine behaves in a similar way.—*Zeitschrift für analyt. Chemie*, 1880, p. 106.

constituent of the hop, and may be obtained by distilling the fresh flowers or the lupulin.

The resin and bitter principle of the hop are to the brewer the most important constituents, its commercial value being directly dependent on them.¹ According to some analyses made by Mr. Porter, the substances capable of solution in ether (viz., the oil, resin, and bitter principle) vary in different samples from 8.8 to a little over 17 per cent.²

Lupulite, or true Lupulin (a substance isolated by Lerner in 1863), can be obtained in rhombic prisms by treating fresh hops with four times their weight of ether, distilling the ether off, then adding to the extract alcohol of 90 per cent. (which leaves the wax undissolved), and again taking up the alcoholic extract with ether. The ethereal solution is repeatedly shaken with strong potash-lye to get rid of the resin, and lastly with pure water to take up the bitter matter. It is precipitated from the aqueous solution by sulphate of copper, the composition of the precipitate being $C_{10}H_{25}CuO_4$; and crystals are obtained by dissolving the precipitate in ether, decomposing with SH_2 , and evaporating the ethereal filtrate in a stream of carbonic acid gas.

A principle very bitter, but not crystalline, can also be obtained by adding to the aqueous extract of the lupulin grains a little lime, and then treating with alcohol. The solution is to be evaporated, the mass treated with water, and the solution again evaporated to dryness. On washing this residue with ether, a white uncrystallisable, bitter principle--soluble in 20 parts of water, very soluble in alcohol, and but slightly so in ether--is obtained; it is probably a derivative of Lerner's lupulite.

Lerner's Lupulite crystallises in glittering, rhombic, brittle prisms, tasteless in themselves, but with a very bitter flavour, and an acid reaction when dissolved in alcohol. They are insoluble in water, but dissolve easily in alcohol, ether, chloroform, bisulphide of carbon, benzole, and turpentine; exposed to the air, they soon become yellow and partly amorphous.

Bungener³ has extracted, by means of light petroleum, a substance forming colourless, prismatic crystals, melting at 92° - 93° , insoluble in water, and having the composition of $C_{50}H_{70}O_8$; he has named it lupulic acid. On treating the ethereal solution with an aqueous solution of copper acetate, there is a green crystalline precipitate of copper lupulate,

¹ Choline, $(CH_3)_3C_2H_4OH, N, OH$, has been discovered in hops, and has also been separated from beer, by Peter Griess and G. H. Harrow (*Journ. Chem. Soc.*, No. cclxx., May 1885, 298). The method used was to precipitate the concentrated extract acidified with hydrochloric acid, by a sufficient quantity of a solution of iodine in hydriodic acid. The choline periodide thus obtained is transformed into the very soluble choline iodide by boiling with water and filtering. The filtered liquid is next treated with silver oxide, and the solution is filtered and evaporated. To purify it further, it should be converted into the aurichloride, this latter compound decomposed by SH_2 , and decomposing the choline chloride by silver oxide. The base freed from water by evaporation forms a thick alkaline syrup, which solidifies in the desiccator to an extremely hygroscopic, friable, crystalline mass; it quickly absorbs carbonic anhydride, has a caustic and somewhat bitter taste, is quite odourless, and is decomposed at a high temperature with formation of trimethylamine. Choline chloride is also extremely deliquescent, and yields a characteristic platino-chloride, almost insoluble in alcohol, but very soluble even in cold water, crystallising out in fine orange tables. The chloride is also precipitated by gold chloride, the precipitate is yellow, easily soluble in boiling water and alcohol, and crystallising from these liquids in small needles or prisms; the formula for the gold salt is $(CH_3)_3C_2H_4OH, N, Cl, Au, Cl_3$ —it therefore contains 44.48 per cent. of gold.

² "On the Examination of Hops," by W. E. Porter, F.C.S. *Analyst*, August, 1877; January, 1878.

³ *Bull. Soc. Chim.*, xlv., 487-496.

$C_{50}H_{68}O_8Cu$. Lupulic acid reduces ammoniacal silver solutions, and is oxidised by exposure to the air to a bitter resin soluble in water. There is also in hops a narcotic alkaloid (hopeine) very similar in its effects to morphine; its composition is $C_{18}H_{20}NO_4 + H_2O$; it dissolves readily in alcohol, but is sparingly soluble in water.

M. Issleib has made an examination of the bitter principles of the hop. His method of separation was to extract with cold water, and then to treat the cold extract with animal charcoal, exhausting the latter after drying with 90 per cent. alcohol. This gave a yellow solution, from which a brown resin was precipitated on concentration, leaving in solution the bitter principle and another substance (C). The bitter may be removed by ether, and is pale yellow, amorphous, non-nitrogenous, and slightly acid, and is dissolved by alkalis with the production of a fine yellow colour. He ascribes to it the composition of $C_{29}H_{46}O_{10}$, and states that it splits up under the influence of sulphuric acid, thus—



The resin has the composition of $C_{10}H_{24}O_{31}$, and is presumed to be formed by the oxidation of oil of hops; lastly, the substance C is considered to be an oxidation product of the oil of hops, and has the composition of $C_{16}H_{18}O_6$.¹ It is probable that these substances are Bungener's lupulic acid.

Oil of Hops, according to the researches of Wagner, consists chiefly of an oxygen-holding oil, $C_{10}H_{18}O$, containing in solution a camphor boiling at 175° . The specific gravity of the oil itself is 0.968. It begins to boil at 125° , and successive portions can be separated by fractional distillation, the last passing over at 235° ; one-sixth distils over between 125° and 175° , and is of a pale colour and quite clear; about half passes over between 175° and 225° , and is also very clear; but the remainder is yellow, and the residue in the retort has the appearance of a brown turpentine.

The crude oil is brownish-yellow, and very sparingly soluble in water, one part requiring more than 600 times its weight for complete solution. It does not contain sulphur, as previously asserted by Payen and Chevallier, nor does it reduce ammoniacal solution of silver. Oxidation by nitric acid produces valerician acid and a yellow acid resin; if dropped on melted caustic potash, valerianate, carbonate of potash, and a hydrocarbon are the results. The oil appears to be neither narcotic nor poisonous, twenty drops having had no action whatever on a kitten (*W. Keil*); but the wonderful preservative properties ascribed to it by older writers are very problematical.

The ash of the hop differs but little from the ashes of foliar organs generally, the relative proportion of each constituent varying within somewhat wide limits, according to the particular variety. (See Table XLV.)

A few other bitter principles, some of which have actually been found, and the remainder are supposed to exist, may be now conveniently described. (For *Picrotoxin* and *Colchicine* the reader is referred to the work on 'Poisons,' by A. Wynter Blyth.)

§ 298. *Absinthin*, $C_{40}H_{28}O_8HO$.—The bitter principle of wormwood, *Artemisia absinthium*. It may be extracted from the hot aqueous extract of wormwood by precipitation with tannic acid, evaporating the tannate to

¹ *Arch. Pharm.* [3], xxi., 345-363; *Journ. Chem. Soc.*, March, 1881.

dryness with oxide of lead, and extracting the dried residue by alcohol, and subsequent purification with animal charcoal. Absinthin dissolves easily in alcohol and ether, with difficulty in hot water, and is scarcely soluble in cold. When obtained from alcoholic solution it is usually in the form of pale-yellow drops, which gradually become crystalline; it has a peculiar odour and extremely bitter taste, and its reaction is neutral. It reduces a hot ammoniacal solution of silver nitrate, but an alkaline tartrate of copper solution remains unchanged by it. It is dissolved by caustic potash with the production of a brown colour. Concentrated sulphuric acid produces first a brown colour, passing into a green-blue; as water is added, the colour becomes darker, until grey flocculent particles are separated. Boiling with dilute acids produces resinous products, but no sugar.

TABLE XLV.—CONSTITUENTS OF THE ASH OF HOP CONES.

	WAY AND OGSTON.			E. WATTS.
	Bentley Variety.	Golding Variety.	Grape Variety.	Grape Variety.
Potash,	11.98	24.88	25.56	19.41
Soda,	0.70
Lime,	17.93	21.59	18.47	14.15
Magnesia,	5.94	4.69	5.27	5.34
Alumina,	1.18
Ferric oxide,	1.86	1.75	1.41	2.71
Sulphuric acid,	7.01	7.27	11.68	8.28
Chlorine,	2.26
Silica,	22.97	19.71	9.99	17.88
Carbonic acid,	5.44	2.17	4.54	11.01
Phosphoric acid,	21.38	14.47	17.58	14.64
Chloride of potassium,	5.45	...	4.34	...
„ sodium,	3.42	0.12	...
Charcoal and loss,	2.44
	99.96	99.95	98.96	100.00
Ash per cent. of dry hops,	8.07	5.95	7.21	...
„ „ fresh „	7.27	5.22	6.22	6.5

The bitter principle, according to some observers, is a direct cerebral excitant; according to others, a pure tonic. It is used occasionally in medicine, in doses from $\frac{1}{2}$ grain to 2 grains, and the herb producing it is officinal in the Continental pharmacopœias.

§ 299. *Aloin*— $C_{16}H_{18}O_7$ (TILDEN), $C_{17}H_{18}O_7$ (STENHOUSE), $C_{15}H_{16}O_7$ (E. SCHMIDT)—may be obtained by treating aloes with dilute sulphuric acid, removing the deposit of resin, and evaporating to a syrup. The crystals thus obtained are yellow needles, soluble in water and alcohol, and possessing a bitter taste. The amount of crystallisation water varies from 5.89 to 14.29 per cent.; if anhydrous, the melting point of the crystals is from 70° to 80° [146° to 148° F.]; if hydrous, sulphuric acid colours it first red, then orange, and it is dissolved by caustic potash with the production of a purple-red colour. The action of nitric acid produces chrysammic, picric, and oxalic acids, with the addition of carbonic anhydride; that of zinc dust, methylantracene and anthraquinone.

§ 300. *Cnicin*, $C_{42}H_{56}O_{15}$, was discovered by Nativelle in the leaves of

the *Cnicus benedictus*; it is found also in the *Centaurea calcitrapa*, and in other composite plants. It forms transparent silky crystals, neutral, without smell, of very bitter taste, melting at a high temperature, but not subliming. Its solution turns the plane of polarisation to the right $[\alpha]_D^{20} = 130^{\circ}68$. It is scarcely soluble in cold water, more so in boiling, soluble in all proportions in alcohol and wood spirit, almost insoluble in ether. Cold sulphuric acid gives with cnicin a red solution, becoming violet on the addition of water, and yellow on the addition of ammonia. Cold concentrated hydrochloric acid dissolves it with the production of a green colour; on heating, brown drops separate, which solidify into a yellow resin.

§ 301. *Daphnin*, $C_{15}H_{16}O_9 + 2H_2O$, a crystalline principle discovered by Vauquelin, 1817, in the bark of the *Daphne alpina*, L., and *D. mezereum*, L. It forms long rectangular prisms, or fine needles, and crystallises with two atoms of water. Its reaction is neutral, its taste bitter. When anhydrous it melts at about 200° to a colourless fluid, which again becomes crystalline on cooling; if the heat is continued, it sublimes as 3 : 4 dioxycumarin (daphnetin). This substance presents itself in fine, colourless, rhombic prisms, melting at 255° ; it has a bitter taste, with a weak acid reaction, and is easily soluble in boiling water, with the production of a yellow colour; it dissolves also in boiling alcohol, but is very little soluble in ether.

Daphnin dissolves slightly in cold water, easily in hot; it is also very soluble in hot alcohol, but is not dissolved at all by ether. In solutions of the caustic and the carbonated alkalis it dissolves with the production of a yellow colour, and is also easily soluble in acetic acid. On boiling with a dilute acid, daphnin breaks up into daphnetin and sugar, and emulsin and fermentation with yeast have a similar effect. An aqueous solution of chloride of iron produces, when cold, a blue colour, and if the liquid is boiled, a dark yellow precipitate. Nitric acid colours it red.

§ 302. *Gentianin*, $C_{14}H_{10}O_5$, discovered by Henry and Caventon in 1821, but first prepared pure by Trommsdorff, is found, like gentiopierin, in the root of the *Gentiana lutea*, L. It forms long, pale-yellow, silky needles, without smell or taste, which may be sublimed above 300° without decomposition. Its solubility, according to Leconte, is as follows:—1 part requires of cold water 5000 parts, of boiling 3850 parts; of cold absolute alcohol 455 parts, of boiling 62.5 parts; of cold ether 2000 parts, for solution. Concentrated sulphuric acid dissolves it with a yellow colour; on dilution with water it separates unchanged; on being boiled with dilute sulphuric acid there is no change. If treated with pure nitric acid (1.43 specific gravity) a dark-green solution is obtained, and on adding water carefully dinitro-gentianin, $C_{14}H_8(NO_2)_2O_5 + H_2O$ separates out as a green powder. If similarly treated with strong nitric acid, and subsequent addition of water to the solution, yellow microscopical prisms are separated, probably *trinitro-gentianin*. Gentianin reduces nitrate of silver.

§ 303. *Gentiopierin*, $C_{20}H_{30}O_{12}$, first prepared pure by Kromayer in 1862, is a glucoside found in the fresh root of the *Gentiana lutea*, crystallising in colourless needles, with one molecule of water of crystallisation. The crystals effloresce in the air, and lose their transparency, becoming white and opaque. The anhydrous crystals melt between 120° and 125° to a brown fluid, which coagulates amorphously, and at higher temperatures is fully destroyed. Water dissolves it easily, but it is insoluble in absolute alcohol and in ether, though, on the other hand, weak alcohol is an excellent

solvent for it. One of the best tests for its presence is the action of concentrated sulphuric acid, giving in the cold a colourless solution, but producing with slight warming a carmine-red colour, and precipitating on the addition of water in grey flocks. It reduces an ammoniacal solution of silver nitrate, and on boiling with dilute sulphuric acid splits up into sugar and gentiogenin.

Gentiogenin, $C_{14}H_{16}O_5$, is an amorphous, yellow-brown powder, of neutral reaction and bitter taste, not easily soluble in cold water, but dissolving readily in alcohol and ether.

§ 304. *Menyanthin*, $C_{30}H_{46}O_4$, a glucoside obtained pure by Ludwig and Kromayer in 1861 from the *Menyanthes trifoliata*, L. As hitherto prepared, menyanthin is an amorphous, terebinthinate mass, becoming slowly solid on drying over sulphuric acid. It has a neutral reaction, and its taste is strongly and purely bitter. It softens at 60° to 65° , and melts at 100° to 115° to a thin, clear fluid, which again solidifies to a hard transparent mass; by stronger heating it is entirely destroyed. Concentrated sulphuric acid gives with it a yellow-brown colour, which on standing becomes violet-red, and grey flocks are separated on the addition of water. By heating with dilute sulphuric acid it splits up into sugar and menyanthol.

Menyanthol is an oil having an acid reaction, and an odour like that of oil of almonds; it is changed by the air (as well as by melting with potash) into a crystalline acid capable of being sublimed.

§ 305. *Quassin*, $C_{10}H_{12}O_3$, a bitter principle, discovered in 1835 by Winckler, in the bark of the *Quassia amara*, L., and *Picrena excelsa*, L. It forms white, opaque, glittering crystals, without odour, and of extremely bitter taste. On heating, it melts and solidifies as a transparent yellow mass; at decomposition temperatures it burns like resin, if exposed to the air. Tannic acid precipitates it from an alcoholic solution in thick, white flocks; cold concentrated sulphuric acid dissolves it, without the production of colour; and on dilution with water it is apparently precipitated without change.

§ 306. *The Ash of Beer*.—The ash of beer contains the mineral constituents that previously existed partly in the water, partly in the hop, and partly in the malt used. It would appear that the ferric oxide, a certain proportion of phosphoric acid, a small portion of the lime and magnesia, with a great part of the silica, remain undissolved, and do not pass into the beer; the rest are dissolved.

The following table gives the average composition of the beer ash of commerce:—

	Beer Ash.
Potash,	37·22
Soda,	8·04
Lime,	1·93
Magnesia,	5·51
Iron oxide,	traces
Sulphuric acid,	1·44
Phosphoric acid,	32·09
Chlorine,	2·91
Silica,	10·82

Table XLVI. may be also useful, showing analyses of ash by Walz and Dickson ("Dictionary of Chemistry, Arts, and Manufactures," edited by Vincent).

§ 307. *Analysis of Beer.*—The ordinary full analysis of beer determines—

- (1.) The alcohol.
- (2.) The carbonic acid.
- (3.) The volatile and fixed acids.
- (4.) The percentage of malt extract, and, if necessary, its composition.
- (5.) The hop resin and glycerin.
- (6.) The nature of the bitter used.
- (7.) The general composition of the ash, and especially the amount of chlorine.

(1.) *The alcohol* is found most accurately by the distillation process described at p. 387, but it is often determined in the following manner:—Shake up the beer in a flask, so as to deprive it of as much carbonic acid

TABLE XLVI.—ASH OF BEERS.

	London Beer.	Munich Beer.	Speyer Beer.	Scotch Ale, 14 Samples.	Scotch Porter, 2 Samples.	Dublin Porter, 2 Samples.	London Porter, 5 Samples.
Potash, . .	38.35	36.58	37.68	3.2 - 29.8	18.9 - 20.9	21.4 - 32.0	4.9 - 31.1
Soda, . . .	7.68	9.83	6.50	20.9 - 38.5	33.8 - 38.8	24.0 - 42.7	21.8 - 50.8
Lime, . . .	2.45	1.48	2.98	0.2 - 2.0	1.3 - 1.6	0.8 - 1.5	0.8 - 6.9
Magnesia, .	3.78	5.64	4.66	0.1 - 5.6	0.2 - 1.4	0.2 - 1.2	0.1 - 1.2
Sulphuric acid, . }	1.36	1.68	2.56	1.6 - 19.2	2.2 - 6.4	2.8 - 10.1	1.6 - 12.2
Chlorine, . .	2.75	3.14	2.14	4.3 - 18.25	7.4 - 11.4	6.9 - 10.1	6.5 - 14.5
Silica, . . .	9.87	9.96	10.29	4.6 - 19.1	13.3 - 18.6	6.9 - 19.7	8.25 - 19.7
Phosphoric acid, . }	33.76	31.69	33.19	6.0 - 25.7	12.2 - 18.8	7.9 - 20.0	9.3 - 20.6
	100.00	100.00	100.00				

as possible, then take the specific gravity at 15°·5, and boil a third of the beer away in an open beaker. Make up the boiled beer to exactly the original quality again, take the specific gravity, and calculate as described at p. 387.

(2.) *The carbonic acid* which the beer holds dissolved is most readily estimated by placing 100 c.c. (or any convenient quantity) in a flask in connection with a mercury pump; collecting the gases, and estimating the carbon dioxide in the usual way by absorbing it by caustic potash, and measuring the volume of gas before and after absorption. Another method is as follows:—A flask provided with a caoutchouc stopper, and carrying a tube twice bent at right angles, is connected with a smaller flask, containing strong ammonia water free from CO₂, into which the tube dips. This second flask must also carry a caoutchouc stopper, which should be doubly perforated, the one hole for the tube already mentioned, the other provided with a short, wide tube, packed with glass wool moistened with ammonia water. The beer must first be gently warmed and ultimately boiled, when the whole of the carbonic acid is absorbed by the ammonia. A sufficient quantity of a solution of calcium chloride is now added, the liquid boiled until all free ammonia is expelled, and the calcium carbonate thrown on a filter and washed. Lastly, the washed calcium carbonate is dissolved in either standard or decinormal acid, according to the quantity, a little cochineal solution is added, and the acid titrated back. 2 c.c. of decinormal

acid equal 10 mgrms. of carbonate of lime, or 4.4 mgrms. of CO_2 . This simple process is applicable to all liquids from which carbonic anhydride can be expelled by boiling.

(3.) *Volatile and Fixed Acids.*—The acetic acid is obtained by distilling the beer nearly to dryness, and estimating the acidity of the distillate by decinormal solution of soda. Should the residue in the flask or retort be still acid, a little water should be added, and the distillation again continued to dryness; any acid now remaining is certain to be a fixed acid, probably lactic. It may be estimated by titration, and returned as lactic. The equivalent of anhydrous lactic acid is 90; hence 1 c.c. of d. n. soda = 9 mgrms. of lactic acid. Should it be specially necessary to determine the percentage of lactic acid a sufficient quantity of beer—say 300 c.c.—is taken, evaporated to a small bulk, diluted with water, filtered, and mixed with a little sulphuric acid; pure carbonate of baryta is now added, and the whole warmed on the water-bath for some time. The liquid is then freed from the precipitate of sulphate of baryta by filtration, and the precipitate well washed with hot water. This filtrate is evaporated to a syrup, and treated, when cold, in a tube or separating-funnel with a mixture of one part of sulphuric acid, one of water, one of alcohol, and ten of ether; the ethereal layer is separated in the usual way, and evaporated. The lactic acid thus obtained is still impure, and it is best to dissolve in water, saturate with freshly-precipitated carbonate of zinc, and estimate as zinc lactate, the latter containing 54.49 per cent. of anhydrous lactic acid. In most cases, however, the error will not be great, if the total acidity of the beer is taken directly without distillation, and returned as acetic acid.

(4.) *The Malt Extract.*—The extract of beer can be determined by evaporating down a carefully measured quantity, and weighing the dry residue. In order to do this with any approach to accuracy, the smallest possible quantity should be taken—5 c.c. or 5 grms. is quite sufficient. This small quantity, spread out as a thin film on the bottom of a tolerably capacious platinum dish, can be thoroughly dried over the water-bath in two or three hours, while if such quantities as 25 c.c., 50 c.c., or 100 c.c. are taken, to get the extract completely dry is very tedious, and usually requires a higher temperature than 100° . It is, however, found in practice much more convenient to dispense with this drying altogether, the alcohol and carbonic acid being driven off, as before described, the beer made up to its first bulk, the specific gravity taken, and the amount of malt extract determined by the aid of the following tables. If the beer has been distilled, the residue in the retort or flask can be made up to the original bulk, brought to the proper temperature, and treated as just described.

The alcoholic strength, the acetic acid, and the amount of malt extract being known, the analyst can now give a fairly approximate estimate of the amount of malt originally used in the brewing of the beer. To do this it is necessary to calculate the 'original gravity' of the beer. The specific gravity of the alcoholic distillate (or, if an indirect process has been used, the specific gravity of the alcoholic strength) subtracted from 1000, gives a number called the 'spirit indication.' The degrees of gravity lost are then ascertained by the aid of the following tables, using the first if the beer has been distilled, and the second if the evaporating process has been used. The degrees of gravity thus found are added to the specific gravity of the boiled beer, and the number thus obtained is called 'the original gravity of the wort.' On reference to Table XLVII., the amount of malt extract is determined, which corresponds to this original gravity.

TABLE XLVII.—SPECIFIC GRAVITY AND STRENGTH OF MALT EXTRACT.

Specific Gravity.	Malt Extract. Per cent.	Specific Gravity.	Malt Extract. Per cent.	Specific Gravity.	Malt Extract. Per cent.	Specific Gravity.	Malt Extract. Per cent.
1,0000	0,000	1,0057	1,425	1,0114	2,850	1,0171	4,275
1,0001	025	58	450	115	875	172	300
2	050	59	475	116	900	173	325
3	075	1,0060	500	117	925	174	350
4	100	61	525	118	950	175	375
5	125	62	550	119	975	176	400
6	150	63	575	1,0120	3,000	177	425
7	175	64	600	121	025	178	450
8	200	65	625	122	050	179	475
9	225	66	650	123	075	1,0180	500
1,0010	250	67	675	124	100	181	525
11	275	68	700	125	125	182	550
12	300	69	725	126	150	183	575
13	325	1,0070	750	127	175	184	600
14	350	71	775	128	200	185	625
15	375	72	800	129	225	186	650
16	400	73	825	1,0130	250	187	675
17	425	74	850	131	275	188	700
18	450	75	875	132	300	189	725
19	475	76	900	133	325	1,0190	750
1,0020	500	77	925	134	350	191	775
21	525	78	950	135	375	192	800
22	550	79	975	136	400	193	825
23	575	1,0080	2,000	137	425	194	850
24	600	81	025	138	450	195	875
25	625	82	050	139	475	196	900
26	650	83	075	1,0140	500	197	925
27	675	84	100	141	525	198	950
28	700	85	125	142	550	199	975
29	725	86	150	143	575	1,0200	5,000
1,0030	750	87	175	144	600	201	025
31	775	88	200	145	625	202	050
32	800	89	225	146	650	203	075
33	825	1,0090	250	147	675	204	100
34	850	91	275	148	700	205	125
35	875	92	300	149	725	206	150
36	900	93	325	1,0150	750	207	175
37	925	94	350	151	775	208	200
38	950	95	375	152	800	209	225
39	975	96	400	153	825	1,0210	250
1,0040	1,000	97	425	154	850	211	275
41	025	98	450	155	875	212	300
42	050	99	475	156	900	213	325
43	075	1,0100	500	157	925	214	350
44	100	101	525	158	950	215	375
45	125	102	550	159	975	216	400
46	150	103	575	1,0160	4,000	217	425
47	175	104	600	161	025	218	450
48	200	105	625	162	050	219	475
49	225	106	650	163	075	1,0220	500
1,0050	250	107	675	164	100	221	525
51	275	108	700	165	125	222	550
52	300	109	725	166	150	223	575
53	325	1,0110	750	167	175	224	600
54	350	111	775	168	200	225	625
55	375	112	800	169	225	226	650
56	400	113	825	1,0170	250	227	675

TABLE XLVII.—*continued.*

Specific Gravity.	Malt Extract. Per cent.	Specific Gravity.	Malt Extract. Per cent.	Specific Gravity.	Malt Extract. Per cent.	Specific Gravity.	Malt Extract. Per cent.
1,0228	5,700	1,0286	7,122	1,0344	8,536	1,0402	9,950
229	725	287	146	345	560	403	975
1,0230	750	288	170	346	584	404	10,000
231	775	289	195	347	609	405	023
232	800	1,0290	219	348	633	406	047
233	825	291	244	349	657	407	071
234	850	292	268	1,0350	681	408	095
235	875	293	292	351	706	409	119
236	900	294	316	352	731	1,0410	142
237	925	295	341	353	756	411	166
238	950	296	365	354	780	412	190
239	975	297	389	355	804	413	214
1,0240	6,000	298	413	356	028	414	238
241	024	299	438	357	853	415	261
242	048	1,0300	463	358	877	416	285
243	073	301	488	359	901	417	309
244	097	302	512	1,0360	925	418	333
245	122	303	536	361	950	419	357
246	146	304	560	362	975	1,0420	381
247	170	305	584	363	9,000	421	404
248	195	306	609	364	024	422	428
249	219	307	633	365	048	423	452
1,0250	244	308	657	366	073	424	476
251	268	309	681	367	097	425	500
252	292	1,0310	706	368	122	426	523
253	316	311	731	369	146	427	547
254	341	312	756	1,0370	170	428	571
255	365	313	780	371	195	429	595
256	389	314	804	372	219	1,0430	619
257	413	315	828	373	244	431	642
258	438	316	853	374	268	432	666
259	463	317	877	375	292	433	690
1,0260	488	318	901	376	316	434	714
261	512	319	925	377	341	435	738
262	536	1,0320	950	378	365	436	761
263	560	321	975	379	389	437	785
264	584	322	8,000	1,0380	413	438	809
265	609	323	024	381	438	439	833
266	633	324	048	382	463	1,0440	857
267	657	325	073	383	488	441	881
268	681	326	097	384	512	442	904
269	706	327	122	385	536	443	928
1,0270	731	328	146	386	560	444	952
271	756	329	170	387	584	445	976
272	780	1,0330	195	388	609	446	11,000
273	804	331	219	389	633	447	023
274	828	332	244	1,0390	657	448	047
275	853	333	260	391	681	449	071
276	877	334	292	392	706	1,0450	095
277	901	335	316	393	731	451	119
278	925	336	341	394	756	452	142
279	950	337	365	395	780	453	166
1,0280	975	338	389	396	804	454	190
281	7,000	339	413	397	828	455	214
282	024	1,0340	438	398	853	456	238
283	048	341	463	399	877	457	261
284	073	342	488	1,0400	901	458	285
285	097	343	512	401	925	459	309

TABLE XLVII.—*continued.*

Specific Gravity.	Malt Extract. Per cent.	Specific Gravity.	Malt Extract. Per cent.	Specific Gravity.	Malt Extract. Per cent.	Specific Gravity.	Malt Extract. Per cent.
1,0460	11,333	1,0518	12,714	1,0576	14,095	1,0634	15,464
461	357	519	738	577	119	635	488
462	381	1,0520	761	578	142	636	511
463	404	521	785	579	166	637	534
464	428	522	809	1,0580	190	638	557
465	452	523	833	581	214	639	581
466	476	524	857	582	238	1,0640	604
467	500	525	881	583	261	641	627
468	523	526	904	584	285	642	650
469	547	527	928	585	309	643	674
1,0470	571	528	952	586	333	644	697
471	595	529	976	587	357	645	721
472	619	1,0530	13,000	588	381	646	744
473	642	531	023	589	404	647	767
474	666	532	047	1,0590	428	648	790
475	690	533	071	591	452	649	814
476	714	534	095	592	476	1,0650	837
477	738	535	119	593	500	651	860
478	761	536	142	594	523	652	883
479	785	537	166	595	547	653	907
1,0480	809	538	190	596	571	654	930
481	833	539	214	597	595	655	953
482	857	1,0540	238	598	619	656	976
483	881	541	261	599	642	657	16,000
484	904	542	285	1,0600	666	658	023
485	928	543	309	601	690	659	046
486	952	544	333	602	714	1,0660	070
487	976	545	357	603	738	661	093
488	12,000	546	381	604	761	662	116
489	023	547	405	605	785	663	139
1,0490	047	548	428	606	809	664	162
491	071	549	452	607	833	665	186
492	095	1,0550	476	608	857	666	209
493	119	551	500	609	881	667	232
494	142	552	523	1,0610	904	668	255
495	166	553	547	611	928	669	278
496	190	554	571	612	952	1,0670	302
497	214	555	595	613	976	671	325
498	238	556	619	614	15,000	672	348
499	261	557	642	615	023	673	371
1,0500	285	558	666	616	046	674	395
501	309	559	690	617	070	675	418
502	333	1,0560	714	618	093	676	441
503	357	561	738	619	116	677	464
504	381	562	761	1,0620	139	678	488
505	404	563	785	621	162	679	511
506	428	564	809	622	186	1,0680	534
507	452	565	833	623	209	681	557
508	476	566	857	624	232	682	581
509	500	567	881	625	255	683	604
1,0510	523	568	904	626	278	684	627
511	547	569	928	627	302	685	650
512	571	1,0570	952	628	325	686	674
513	595	571	976	629	348	687	697
514	619	572	14,000	1,0630	371	688	721
515	642	573	023	631	395	689	744
516	666	574	047	632	418	1,0690	767
517	690	575	071	633	441	691	790

TABLE XLVII.—*continued.*

Specific Gravity.	Malt Extract. Per cent.	Specific Gravity.	Malt Extract. Per cent.	Specific Gravity.	Malt Extract. Per cent.	Specific Gravity.	Malt Extract. Per cent.
1,0692	16,814	1,0700	17,000	1,0707	17,158	1,0714	17,318
693	837	701	022	708	181	715	340
694	860	702	045	709	204	716	363
695	883	703	067	1,0710	227	717	386
696	907	704	090	711	250	718	409
697	930	705	113	712	272	719	431
698	953	706	136	713	295	1,0720	17,454
699	976						

TABLE XLVIII.—FOR USE IF THE DISTILLATION PROCESS HAS BEEN USED.

Degrees of Spirit Indication.	·0	·1	·2	·3	·4	·5	·6	·7	·8	·9
0	0·0	0·3	0·6	0·9	1·2	1·5	1·8	2·1	2·4	2·7
1	3·0	3·3	3·7	4·1	4·4	4·8	5·1	5·5	5·9	6·2
2	6·6	7·0	7·4	7·8	8·2	8·6	9·0	9·4	9·8	10·2
3	10·7	11·1	11·5	12·0	12·4	12·9	13·3	13·8	14·2	14·7
4	15·1	15·5	16·0	16·4	16·8	17·3	17·7	18·2	18·6	19·1
5	19·5	19·9	20·4	20·9	21·3	21·8	22·2	22·7	23·1	23·6
6	24·1	24·6	25·0	25·5	26·0	26·4	26·9	27·4	27·8	28·3
7	28·8	29·2	29·7	30·2	30·7	31·2	31·7	32·2	32·7	33·2
8	33·7	34·3	34·8	35·4	35·9	36·5	37·0	37·5	38·0	38·6
9	39·1	39·7	40·2	40·7	41·2	41·7	42·2	42·7	43·2	43·7
10	44·2	44·7	45·1	45·6	46·0	46·5	47·0	47·5	48·0	48·5
11	49·0	49·6	50·1	50·6	51·2	51·7	52·2	52·7	53·3	53·8
12	54·3	54·9	55·4	55·9	56·4	56·9	57·4	57·9	58·4	58·9
13	59·6	60·0	60·5	61·1	61·6	62·2	62·7	63·3	63·8	64·3
14	64·8	65·4	65·9	66·5	67·1	67·6	68·2	68·7	69·3	69·9
15	70·5

TABLE XLIX.—FOR USE IF THE EVAPORATION PROCESS IS ADOPTED.

Degrees of Spirit Indication.	·0	·1	·2	·3	·4	·5	·6	·7	·8	·9
0	0	0·3	0·7	1·0	1·4	1·7	2·1	2·4	2·8	3·1
1	3·5	3·8	4·2	4·6	5·0	5·4	5·8	6·2	6·6	7·0
2	7·4	7·8	8·2	8·7	9·1	9·5	9·9	10·3	10·7	11·1
3	11·5	11·9	12·4	12·8	13·2	13·6	14·0	14·4	14·8	15·3
4	15·8	16·2	16·6	17·0	17·4	17·9	18·4	18·8	19·3	19·8
5	20·3	20·7	21·2	21·6	22·1	22·5	23·0	23·4	23·9	24·3
6	24·8	25·2	25·6	26·1	26·6	27·0	27·5	28·0	28·5	29·0
7	29·5	30·0	30·4	30·9	31·3	31·8	32·3	32·8	33·3	33·8
8	34·3	34·9	35·5	36·0	36·6	37·1	37·7	38·3	38·8	39·4
9	40·0	40·5	41·0	41·5	42·0	42·5	43·0	43·5	44·0	44·4
10	44·9	45·4	46·0	46·5	47·1	47·6	48·2	48·7	49·3	49·3
11	50·3	50·9	51·4	51·9	52·5	53·0	53·5	54·0	54·5	55·0
12	55·6	56·2	56·7	57·3	57·8	58·3	58·9	59·4	59·9	60·5
13	61·0	61·6	62·1	62·7	63·2	63·8	64·3	64·9	65·4	66·0
14	66·5	67·0	67·6	68·1	68·7	69·2	69·8	70·4	70·9	71·4
15	72·0

TABLE L.—FOR ASCERTAINING THE VALUE OF ACETIC ACID.

Excess per cent. of Acetic Acid in the Beer.	CORRESPONDING DEGREES OF SPIRIT INDICATION.									
	·00	·01	·02	·03	·04	·05	·06	·07	·08	·09
·0	...	·02	·04	·06	·07	·08	·09	·11	·12	·13
·1	·14	·15	·17	·18	·19	·21	·22	·23	·24	·26
·2	·27	·28	·29	·31	·32	·33	·34	·35	·37	·38
·3	·39	·40	·42	·43	·44	·46	·47	·48	·49	·51
·4	·52	·53	·55	·56	·57	·59	·60	·61	·62	·64
·5	·65	·66	·67	·69	·70	·71	·72	·73	·75	·76
·6	·77	·78	·80	·81	·82	·84	·85	·86	·87	·89
·7	·90	·91	·93	·94	·95	·97	·98	·99	1·00	1·02
·8	1·03	1·04	1·05	1·07	1·08	1·09	1·10	1·11	1·13	1·14
·9	1·15	1·16	1·18	1·19	1·21	1·22	1·23	1·25	1·26	1·28
1·0	1·29	1·31	1·33	1·35	1·36	1·37	1·38	1·40	1·41	1·42

From the malt extract the amount of malt used is calculated, 32·0 of malt equalling 21·0 of extract; or, instead of referring to the malt extract tables, 1000 may be subtracted from the original gravity, and the remainder multiplied by ·025, which will equal the total amount of malt extract per gallon in pounds weight.

EXAMPLES.

(1.) An ordinary Mild Ale.

Specific gravity of alcoholic distillate	=	1000·0
		993·6
Spirit indication,		6·4

This spirit indication, on reference to Table XLVIII., equals 26·0.

Gravity of boiled beer,	=	1014·2
Add,		26·0
Original gravity,		1040·2

Now, on reference to Table XLVII., 1040·2 correspond to 9·950 parts of extract in 100, or ·9950 lb. per gallon.

Or, if from the original gravity,	1040·2
Be subtracted,	1000
And the number obtained	40·2
Be multiplied by	·025
The product equals	1·005

which does not materially differ from the amount obtained from the tables.

(2.) A beer, by the evaporation process, indicated 6·1 per cent. of alcohol. On reference to the specific gravity table (Table XL.), p. 380, the specific gravity corresponding to this strength is ·9896.

	1000·0
	989·6
Spirit indication	= 10·4

This, according to Table XLIX., corresponds to 47·1 ;

The gravity of the boiled beer was	1016·2
Add	47·1
	1063·3
Original gravity,	1063·3

corresponding, according to Table XLVII., to 1·5441 lbs. of malt extract to the gallon. The amount of malt used in the two examples is found thus—

$$(1.) \frac{.9950 \times 320^1}{210} = 1·516 \text{ lbs. of malt to the gallon.}$$

$$(2.) \frac{1·5441 \times 320^1}{210} = 2·3528 \text{ lbs. of malt to the gallon.}$$

If the beer should have a somewhat large proportion of acetic acid (as in old and hard beers), it will be necessary to take into account the loss of gravity by acetic acid. This complicates the calculation, and is only occasionally required.

The loss of gravity by acetic acid is obtained by the use of Table L. Let us suppose ·36 per cent. of acetic acid to have been found in Example 1; from this subtract ·10 per cent., the amount which may be taken as incidental to healthy fermentation, and allowed for in the table. Thus,

Total acetic acid present,	·36
Deduct,	·10
	·26
Acetic acid to be taken account of,	·26

This ·26, according to the table, equals ·34, the number to be added to the spirit indication.

Spirit indication from alcohol,	6·4
„ „ from acetic acid,	·34
	6·74
Total spirit indication,	6·74

The rest of the calculation is as before.

It may be desirable to examine the malt extract further, and specially with a view to ascertaining whether grape sugar has been used in place of malt or not. This may be done, according to Haarstick, by taking advantage of the fact that most, perhaps all, of the grape sugars of commerce contain Béchamp's amylin, a substance of great rotatory power, and one, moreover, not destroyed by fermentation.

The samples of beer were examined by Haarstick as follows :—1 litre of beer was evaporated to a syrup, to which alcohol of 90 per cent. was added, drop by drop, from a burette with constant agitation, until a volume of about 300 c.c. had been used. The separation of dextrin was completed with 95 per cent. alcohol, until the filtrate did not show the least turbidity when mixed with an equal bulk of 95 per cent. alcohol. The mixture was then left at rest for twelve hours, filtered, and the greater part of the alcohol distilled off. The remainder was evaporated to dryness on a water-bath, the solid portion dissolved in distilled water, and the solution diluted to a litre, and fermented with yeast at 20°, until all the sugar had been

¹ Or, of course, the factor $\frac{32}{21} = 1·524$ may be used, and then the two equations are as follows :—(1.) $.995 \times 1·524 = 1·516$, and (2.) $1·5441 \times 1·524 = 2·3532$.

destroyed. The fermentation was completed on the fourth day, and the result of the process was, that beers prepared without grape sugar gave a solution of no rotatory power, while those prepared with grape sugar turned the plane of polarisation $2^{\circ}0$ to $3^{\circ}4$ to the right, as observed in Hoppe's scale.

(5.) *The Hop Resin and Glycerin.*—Griessmayer¹ estimates the hop resin and glycerin by concentrating the beer to one-third of its bulk, and shaking it up with petroleum ether; this, on separation and evaporation, leaves the hop resin. The liquid, now freed from hop constituents, is made alkaline by baryta water, or, better, by barium alcoholate, and shaken up several times with a mixture of two parts of alcohol and three of ether; the latter, on being separated and evaporated in the usual way, leaves the glycerin.

Clausnizer² gives, as the result of his elaborate experiments on the various processes for the estimation of glycerin, the following method, which would appear to be the best hitherto published: 50 c.c. of beer are warmed on the water-bath to get rid of CO_2 , and then mixed with 3 grms. of hydrate of lime and evaporated to a syrup; 10 grms. of powdered marble are then added, the mass frequently stirred, and the whole dried on the water-bath. The dish with its contents is now weighed, and an aliquot part (about two-fifths to three-quarters) put in the extraction apparatus figured at p. 50, and exhausted with 20 c.c. of alcohol of 80 to 90 per cent. The alcoholic extract (now about 15 c.c.) is mixed with 25 c.c. of anhydrous ether, the precipitate separated, and the ether-alcohol received into a weighed flask, the precipitate also being washed with alcohol-ether [2:3]. The almost colourless filtrate is freed from alcohol and ether by very slow evaporation. Lastly the glycerin is dried at 100° until in two hours no more than 2 mgrms. are lost. This process will take from two to four hours for normal beer, from four to six for beer to which glycerin has been added. It may be well to burn up the glycerin and subtract the ash found. Clausnizer gives the percentage of glycerin in common German beers as follows:—

	Alcohol.	Extract.	Glycerin. ³
Wagener's Boekbier,	3.1	6.4	.208
„ Beer,	3.0	5.2	.206
Rhine beer,	2.8	3.9	.237
Bavarian beer,	4.4	6.0	.220
„ „	3.7	4.5	.264

(6.) *The Nature of the Bitter used.*—This is the most difficult part of the investigation, and requires a very considerable amount of practical knowledge. Occasionally, if the beer is put in the tube figured at p. 407, small fragments of quassia, calumba, and similar substances will be observed to sink to the bottom as a sediment, and may be detected microscopically; indeed, it is probable that were it possible to obtain the sediment from the beer casks, many foreign matters might in this way be detected.

¹ *Ber. d. d. chem. Gesells.*, xi., 292, 293.

² Clausnizer, F.: Zur Glycerine Bestimmung in Bier. *Zeitschrift für analytische Chem.*, xx., 80.

³ E. Borgmann (*Zeitschr. anal. Chem.*, xxii., 533, 534) gives very different results, for in the examination of a large number of beers, both German and English, he found the ratio of alcohol and glycerin to average 100 alcohol: 4.8 glycerin—the extremes were $\frac{1.00}{5.5}$ and $\frac{1.00}{4.2}$. Glycerin can be estimated in beer also by Wanklyn's process (see p. 300 and p. 455).

In most cases it is sufficient to use some of the special processes here-after described, and restrict the inquiry to proving the absence of picric acid, of picrotoxin, and of any other specially noxious substance which may be suspected. Should, however, the analyst desire to examine the beer generally for various organic principles, the elaborate process as worked out by Dragendorff and others, fully described in the work on 'Poisons,' by A. Wynter Blyth, must be used. About 600 to 1000 c.c. of the beer are evaporated to a syrupy consistence, and sufficient strong alcohol is added to precipitate the dextrin. The whole is filtered, and after standing some hours, the filtrate, acidulated with sulphuric acid, and shaken up successively with petroleum ether, benzine, chloroform, and (if salicin be sought) amylic alcohol.

Or the beer may be precipitated with acetate of lead, filtered, the excess of lead thrown out by sulphuric acid, and treated with the solvents as described. If inquiry be made as to the nature of the residue left by these different solvents in beer simply made from hops and malt, it appears that—

(1.) The *petroleum ether* extracts—

- (a.) An amorphous, slightly bitter substance, soluble in ether and alcohol, and partially soluble in water; this is derived from both hops and malt;
- (b.) A substance precipitable by basic acetate of lead; and one
- (c.) Becoming red by Fröhde's reagent; both derived from the hop.
- (d.) A substance derived from the hop alone, becoming red with sulphuric acid and sugar.

(2.) *Benzine* extracts the same substances, and in addition—

- (e.) A substance derived from the hop, which precipitates tannin;
- (f.) A substance derived from the malt, becoming dark-brown on the addition of sulphuric acid.

(3) *Chloroform* extracts (a.), (b.), and (e.), and also (f.), a substance partly precipitated by potassium iodide and phosphomolybdic acid;

- (g.) A substance reducing ammoniacal solution of silver nitrate;
- (h.) A substance crystallisable from ether. All of these are derived from the malt.

The normal reactions of the substances being known, the following scheme will be found useful, always bearing in mind that few chemists, should they obtain any of the reactions mentioned, would conclude from this alone that the substance is actually present. The reaction would be considered as an *indication only, to be supplemented by other evidence*. Unless this is remembered and acted upon, the most unfortunate errors may be committed by the inexperienced. The beer is most conveniently shaken up with the solvent in the tube figured p. 223; the ether and benzine will float at the top, the chloroform will gravitate to the bottom; in either case separation is tolerably easy. It will, however, be found a good plan, in the first place, to separate the liquids rather roughly—*i.e.*, to draw off the ether, benzine, and chloroform layers, with some of the adjacent liquid, to wash this in the same tube with water, to withdraw the solvent from the water as completely as possible by the separating tube; and if this (as sometimes happens) is not very feasible, to evaporate the impure liquid to dryness in a water-bath, and exhaust the residue with the original solvent. The latter may be, in the first place, concentrated over hot water, and then portions distributed between two or three watch-glasses, and evaporated to dryness.

I. THE ACID SOLUTION.

PETROLEUM RESIDUE.

(a.) It is amorphous, colours sulphuric acid first brown, then violet, and finally red violet.
Traces of Absinthin.

(b.) It is amorphous, colourless, having a hot taste, reddening the skin, and colouring sulphuric acid brownish-red.
Traces of Capsicin.

(c.) It is amorphous, green, is coloured by sulphuric acid and sugar, and gives no precipitate with ammoniacal silver solution.
Resin of the Juniper berries.

(d.) It is crystalline, yellow, and becomes blood-red on warming with cyanide of potassium.
Picric acid.

BENZINE RESIDUE.

A. Crystalline residue.

It is not bitter ; caustic potash colours it purple-red, sulphuric acid first red, then orange.
Aloetin.

B. Amorphous residue.

a. *The residue soluble in water ; does not trouble or reduce gold chloride solution in the cold.*

(a.) Tannin does not precipitate the solution in water ; residue sharp tasting.

1. Sulphuric acid colours it red brown. *Capsicin.*

2. Sulphuric acid colours it brown. *Daphne bitter.*

(b.) Tannin precipitates the solution in water ; the residue somewhat bitter.

I. Basic lead acetate causes a weak turbidity, sulphuric acid and sugar scarcely redden.

1. Iron chloride colours the watery solution, on warming, brownish-green ; taste slightly bitter. *Gentian bitter.*

2. Iron chloride colours the watery solution brown ; taste peculiar, almost un-supportably bitter. *Quassia.*

II. Basic lead acetate strongly precipitates, sulphuric acid and sugar colour it gradually a beautiful cherry-red ; taste bitterish. *Cnicin.*

b. *The residue soluble in water ; does not trouble solution of gold chloride in the cold, but reduces it on warming.*

(a.) Tannin causes a faint turbidity in the water solution, ammoniacal silver solution is not reduced. Heated with diluted sulphuric acid, an ericinol smell is developed.

Fröhde's reagent colours it black-brown, sulphuric acid and sugar a beautiful red. *Ledum bitter.*

(b.) Tannin precipitates the watery solution ; ammoniacal solution of silver is reduced. Heated with diluted sulphuric acid, a weak smell of menyanthol is developed. *Trifolium bitter.*

c. *The residue soluble in water, precipitates in the cold chloride of gold, but does not reduce it upon warming.* Heated with diluted sulphuric acid (1 of acid to 5 of water), it gives a weak benzoic acid smell. *Centaurea bitter.*

d. *The residue soluble in water, precipitates in the cold chloride of gold, which it reduces upon warming.* Sulphuric acid dissolves it first brown, then gradually violet ; after the addition of water quickly a beautiful violet. Hydrochloric acid of 1.135 specific gravity colours it first green, then a beautiful blue. *Absinthin.*

CHLOROFORM RESIDUE.

A. Chloride of gold does not precipitate, and is not reduced.

a. Tannin gives no precipitate ; the residue has a pungent taste. Sulphuric acid colours it dark-brown red ; it reddens the skin. *Capsicin.*

b. Tannin precipitates.

(a.) Basic lead acetate gives a decided precipitate. Heated with diluted sulphuric acid it is first troubled, then it becomes brown-red, and develops a weak smell of benzoic acid. *Cnicin.*

(b.) Basic acetate of lead gives little or no precipitate.

I. *Sulphuric acid colours brown.*

1. Residue bitter.

aa. Strongly bitter.

bb. Residue somewhat bitter.

2. Residue tasting pungent.

Quassia.
Gentian bitter.
Daphne bitter.

II. *Sulphuric acid colours but slightly yellow or not at all. Colocynth bitter.*

B. Chloride of gold does not precipitate in the cold, but is reduced in the warm.

a. Tannin does not precipitate.

1. Intoxicates fish, tastes bitter.

Picrotoxin.

2. Is tasteless or slightly bitter; caustic potash colours it red-brown.

Constituent of Aloes.

b. Tannin precipitates.

(a.) Ammoniacal solution of silver is reduced. Heated with dilute sulphuric acid, as well as with Fröhde's reagent, there is a strong smell of menyanthol.

Menyanthin.

(b.) Ammoniacal solution of silver is not reduced. With concentrated sulphuric acid and sugar, after long standing, a splendid carmine red develops; heated with diluted sulphuric acid, as well as with Fröhde's reagent, an intense ericolin smell is developed.

Ericolin.

C. Chloride of gold precipitates in the cold, and is not reduced by the application of heat. Nitric acid colours violet.

Colchicine.

Heated with sulphuric acid, an odour somewhat like trifolium is developed, then the solution becomes red-brown, and the smell similar to benzoic acid.

Centaurea bitter.

D. Chloride of gold precipitates in the cold and reduces in the warm. Sulphuric acid colours brown, then the solution becomes gradually dirty violet.

Wormwood bitter.

If necessary to go further, search may be made for the alkaloids by rendering the liquid weakly alkaline by carbonate of soda or by ammonia.

II. THE ALKALINE SOLUTION.

I. RESIDUE OBTAINED BY SHAKING UP WITH BENZINE.

(1.) It dilates the pupil.

(a.) Platin. chloride does not precipitate the aqueous solution, A solution in sulphuric acid gives on warming a peculiar smell.

Atropine.

(b.) Platin chloride precipitates.

Hyoscyamin.

(2.) It does not dilate the pupil.

(a.) The sulphuric acid solution gives with oxide of cerium or bichromate of potash a blue colour.

Strychnine.

(b.) The sulphuric acid solution gives a red colour with nitric acid solution.

Brucine.

II. RESIDUE OBTAINED FROM SHAKING IT UP WITH AMYLIC ALCOHOL.—(This need only be done if salicin be suspected.) On warming with sulphuric acid and bichromate of potash, a smell of salicylic acid is developed.

Salicin.

§ 308. Dr. Adams¹ has found that the bitter of new hops is destroyed by boiling with dilute sulphuric acid. He operates as follows:—The beer is boiled, and basic lead acetate solution added until on the point of saturation. The boiling is continued for some time, after which the precipitate is filtered off and the filtrate is acidified with sulphuric acid and filtered from the lead sulphate; the clear acid filtrate is concentrated to a small bulk, chalk is added to neutralise the acid, and the liquid again filtered. The filtrate is now devoid of bitter taste, if hops alone have been used.

A second process suitable for detecting either old hops or new is on the same principles. The boiling beer is neutralised by barium hydrate, or, rather, made alkaline. To the filtrate sulphuric acid is added until made just acid. Then the lead treatment as in the first process is followed, and a filtrate ultimately obtained devoid of bitterness, if hops alone have been used. On the other hand, most bitters, such as quassia, gentian, chiretta, and many others, are but little affected by the boiling with sulphuric acid.

A very good process, only aiming at the identification of a few principles, is recommended by Enders; it is as follows:—

¹ *Analyst*, July, 1890.

The beer is evaporated to a syrup, the dextrin separated by mixing it with three or four times its volume of alcohol, the liquid filtered, and the sugar precipitated by ether. The filtered ether-alcohol solution is evaporated, the residue dissolved in alcohol, mixed with water, and precipitated by means of acetate of lead. The precipitate is filtered, the filtrate put on one side. The washed precipitate is then separated from lead by SH_2 , the lead sulphide filtered and washed with alcohol, and the filtrate (as well as the alcohol washing of lead sulphide) evaporated together. The residue is dissolved in chloroform, and the solution warmed with water until all the chloroform is driven off. The *hop bitter*, which remains insoluble, is filtered off, and the filtrate evaporated to dryness. The lupulin in it should taste bitter and have an acid reaction; it is soluble in alcohol, ether, and chloroform; is not precipitated when in solution in weak spirit by tannic acid, but is precipitated by acetate of lead. Ammoniacal solution of silver is not reduced by it. The filtrate of the first lead precipitate is freed from lead by SH_2 , the lead sulphide filtered off and washed with hot water, the excess of SH_2 driven off by warming, and then tannin added to the filtrate. If no precipitate occurs, absinthin, quassia, and menyanthin are absent. Any precipitate is filtered, dried with carbonate of lead, boiled with alcohol, evaporated, and, lastly, treated with ether. The latter agent dissolves absinthin, which is also soluble in alcohol, and in much hot water; from the latter solution it is precipitated by tannic acid, but not by lead acetate; it is soluble in sulphuric acid, and on careful addition of water to this solution a *violet-blue colour* is produced. Absinthin reduces an ammoniacal solution of silver. Ether leaves menyanthin and quassia undissolved. Both are soluble in alcohol, and the latter behaves towards tannic acid and acetate of lead like absinthin. Menyanthin reduces ammoniacal solution of silver; quassia does not.

Picrotoxin may be specially tested for by some one of the following processes:—

Herapath's Process.—Mix the beer with acetate of lead in excess; filter, and transmit sulphuretted hydrogen through the filtrate. Filter again, concentrate the filtrate, and treat it with animal charcoal, which has the property of absorbing the picrotoxin. Wash the animal charcoal, dry at 100° , and boil with alcohol; this dissolves out the picrotoxin, from which it may be obtained in tufts of crystals.

Depaire's Process.—Mix with one litre of beer finely powdered rock salt (which throws down the resinous and extractive matters), and shake the liquid with ether; an impure picrotoxin crystallises on separating the ether and evaporating it: or the beer may be simply acidulated with hydrochloric acid and agitated with ether, the ether separated and evaporated as before.

Schmidt's Process.—1. Evaporate the beer in a water-bath to a syrupy consistence, mix it with tepid water till it is perfectly liquid, so as to bring the volume to a third of the liquid used; heat and shake with animal charcoal. Let it stand several hours, filter, and heat slightly; precipitate by basic acetate of lead, and again filter. The liquid should now be of a yellow wine-colour; if not, re-filter through animal charcoal. Add from 5 to 10 cubic centimetres of amylic alcohol, and shake briskly several times at intervals; after twenty-four hours the amylic alcohol, containing the greater part of the picrotoxin, collects on the surface. The remainder is subsequently eliminated by fresh treatment with amylic alcohol. Collect the limpid layers of this alcohol, and leave the rest to

evaporate spontaneously. On the sides of the capsule a yellowish ring forms, and this contains the picrotoxin mixed with resinous substances.

2. *Isolation of the Picrotoxin.*—First, dissolve the resinous product in weak alcohol, evaporate to dryness, recover by a little boiling water containing a small quantity of H_2SO_4 , boil to expel any volatile matter, add a little animal black to eliminate all extractive and resinous matter, and, lastly, filter. Evaporate inodorous liquid, and when a fresh bitter taste is developed, shake up with ether; this redissolves the picrotoxin, and collects into a distinct layer on the surface of the liquid. Treat again with ether, and the whole of the picrotoxin is eliminated; finally, the ethereal liquids are mixed, a little alcohol is added, and the whole is evaporated. The white or yellowish ring formed consists of picrotoxin, which then has only to be dissolved in alcohol to furnish the immediate principle in the form of well-defined crystals. These crystals, however, will not be obtained unless the solution be quite free from resinous substances; if not free, and if, for instance, the ethereal solution is of a yellow colour, it must be recovered with water and treated by charcoal, as above described.

Schmidt was able to separate by this process 0.04 grain of picrotoxin in a bottle of beer which had been adulterated with eight grains of Indian berry.¹

Dragendorff has modified the method of Schmidt, by adding an excess of acetate of lead, precipitating this by SH_2 , and with the lead sulphide getting rid of some colouring matter. The sulphide of lead may be washed and dried, and then boiled with ether, to recover any picrotoxin which has gone down with it.

By the following process the animal charcoal may be dispensed with altogether:—The beer is first evaporated to a syrup, then 4 to 5 vols. of alcohol of 90 to 94 per cent. are added. After maceration for twenty-four hours at a low temperature, the alcoholic extract is evaporated, the residue acidified with diluted SO_4H_2 , and treated several times with amyl alcohol or chloroform; but previously to this the fluid should be shaken up several times with benzine, which does not remove any picrotoxin. The rest of the process is similar to that of Schmidt.

§ 309. *Special Tests for Picric Acid.*—If picric acid alone should be sought for, the quickest way is to agitate the beer in the separating tube, described p. 223, with half its volume of amyl alcohol. On separating and evaporating the alcohol, if picric acid be present a yellow residue will be left, and can be identified by the action of potassic cyanide, as described below. It will not be worth while testing specially for picric acid, if the beer, on being treated with subacetate of lead, loses nearly all its bitter taste; but if, on the contrary, it continues bitter, picric acid, or some other bitter not precipitable by acetate of lead, is present.

Another test frequently proposed for picric acid is to soak some pure wool in the beer, first gently warmed over the water-bath and acidulated with HCl, when the picric acid will stain the wool yellow. The wool thus stained may next be warmed with aqueous ammonia, the liquid filtered, concentrated to a small bulk, and a few drops of a solution of potassic cyanide added, when, if picric acid be present, a red colour (potassium isopurpurate) will be produced. Picric acid may be also recognised, if present, by spectroscopic examination: the dried extract is exhausted either by amyl alcohol or ether, and the solution examined, comparing any spectrum obtained with that given by a solution of the acid.

¹ M. Schmidt: *Chem. News*, March 12, 1864, p. 122.

The best method, according to W. Fleck, of detecting picric acid in beer, is to evaporate down, say half a litre of the beer to a syrup, mix the residue with ten times its volume of absolute alcohol, filter, wash, evaporate the alcoholic solution to dryness, treat this dry extract with water as long as the water is coloured, evaporate down the watery extract to dryness, and extract with ether. The ether will contain any picric acid in a state pure enough to weigh, if not absolutely pure. The ether may be driven off and the picric acid taken up by chloroform or benzole, and crystallised out from either of those solvents.¹

§ 310. Mr. Sorby has endeavoured to discover by spectral analysis differences between picric acid, gentian, calumba, and chiretta. The characters of the pure colouring-matters of malt and hops, when single and unfermented, have been already described. They are, however, modified somewhat by fermentation and by keeping in the cask, and the following is a description of what may be found in pure beers. The colouring-matter is first separated by evaporating down and precipitating by alcohol; the alcoholic filtrate is in turn evaporated, and the residue dissolved in water. On now adding to this aqueous solution sodic hypochlorite, it becomes gradually flesh-coloured, and if it is strong a spectrum can be obtained. This spectrum is lightly shaded to $C\frac{2}{3}D$, then dark to $D\frac{1}{2}E$, afterwards very dark; and when most advantageously seen half dark from $D\frac{2}{3}E$ to F , then easily shaded to $F\frac{1}{2}G$, farther on very dark; and there is a broad absorption-band from $E\frac{1}{2}b$. If more hypochlorite is added, it becomes deeper orange, and finally orange yellow. No difference could be discovered spectroscopically between gentian, chiretta, and hops. Calumba root it is possible to detect as follows:—

A sample of pure and one of suspected beer are evaporated down. [A beer may be suspected if the extract is bright yellow.] Two observation tubes are taken, and the pure beer diluted until it gives a spectrum easily shaded from $D\frac{1}{2}E$ to $E\frac{1}{2}b$, then moderately dark from $b\frac{2}{3}F$, afterwards dark. In the other tube is placed as much of the suspected sample as will give a colour of about equal intensity, and in both cases about one-fourth of the mixture must be alcohol, in order to avoid a precipitate. Sodic hypochlorite is now added to both slowly, and in about equal proportion; when pure, beer will become first red, and then flesh-coloured, with a spectrum moderately dark from $b\frac{2}{3}F$ to $F\frac{1}{2}g$, then half dark to $F\frac{1}{2}g$, afterwards dark; whilst beer containing calumba root is coloured orange-red, and gives a spectrum lightly shaded from D to $D\frac{1}{2}E$, afterwards half dark from $D\frac{2}{3}E$, then dark. Too large an amount of hypochlorite must not be added, or the colour is then too like that of pure beer. Picric acid, when in beer, gives a spectrum which is easily shaded from $F\frac{1}{2}g$ to $F\frac{3}{4}g$, then half dark to about g , afterwards very dark. On addition of sulphuric acid the spectrum is lightly shaded from $F\frac{1}{2}g$ to $F\frac{3}{4}g$, then half dark to beyond g ; afterwards very dark.

Salicylic Acid is used occasionally in brewing. If the beer, concentrated to a small bulk, is acidified with hydrochloric acid and shaken up with ether, the latter solvent will extract the organic acid, and it may be identified by the colour it gives with ferric chloride, and by its physical properties.

§ 311. (7.) *The Ash*.—The analysis of the ash of beer differs in no degree from an analysis of ash in general, and is carried out on the principles detailed, p. 93, *et seq.* The substances to which the analyst's

¹ H. Fleck, *Correspondenz-Bl. d. Vereins. Anal. Chem.*, iii., 77.

attention is specially directed are—the amount of salt, the presence or absence of iron (often added in the form of sulphate to porter), alum and lead.¹ Of these the only one necessary to allude to further is the salt.

The salt in beer is determined with sufficient accuracy by charring in a platinum dish the extract from 70 c.c. of beer (it is not well to burn to a complete ash, for in doing so there is always a considerable loss of chlorides). The charred mass is boiled up with successive portions of distilled water, filtered, and the filtrate made up to a known bulk, from which a convenient fractional portion is taken, and titrated with a solution of nitrate of silver (4.79 grms. to the litre), 1 c.c. = 1 mgrm. of chlorine, using as an indicator neutral chromate of potash. The chlorine found is calculated and expressed as common salt, every mgrm. of salt, when 70 c.c. are taken, being equivalent to 1 grain in the gallon. Mr. Griffin's convenient measure, the septem or one-hundredth of a gallon, may, of course, be used, and grains instead of grammes—each grain measure of nitrate of silver being made to correspond to one-tenth of a grain of common salt per gallon. Should the amount thus found indicate more than 50 or 60 grains per gallon, a second determination of chlorine should be made by the more accurate gravimetric method of weighing the chlorine as chloride of silver. It is always advisable then to make a qualitative and quantitative examination of the soluble portion of the ash; for, if called as a witness, the analyst must be prepared to state positively the amount of chloride of *sodium*; nor can he well do this simply from the soluble chlorine, for that may represent other chlorides besides chloride of sodium.

The amount of salt derived from the hops and malt can approximately be determined, as Mr. Gatehouse has shown,²—First, for the malt: by taking the original gravity, as before described (p. 419), obtaining thence the quantity of malt originally used in the beer, and reckoning the malt to contain .025 per cent. of salt (and certainly no malt will be found to exceed this). Then, for the hops: the possible maximum of salt in hops is .345 per cent. The quantity used in brewing being seldom, if ever, more than 20 lbs. per quarter of malt for bitter beer, and generally less than half this amount for strong beer, and this weight of malt giving at least 72 gallons, the salt derived from the hops cannot in bitter beer exceed 6.7 grains, and in strong beer 3.35 grains per gallon.

Thus, to take the first example (p. 424), the original gravity of which was 1040.2, corresponding by the tables to .9950 lbs. of malt extract per gallon. Since 320 of malt equal 210 of extract, and there are 7000 grains in the pound—

$$\frac{.9950 \times 320 \times .025 \times 7000}{210 \times 100}$$

= 2.65 grains of salt per gallon as the possible maximum from the malt, the beer in question not being a bitter beer. Add to this 3.35 as possible maximum amount of salt from the hops—

2.65 from malt,
3.35 from hops,

6.00 salt possible from both malt and hops.

¹ Many firms now use large copper coolers, but no injurious amount of copper appears to have been as yet detected in beers.

² "The Amount of Salt in Beer." *Analyst*, No. 20, 1877.

Now, if the composition of the water used in brewing be known, one-third more than the actual quantity of salt in the water present may be added to the number representing the salt from the hops and malt, and the data are then complete for the analyst to form his judgment. The amount of salt in the water used will, however, only occasionally be known.

The numbers used in the above calculations being constants, the process is shortened by simply deducting 1000 from the original gravity, and multiplying by '066, the result giving the salt in grains per gallon. Thus taking an example from Mr. Gatehouse's excellent paper:¹—

	Per cent.
Beer found to contain alcohol,	5·2
Malt extract	7·38
Specific gravity of alcohol equals '9911 ; the spirit indication therefore equals 8·9, giving an original gravity,	38·6
Gravity of boiled beer,	1030·6
<hr/>	
Original gravity of wort,	1069·2
Salt due to malt alone, $69·2 \times '066 =$	4·567
Salt due to hops,	3·350
Salt due to water, unknown.	
Possible total due to malt and hops,	7·917
The amount of salt actually found being 8·55.	

It has been suggested that the concrete sugar so largely and legitimately used by brewers sometimes contains a considerable amount of chlorides. If samples are found, occasionally, with more than a trace of chloride, it is because the sugar itself has been prepared for the brewers; nor can there be any difference whether the brewer adds the salt as salt, or first mixes it with sugar. However, analytical proof of sufficient chlorides *naturally* present in concrete sugar, to raise the salt in beer made from ordinary materials to over 50 grains per gallon, is wanting; and all the evidence in its favour has been derived from loose statements.

§ 312. *Adulteration of Beer with Sugar.*—In 48 and 49 Vict., c. 51, Sect. 8, it is enacted—

(1.) A brewer of beer for sale shall not adulterate beer, or add any matter or thing thereto (except finings for the purpose of clarification, or other matter or thing sanctioned by the Commissioners of Inland Revenue), before the same is delivered for consumption, and any beer found to be adulterated or mixed with any other matter or thing (except as aforesaid) in the possession of a brewer of beer, for sale, shall be forfeited, and he shall incur a fine of fifty pounds.

(2.) A dealer in, or retailer of, beer shall not adulterate or dilute beer, or add any matter or thing thereto (except finings for the purpose of clarification), and any beer found to be adulterated or diluted, or mixed with any other matter or thing (except finings), in the possession of a dealer in, or retailer of, beer shall be forfeited, and he shall incur a fine of fifty pounds.

There have been a great number of prosecutions by the Excise under this Act, either for the offence of adding sugar to or diluting the beer. It is not practicable for the public analyst, in the absence of definite standards, to certify to either sugar or water, save, of course, in some extreme cases. The Excise authorities are able to detect sugar and water, because they have power to go down in the publican's cellar and take samples of beer as it has been delivered from the wholesale house, and also of the same beer

¹ *Op. cit.*

prepared for retail. The authorities are thus able to take for their standard the undoctored beer, and to calculate the amount of water and sugar per barrel (36 galls.) which has been added to the doctored beer.

The Detection of Arsenic in Beer, Sugar, and other Articles of Food.

§ 313. So long ago as 1879, in the first edition of this work, then published under the title of "A Manual of Practical Chemistry, the Analysis of Foods, and the Detection of Poisons," attention was called to the possibility of the contamination of beer by arsenic in the following words:—

"It is possible for arsenic to be found in beers manufactured from glucose, for in certain kinds of the latter arsenic is occasionally discovered, the substance probably having been introduced by the use of an arsenical sulphuric acid in the process of manufacture. The darker the colour of the glucose, the more likely is it to be arsenical; thus Mr. Ritter found:—

	Grm.	
In white glucose,	0·0165	arsenic per kilogram.
In yellow "	0·0170	" "
In black "	0·1094	" "

"Clouet, in the examination of a very large number of arsenical glucoses, found as a minimum 0·0025 gm., as a maximum 0·0070 gm., as a mean 0·0051 gm. metallic arsenic per kilogramme (T. Clouet, "Du glucose arsenical," *Ann. d'Hygiène Publique*, xlix., Jan. 1878)."

It was not suspected that the danger was more than a possibility. In the latter part of 1900, however, a remarkable epidemic of peripheral neuritis, in many cases fatal, broke out in Lancashire and Staffordshire. The numbers affected will never be exactly known, but in Manchester and Salford alone it was estimated that 3000 persons suffered.

In November 1900, Dr. E. S. Reynolds, suspecting the disease was due to arsenic, and observing that the sufferers were all beer drinkers and most, though not all, large beer drinkers, had the beer at once submitted to careful examination, with the result that in the beer drunk by the patients arsenic was discovered in dangerous proportions.

It was next proved that the epidemic was limited practically to persons who drank beer from a particular source, and farther that this beer was brewed from brewing sugars supplied by a particular Liverpool firm.

Various samples of this glucose taken from breweries showed quantities of arsenic calculated as arsenious oxide of from ·56 to 9·17 grains per lb. Invert sugar from the same source was found to contain from 1·66 to 3·21 grains per lb.

It was found farther that these sugars had become contaminated by arsenic by the use of an arsenical acid supplied by a Leeds firm; various specimens of this acid contained from 1·4 to 2·6 per cent. of arsenious oxide.

The epidemic excited such general attention that a Royal Commission¹ was appointed in February 1901, and issued a valuable first report in the same year, in which various recommendations were made. With regard to the mode in which arsenic may gain access to beer the Commission found that this may occur in the following three ways:—

¹ "First Report of the Royal Commission appointed to enquire into Arsenical Poisoning," 1901.

1. "*By way of Brewing Sugars.*—If an arsenical mineral acid is used in the manufacture of glucose from starch or invert sugar from cane sugar, these products are liable to be contaminated with arsenic.

2. "*By way of Malt.*—Grain exposed to the products of combustion is liable to be contaminated with arsenic.

3. "*In other ways.*—Other ingredients of beer, such as chemical substances used for various purposes, are liable, from their mode of preparation, to contain arsenic, and we have found minute quantities have been in some instances introduced in this way."

§ 314. *Gutzeit's Test for Arsenic.*—Arseniuretted hydrogen produces a yellow to dark orange stain if passed over paper which has been treated with mercuric chloride; the depth of colour has a relationship to the amount of arseniuretted hydrogen.

This is the principle of Gutzeit's test.

The method of performing the test is as follows:—Squares of thin Swedish filter-paper, of about 4 cm. sq., are cut and prepared by dropping on the centre of each square a large drop of, say, a 5 per cent. solution of mercuric chloride; the squares are allowed to dry, and are then ready for use. A suitable quantity of the liquid to be tested, *e.g.*, 50 to 100 c.c., are acidified by adding from 5 to 10 c.c. of arsenic-free hydrochloric acid and half a c.c. of a 15 per cent. solution of cuprous chloride. A rod (or some pieces) of arsenic-free zinc is added, the resulting gas passed through sugar of lead solution and allowed to impinge upon the mercuric chloride paper. The test may be made to a certain extent quantitative by evolving arseniuretted hydrogen from known quantities of arsenious acid. Mr. Edward Dowzard¹ has devised a very neat series of absorption cells which are fitted into the neck of an ordinary flask; the cells are charged with lead acetate solution, and thoroughly free the gas from sulphuretted hydrogen; the last cell is capped with the prepared mercuric chloride paper. The test is not affected by antimony, selenium, or tellurium; it is capable of detecting less than one 200th of a mgrm. of As_2O_3 .

§ 315. *Reinsch's Test for Arsenic.*—The test is by far the most convenient for detecting arsenic, whether in organic or inorganic liquids. When properly performed it is capable of detecting with certainty less than one tenth of a milligram. of arsenic; hence it may be confidently recommended to the Food Analyst for the purpose of sorting out a number of samples into "practically arsenic free" and otherwise.

As a qualitative test Reinsch's method is performed as follows:—copper gauze or copper foil is oxidised in the air by heating it in a tube, open at both ends, to a gentle red heat. The oxide film is then dissolved off by a few seconds' contact with nitric acid; the copper, washed with water, is then chemically clean, and should show a bright surface. A piece of copper thus prepared, about 1 inch \times $\frac{1}{4}$ inch, is suspended by a thin platinum wire in, say, from 50 to 100 c.c. of the liquid to be examined, acidified with dilute pure hydrochloric acid, and very gently boiled for 20 minutes. The copper is from time to time examined, and should a dark brown or black stain be produced, this may be caused by sulphur, by antimony, by arsenic, by organic matter, and by other substances. The copper is washed with alcohol and with water and dried at a gentle heat. It is then dropped into a glass tube closed at one end, or, better still, placed in a subliming cell consisting of a ring of glass closed by a circle of thin microscope covering glass, supported on a porcelain crucible lid, and the

¹ *Journ. Chem. Soc. (Trans.)*, 1901.

copper heated almost to a dull red heat. If arsenic be present, the arsenic sublimes as arsenious acid crystals.

These crystals are for the most part regular octahedra, with, it may be, a mixture of rectangular prisms; irregular and confused forms also are not unfrequent, but, under the circumstances, a distinct crystalline sublimate from copper coated in the manner described can be nothing else but arsenious acid.

Should an attempt be made to make the test a quantitative one, the process suggested by Mr. John Clarke¹ is convenient. The strip of copper coated with arsenic is removed from the liquid and a second slip inserted. The coated slip is washed with alcohol and then with water. It is then placed in a mixed dilute aqueous solution of potash and peroxide of hydrogen, and allowed to remain in the cold until the film is dissolved and the copper has regained its original brightness. If the second slip of copper is stained, this is digested in the same liquid, while the first cleaned slip is gently boiled with the liquid under examination, and the process is repeated alternately with the two slips of copper until no more staining is observed. The alkaline oxidising liquid is now boiled, and the small amount of CuO that may be formed is filtered off. The filtrate is boiled to a small bulk. This concentrated liquid is washed into a small distilling flask with strong arsenic-free hydrochloric acid, some ferrous chloride added, the flask fitted with a safety tube and connected with a small worm condenser, an inverted calcium chloride tube attached, and the chloride of arsenic distilled over. It is advisable to distil down twice with strong hydrochloric acid. Then hydric sulphide gas is passed into the distillate; there will be a precipitate if more than one-tenth of a milligramme be present; a yellow colour, more or less deep, according to quantity, if less than one-tenth be present. Even so little as one-thousandth of a milligramme of arsenic sulphide gives a faint yellow colour. These small quantities may be estimated colorimetrically by having a comparison solution containing known small quantities of arsenious acid and imitating the colour in the same manner as in the Nessler process (described in the chapter on 'Water').

§ 316. *The Marsh Berzelius Process*, as recommended by the Joint-Committee of the Society of Public Analysts and the Society of Chemical Industry, is as follows:—

(1) Purification of Materials.

Hydrochloric Acid.—The original report suggested the dilution of the acid before treatment, but this dilution had the effect of partially retaining any arsenic. It is now² recommended to use the strong acid of commerce, to add about 5 c.c. of bromine per litre, then sulphur dioxide in excess either as gas or in solution. The liquid is allowed to stand twelve hours, and one-fifth is then distilled over. The remainder in the retort is arsenic-free.

Sulphuric Acid.—Half a litre of the purest sulphuric acid obtainable is diluted with four volumes of water, a few grms. of sodium chloride added and distilled; the first 50 c.c. contains any arsenic present, and is rejected.

Zinc.—Arsenic-free zinc may be obtained by melting zinc and adding sodium to the molten mass. The whole of the sodium must be got rid of by volatilisation and by skimming off the scum from time to time.

¹ *Journ. Chem. Soc. (Trans.)*, 1893.

² *Analyst*, Feb. 1902, xxvii., 48, 210.

Mr. William Thomson, among others,¹ criticises the methods of the Committee for the purification of the two acids. He prefers to dilute hydrochloric acid with one-third of its volume of water, to add one gm. of chromic acid per litre and to distil. The distillate is arsenic-free; it, however, contains chlorine. This is got rid of by passing dry filtered air through it for about half an hour.

Sulphuric Acid is similarly purified by adding either 1 gm. of chromic acid or the same quantity of potassium permanganate and distilling, the first portion being rejected on account of possible contamination of the neck of the non-tubulated retort in which the operation is performed. The distillate is arsenic-free.

Apparatus.—The apparatus recommended by the Joint-Committee presents nothing special. A 200 c.c. flask is fitted with a doubly perforated cork or indiarubber stopper or a glass stopper ground in. The one perforation carries a tapped funnel holding about 50 c.c. and an exit tube. To the exit tube is fitted a wide bit of tubing carrying in succession a roll of blotting-paper which has been soaked in acetate of lead solution and dried, a wad of cotton-wool, granulated calcium chloride, and a second wad of cotton-wool. To this tube is fitted a hard glass tube having an external diameter of .092 inch, as in the ordinary Marsh apparatus.

The operation is performed as follows:—20 grms. of arsenic-free zinc, well washed, are placed in the bottle, and a sufficient quantity of acid run in to cause a brisk evolution of hydrogen. After all air has been expelled an inch of the tube close to the constriction is brought to a red heat. To assist the distribution of the heat the tube may be conveniently supported and protected by copper gauze. The evolution of gas is kept up by adding more acid from time to time, and the gas allowed to stream through the red hot tube for half an hour. No mirror should result if the materials are arsenic-free. Standard mirrors are made by taking known quantities of arsenious oxide in solution, adding them to the flask, and evolving the gas as thus described. The quantities recommended for standards are .004 mgrm., .006 mgrm., .008 mgrm., and .01 mgrm. of arsenious acid.

These mirrors are sealed off while the hydrogen gas is flowing, and mounted on cardboard. It is pointed out by the Committee that HCl gives better results than sulphuric acid, and that if sulphuric acid is used standard mirrors must then be made by the use of H_2SO_4 . In other words, standard mirrors are not reliable, save when they are produced under the exact conditions to be employed when testing a solution of unknown strength.

The Committee state that organic materials may be tested by the hydrochloric acid method without previous destruction, though it is preferable to destroy organic matter.

Liquids to be tested without destruction of organic matter, such as beer, must be added in very small quantities at a time.

Malt is tested by placing 50 grms. in a separator funnel furnished with a stopcock, and adding 50 c.c. of HCl and 50 c.c. of water warmed to 50° C. The whole is allowed to digest for about twenty minutes, with frequent shaking, and then the acid solution separated. Every 20 c.c. contains the arsenic from 10 grms. of malt.

In a similar manner 20 grms. of hops may be tested.

In all these cases 10 c.c. of the liquid and 10 c.c. of HCl are added to

¹ *British Food Journal*, iv., 170.

the flask containing the zinc and hydrogen, and the gas is developed during twenty minutes.

If no mirror appears, another 10 c.c. are added, and so forth.

Destruction of Organic Matter.—There are two methods—(1) destruction by an acid; (2) burning up the organic solid mixed with lime or magnesia.

(1) *The Acid Method.*—10 grms. of the substance are placed in an 8 centimetre porcelain crucible, and covered with from 10 to 15 c.c. of pure redistilled nitric acid. The crucible is heated on a sand-bath until all the acid is expelled and the crucible contains a black perfectly charred and nearly dry mass; the contents of the crucible are now extracted with a few c.c. of dilute HCl, the whole being allowed to digest on the water-bath for half an hour. The contents are filtered, the charred mass washed with hot water, and the filtrate concentrated down to about 30 c.c., cooled, and tested in the Marsh apparatus.

(2) *The Basic Method.*—The materials are mixed with pure lime or magnesia, dried and incinerated. The proportions recommended for such liquids as beer are 1 gm. for every 20 c.c. The ash is dissolved in HCl, and the solution tested as before. The Committee do not recommend this method for hops.

To prove that the mirror is due to metallic arsenic, the sealed-up tube is broken. The portion of the tube containing the mirror is cut off, the hydrogen replaced by air and carefully sealed up; the sealed-up tube is drawn backwards and forwards through the Bunsen flame until the mirror disappears. On cooling, minute crystals of arsenious oxide are deposited, which are readily identified under the microscope by their crystalline form.

The delicacy of the test is, with quantities of 20 grms. or 20 c.c., one part of arsenious oxide in 7,000,000.

§ 317. Mr. William Thomson has introduced various modifications into the method of the Committee. Electrolytic zinc, usually contaminated with iron, is granulated, treated with acid, and the feathery portions (which do not become black on the surface when treated with an acid, and yield very little effervescence) are picked out. These feathery portions are, as a rule, free from iron and arsenic.

On the addition of an acid the evolution of hydrogen is slow, but to remedy this defect Mr. Thomson adds a small quantity of sulphate of copper solution, which, by electrolytic action, produces a brisk stream of gas.

He uses a smaller flask (50 c.c.) than that recommended by the Committee, and instead of a tap grinds the end of a solid rod into the thistle funnel (see fig. 66a). The gas passes over blotting-paper which has been soaked in lead acetate solution and dried in a similar way to that recommended by the Committee.

In testing beers and organic solutions generally, he destroys the organic matter as follows:—

50 c.c. of beer are evaporated to a syrup in a 200 c.c. Jena glass flask and 25 c.c. of strong pure nitric acid, and 5 c.c. of strong sulphuric acid are added. The whole is heated cautiously on a sand-bath, the fumes being sucked through a solution of soda by means of a water pump. When the liquid darkens, successive quantities of 3 c.c. of nitric acid are added until the fluid is colourless and fumes of sulphuric acid are given off.

The liquid is cooled, 10 c.c. of water are added, and the liquid is boiled

in order to break up the nitro sulphuric acid formed. The liquid is then cooled, diluted with 10 c.c. of water, and delivered into the Berzelius-Marsh apparatus.

Standard mirrors are made in the usual way, strictly following out the details laid down. A photographic representation of Mr. Thomson's mirrors are appended (see fig. 66*b*).

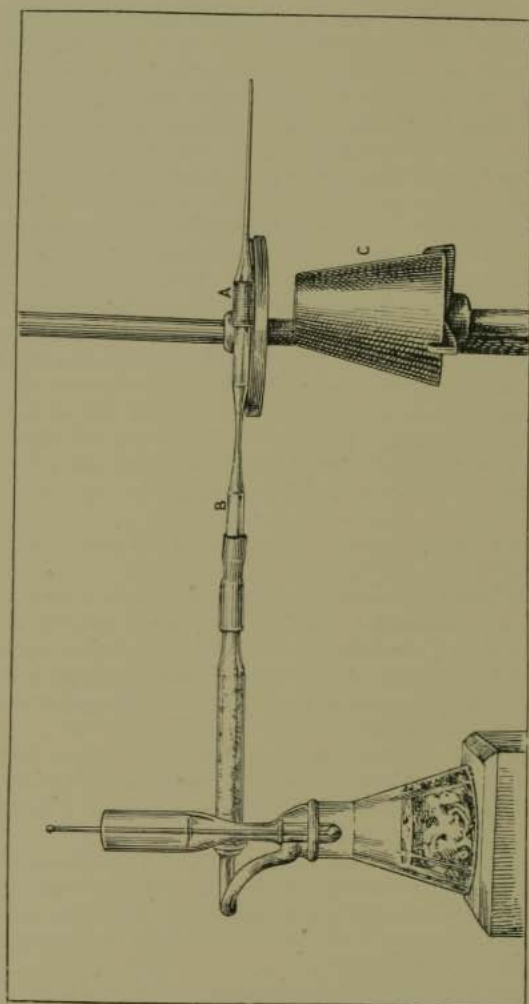


FIG. 66*a*.—Thomson's apparatus for detection of arsenic.

§ 318. *Berntrop's Method*.—J. C. Berntrop¹ has proposed the following process:—To a litre of the beer add a few drops of bromine, and allow to stand twenty-four hours. Then make the beer strongly alkaline with ammonia, add 5 c.c. of a saturated solution of sodium phosphate and 10 c.c. of the ordinary magnesia mixture, and allow to settle for twenty-four

¹ *Chem. News*, March 14, 1902. *Zeit. f. anal. Chemie*, 1902, 11.

hours. The clear liquid is poured off, the precipitate transferred to a filter as far as possible, then dissolved off the filter by a warm solution of 20 per cent. sulphuric acid. The solution is run into the flask which contained the original precipitate for the purpose of dissolving adhering crystals. The solution is now heated in a Kjeldahl flask, a little nitric acid being added from time to time. The heating is continued until sulphurous acid begins to escape and the liquid is colourless.

The solution is now placed in a Marsh's apparatus, and any arsenic present detected in the usual way.

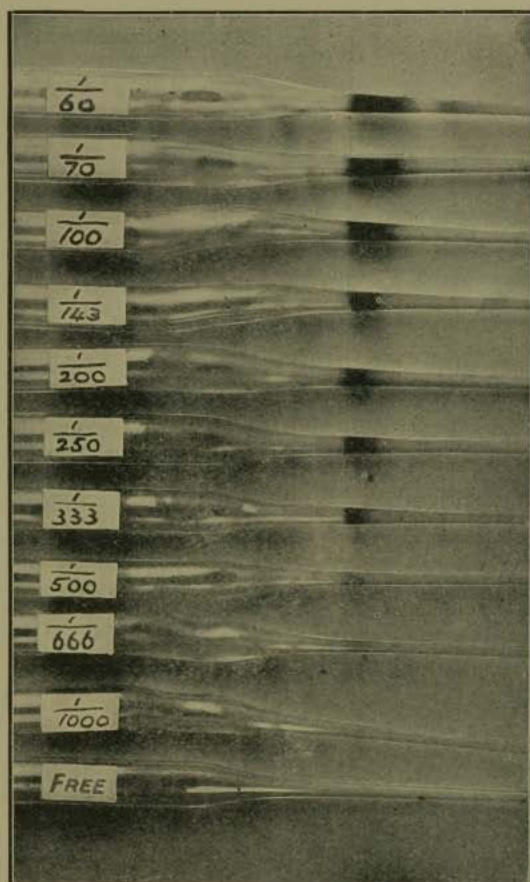


FIG. 66b.

The advantages of the method are obvious. Considerable quantities of organic liquids can be dealt with. The arsenic, if present, is concentrated as ammonio-magnesia arsenate, and falls with the ammonio-magnesian phosphate. The ultimate solution contains no organic matter. On the other hand, it is not to be relied upon as a quantitative process, the ammonio-magnesia arsenate not being absolutely insoluble in the presence of alkaline chlorides, which always occur in beer.

WINE.

§ 319. *Constituents of Wine.*—Wine is the fermented juice of the grape, with such additions only as are essential to the stability or keeping of the liquid (*Dupré*). The constituents of grape juice and wine may be arranged and compared as follows:—

MUST.	WINE.
Water.	Water.
Grape sugar.	Grape sugar (0 to several per cent.)
	Alcohols (mainly ethylic, but also small quantities of the higher alcohols, such as propylic, butylic, amyllic, and others).
	Residues of albuminoid bodies.
Albuminoid bodies.	Aldehydes (mainly ethylic).
	Isobutylglycol. ¹
	Acetal.
	Furfurol.
	Acetic acid.
	Succinic acid.
Hydro-potassic tartrate.	
Tartrate of lime.	{ Tartrate of lime, } In smaller propor-
	{ Tartaric acid, } tions than in 'Must.'
Vegetable mucus.	
Gum.	Gum.
Malic acid (in bad seasons).	Malic acid (in bad seasons).
Salts of ammonia and of similar bases.	
Small mixtures of colouring-matters.	Colouring-matters.
	Glycerin.
Organic acids in combination and certain extractive matters.	Organic matters in combination and certain extractive matters.
	Esters—Acetic, caproic, caprylic, butyric, and tartaric esters have been identified. Recently small quantities of salicylic acid (0·8 mgrm. per litre) have been detected in Portuguese absolutely genuine wines. ²
	Tannin.
Mineral matters. ³	Mineral matters.
	A few ferment cells and similar forms.

¹ A. Henninger, operating on 50 litres of Bordeaux, succeeded in isolating, by fractional distillation, 6 grms. of isobutylglycol, boiling point 178·5 C. Making a correction for the glycol carried away by aqueous vapour, he considers the amount in wine to be equal to 0·05 per cent., or about one-fifteenth of the glycerin. (*Comptes Rendus*, xcv., 94-96.)

² *Zeit. f. analytische Chemie*, 1902, 64.

³ Among the mineral matters of the grape, and, therefore, generally also in wines, is a small quantity of boric acid. The mineral matters of the ash of eighteen samples of grape juice have been quantitatively determined by Mr. Carter Bell, *Analyst* (November, 1881); the chief results are as follows:—

	Potash.	Soda.	Chlorine.	Sulphuric Acid.	Phosphoric Acid.	Lime.	Magnesia.	Iron Phosphate.	Alumina Phosphate.	Lime Phosphate.	Silica.
Maximum,	54·24	10·54	3·39	13·68	7·28	13·99	12·57	1·68	3·85	23·73	·98
Minimum,	31·23	·29	·29	3·14	·22	·65	·96	·05	·05	3·13	·08
Mean,	42·14	3·37	1·09	9·14	3·00	4·55	9·67	·63	·87	12·78	·29

This may be compared with the mineral constituents of wine given at p. 468.

§ 320. *Changes taking place in Wine through Age.*—Berthelot¹ has made several analyses of wines 100 and 45 years old respectively, which are interesting as contributing to more accurate knowledge regarding the effect of age upon wine. The wines were both samples of Port. The one 100 years old had a large deposit of colouring-matter, and was yellow; the colour of the second sample was dark, but yet lighter than that of new wine. The results of the analyses are as follows:—

	Port wine, 100 years old.	Port wine, 45 years old.
Specific gravity (at 10° C.),	·988	·991
Total residue at 100° C.,	3·36	5·30
Sugars, reducing,	1·25	3·15
Sugars, after the action of dilute acid,	1·29	3·68
Acid, calculated as tartaric acid, grms. per litre,	5·17	5·46
Tartaric ether,	1·11	1·17
Cream of tartar,	·27	·42
Alcohol, per cent.,	15·9	16·1

The analyses of the deposits gave the following results:—

	Wine, 100 years old.	Wine, 45 years old.
Sugar, reducing,	1·25	3·15
Cane-sugar,	·04	·53
Pure acids,	·51	·52
Acids as ethers,	·27	·28
Cream of tartar,	·03	·04
	2·10	4·52
Glycerin and other matters,	1·16	·98

This research of Berthelot's, as well as the more recent investigations of Schmidt,² show that there is a gradual deposit of the colouring-matter, and that some of the sugar has disappeared from the old wine, which gives a smaller residue. Cane-sugar is practically absent in the sample 100 years old, a fact which Berthelot interprets as confirmatory of his observation of the slow invertive action of inorganic acids on cane-sugar. The alcohol is lower in old wines than new, and the acidity tends to diminish, the acids combining with alcohols to produce esters. The experiments of Macagno may also be here cited, from which it appears that in wines of the same class the tannin decreases through age, while the glycerin increases.

§ 321. *Adulterations of Wine.*—The adulterations of wine are as follows:—Watering, fortifying with spirit, fortifying and watering, the addition of various fermented liquids, such as wines of low value to those of high value—that is to say, alcoholic liquids made from the fermentation of glucoses or various sugars, or wines made from raisins or figs to wines made from the grape—the mixing or blending of wines (this may be a necessary operation in some cases, in others it takes the form of sophistication, when wines of higher quality are mixed with wines of low quality and sold as wines of the higher quality), plastering, the addition of bitartrate of potash and ethers (such as *œnanthic ether*) to give a fictitious appearance of age, the addition of alum to brighten the colour, the artificial colouring of wines, the addition of antiseptics (such as salicylic acid), and the addition of fluoborates or fluosilicates.

§ 322. *The Analysis of Wine.*—The analysis may be divided conveniently into:—I., Physical characters; II., the estimation and qualitative detection of constituents volatile at or below 100° C.; III., estimation and

¹ *Comptes Rendus*, lxxxviii., 1879, 626.

² *Die Weine*. Berlin, 1893.

identification of matters not volatile at 100° C.; IV., the estimation of the total constituents of the mineral matters of the ash, and, similarly, the identification and estimation of the separate constituents of the ash.

I. *Physical Characters*.—This embraces the specific gravity and the action on polarised light.

II. *Constituents Volatile at or below 100° C.*—This embraces the estimation of alcohol, volatile esters, sulphurous acid, aldehyde-sulphurous acid, volatile acid, and, as far as practicable, the identification of the constituents comprised under those names.

III. *Constituents not Volatile at or below 100° C.*—This embraces total extract,¹ total fixed acid, free tartaric acid, sugars, potassic tartrate, fixed esters, glycerin, potassic sulphate, tannin, and colouring matters.

IV. *The Ash*.—Special processes may be required for the detection and estimation of some of the adulterants mentioned.

The Table LI. gives, according to this plan, the results of the analysis of a number of wines of considerable age and high price analysed by Dr. Conrad Schmidt.² The Table LII. gives a number of analyses by Dr. Dupré.

I. PHYSICAL CHARACTERS.

The specific gravity and polarimetric estimations are made on similar lines to those already detailed under 'alcohol' or 'sugar.'

II. CONSTITUENTS VOLATILE AT OR BELOW 100° C.³

Alcohol.—Wines, in regard to their alcoholic content, may be divided into two classes—viz., *natural wines*, the strength of which has not been increased by the addition of spirit; and *fortified wines*, such as those of

¹ E. Riegler (*Zeit. f. anal. Chemie*, 1896, 27) has published an easy and rapid method of determining the amount of alcohol and extract in wine. The refraction is taken of the wine by means of an Abbe's or a Pulfrich's refractometer; the alcohol is next got rid of by boiling a measured quantity; after cooling and making up with water to the original bulk the refraction is again taken. He finds that a gramme of extract in 100 c.c. of wine raises the refraction beyond that of water 0.00145, and 1 gm. of alcohol in 100 c.c. of wine raises the refraction 0.00068. If N = the refraction of the undistilled wine; a , the refraction of the distilled pure water; $a + b$, the refraction of the wine freed from alcohol and made up to the original volume, then $\frac{N - (a + b)}{0.00068}$ = alcohol in

grms. per 100 c.c. of wine and $\frac{b}{0.00145}$ extract in 100 c.c. wine.

² "Die Weine des herzoglich nassauischen Cabinetskeller," by Hofrath Dr. Conrad Schmidt. Berlin, 1893.

³ THE GENERAL PROCESS OF J. NESSLER AND M. BARTH (*Zeit. anal. Chem.*, 1882, 43).

Solid Residue.—The authors evaporate two separate quantities of wine, the one with the addition of a measured quantity of titrated baryta-water, the other without any addition; the baryta-water mixture is dried at 110°–115° C. for eight hours—the addition fixes volatile acid and glycerol. The second portion is simply dried at 100° C. in a current of dry air; the difference between the two determinations, correction being made for the baryta, represents glycerol and volatile acid.

Effects on Polarised Light.—Neubauer has shown that perfectly fermented wine scarcely polarises light, but if the wine has been imperfectly fermented the plane of polarisation is turned to the left. On the other hand, wines sweetened with potato-sugar contain a considerable proportion of dextro-rotatory non-fermentable substances. The dextro-rotatory substances natural to wine are insoluble in strong alcohol, whilst the

TABLE LI.—GIVING THE CHIEF RESULTS OF SCHMIDT'S ANALYSES (1888-1892) OF CERTAIN FINE AND EXCEPTIONALLY VALUABLE OLD WINES.

Parts by weight per litre.

	Hoch-eimer. 9 samples. (1706-1868).	Stein-berger. 19 samples. (1811-1873).	Marko-brunner. 7 samples. (1823-1868).	Rudes-heimer. 11 samples. (1831-1880).
Physical Characters.				
Specific gravity.				
Min.,	0·9981	0·9982	0·9983	0·9963
Max.,	1·0044	1·0025	1·0018	1·0026
Mean,	1·0105	0·9990	0·9997	0·9995
Polarisation—200 mm. tube. (Expressed in Wild's degrees.)				
Min.,	+0·16°	+0·05°	+0·30°	+0·05°
Max.,	+0·49°	+1·50°	+1·32°	+1·55°
Mean,	+0·33°	+0·39°	+0·59°	+0·45°
Constituents volatile below 100° C.				
Alcohol.				
Min.,	37·6 (1706)	43·7	49·4	46·9
Max.,	76·7	92·9	90·0	93·6
Mean,	56·3	67·7	74·4	71·7
Volatile Esters. (Expressed in c.c. of d.n. KHO.)				
Min.,	27·6	17·2	15·6	27·2
Max.,	50·4	48·0	49·2	50·4
Mean,	32·6	32·7	34·4	36·8
Aldehyde-sulphurous acid. (In grms. of SO ₂ .)				
Min.,	0·059	0·069	0·059	0·077
Max.,	0·223	0·245	0·214	0·260
Mean,	0·125	0·170	0·172	0·160
Volatile acid. (Expressed as acetic acid.)				
Min.,	1·01	1·10	1·20	1·04
Max.,	2·32	2·42	2·15	2·29
Mean,	1·44	1·52	1·67	1·47
Constituents not volatile at 100° C.				
Extract.				
Min.,	25·43	20·98	23·48	21·80
Max.,	27·79	42·31	44·06	47·37
Mean,	27·50	29·5	31·78	30·54
Total Tartaric acid.				
Min.,	2·31	1·74	2·03	1·80
Max.,	3·96	3·32	3·09	3·15
Mean,	2·72	2·51	2·26	2·46
Free Tartaric acid.				
Min.,	0·63	0·51	0·39	0·39
Max.,	1·35	1·02	0·72	1·41
Mean,	0·84	0·71	0·67	0·71

TABLE II.—*continued.*

	Hoch- eimer. 9 samples. (1706-1868).	Stein- berger. 19 samples. (1811-1873).	Marko- brunner. 7 samples. (1822-1868).	Rudeg- heimer. 11 samples. (1831-1880).
Constituents not volatile at 100° C.—<i>continued.</i>				
Total fixed acid reckoned as tartaric acid.				
Min.,	2.97	4.10	4.13	3.80
Max.,	5.94	6.44	5.16	5.40
Mean,	4.62	4.62	4.78	4.67
Tannin and colouring matters.				
Min.,	0.19	0.22	0.23	0.16
Max.,	0.71	0.45	0.68	0.66
Mean,	0.39	0.36	0.42	0.28
Fixed ester. (Expressed in c.c. of d. n. KHO.)				
Min.,	134.1	144.4	128.4	118.8
Max.,	257.6	488.8	446.4	496.0
Mean,	188.7	233.9	268.5	231.8
Glycerin.				
Min.,	13.06	10.51	11.70	9.79
Max.,	15.81	18.76	22.55	24.46
Mean,	12.62	14.64	16.29	14.63
Potassic sulphate.				
Min.,	0.76	0.59	0.44	0.47
Max.,	1.63	1.19	1.52	1.20
Mean,	1.15	.86	0.78	0.73
Ash.				
Min.,	2.19	1.78	2.13	1.87
Max.,	2.67	2.59	2.45	2.60
Mean,	2.55	2.24	2.26	2.23
NUMERICAL RELATIONS OF CERTAIN CONSTITUENTS.				
Glycerin : Alcohol. (Alcohol = 100.)				
Min.,	15.9	16.8	18.3	14.1
Max.,	30.7	32.3	26.1	29.5
Mean,	23.5	21.5	21.5	20.5
Volatile acid : Total acid. (Total acid = 100.)				
Min.,	15.6	12.6	18.2	16.7
Max.,	34.8	38.4	31.6	31.3
Mean,	21.7	23.4	25.8	22.2
Volatile ester to alcohol. (Alcohol = 100.)				
Min.,	52.9	33.5	17.4	41.6
Max.,	64.7	61.7	69.6	64.0
Mean,	56.3	48.7	49.6	52.3
Volatile ester to volatile acid. (Volatile acid = 1.)				
Min.,	18.2	15.0	9.5	21.9
Max.,	26.3	30.0	29.5	34.0
Mean,	22.8	22.2	20.9	25.7
Total ester to total acid. (Total acid = 1.)				
Min.,	21.5	29.9	24.6	25.5
Max.,	46.2	73.3	63.3	69.2
Mean,	33.3	41.4	41.7	40.6

TABLE LIII.—WEIGHT IN GRAMMES OF SOME OF THE CHIEF CONSTITUENTS OF 1 LITRE (1000 C.C. OR 1000 VOLUMES) OF THE UNDERMENTIONED WINES (DUPRÉ).

Particulars of Wines Analysed.	Doz.	Vintage.	Absolute Alcohol.	Free Fixed Acid as Tartaric Acid.	Free Volatile Acid as Acetic Acid.	Total Free Acid as Tartaric Acid.	Real Tartaric Acid.	Total Dry Residue.	Grape and Fruit Sugar.	Total Amount of Ash.	Carbonate of Potassium.	Sulphates and Chlorides.	Phosphate and Carbonate of Calcium.	Total Amount of Phosphoric Acid.	Alcohol in Fixed Ethers.	Alcohol in Volatile Ethers.	Total Alcohol in Ethers found.	Total Alcohol in Ethers calculated.	Proportion per cent. of Alcohol found to Alcohol calculated.
Hook, white,	30s.	1862	95.6	3.48	0.57	4.20	...	18.63	...	1.95	0.58	0.76	0.60	0.92	0.132	0.230	0.362	0.360	100.5
"	40s.	1859	92.0	4.20	1.14	5.62	2.550	18.55	0.12	1.70	0.07	0.78	0.85	0.30	0.199	0.239	0.438	0.458	95.7
"	120s.	1857	104.4	4.31	0.93	5.37	0.875	20.60	1.12	1.45	0.14	0.46	0.85	0.35	0.225	0.289	0.464	0.493	94.3
Claret,	15s.	1865	85.3	4.24	1.47	6.08	0.875	21.40	4.31	2.08	0.66	0.95	0.48	0.33	0.155	0.197	0.352	0.476	74.0
"	48s.	1865	120.0	4.24	1.74	6.41	1.875	24.33	2.04	2.25	0.66	1.05	0.55	0.30	0.186	0.248	0.430	0.581	74.6
"	66s.	1861	85.3	3.23	1.80	5.48	1.838	18.00	0.95	2.00	0.38	0.99	0.63	0.30	0.166	0.216	0.382	0.429	88.8
Hungarian, red,	21s.	...	113.6	3.56	2.49	6.68	0.600	20.85	1.47	1.85	0.41	0.91	0.53	0.35	0.151	0.358	0.509	0.656	77.6
"	34s.	...	95.4	5.83	1.47	7.16	0.875	18.20	0.61	1.75	0.14	0.81	0.80	0.25	0.186	0.271	0.457	0.613	74.5
"	white,	...	94.9	4.74	1.80	6.99	0.375	18.13	0.24	1.88	0.12	0.90	0.85	0.25	0.162	0.273	0.435	0.596	73.0
Greek wine,	20s.	...	107.2	3.41	3.00	7.16	0.675	25.30	2.00	2.25	0.07	1.18	1.00	0.25	0.224	0.214	0.438	0.690	63.6
"	28s.	...	124.5	4.54	1.68	6.64	...	24.42	1.12	3.05	0.41	2.01	0.62	0.25	0.384	0.179	0.563	0.707	79.6
"	30s.	...	138.9	2.33	1.77	4.54	0.300	25.50	3.64	3.75	0.21	2.49	1.05	0.45	0.245	1.207	0.453	0.530	85.1
Sherry,	high price,	1865	994.09	172.0	2.70	1.53	4.61	48.00	25.65	4.50	0.07	3.63	0.80	0.18	0.206	0.216	0.422	0.639	68.1
"	"	1860	997.93	178.1	3.08	1.68	5.18	262.53	29.70	5.50	0.18	4.41	0.95	0.25	0.260	0.391	0.681	0.749	90.8
"	"	1857	998.30	184.0	2.81	1.62	4.84	150.56	35.10	5.13	0.07	4.18	0.88	0.13	0.262	0.439	0.731	0.772	101.2
Madeira, E. I.,	60s.	1812	177.5	3.26	1.68	5.36	0.300	43.47	20.80	3.90	0.27	2.52	1.10	0.42	0.305	0.382	0.687	0.774	88.7
"	high price,	1812	180.0	4.20	3.27	8.25	...	45.41	16.29	3.59	0.17	1.93	1.49	0.50	0.460	0.773	1.233	1.207	102.1
"	32s.	1854	185.6	3.08	0.84	4.13	0.225	75.57	43.31	2.48	0.68	1.34	0.65	0.35	0.302	0.228	0.430	0.620	69.4
Port,	high price,	1854	175.3	3.54	1.07	4.88	0.225	53.90	22.84	2.58	0.66	1.37	0.55	0.33	0.361	0.220	0.571	0.697	84.9
"	"	1842	182.6	2.66	1.08	4.01	0.150	31.01	10.10	0.69	0.89	0.86	0.45	0.18	0.283	0.331	0.614	0.335	103.2
"	"	old	187.1	1.88	1.21	3.26	...	49.83	32.40	2.25	0.21	1.54	0.50	0.33	0.256	0.189	0.445	0.447	99.3
Marsala,	20s.	very old	108.9	2.25	1.38	3.98	0.150	57.48	37.60	3.13	0.55	1.92	0.65	0.23	0.333	0.216	0.549	0.550	99.8

Spain and Portugal, which absolutely *require* the addition of a certain amount of spirit to preserve them. Natural wines contain as a minimum 6 per cent., and as a maximum a little over 12 per cent., of absolute alcohol by weight. Even under favourable conditions scarcely more than 15 per cent. of alcohol can be obtained by fermentation. The percentage of alcohol in fortified wines depends, of course, entirely on the operator; it appears to range usually from 12 to 22 per cent. by weight. The alcohol is returned as ethylic, but there are always traces of the higher homologous alcohols—*e.g.*, propylic, butylic, and amylic. It would be highly desirable to have a number of determinations of the higher alcohols in genuine, and sophisticated wines; this has never yet been done on any scale. The methods to be followed differ in no essential way from those already detailed at pp. 393–397.

§ 323. *Volatile Acids.*¹—All wines possess an acid reaction, due to

impurities in potato-sugar are mostly soluble in the same. Nessler and Barth have modified Neubauer's process as follows:—The wine is evaporated to one-fifth of its bulk; the tartaric acid is separated by precipitation with potassium acetate; 90 per cent. alcohol is next added to complete precipitation; the whole is filtered, and the filtrate mixed with ether; all optically active substances will now be found in the lower stratum.

Chlorine.—The proportion of chlorine in genuine wines lies between '002 and '0028 per cent., and never exceeds '006 per cent.; it should not be estimated in the ash, but directly in the wine.

50 c.c. of the wine are acidulated with nitric acid, an excess of standard silver solution added, and then standard thiocyanate solution is run in until a drop of the liquid, when mixed in a plate with ferric sulphate, first shows a pink coloration.

Estimation of Free Tartaric Acid.—100 c.c. of the wine are evaporated to a thin syrup, and mixed with alcohol so long as a precipitate appears. The cream of tartar, after a few hours' standing, is separated. To the filtrate, from 1½ to 2 c.c. of calcium acetate are added. Wines destitute of free tartaric acid show no turbidity. Any weighable quantity of acid-tartrate is, after standing, filtered off.

Citric Acid.—In falsified wines citric acid is sometimes met with.

100 c.c. of wine are evaporated to 7 c.c., and precipitated with 80 per cent. alcohol, and filtered. The filtrate is partly neutralised by milk of lime, and the filtrate from this is diluted to the original bulk taken, *viz.*, 100 c.c. About 1 c.c. of a cold neutral saturated solution of lead acetate is added, and the precipitate (containing phosphoric, sulphuric, tartaric, and part of the malic acid) is collected, decomposed with hydrogen sulphide, and the solution of the free acid rendered alkaline with lime. The calcium phosphate is separated by filtration, the filtrate is slightly acidified by acetic acid, tartrate of lime separating. From the filtrate, calcium citrate separates on prolonged boiling. On drying at 100° C., it is weighed— $(C_6H_8O_7)_2 Ca_3 + 4H_2O$.

¹ The acids in wine are estimated by C. Schmidt and C. Hiepe (*Zeits. für anal. Chemie*, xxi., 534–541) as follows:—200 c.c. of wine, concentrated by evaporation to half, are precipitated by basic lead acetate. The precipitate is filtered off, suspended in water after being well washed with cold water, and then decomposed by SiH_4 . The solution of the acids thus obtained is filtered from the lead sulphide, concentrated to 50 c.c., neutralised with KHO, and still further concentrated. An excess of a saturated solution of calcium acetate is then added, and the whole allowed to stand for several hours; the precipitate of calcium tartrate is then filtered, washed, ignited, and the alkalinity titrated with standard hydrochloric acid. The result is calculated into tartaric acid, a correction of '0286 gm. being added for the solubility of calcium tartrate. The filtrate from the calcium precipitate is again concentrated to 20–30 c.c., and 60–90 c.c. of 96 per cent. alcohol added. The precipitate consists of calcium malate, succinate, sulphate, and a small quantity of calcium tartrate. It is filtered off, dried at 100° C., and weighed. It is then dissolved in the minimum quantity of hot dilute hydrochloric acid, slightly alkalisied by potassium carbonate, and the precipitated calcium carbonate removed by filtration. After neutralisation by acetic acid, the filtrate is concentrated to a very small bulk, and precipitated hot by barium chloride, which throws down barium succinate and sulphate. The precipitate is treated with hot dilute hydrochloric acid, which leaves the barium sulphate undissolved, and which therefore can be filtered off, ignited, and weighed. To the hydrochloric solution of barium succinate, a sufficient

acids, which are conveniently divided into *volatile* and *fixed*. Dr. Dupré puts the amount of volatile acid, expressed in terms of acetic acid, as 0.3 to 0.6 per cent. by weight in volume. About one-fourth of the total acidity in white natural wines should be due to volatile acids, and in red and fortified wines the volatile should not amount to more than about one-third of the total acidity. The non-volatile acids appear to be chiefly malic and tartaric (sometimes part of the tartaric being replaced by succinic); the former, according to Dupré, predominating in pure natural wines, and largely so in fortified liquors; whilst in plastered wines it is often present to the total exclusion of tartaric acid.

In artificial wines, it is common enough to find a considerable amount of free tartaric acid; but the mere detection of free tartaric acid is not enough to prove adulteration, since this is found in small quantity in many natural wines. If, however, with a small amount of free acid there is a preponderance of tartaric acid, then sophistication may be suspected. It has been suggested that such free acid may be recovered from the wine by agitation with ether, but J. Nessler, in a direct experiment, could only recover 3.93 per cent. of the free tartaric acid present when the wine was directly treated with ether; 25 per cent. when the wine was evaporated to a syrup. He recommends the following processes:—

The wine is agitated with tartar and divided into two parts, to one of which a few drops of concentrated acetate of potassium solution is added, and the mixture carefully observed, noticing whether any tartar crystals form. Errors are avoided by comparing the one portion to which a few drops of the potassium acetate has been added, with the other portion to which no acetate has been added, the separation of the tartar crystals being a proof of free tartaric acid in the wine. Free sulphuric acid may be detected by means of the methods described in the article on Vinegar.

The chief volatile acid is acetic; white German and French wines seldom contain more than .8 grm. per litre of volatile acid reckoned as acetic, and red more than 1.2 per litre. Red wine containing as much as 1.6 grm. per litre is considered sour and unfit; it is, therefore, curious to find that the very high-class wines in Table LI. may yield as much as 2.15 to 2.42 per litre.

The general method of estimation is to take from 10 to 20 c.c. of the wine suitably diluted, and titrate with d. n. soda, using tincture of logwood as an indicator, the result being the *total acidity*. On now evaporating the wine on the water-bath to a syrupy extract, diluting and again titrating, the loss of acidity corresponds to the *volatile acid*, the latter being expressed in terms of acetic acid, the non-volatile as tartaric acid. L. Weigert¹ has shown that by distilling in a vacuum, the whole of the acetic acid can be obtained; 40 or 50 c.c. of the wine are in this way boiled to dryness, water added to the dry residue, and the process thrice repeated.

A method of diagnosing and estimating volatile acids has been proposed by E. Duclaux, based upon the more or less regular way in which volatile acids distil each after its own manner, and the estimation of acidity in

quantity of sulphuric acid is added to precipitate the barium as sulphate, and from the weight of the latter the amount of succinic acid is calculated, 233 of $\text{BaSO}_4 = 118$ of $\text{C}_4\text{H}_6\text{O}_4$. The weights of the sulphuric, succinic, and tartaric acids are calculated as calcium salts, and subtracted from the weight of the lime precipitate, the difference being reckoned as malate, 172 parts of calcium malate = 134 malic acid.

¹ *Zeitsch. für analyt. Chemie*, 1879, 207.

successive fractions of the distillate. This was described in detail in a former edition, but the method has not found favour among practical chemists and the accuracy of the calculations has been questioned.¹

Detection and Estimation of Free Sulphurous Acid and of Aldehyde-sulphurous Acid.—Dr. Conrad Schmidt has shown that when a wine is sulphured the sulphurous acid gradually disappears, a small part being oxidised to sulphuric acid, the larger part uniting with aldehyde. According to Schmidt free sulphurous acid is injurious to health, whereas a fair proportion of aldehyde-sulphurous acid can be taken without any injury whatever. The limit for free sulphurous acid on the Continent is 20 mgrms. per litre. Schmidt has devised a process of estimating free sulphurous and aldehyde-sulphurous acid.

A stream of carbon dioxide is led into a 100 c.c. flask, and when the air has been displaced 50 c.c. of the wine are added, 5 c.c. of sulphuric acid (1 : 3) and a little starch solution. A decinormal solution of iodine is dropped in until the blue is permanent, and the number of c.c. of iodine used is translated into sulphur dioxide, each c.c. of decinormal iodine solution being equal to 2.3 mgrms. of SO₂.

This determination gives the sulphurous acid existing in a free state. In a 200 c.c. flask, 25 c.c. of KHO solution and 50 c.c. of wine are mixed, and after standing fifteen minutes 10 c.c. of sulphuric acid (1 : 3) and a little starch are added, and the liquid titrated as before. The result is total sulphurous acid, and by subtracting the amount of free sulphurous acid from the total, that which has been in combination with aldehyde is known.

It is, therefore, only new wine which is likely to have any free sulphurous acid; in old wine the sulphurous acid will be in combination.

§ 324. *Estimation of Esters in Wine.*—The compound esters in wine may be divided into volatile and non-volatile. The volatile esters give the bouquet or odour, the fixed esters the taste to wine. The proportion of volatile to fixed esters is very small in unfortified wines, but the reverse is the case with fortified wines. The total amount of esters is extremely small; Dr. Dupré gives about one part of compound ester in 300 parts of wine as the highest proportion he has yet met with. The esters themselves are, of course, derived from conversion of the alcohols, the ultimate amount depending entirely on the relative proportion of alcohols, acid, and water present, and not being dependent on the nature of the alcohols or acids. If, as sometimes happens, an excess of compound ester is added to a wine, decomposition will at once begin, until ultimately the wine will contain no more than it would otherwise have reached in the natural order of things. An estimation of the esters is, therefore, of the greatest possible importance, as it enables the analyst to judge of the age, character, etc., of the wine.

Berthelot has given the following formula for the calculation of the amount of alcohol present in the compound esters of wine :—

$$y = 1.17 A + 2.28$$

$$x = \frac{y \times a}{100}$$

A is the percentage of alcohol by weight in the wine; a the amount of alcohol equivalent to the total free acid (reckoned as acetic) contained in 1

¹ See a paper by Mr. Droop Richmond, *Analyst*, September and October, 1895.

litre. y = the proportion per cent. of a present as compound ether in 1 litre when the alcoholic strength is A . x = alcohol present in compound ethers of wine. It hence follows that if the amount of alcohol present as ester, found by experiment, fairly agrees with the calculated amount, etherification is complete, and the wine must be of a certain age; if the compound esters exceed the proper amount, the probability is that it is an artificial wine; and, lastly, if the amount of esters is below the theoretical standard, either etherification is not complete, on account of its youth, or alcohol has been recently added.

Since in small quantities of wine the esters cannot be satisfactorily identified, Dr. C. Schmidt's method of expressing the esters in terms of decinormal alkali is to be recommended. The wine is first carefully neutralised and 100 c.c. are distilled until the distillate measures 90 c.c.; the 90 c.c. are made up to 100 c.c. by the addition of distilled water; 25 c.c. of the 100 are made up to 50 c.c. by the addition of neutral absolute alcohol, 25 c.c. of d. n. soda added, and the whole allowed to stand for an hour; the alkaline liquid is then titrated, using phenol-phthalein as an indicator, and a certain loss of alkalinity owing to the saponification of the volatile esters will be found; this difference is returned as so much volatile ester in terms of decinormal alkali; by treating quite similarly the residue in the retort the fixed esters are estimated.

The determination of the volatile esters is more important than that of the fixed esters, because the bouquet and properties of a wine are dependent to a considerable extent upon the nature and amount of these esters. Dr. C. Schmidt claims to have synthetised an ester which, added to young wine, gives it the same flavour as an old wine, but no details of his experiments have been published. Without a doubt, certain essences used by the wine producers are rich in volatile esters.

It is, however, only by an intimate knowledge of the amount of volatile esters which a natural wine may possess at different periods of time that it is possible to be certain of the fraudulent addition of esters.

III. ESTIMATION AND IDENTIFICATION OF MATTERS NOT VOLATILE AT 100° C.

§ 325. EXTRACT OR SOLID RESIDUE.—The dry extract in pure natural wines is usually given as from 1.5 to 3 per cent., some of the Rhine wines of undoubted purity give, however, an extract of nearly 5 per cent.; the presence of sugar in fortified wines may raise the extract to 6.0 or 10 per cent. The solid residue may be taken by simply evaporating 10 c.c. to dryness, which can be done rapidly without any decomposition of the solids, by using a large flat platinum dish, and thus spreading the 10 c.c. out in a thin layer. This method is, however, somewhat inconvenient, and causes a loss of glycerin; therefore the indirect process for beer, given at p. 419, may be employed instead, wine extract being considered equal in density to malt extract.¹ But in wines containing much ash (since the mineral constituents of the latter seriously affect specific gravity, containing in a

¹ See also Riegler's method (p. 444). A. Gautier (*Annales d'Hygiène Publique*, xlvii., 118, 1877) has recommended in all cases the evaporation of 5 c.c. of wine on a watch-glass, in a vacuum, without the application of artificial heat. This method takes from two to six days, according to the temperature, for completion, so that it is scarcely applicable for technical purposes; but it is evident that a heat of 30° C., whilst greatly expediting the process, would in no way impair its accuracy.

given specific gravity about twice as much substance in solution as a sugar solution of the same gravity), it is necessary to subtract from the percentage of extract thus estimated, the percentage of ash found in the same wine; or if the amount of extract without the ash is required, twice the percentage of ash has to be subtracted from the percentage found. Dupré and Thudichum give the following examples :

ROSENTHALER, 1859 (£15 Ohm).		Per cent.
Specific gravity of de-alcoholised wine,		1016·48
Percentage of extract (see table, p. 420),		4·122
Percentage of ash found,		0·170
Total solid constituents,		3·952
To find total solids minus ash, subtract again		0·170
Total solid constituents,		3·782
SHERRY, 1865.		
Specific gravity of de-alcoholised wine,		1017·87
Percentage of extract from specific gravity (see table, p. 420),		4·467
Percentage of ash found,		0·515
Total solid constituents,		3·952
Subtract ash,		0·515
Total solid constituents,		3·437

H. Hager,¹ after evaporating off the alcohol, and making up the wine to its original volume by means of water, determines the amount of extract from the following table, which is based on his own experiments, and differs a little from the malt extract table, p. 420.

TABLE LIII.

Per cent. of Extract.	Specific gravity, 15°·0 C. Water = 1·000.	Per cent. of Extract.	Specific gravity, 15°·0 C. Water = 1·000.	Per cent. of Extract.	Specific gravity, 15°·0 C. Water = 1·000.
0·50	1·0022	5·25	1·0239	10·00	1·0461
0·75	1·0034	5·50	1·0251	10·25	1·0473
1·00	1·0046	5·75	1·0263	10·50	1·0485
1·25	1·0057	6·00	1·0274	10·75	1·0496
1·50	1·0068	6·25	1·0286	11·00	1·0508
1·75	1·0079	6·50	1·0298	11·25	1·0520
2·00	1·0091	6·75	1·0309	11·50	1·0532
2·25	1·0102	7·00	1·0321	11·75	1·0544
2·50	1·0114	7·25	1·0332	12·00	1·0555
2·75	1·0125	7·50	1·0343	12·25	1·0567
3·00	1·0137	7·75	1·0355	12·50	1·0579
3·25	1·0148	8·00	1·0367	12·75	1·0591
3·50	1·0160	8·25	1·0378	13·00	1·0603
3·75	1·0171	8·50	1·0390	13·25	1·0614
4·00	1·0183	8·75	1·0402	13·50	1·0626
4·25	1·0194	9·00	1·0414	13·75	1·0638
4·50	1·0205	9·25	1·0426	14·00	1·0651
4·75	1·0216	9·50	1·0437	14·25	1·0663
5·00	1·0228	9·75	1·0449		

[N. B.—The specific gravity increases or diminishes ·00024 for each degree.]

¹ *Chem. Centrbl.*, 1878, 415.

The extract and amount of alcohol being known, it is, in certain instances, possible to detect the *watering* of wine, although such a diagnosis can only be made when the analyst is intimately acquainted with the kind of wine under examination, and in some cases with the characters of the particular vintage. The Bordeaux wines, according to Girardin and Pressier, give almost always the same amount of extract, varying only within the limits of 20 to 20·8 grms. the litre; and the proportion of alcohol also is fairly constant—viz., from 5 to 15, the mean being 10 per cent. From these data they calculated the amount of genuine wine present in any samples. Thus, supposing the extract in a Bordeaux wine to be 14·5, then

$$\frac{1000 \times 14.5}{20.9} = x, \text{ or } 725.00$$

i.e., the litre contains 725 c.c. of wine, the rest being alcohol and water. To know the quantity of alcohol added, it is necessary to ascertain how much the 72·5 parts of wine contain of absolute alcohol;

$$100 : 10 : : 72.50 : x$$

$$x = 7.25.$$

If the absolute alcohol is found, for example, to be 11, then, subtracting 7·25 from 11, it is supposed that 3·75 of alcohol has been added.

That this process, as applied to the Bordeaux wines, is in the main correct, is supported by the fact that the Rouen wine-merchants have frequently paid duty on the excess of alcohol, etc., which Girardin and Pressier found in their wines.¹

In the Municipal Laboratory, Paris, the chemists determine the watering and fortifying of wine by a calculation of the relationship of what they call 'the reduced extract' to alcohol.

The 'reduced extract' is the total extract diminished by the number of grammes less 1 of the potassic sulphate and reducing sugar; thus, if the total extract should be 29·7, the potassic sulphate 3·1, and the sugar 4·5, 2·1 + 3·5 = 5·6; and subtracting 5·6 from the total extract gives the reduced extract as 24·1. The weight of the alcohol for red wines they consider should not exceed four and a-half times the extract, and when this relation is exceeded the wine has been fortified. To determine this relation, the alcohol by weight is divided by the reduced extract.

For white wines the relation between alcohol and reduced extract is fixed at 6·5. Wines watered down and then the alcoholic strength brought up by the addition of alcohol are detected in the French Laboratory as follows:—

In all normal wines the sum of the alcohol per cent. by volume added to the total acidity per litre (alcohol-acid number), calculated as sulphuric acid, is seldom below 12·5. Water lowers this number, alcohol increases it. First, then, the relation of the alcohol to the reduced extract is obtained; if it exceed 4·5, then by calculation on the standard of 4·5, the amount of alcohol that the natural wine may be supposed to have had originally is obtained; and the difference between this and the amount found represents

¹ In Paris a commercial standard has been arrived at, based on the analysis of 6000 samples, and it is laid down that "the amount of added water in all wine which is not sold as being of any special brand, shall be calculated on the basis of 12 per cent. of alcohol by volume, and 24 grms. of dry extract per litre."

the alcohol added. Next, the alcohol-acid number is obtained, and if this is below 12.5 the presumption is that the wine has been watered.

An example will make this clear. A red wine gave the following :—

Dry extract per litre,	14.2
Acidity per litre,	3.100
Alcohol per cent. (volume),	16.0
The relation by weight of alcohol-extract,	= 9.01
The alcohol-acid number,	= 19.1
Now calculating on a standard of,	4.5
Natural alcohol in the wine,	$14.2 \times 4.5 = 63.9$
Reducing this by dividing by 8.0, the volume of alcohol is equal to,	7.99
Alcohol in excess,	$16 - 7.99 = 8.01$
Alcohol-acid figure,	$7.99 + 3.1 = 11.09$

Hence, the alcohol-extract number being superior to 4.5 and the corrected acid-alcohol number being below 12.5, there is a presumption of both watering and fortifying.

This calculation being the results of prolonged experience with regard to ordinary wines is a fairly safe guide to the analyst ; but with regard to certain exceptional wines caution must be exercised in the interpretation of results.¹

§ 326. *Estimation of Succinic Acid and Glycerin.*—Half a litre to a litre of wine is decolorised with animal charcoal, filtered, and the charcoal well washed with water ; the filtrate and washings are then evaporated down in the water-bath, and the drying finished in a vacuum. The residue, when dry, is treated with a mixture of 1 part of strong alcohol and 2½ parts of rectified ether. The latter is driven off by floating the dish in warm water, and the whole evaporated again on a water-bath. The residue is now neutralised with lime-water, which combines with the succinic acid, and forms succinate of calcium. The glycerin is dissolved out by alcohol and ether, and weighed either directly or by loss. The succinate of calcium remaining behind is impure, and should be well washed with spirit before weighing. Every 100 parts of calcic succinate equals 75.64 of succinic acid ($H_2C_4H_2O_4$) ; and since Pasteur has shown that 112.8 parts of grape sugar (107 of cane) yield about 3.6 of glycerin and 0.6 part of succinic acid, it follows that in a natural wine the glycerin would amount to about one-fourteenth part of the alcohol present.²

It has, indeed, hitherto been generally accepted that for every 100 parts of alcohol there should be not less than 7 nor more than 14 of glycerin, and it has been held that deviations from this standard mean addition of alcohol or glycerin ; but the highest quality of the Rhine wines (Table LI.) vary between 16 and 31 parts of glycerin to 100 of alcohol, and, therefore, the views hitherto held demand modification.

¹ E. Egger detects the watering of wine by the presence of nitrates, the grape being stated to be absolutely destitute of nitrates ; white wines are evaporated to a syrup and absolute alcohol added ; so long as it produces a cloud the mixture is filtered, decolorised, and tested with diphenylamine and sulphuric acid. Red wines are precipitated with lead acetate and then magnesium sulphate added before evaporation (*Chem. Centr.*, 1885, 71, 72).

² According to E. Borgmann (*Zeitsch. für anal. Chemie*, xxii., 58-60) the ratio of glycerin to alcohol in pure wines is never less than 7.3 : 100. Analyses of white wines by R. Fresenius and E. Borgmann give the following ratios of glycerin and alcohol :—

	Alcohol.	Glycerin.
Maximum,	100	: 14.4
Minimum,	100	: 7.3
Mean,	100	: 10.4

(*Zeit. f. anal. Chemie.*, xxiii., 48).

Schmidt¹ evaporates a known bulk of the wine with hydrated calcium oxide, extracts the residue with 96 per cent. alcohol, evaporates the clear filtrate, then dissolves this last residue in 15 c.c. absolute alcohol, precipitates with 25 c.c. of ether, filters and evaporates the alcohol-ether solution, and, after one and a-half hours' drying, weighs.

Stierlin² evaporates the liquid without any addition to one-fifth or one-sixth of its volume, extracts with hot absolute alcohol, and uses this alcoholic extract for the estimation of the sugar, non-volatile acids, bitter matters, alkaloids, and glycerin. For the estimation of the last, a measured portion of the alcoholic extract is freed from alcohol by evaporation, and then evaporated down to dryness with slight excess of caustic lime. The glycerin is extracted either with alcohol and ether (2 : 3), or with alcohol and chloroform. (See also the process for extracting glycerin from beer, p. 426.)

Raynaud has pointed out that although the processes in use for the estimation of glycerin are fairly exact, yet with plastered wines too high results are obtained; for if there is any considerable amount of sulphate of potash, it is decomposed by lime, and hydrate of potash is formed, which is dissolved by glycerin in the presence of alcohol, and is weighed with it. He therefore recommends the following process:—The liquid operated upon is evaporated to about one-fifth of its volume, and the potash precipitated by hydrofluosilicic acid and filtered. The filtrate is made weakly alkaline by the addition of hydrate of baryta; sand is also added, and the mass is evaporated to dryness in a vacuum; the dry residue is then extracted with a very large volume of absolute alcohol and ether, as much as 300 c.c. for 250 c.c. of wine being recommended. With the improved processes of extraction which we now possess, however, this is quite unnecessary, and 50 to 100 c.c. in a Soxhlet's apparatus (see p. 49) will have quite the same effect as a much larger quantity. On the evaporation of the alcohol and ether, the glycerin is allowed to stand for twenty-four hours in a vacuum over phosphoric anhydride; finally, it is put into a tube, a perfect vacuum formed, and distilled into the cool part of the tube by a temperature of 180° C.

Probably the best method of estimating glycerin is to separate it from most volatile substances by distillation in a vacuum, and then to oxidise it into oxalic acid, as described under the article 'Butter' (p. 300). A. Partheil³ effects this in the following manner:—50 c.c. of the liquid to be examined, first neutralised by adding a little calcium carbonate, are evaporated down to 15 c.c. and introduced into a small retort. This retort is enclosed in an air bath, the bottom of the bath being made of sheet iron, the sides and top of asbestos card. The neck is connected with a globular receiver, the second opening of the receiver being joined to an inverted condenser, and then to a pump. The receiver is also kept cool. The liquid is first distilled almost to dryness, at ordinary pressure, at a temperature of 120° C. It is then cooled to about 60° C., and the pressure reduced by means of the pump, the temperature raised to 180° C., and the distillation continued for one and a-half hours; the pressure at the end of that time is released, the retort cooled, 10 c.c. of water added, and distillation again proceeded with at the ordinary pressure at a temperature in the bath of 120° C. The distillate is diluted to about 200 c.c., 8 to 10 grms. of caustic soda dissolved in it, and 5 per cent. potassic permanganate added until the colour remains a decided blue-black. The whole is heated for an hour, decolorised with SO₂, 20 c.c. of acetic acid added, the SO₂ driven off by heat, and the oxalic acid precipitated by calcium chloride.

§ 327. *Estimation of Tartaric Acid and Bitartrate of Potash.*—This is best estimated by the method suggested by Berthelot:—20 c.c. of wine are mixed with 100 c.c. of equal volumes of alcohol and ether in a well-stoppered flask. The same process is employed with another 20 c.c. to which has been added sufficient potash to neutralise about one-fifth of the free acid present. Both bottles are allowed to stand two or three days, and at the end of the time, owing to the insolubility of bitartrate of potash in strong alcohol, there will be a deposit of that salt in both bottles. The first will represent the bitartrate of potash present as such; the second, the whole of the tartaric acid which the wine contains. There is, however, always a small quantity of bitartrate in solution, about .004 grm., equalling .28 d. n. soda, and this amount must be added to that found. The precipitates from both bottles are collected on separate

¹ *Op. cit.*

² Stierlin, "Das Bier," etc. Berne, 1879.

³ *Arch. Pharm.*, 1895, ccxxxiii., 391.

filters, washed with the alcohol-ether mixture, dissolved in water, and titrated with soda solution.

Direct Estimation of Malic Acid.—100 c.c. of wine are precipitated with lime-water, added only in slight excess; the filtrate is evaporated down to one-half, and absolute alcohol added in excess; the resulting precipitate, consisting of malate and sulphate of lime, is then collected on a filter, washed, and weighed. If, now, the sulphate of lime in this sample be estimated by solution in water, and precipitation of the sulphuric acid by baric chloride, etc., and the amount subtracted from the total weight of the precipitate, the remainder equals malate of lime.

The Estimation of Sugar in Wine is carried out on the principles described at pp. 111–120.¹

Albuminoid Substances.—The albuminoid substances in wine may be estimated by Mr. Wanklyn's well-known ammonia process:—5 c.c. of the wine are put in a half-litre flask, and made up with water to 500 c.c. : $\frac{1}{100}$ (i.e., 5 c.c.) of this is distilled with a little water and pure carbonate of soda (ammonia free), and the ammonia in the distillate estimated by the colorimetric process known as Nesslerising. An alkaline solution of permanganate of potash is then added, and the operation repeated—the ammonia coming over now being the result of the breaking-up of albuminoid bodies. It would appear that in *white* wines, the albuminous matters are very small in amount; while in *red* and most *young* wines, there is an excess of albuminous matters, which decreases with age; hence, in experienced hands, a determination of this kind may help to distinguish between old and new.

Thudichum and Dupré found in certain wines the following amounts of ammonia:—

	Ammonia free. per cent.	Ammonia albd. per cent.
Ingelheimer, red,	0·0051	0·3730
Port, 1851,	0·0046	0·0888
Sherry, 30 years in bottle,	0·0073	0·1807
Madeira,	0·0021	0·1581
Merstemer,	0·0021	0·3550
Natural Port,	0·0019	0·0527
Port, 1865,	0·0012	0·1760

§ 328. *Astringent Matters.*—Schmidt estimates the astringent and colouring-matters of wine by oxidising with potassic permanganate before and after treatment with animal charcoal.

A. Girard (*Compt. Rend.*, xcv., 185–187) employs sheep-gut for the estimation of tannin in wine. The gut is well washed and cleaned; it is then treated with alkalies, and bleached by the action of potassic permanganate and sulphurous acid. It is then twisted into cords, and again

¹ A special process is in use in the Paris Laboratory for the detection of fictitious claret. The liquid is known as piquette of raisins and dried fruits, and it is often added as an adulterant to the genuine article. 300 c.c. of the wine are fermented fully with yeast at 29°·5 C. The liquid is then filtered, and placed in a dialysing apparatus, in which the outer water is constantly renewed automatically. The water is examined from time to time by the polariscope, and, when the polarisation is constant, the dialysis is stopped. The liquid is neutralised by boiling with chalk, and evaporated to dryness on the water-bath. The residue is treated with 50 c.c. of absolute alcohol, and twice washed with 25 c.c. of the same. The alcoholic extract is next decolorised by charcoal, evaporated to dryness, and the residue taken up with 30 c.c. of water and examined by the polariscope. True claret gives no rotation, or is only very slightly dextrogyrate, while wines mixed with piquette of fruit or glucose are respectively strongly levo- or dextrogyrate.—Dr. Muter in *Analyst*, October, 1885.

bleached by sulphurous anhydride. From 3 to 5 grms. of the gut-cords (the water in which has been previously determined) are soaked in water for four or five hours, and, after being untwisted, are added to 100 c.c. of the wine. After from twenty-four to forty-eight hours, the whole of the tannin is absorbed, and is expressed by the gain in weight of the gut after being washed and dried at 100° C.

§ 329. *Estimation of the Colouring-matter of Wine.*—The colouring-matter of red wines has been termed *œnolin*, or *œnocyanin*, and has also received other names. Glenard has assigned to it the formula $C_{10}H_{10}O_5$; but it is doubtful whether it has ever yet been separated in a state of absolute purity. The process used by Glenard was—precipitation with lead acetate, exhaustion of the washed, dried, and powdered precipitate, first, with anhydrous ether saturated with HCl, then with pure ether; and, lastly, extraction with alcohol, from which the œnolin was obtained by evaporation as a bluish-black powder insoluble in ether, almost insoluble in pure water, but more readily dissolved in acidulated water, acidulated alcohol dissolving it easily. The blue colour is turned red by acid. Œnolin, according to Vasérine,¹ may be separated from wine by mixing the latter with lime to the consistency of a paste, which is drained on a funnel. The residue, containing the colouring-matter, is mixed with alcohol of 95 per cent., and treated with sufficient sulphuric acid to neutralise the lime and decompose the compound of lime with the colouring-matter. The solution is filtered from calcium sulphate, and on evaporation leaves œnolin as a black powder.² Solutions of œnolin show, when examined by the spectroscope, certain bands.

The colour of white wines is due to oxidised tannin; it takes long to develop; hence the manufacturer not unfrequently adds a little caramel. Should this be the only addition, it would be injudicious to consider the wine adulterated.

The artificial colouring of wines by elder-berry, logwood, cochineal, aniline, etc.,³ is said to exist, at all events, on the Continent; and it is a fact that a few home-made, low-priced wines, almost entirely fictitious, are passed off by the aid of the same or similar substances; but with regard to the ordinary foreign wines in English commerce, there is no reliable evidence whatever that any adulteration of this kind has been practised. Nevertheless, it is absolutely necessary to be acquainted with the best and most recent methods for the discovery of such frauds.

The substances actually found to be used fraudulently as artificial colouring are—(1.) Bordeaux verdissant, which is a compound of methylene blue, diphenylamine orange, and the acid-sulpho-derivative of fuchsine; (2.) a mixture of amido-benzene, methyl-violet, and the acid-sulpho-derivative of fuchsine.

In the Paris Laboratory⁴ they use three preliminary tests, and consider

¹ *Bull. Soc. Chim.* [2], xxix., 109, 110.

² According to L. M. Krohn, the red colouring-matter of wine may be obtained by electrolysis as a deposit on the positive pole. The colouring-matters used to adulterate wines do not give this deposit (*Journ. Pharm.* (5), ix., 298-300).

³ Soubeiran says—"At Fismes, in the neighbourhood of Rheims, there has been manufactured for more than a century (since 1741) a colouring agent composed of elderberries, alum, and water, in different proportions, the prolonged use of which can only have injurious consequences on the health, on account of the alum. Unfortunately, the production of this colouring agent (*teinte*) was encouraged by a royal decree of 1781. . . . Recent analyses have shown that this *liqueur de Fismes* contains from 20.8 to 57.56 grms. per litre of alum."—*Dict. des Falsifications*. Paris, 1874.

⁴ "Report on processes in use at the Municipal Laboratory of the city of Paris." By Dr. Muter. *Analyst*, 1885.

wines genuine as to colour if they respond to these tests. On the other hand, should the results be unfavourable, the analyst possesses a valuable guide to the class of colouring-matters present. The three methods are as follows:—

(1.) Sticks of chalk are steeped in a 10 per cent. solution of egg-albumen, and dried first in the air and then at 100° C. The wine is tested by allowing two drops to fall on a surface of the chalk from which the excess of albumen has been removed by scraping; genuine wine gives a gray colour, and young and highly-coloured wine may give a somewhat bluish tint, but there should be no trace of green, violet, or rose.

(2.) The wine is alkalisied by baryta-water until it is of a greenish hue; it is then shaken up with acetic ether or amylic alcohol. If the wine is pure there is no colour in the upper layer, with or without the addition of acetic acid. On the other hand, coal-tar colours of a *basic* nature colour the solvent, and give indications suggestive of amido-benzene, fuchsine, safranine, chrysoidine, chrysaniline, mauveine, methyl-violet, and Beibrich red.

(3.) 10 c.c. of the wine are alkalisied until the wine becomes of a green colour by the addition of 5 per cent. potassium-hydrate solution. To this are added 2 c.c. of a solution of mercurous acetate; the whole, after shaking, is filtered. With pure wine the filtrate is colourless, both in itself and after acidulating with hydrochloric acid, while coal-tar colours of an *acid* nature tint the filtrate red or yellow.

If either of the two last tests responds, the wine is further examined as follows:—

A.—THERE IS EVIDENCE FROM (2.) OF AN ANILINE BASIC COLOUR.

The amylic alcohol or acetic ether solvent is separated and divided into two parts, W. and S.—the one, W., is evaporated down with threads of wool, the other, S., on threads of silk. The wool and silk are both coloured red, hydrochloric acid discolours it to a dirty brown, but water restores the red.

The silk is coloured red, but not the wool; the reaction with hydrochloric acid does not take place.

Violet, and when treated with HCl, becomes bluish-green, changing to yellow, but water in excess restores the violet.

Indigo-blue, when treated with HCl, but on dilution with water, reddish-brown.

Threads scarcely affected with HCl, but decolorised by boiling with powdered zinc, the colour returning on exposure to air.

Straw-yellow threads, becoming poppy-red with strong sulphuric acid.

Orange-yellow threads, becoming scarlet with sulphuric acid.

Threads of a reddish colour, decolorised instantly by a few drops of sodium bisulphite.

Dyes analogous or allied to Rosaniline.

B.—THERE IS EVIDENCE FROM (3.) OF A TAR DYE OF AN ACID NATURE.

The wine is strongly acidulated with hydrochloric acid, and shaken up with acetic ether (or amylic alcohol). The wine is saturated with a slight excess of ammonia, and shaken up with the same solvent. The ethereal fluids are mixed, evaporated to dryness, and tested with a drop of strong sulphuric acid.

Parma, violet colour.

Maroon.

Blue.

Scarlet.

Red.

Green to violet.

Fuchsine red.

Orange yellow, or on dilution with water a transient poppy-red.

Brownish-yellow.

Yellow.

Roccelline.

Foundation Red.

Bordeaux R. and B.

Ponceau R.

„ B.

The Beibrich Red.

Tropoline 000.

Tropoline O., or Chrysoine.

Helianthine.

Eosine B., or J. J.

Safranine.

Ethyleosine.

Dr. Dupré, taking advantage of the fact that the colouring-matter of wine¹ only dialyses to a minute extent, and that the colouring-matters of Brazil, logwood, and cochineal, readily dialyse, separates the latter colouring-matters from the wine by dialysis. The same chemist has suggested a still more convenient and practical process—viz., the staining of a jelly. The jelly is made by dissolving 5 grms. of gelatin in 100 c.c. of warm water, and pouring the solution into a square flat mould made of paper. From this cake cubes about $\frac{3}{4}$ inch square are cut with a sharp wet knife, and are immersed in the wine, taken out after the lapse of from twenty-four to forty-eight hours, washed slightly, and sections cut, in order to see how far the colouring principles have penetrated. If the wine is pure, the colour will be confined almost entirely to the edges of the slice, or will not have penetrated beyond $\frac{1}{16}$ to $\frac{1}{8}$ inch; most other colouring-matters rapidly permeate and colour the jelly.

(1.) *Colouring-matters penetrating slowly into the jelly:—*

Colouring-matter of pure wine.
“ “ Rhatany root.

(2.) *Colouring-matters penetrating rapidly into the jelly:—*

Rosaniline.	Litmus.
Cochineal.	Red Cabbage.
Logwood.	Beetroot.
Brazil-wood.	<i>Malva sylvestris.</i>
Indigo.	<i>Althaea officinalis.</i>

The jelly may be examined spectroscopically, good results being obtained in the case of rosaniline, red cabbage, and beetroot; and may be also tested with reagents—*e.g.*, dilute ammonia dissolves much colour from the slice, if the colour should be derived from logwood or cochineal; on the other hand, the ammonia remains colourless in the case of rosaniline, red cabbage, and beetroot.

A simple method for the detection of certain colouring-matters is that of Lammatine:—Shake 100 parts of the wine with 100 of coarsely powdered peroxide of manganese, and then pass through a double filter; if pure, a colourless filtrate will result. The process is said to answer well in the case of logwood and cochineal, but to fail with aniline.

A general method, applicable to fuchsine and other colouring-matters, is based upon the fact that a great many of the colouring-matters which may be used for purposes of wine adulteration (such as caramels, ammoniacal cochineal, sulphindigotic acid, logwood, and the lichen reds), are precipitated by acetate of lead; whilst fuchsine, if present in considerable quantity, is only partially thrown down. Those which are not precipitated may be separated by agitating the filtrate with amyl alcohol.

The lead precipitate may be treated by dissolving out cochineal, sulphindigotic acid, and fuchsine, by a solution of potassic carbonate [2:100]. From this liquid the fuchsine is separated by neutralisation with acetic acid and agitation with amyl alcohol, the rose-coloured liquid obtained being identified as a solution of fuchsine by the spectroscope. On

¹ The colouring-matter of Rhatany root has the same property.

now acidifying the liquid with sulphuric acid, the carminic acid from the cochineal is removed by means of amyl alcohol, and identified by its three bands—viz., one between D and E in the red, the second in the green, and the third in the blue. The indigo remaining may be detected by the blue colour, and absorption-band between C and D. The original lead-precipitate, insoluble in potassic carbonate, is treated with a two per cent. solution of potassium sulphide, which dissolves the colouring-matter of logwood and that of the wine itself. Logwood may, however, be tested for directly in the wine by the addition of calcium carbonate and two or three drops of lime-water. In the case of a natural wine, the filtered liquid is almost colourless, but is of a fine red colour if logwood is present. Lastly, the lichen red may be obtained by washing the insoluble portion left after treatment with potassium sulphide, and dissolving it in alcohol, when a red colour and a definite absorption-band reveal its presence.

For the detection of fuchsine simply, Bouillon (*Comptes Rendus.*, lxxxiii., 858, 859) recommends half a litre of the wine to be evaporated down to 120 c.c., with the addition of 20 grms. of barium hydrate. It is then filtered, and the filtrate shaken up with ether; the ether is separated, a drop of acetic acid, a little water, and a small piece of white silk, are added, and (if an appreciable amount of fuchsine is present) the silk assumes a pink colour immediately; if not, the liquid must be concentrated nearly to dryness.

F. König¹ has a process for detecting fuchsine: 50 c.c. of the wine are treated with ammonia in slight excess, and boiled with a little pure wool [.5 gm.], until all the alcohol and ammonia are evaporated. The wool is washed and directly moistened with strong potash, and heated until it dissolves into a more or less brown fluid. After cooling, to this is added half its volume of pure alcohol, and then an equal volume of ether; it is strongly shaken. The smallest trace of fuchsine is taken up by the ether, and is coloured red by acetic acid. 0.4 mgrm. fuchsine in a litre of wine is said by this means to be discovered readily. The process destroys the natural colour of the wine.

§ 330. Vogel and others have studied the detection of the colouring-matters of wine by means of the spectroscope.

The following curves are examples of various colouring-matters (see fig. 66 c):—

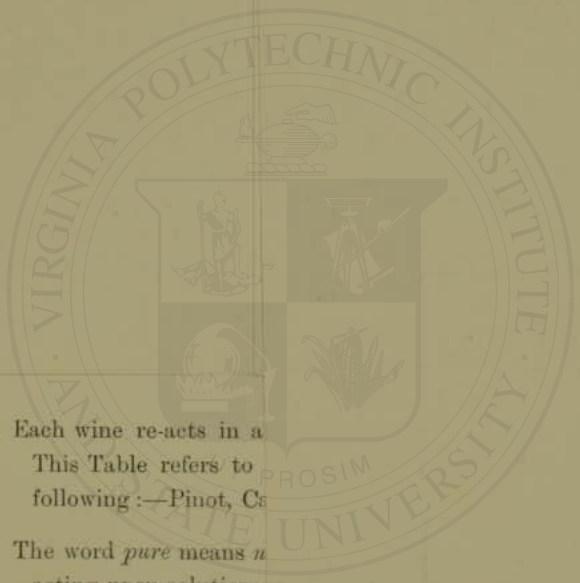
- No. 37. Wine colouring-matter, (I.) pure, (II.) diluted.
- No. 38. Wine colouring-matter, with ammonia.
- No. 39. (I.) Mallow colouring-matter concentrated, (II.) elderberry concentrated.
- No. 40. Acid cherry. (b) Acid cherry with the addition of tannin.
- No. 41. Mallow colouring-matter with the addition of alum.
- No. 42. Indigo solution.

With carefully made comparison solutions, there can be little doubt that the spectroscopical method of identifying colouring-matters will be found of great value.

Lastly, M. A. Gautier² has proposed a method aiming at a systematic detection of every probable colouring-matter likely to be added to wine. How far the whole, or any portion, of this elaborate system will be followed and confirmed by chemists remains to be seen.

¹ *Ber. der deutsch. chem. Gesellsch.*, Berlin, xiii., 2263.

² *J. Pharm. Chim.* [4], xxv., 8-12 and 102-106.



Black e
Wine,

DWARF E

Dwarf e
Wine,

PRIVET, I

Privet,
Wine,

WHORTLI
pure.

Myrtill
Wine,

INDIGO, I

Indigo,
Wine (C
clarifi

1. Each wine re-acts in a
This Table refers to
following:—Pinot, Cs
2. The word *pure* means *n*
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3. *Brazil-wood*, 1 part, *win*
examined resulted fro
in the proportions na
coloration, and repres
ating substance.

TABLE A.

[To face p. 460.]

Nature of the Liquid examined.	A.	B.	C.	D.	E.	F.	G.	H.	K.	L.	M.	N.	O.	P.
	To 1 c.c. of the unclarified wine, add 3 to 5 c.c. of a 1-200 solution of sodium carbonate, according to the acidity and intensity of colour.	Heat the preceding to boiling.	To 2 c.c. of clarified wine add 1.5 to 2 c.c. of an 8 per cent. solution of sodium bi-carbonate charged with carbonic acid. Examine the colour after 2 minutes.	To 2 c.c. of clarified wine add 3 c.c. of dilute ammonia (1 of ammonia to 10 of water); then dilute with 5 c.c. of water.	2 c.c. of clarified wine are treated with 2 c.c. of saturated baryta water, and filtered after 15 minutes. Colour of the filtrate.	The filtrate E is acidulated with acetic acid.	To 2 c.c. of clarified wine add 3 to 4 c.c. of saturated solution of borax (depending upon the depth of the colour of the wine).	To 4 c.c. of clarified wine add 1 c.c. of 10 per cent. solution of alum, and 1 c.c. of a 10 per cent. solution of sodium carbonate. Colour of the lake.	Colour of the filtrate from H.	To 2 c.c. of clarified wine add 1 c.c. of sub-acetate of lead, of 15' B. Shake. Filter. Colour of precipitate.	Colour of filtrate from L.	To 1 c.c. of clarified wine add 1 c.c. of aluminium acetate of 2' B. Colour of liquid.	To 1 c.c. of clarified wine add 4 drops of potassium aluminate (obtained by adding alum to a solution of alumina in potash). Filter the mixture.	To 0.1 gm. of barium biniodide add 5 drops of a 5 per cent. solution of tartaric acid, and then 3 c.c. of clarified wine. Examine the colour 18 hours afterwards.
PURE WINE.	Coloration bluish-green; grey, slightly greenish, according to the wine. With ammon. lilac.	The liquid becomes yellow; has a tendency to become decolorised. Any trace of lilac present disappears.	Deep grey, with a little green, or sometimes violet.	Greenish grey-blue, or greenish-grey, faintly lilac.	Dirty yellow, with a little green.	Liquid faintly roseate; in certain cases nearly colourless.	Liquid grey-blue or greenish; flax blossom; sometimes a trace or more of lilac.	Lake bluish-green, or greenish.	Clear bottle-green, almost colourless with certain rare varieties.	Ashy blue-green, rarely apple-green, or yellowish-green.	Decolorised.	Wine lilac. <i>Aramon</i> is nearly decolorised.	Lilac, faintly rose, tendency to decolorise.	Liquid scarcely rose-tinted; a trace (or none) of orange deposit in contact with the biniodide.
PURE BRAZIL-WOOD.	Red currant.	"	Red currant.	Red currant.	"	"	Red currant.	"	"	Brown lilac.	If an excess of lead-salt is added and the fluid heated: Red currant.	Preserves its original colour.	"	"
Brazil-wood, 1 part. } Wine, 4 " }	Brown-lilac, or brown tinged with maroon.	Wine colour.	Wine lilac.	Lilac, soiled with grey, or maroon.	Red-brown.	Colour of onion skin, or nearly colourless yellow.	Wine lilac.	Lilac-lake, running into rose or brick-red.	Grey, with a little maroon.	Precipitate ash-blue, tinged with yellow or brick-red.	Decolorised, or very slightly reddish.	Old wine-red, or roseate.	Colour of onion skin, or slightly rose.	Liquid yellow, without rose colour even in 10 hours. Large orange deposit in contact with the biniodide.
PURE LOGWOOD.	Red-purple, or violet.	"	Rose wine-colour.	Violet-lilac. The violet tends to disappear with an excess of re-agent, and to return on heating.	"	"	Rose wine colour.	Lake violet-blue.	"	Somewhat violet.	Colourless; or but slightly lilac.	Becomes violet-blue.	Precipitate lilac, or somewhat violet.	"
Logwood, 1 part. } Wine, 4 " }	Same colorations as pure wine.	Lilac, or wine violet.	Greenish deep grey.	Greenish-grey.	Dirty greenish-yellow.	Scarcely roseate.	Grey-blue (flax blossom), slightly tinged with maroon.	Lake bluish-green tinged with violet, becoming more violet by drying in the air.	Clear bottle-green.	Blue, rather more violet than with wine only.	Decolorised, or but very slightly yellow.	Violet or lilac.	Rose, tinged with violet.	Like the preceding mixture.
PURE COCHINEAL.	Lilac.	"	Lilac.	Violet-lilac.	"	"	Lilac.	Lake-rose.	Liquid always rose, more or less deep.	Deep violet lilac.	Lilac, almost colourless.	Rose lilac.	Rose lilac.	"
Cochineal, 1 part. } Wine, 4 " }	Grey flax-blossom, or grey with tinge of lilac.	The grey tinged with lilac is unchanged.	Grey, tinted with lilac or violet.	Greenish-grey, or greenish-grey brown.	"	Very distinctly rose.	Lilac, or bluish-grey tinged with lilac.	Lake - bluish, slightly roseate.	Rose lilac.	Ashy-blue; clear green.	Decolorised.	Wine lilac.	Rose.	Liquid rose, with tinge of orange-yellow in contact with biniodide.
PURE FUCHSINE.	Rose.	The rose-lilac or rose-violet tint disappears.	Rose.	Rose, decolorised by an excess of re-agent.	Dirty yellowish-green.	"	Rose.	"	"	No precipitate.	Rose.	Rose.	"	"
Fuchsine, 1 part. } Wine, 4 " }	Colour greenish-grey, sometimes with lilac tint.	"	Wine, lees roseate.	Greenish-grey, with or without tinge of rose.	"	Distinctly rose.	Bluish-grey with lilac; sometimes the latter is very faint.	Lake-bluish or greenish, slightly roseate.	Clear green.	Ashy-blue; sometimes rose-tinted.	"	Lilac or rose.	"	Liquid scarcely rose-tinted. Orange deposit in contact with biniodide.
PORTUGAL BERRIES, pure.	Violet rose.	"	Violet-rose.	Violet-rose.	Dirty greenish-yellow.	"	Violet-rose.	Lake violet.	"	Violet maroon, slight; soluble in excess.	Decolorised if the lead-salt is not in excess; otherwise yellow-red.	Violet-rose.	"	"
Phytolacca, 1 part. } Wine, 4 " }	Dark violet, or lilac.	Yellow-grey with little maroon, which increases with the amount of phytolacca.	True lilac.	Deep grey, with a little maroon, or lilac.	"	"	Lilac, or grey-blue with little lilac.	Bluish-green, or greenish, slightly roseate, with more phytolacca.	Lilac.	Ashy-greenish-blue.	Decolorised, or only very faintly roseate.	Wine lilac, or distinctly lilac.	Bright rose.	Liquid distinctly rose. Orange deposit in contact with biniodide.
HOLLYHOCK, pure.	Dark bottle-green.	Dark greenish-grey.	Bottle-green.	Bottle-green, darkened with grey.	"	"	Colour of dark infusion of tea.	"	"	"	"	"	"	"
Hollyhock, 1 part. } Wine, 4 " }	Greenish, slightly blue, or greenish-grey.	Partially decolorised, slightly greenish-grey.	Grey, with little green or blue.	"	"	Scarcely roseate.	Greenish blue-grey.	"	"	"	"	"	"	"
BETROOT, pure.	Rose, or yellowish-red, which is persistent.	"	Rose or yellowish-red, which is persistent.	Yellow if the beet-juice was fermented; roseate if new.	"	"	Rose or yellowish-red, according to the age of the infusion.	"	"	"	"	"	"	"
Beetroot, 1 part. } Wine, 4 " }	Yellowish-grey, if the beet-juice was fermented: faint yellowish-red if fresh.	Tends to be decolorised if the beet-juice was old; yellowish-grey touched with red if fresh.	Reddish-yellow or lilac brown.	Dirty yellowish-grey, with a little maroon if the beet-juice was fresh and not fermented.	Clear yellowish.	Yellowish, or colour of onion skin, slightly rose.	Grey; a little brown-violet if the beet-juice was recent.	"	"	"	"	"	"	"
BLACK ELDER, pure.	Violet lilac, changing to dark bluish-grey, then to bluish-green.	"	Wine lilac.	Fine green.	"	"	Wine colour.	"	"	"	"	"	"	"
Black elder, 1 part. } Wine, 4 " }	Dark green with lilac tinge.	Dark greenish-grey.	Remains lilac for an instant, then rapidly changes to grey with greenish blue.	Dirty greenish-grey.	Dirty greenish-yellow.	Liquid is roseate.	Lilac, or greenish-grey-blue, scarcely lilac.	"	"	"	"	"	"	"
DWARF ELDER, pure.	Bottle-green, darkened with grey.	"	Wine red.	Dark bottle-green.	"	"	Colour of port wine.	"	"	"	"	"	"	"
Dwarf elder, 1 part. } Wine, 4 " }	Green with lilac tinge, or grey slightly green.	The green tends to disappear on heating.	Lilac with grey; or grey tinged with maroon.	Dark green with grey, possibly tinged with maroon.	Clear greenish-yellow.	Scarcely roseate.	Greenish - grey - blue, tinged with lilac.	"	"	"	"	"	"	"
PRIVET, pure.	Dark green.	"	Dark grey tinged with maroon.	Dark green.	"	"	Dirty reddish-rose.	"	"	"	"	"	"	"
Privet, 1 part. } Wine, 4 " }	Dark green to greenish-grey.	Changes to dirty yellowish.	Greenish-grey.	Bluish, greenish-blue, or greenish-grey, according to the variety of wine.	Yellowish, with a little green.	"	Greenish - grey - blue, tinged with lilac.	"	"	"	"	"	"	"
WHORTLEBERRIES, pure.	Wine colour, more re-agent changes it to grey-lilac, and grey tinged with lilac.	Becomes yellowish-grey.	Remains lilac. An excess of the re-agent rapidly changes it to grey, slightly red.	Maroon by transmitted light; grey bottle-green, by reflected light.	"	"	Dirty yellow, tinged with lilac.	"	"	"	"	"	"	"
Myrtille, 1 part. } Wine, 4 " }	Yellowish-grey with a little lilac or rose.	Becomes deep grey.	Yellowish-grey, frequently with a little red.	Greenish-grey or yellowish-grey.	Greenish-yellow.	Faintly roseate.	Grey, with a little lilac.	"	"	"	"	"	"	"
INDIGO, pure.	Blue.	"	Liquid blue, tending to become green.	Blue, slowly decolorised.	Becomes green; then slowly decolorised.	"	Blue.	"	"	"	"	"	"	"
Indigo, 1 part. } Wine (not clarified) 4 " }	Greenish, slightly blue.	Tends to become yellow.	Bluish, or greenish-blue.	Oak-leaf green.	Dirty green-grey.	Roseate.	Bluish-green.	"	"	"	"	"	"	"

NOTES TO TABLE A.

- Each wine re-acts in a slightly different manner, according to its variety, age, etc. This Table refers to wines of five to fifteen months old, and particularly to the following:—Pinot, Carignane, Teinturier, Carbenet.
- The word *pure* means *not mixed with wine*. The reactions shown were obtained by acting upon solutions of the substances in water, containing 10 per cent. of alcohol, and were made of such strength that the colours corresponded in intensity with those of the wines being examined.
- Brazil-wood, 1 part, wine 4 parts*, means that the intensity of the colour of the liquid examined resulted from the mixture of the decoction of Brazil-wood and of wine in the proportions named. These proportions refer only to the intensity of the coloration, and represent but a very minute ponderable quantity of the adulterating substance.

The following abstract of M. Gautier's paper is taken from the *Analyst*, i., 1877 :—

M. GAUTIER'S PROCESS FOR DETECTING COLOURING-MATTERS IN WINE.

§ 331. *Preliminary Preparation of the Sample.*—The sample is mixed with $\frac{1}{10}$ its volume of white-of-egg previously diluted with $1\frac{1}{2}$ times its bulk of water, well shaken, and, after standing for half an hour, filtered. If the wine is very poor in tannates, a few drops of a fresh aqueous solution of tannin should be added before agitating with albumen. The

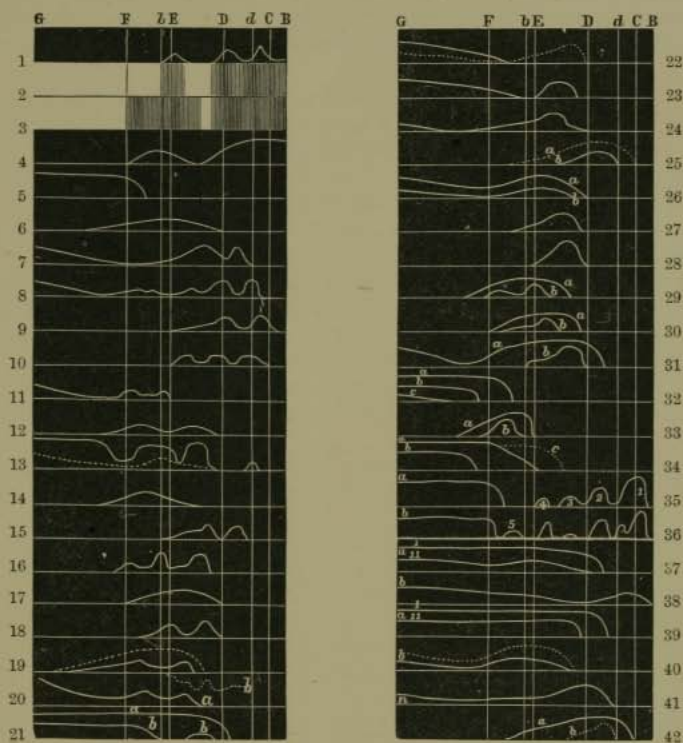


FIG. 66, c.

filtrate is treated with dilute sodium bicarbonate until its reaction is *very* feebly acid. All the reactions of Table B. must be made on this liquid, except those for indigo, which are executed upon the albuminous precipitate.

TABLE B.—SYSTEMATIC PROCESS TO BE FOLLOWED FOR THE DETECTION OF THE NATURE OF FOREIGN COLOURING-MATTERS ADDED TO WINES.

A. Having placed aside the filtrate from the albuminous precipitate, the precipitate is washed until the washings are almost colourless.

Two cases may present themselves:—

(a.) The precipitate after washing remains wine-coloured, lilac, or maroon; *wine, natural, or adulterated with the greater part of the substances usually employed.* Pass on to C.

(b.) The precipitate is of a very deep wine-colour, violet-blue, or bluish; *wines from the deepest coloured grapes, or wines coloured with indigo.* Proceed to B.

B. The precipitate is washed with water, then with alcohol of 25 p.c., a part is then removed and boiled with alcohol of 85 p.c.

(a.) The filtrate is *rose, or wine-coloured.* A portion of the precipitate is removed from the filter, suspended in water, and carefully saturated with dilute potassium carbonate. The colour changes to brown or blackish-brown; *natural wines, or adulterated with substances other than indigo.* Pass to C.

(b.) The filtrate is *blue.* A portion of the precipitate suspended in water and treated with dilute potassium carbonate affords a deep blue liquid, which changes to yellow by an excess of the reagent. *Various preparations of indigo. Indigo.*

C. 2 c.c. of wine are treated with 6 to 8 c.c. of a $\frac{1}{200}$ solution of sodium carbonate, which must be added in slight excess (1 c.c.) after the change of colour.

(a.) The liquid becomes *lilac, or violet*; sometimes the liquid becomes wine-coloured, or dashed with violet. *Brazil-wood, cochineal, Portugal berries, fuchsine, . . . wines of certain sorts, fresh beetroot, logwood, both elders, whortleberries (myrtile), Portugal berries.* Pass to D.

(b.) The liquid becomes bluish-green, sometimes with a faint lilac tint, *wine, hollyhock, privet,¹ whortleberries, logwood, Portugal berries, fuchsine.* Pass to M.

(c.) The liquid becomes greenish-yellow without any blue or violet, *beetroot (old or fermented decoction), whortleberries, certain rare varieties of wine.* Pass to L.

D. The liquid C. (a.) is heated to boiling.

(a.) The liquid remains wine-violet, rose, or wine-lilac, or becomes a brighter lilac; *logwood, Brazil wood, cochineal, certain varieties of wine.* Pass to E.

(b.) The colour disappears, or changes to a yellow, or maroon, or reddish tint, *wine, fuchsine, both elders; whortleberries, Portugal berries, fresh beetroot.* Pass to F.

E. Treat 4 c.c. of the wine with 2 c.c. of each of a 10 per cent. solution of alum, and a 10 per cent. solution of crystallised sodium carbonate. Filter.

(a.) Clear yellowish-green lake (which may be bluish from mixtures of maroon-coloured wines), filtrate colourless, becoming very slightly yellow on warming; its own volume of aluminium acetate at 2° B. almost wholly decolorises it. On acidification with acetic acid, after treatment with its own volume of barium hydrate (saturated solution), the wine

¹ The colouring-matter of privet berries is stated to be used in Saxony for colouring wine; it gives an absorption band at D., and a faint absorption at F. This colouring-matter is extracted by amyl alcohol. Tartaric acid heightens the colour, shutting out all the blue. Alum colours it beautifully blue and broadens the absorption at D., while the absorption of the blue and green is diminished. Tartaric acid annihilates the blue colour, and gives a colour similar to wine; careful neutralisation with ammonia restores the blue colour and the band at D. (*Vogel*).

becomes clear greenish-yellow or maroon, *pure* or *mixed* wines. See Table A.

(b.) Greenish-blue lake, or dirty yellowish-green, according to the varieties present, sometimes very slightly wine-coloured. Filtrate bright-rose, gradually decolorised on warming, though retaining a tinge of lilac; not decolorised by lime-water in the cold. COCHINEAL.

(c.) Wine-violet lake, which darkens on exposure to the air. Filtrate bottle-green, or grey faintly red (if much logwood is present). The filtrate becomes green on warming. LOGWOOD.

(d.) Lilac, or maroon-lilac lake. Filtrate greyish with tint of maroon. On boiling, this filtrate becomes fine old-wine coloured. BRAZIL-WOOD.

F. Treat 4 c.c. of the wine with alum and sodium carbonate (as explained at E.), add to the mixture two or three drops of very dilute sodium carbonate, and filter.

(a.) The filtrate is lilac or wine-coloured, *Portugal berries, fresh beetroot*. Pass to G.

(b.) The filtrate is bottle-green, or reddish-green, *wine, fuchsine, black elder, whortleberries, beetroot*. Pass to H.

G. Treat 2 c.c. of the wine with subacetate of lead solution, of density 15° B. Shake. Filter.

(a.) The filtrate is rose-coloured, which persists even when made slightly alkaline; it slowly disappears on boiling. Lime-water destroys the rose colour. PORTUGAL BERRIES.

(b.) The filtrate is yellowish, or brownish-red. FRESH BEETROOT.

H. The alum-lake obtained from F. (b.) is—

(a.) Deep blue. On treating the clarified wine with a few drops of aluminium acetate solution, it becomes a decided violet, or wine violet. *Both elders*. Pass to I.

(b.) Bluish-green, green, or faintly rose-tinted, *wine, whortleberries, beetroot, fuchsine*. Pass to J.

I. After the test H. (a.) treat a fresh quantity of 2 c.c. with 1.5 to 2 c.c. (according to its acidity and the depth of its colour) of an 8 per cent. solution of sodium bicarbonate charged with carbonic acid.

(a.) The liquid remains lilac for a moment, then changes to greenish-grey blue. Another specimen treated with sodium carbonate (according to C.), and heated to boiling, becomes dark greenish-grey. BLACK ELDER.

(b.) The liquid retains a lilac tint, or becomes grey with mixture of maroon, or dirty lilac. Another specimen treated with sodium carbonate (as at C.) tends to discolour on heating, the green being replaced by red. DWARF ELDER.

J. Treat 5 c.c. of the clarified wine with a slight excess of ammonia, heat to boiling, and after cooling shake with 10 c.c. of ether, decant and evaporate the ether, and treat the residue left on evaporation with acetic acid.

(a.) The liquid becomes red. FUCHSINE.

(b.) The liquid does not become red, *wine, whortleberries, fresh beetroot*. Pass to K.

K. Another specimen is treated according to C. with sodium carbonate.

(a.) The colour darkens or becomes red on heating, *whortleberries, fresh beetroot*. Pass to L.

(b.) The greenish or bluish-green liquid, possibly having a wine tinge, has a tendency to discolour on heating. *Natural wine*.

L. Treated with sodium bicarbonate according to the rules given at I.

(a.) The liquid is deep grey, slightly greenish, green, sometimes green with very slight lilac tint.

The clarified wine, treated with an equal volume of saturated baryta water, and filtered after standing for fifteen minutes, gives a dirty yellow, or slightly greenish filtrate.

With an equal volume of aluminium acetate of 2° B. it gives a lilac wine-coloured filtrate.

With a few drops of aluminate of potash no change of colour. With sodium carbonate, employed as at C., the liquid tends to lose its colour on heating. With barium peroxide, used according to Table A, column P, the liquid is faintly rose-tinted, with or without an orange-coloured deposit on the barium peroxide. NATURAL WINE.

With the general characters above indicated, if with baryta water it affords a madeira-coloured filtrate, changing to buff on acidulation with acetic acid; if with borax it becomes deep-green with a bluish cast; if with alum and sodium carbonate (as at E) a precipitate falls of a deep bottle-green, with bluish tinge, and if with aluminium acetate it remains rose-coloured with no change to violet-blue. TEINTURIER.

(b.) The liquid is reddish-yellow or brown-lilac. By treatment with acetate of alumina the filtrate is clear lilac. With a few drops of aluminate of potash the colour becomes that of the skin of an onion, and with a larger quantity of the reagent the colour is green, tinged with maroon. With sodium carbonate (employed as at C.) the fluid passes to yellowish or greyish-yellow, with tinge of red. With barium peroxide, flesh-coloured liquid with considerable orange-coloured deposit in contact with the peroxide. BEETROOT, *fermented or not*.

(c.) The liquid is yellowish-grey, with tinge of green or red. With baryta water the filtrate is yellowish olive-green. With aluminium acetate the filtrate is bluish violet, or violet-lilac. With aluminate of potash, fresh rose, becoming yellowish-green, with an excess of reagent. With sodium carbonate (as at C.) the fluid becomes deep grey on heating. With barium peroxide the fluid is bleached, or remains but very slightly roseate, with a trace of orange deposit in contact with the peroxide. WHORTLEBERRIES.

M. The mixture of wine and alkaline carbonate C. (b.) is heated to boiling.

(a.) The mixture becomes lilac-violet, or violet. LOGWOOD.

(b.) The mixture tends to become decolorised, or changes to yellowish-green, or dark green, or maroon green, *natural wines, whortleberries, both elders, privet, Portugal berries, fuchsine*. Pass to N.

N. Treat the wine with alum and sodium carbonate, as directed at E., and filter.

(a.) The colour of the filtrate is lilac. *Portugal berries*.

(b.) The filtrate changes to bottle-green, or reddish-green. *Natural wines, whortleberries, hollyhock, privet, both elders, fuchsine*. Pass to O.

O. Treat 2 c.c. of the clarified wine with 3 or 4 c.c. of a saturated solution of borax, according to the intensity of the colour of the wine.

(a.) The liquid remains wine lilac, or with some violet tinge, *both elders, privet, whortleberries*. Pass to P.

(b.) The fluid becomes bluish-grey-flax-blossom, greenish or bluish-grey, with very faint trace of lilac, *pure wine, whortleberries, hollyhock, fuchsine*. Pass to R.

P. Treat a new portion of wine with sodium bicarbonate (as directed at I.).

(a.) The tint, at first lilac, changes afterwards to grey, slightly brownish, or to maroon. If a new portion be treated with sodium carbonate, according to C., and then heated to boiling, it becomes clearer, and loses its green tint.

The lake obtained according to E. is deep blue-green. DWARF ELDER.

(b.) The specimen remains grey, tinged with green, bottle-green, or yellowish. Sometimes (black elder) it acquires a lilac tint, which almost immediately disappears, changing to a greenish-grey-blue, *whortleberries*, *black elder*, *privet*. Pass to Q.

Q. Treat a specimen of the wine with alum and carbonate of soda (as directed at E.). Shake the mixture, and after a few moments throw it on a filter.

(a.) The lake remaining on the filter is deep green-blue; the filtrate is clear bottle-green. A sample treated with sodium carbonate (as at C.) darkens and becomes grey, slightly greenish, on heating to boiling. BLACK ELDER.

(b.) The lake is clear bluish or greenish. The filtrate is clear bottle-green. A sample treated with sodium carbonate (as at C.), and heated to boiling, changes to dirty yellowish. PRIVET.

(c.) The lake is ash-green faintly rose-tinted. The filtrate is bottle-green, with tint of maroon. A sample treated with sodium carbonate (according to C.) becomes deep grey on being heated to boiling. WHORTLEBERRIES.

R. Treat a specimen of the wine with ammonia and ether, as directed at J.

(a.) The ether being decanted and evaporated, the fluid residue becomes rose-coloured on treatment with acetic acid. FUCHSINE.

(b.) The liquid left after the evaporation of the ether does not become red on acidification with acetic acid, *natural wines*, *hollyhock*, *whortleberries*. Pass to S.

S. A sample is treated with its own bulk of a solution of aluminium acetate of 2° B.

(a.) The colour of mixture remains wine-coloured, *natural wines*, *whortleberries*; differentiate between them, as directed at L (a), and L (c).

(b.) The colour of the mixture becomes violet-blue, *hollyhock*, *whortleberries*. Pass to T.

T. A specimen is treated with alum, and sodium carbonate (as at E.), and after a few moments filtered.

(a.) The lake is clear green, slightly bluish, and rose-tinted; filtrate is bottle-green, with little maroon. With borax (as at O), particularly if the sample has been concentrated, the liquid is grey with trace of lilac. 2 c.c. of the liquid treated with 3 c.c. of dilute ammonia (1 vol. of liq. ammonia with 10 vols. of water), and the mixture diluted with its own bulk of water, gives a liquid which is yellowish-grey, greenish, or greenish-grey. The other characteristics as at L. WHORTLEBERRIES.

(b.) The lake is green, slightly bluish, quite free from rose, filtrate clear bottle-green. With borax the liquid is greenish blue-grey. With ammonia (as above), dark bottle-green. With aluminium acetate (as at S.), bluish-violet coloration. HOLLYHOCK.

Although somewhat difficult, this systematic method serves for the discovery of several colouring-matters mixed in one wine, if the indications of

Tables A. and B. are carefully observed and followed. It is always desirable to determine the presence of fuchsine by the special reactions given further on. By means of Table B. the presence of one or several of the colouring-matters may be detected; but before deciding, it is as well to verify by repeating, for the substances so found, the reactions of Table A. on the sample; and also the more special characteristics given further on, for the identification of those substances.

SPECIAL REACTIONS FOR THE DETECTION OF CERTAIN OF THE
COLOURING-MATTERS MIXED WITH WINES.

Brazil Wood.—Even a very strong clarification (two or three times more albumen than mentioned at the head of Table B.) does not wholly decolorise the adulterated wine. It becomes yellow-buff, which on exposure to the air gradually changes to red. If a wine that has been adulterated with Brazil-wood is clarified, and then a skein of scoured silk, washed with dilute tartaric acid, be soaked in it for twenty-four hours, and then withdrawn, washed, and dried at 60° to 70° C., the silk will be found to be dyed lilac-maroon, or red. In pure wine, the skein remains wine-coloured or lilac.

If the dyed silk be now dipped into dilute ammonia, and heated to 100° C. for a moment, it becomes lilac-red, if Brazil-wood were present; but deep grey, with scarcely a tinge of its original colour, if the wine were pure. If the ammonia be replaced by lime-water, the skein changes to ash-grey if Brazil-wood were present; but to a dark, dirty-yellowish-red if the wine were pure. Finally, if the skein be dipped into aluminium acetate, and then heated to 100° C., it retains its wine-red lilac colour. This reaction differentiates Brazil-wood from logwood.

Logwood.—If the colour due to logwood is in excess in the wine, ammonia gives it a shade of violet; if the proportion of logwood is small, the reactions B, L, N, of Table A., which are very delicate, should be tried.

A skein of silk, prepared in the manner described for Brazil-wood, and treated with logwood, becomes dyed lilac-red, or maroon, which dilute ammonia changes to violet-blue tinged with grey, and acetate of aluminium to bluish-violet.

Cochineal.—The lilac, or roseate tints due to the reactions A, B, H, K, of Table A., are very sensitive, the last being very characteristic; the only substance likely to be confounded with it is the phytolacca (Portugal berries), which is differentiated by the reaction B, of the same table.

A skein of scoured silk, mordanted with aluminium acetate soaked in the clarified wine for twenty hours is dyed of a wine violet colour, analogous to that of pure wine, on being dried at 100° C. The colour does not change, even at 100° C., by cupric acetate (exclusion of fuchsine); but if the skein be dipped into a dilute solution of zinc chloride, heated to 100° C., and then wetted with sodium carbonate, washed with water and dried, the colour becomes fine purple, whereas with pure wine the tint would remain sombre grey-lilac.

Cochineal may be discovered by the spectroscope if present in large quantity, but if it amounts to only about 12 per cent. of the total coloration, it cannot be so detected. It rapidly separates from wines, being precipitated in the lees.

Fuchsine.—This should be sought for in all wines found to be adulterated with other substances. The reaction J of Table B., p. 463, is very sensitive.

Great care must be taken to avoid loss of rosaniline from imperfect decomposition of its salts in solution; moreover, arsenic should always be sought for where the wine is found to contain any aniline. Fuchsine rapidly separates from the wines to which it has been added. A skein of silk becomes dyed rose by soaking in a wine adulterated with fuchsine, and its colour passes to yellow on treatment with hydrochloric acid, but to bright red if the wine was pure. The dyed skein treated with dilute cupric acetate, and dried at 100° C., becomes fine deep rose-violet if fuchsine is present, and of a lilac tinged with ash-grey if the wine is pure. This reaction is very sensitive.

Phytolacca.—(Portugal berries). The rose or lilac colorations of the reactions A, G, and especially C of Table A., are very sensitive.

Hollyhock—(*Althaea rosea*), much used. This substance imparts a peculiar flavour, which in a few months becomes actually disagreeable, while the colouring-matter itself rapidly precipitates.

Beetroot.—This is generally employed only to mask other adulterants. The lilac tint of reaction C of Table A., if the beetroot is fresh, and the yellowish colours due to alkalies (reactions D, E and F of Table A.) are very sensitive, even with old decoctions.

Black Elder, Dwarf Elder.—The dwarf elder imparts a faint turpentine odour to the wines. The berries of both varieties are particularly used to communicate a special colour and flavour to port wine. The *teinte de Fismes* (p. 457), largely used at Fismes, Paris, and elsewhere, is made by digesting 250 to 500 parts of elder-berries, and 30 to 60 parts of alum, with 800 to 600 parts of water, and then submitting the mixture to pressure. M. Maumené reports having discovered as much as 4 to 7 grms. of alum per litre in wines adulterated with this substance. Sometimes (though rarely) the alum is replaced by tartaric acid. Wines adulterated with either yield a violet-blue lake (reaction H, Table A). By comparison with pure wine the difference is very marked.

A piece of flannel, or skein of silk, mordanted with aluminium acetate, heated for some time in the suspected wine, then washed, and immersed in water made faintly alkaline with ammonia, becomes green if the wine is pure, but dark brown if black elder is present. Probably the same reaction occurs with dwarf elder.

Privet.—This is seldom used. The general reactions, particularly A and G of Table A., may be referred to (see also footnote, p. 462).

Kino.—Said to be coming into use. It is precipitated by gelatin, and gives no definite absorption bands. According to Etti¹ kino red is the anhydride of kinone, and has the formula $C_{25}H_{22}O_{11}$.

Indigo.—The reactions A (b) and B (b) of Table B., p. 462, are so sensitive that they are alone sufficient to characterise indigo. Wool or silk mordanted with aluminium acetate, heated with 20 to 40 c.c. of the suspected wine nearly to dryness, washed, and then dipped into very dilute ammonia, become dirty green if the wine be pure, but blue if indigo be present. Indigo being often used to mask the too bright colours of cochineal and fuchsine, these should always be sought for after the removal of the indigo by clarification with albumen. Indigo very rapidly separates from wines, and it may frequently be found in the lees, even when the wine itself gives no indication of its presence. Except in such cases as indigo and cochineal, it is only upon a series of concordant reactions that the presence of an artificial colouring-matter should be affirmed.

¹ *Ber. d. d. chem. Gesells.*, xi., 1883, 1879.

§ 332. *Mineral Substances, or Ash.*—The ash of a great many wines, and especially of sherries, imported into this country, consists nearly entirely of sulphates.¹ This is due either to sulphuring or plastering. It is found absolutely necessary to charge many wines slightly with sulphurous acid, some of which becomes sulphuric acid; and in such a case the chlorides and carbonic acids are diminished in the ash, and the sulphuric increased, but the total weight of the ash itself is not materially increased. On the other hand, plastering (by which is meant the addition of plaster of Paris to the grapes before they are crushed) has the effect, by its reaction on cream of tartar, of producing a soluble sulphate of potassium, which may very materially increase the ash of the wine.²

Under absolutely normal conditions, the ash consists of carbonate, sulphate, phosphate, chloride of potassium, chloride of sodium, phosphate and carbonate of calcium, with very small quantities of magnesia, iron, silica, and frequently lithium and manganese.

The ash from a litre of wine examined by Boussingault contained—

	Grms.
Potash, ³	0·842
Lime,	0·092
Magnesia,	0·172
Phosphoric Acid,	0·412
Sulphuric Acid,	0·096
Chlorine,	a trace
Carbonic Acid,	0·250
Sand and Silica,	0·006
	1·870

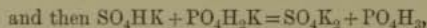
With regard to the analysis of the ash, etc., see p. 93, *et seq.*

¹ The sulphuric acid in sherries ranges from 1·5 to 8 grms. per litre (equal to from 19·0 to 93·8 grains per bottle of $\frac{1}{2}$ gallon).

² A plastered wine contains more potash than one not plastered, for in the latter there is a deposition and separation of the hydro-potassic tartrate, but in plastered wines from double decomposition calcium tartrate is formed and deposited, whilst potassic sulphate passes into solution. According to Hilger, a plastered wine always contains more than '06 per cent. SO_3 , and shows a notable increase of the ash constituents. The general view of the reaction which occurs on the addition of calcic sulphate is that some tartaric acid is also set free according to the following equation:—



This free acid again acts on the potassic sulphate, forming SO_4HK and $\text{C}_4\text{H}_5\text{KO}_6$. R. Kayser, on the other hand, considers that free phosphoric acid is formed— $\text{SO}_4\text{Ca} + \text{C}_4\text{H}_5\text{KO}_6 = \text{SO}_4\text{HK} + \text{C}_4\text{H}_4\text{CaO}_6$,



and that it is this free phosphoric acid which gives the lively tint to red wines. Plastering clears a wine rapidly, because the calcic tartrate quickly separates. The main chemical changes, therefore, which can be traced are, briefly, a decrease in the tartaric acid and an increase in potash and sulphates. The standard in use in the Paris Municipal Laboratory is 2 grms. per litre of potassic sulphate; should a wine contain more than this quantity, it is considered plastered. The maximum amount of potassic sulphate found by M. Marty in genuine wine was '6 gm. per litre. A preliminary examination of the wine is effected as follows:—A solution of 5·608 grms. of baric chloride and 100 c.c. of HCl is made up to a litre with water. Two tubes are charged, each with 20 c.c. of wine, and to the one is added 5 c.c. and to the other 10 c.c. of the barium of chloride solution. If, after the precipitate has subsided, the clear liquid from the 5 c.c. tube gives no precipitate, the wine is not plastered; or, if it gives a precipitate, whilst the second tube gives no precipitate, the plastering is beneath the standard; but if the second tube gives a precipitate, the wine is plastered, and the usual methods of estimation must be adopted.

³ The rule is that nearly half the ash of a natural wine consists of K_2O in combination.

§ 333. *Detection of Fluoborates and Fluosilicates.*—Alkaline fluoborates and sometimes alkaline fluosilicates are used as antiseptics in the manufacture of wine. Their detection is as follows:—100 c.c. of wine are treated with an excess of calcium hydrate and evaporated to dryness and ignited. Should fluoborate have been used the borate of lime is soluble in acetic acid, while both fluoride and silicate of lime are insoluble in acetic acid. Therefore, after ignition the ash is treated with acetic acid and filtered. On evaporating the acetic acid solution to dryness the residue is tested for boric acid as described on p. 234. Silicates are determined in the insoluble portion of the ash remaining on the filter, and fluorides are detected by heating the ash so as to render it anhydrous, and, after mixing with a little sand, transferring it to a test-tube, then adding sufficient strong sulphuric acid to form a paste and closing the mouth of the test-tube with a cork carrying a small U-tube, a single drop of water having been put in the bend. On now heating the test-tube, fluoride of silicon is evolved; but immediately decomposes on passing through the drop of water; hence there is a characteristic deposit of gelatinous silica in the U-tube.

If, therefore, by these processes silicon and fluorine are detected, the amount of silica being in excess of what is usual in the ash, a silicofluoride has been added; or, if fluorine and boric acid have been found, this denotes the presence of a fluoborate.





PART VII.

VINEGAR.

§ 334. *Constituents of Commercial Vinegar.*—Vinegar is a liquid resulting from the acetous fermentation of a vegetable infusion or decoction; it contains acetic acid, acetic ether, alcohol, sugar, gum, extractive matter, alkaline acetates, and tartrates, and a variable amount of salts (depending on the substances from which it has been produced).

Varieties of Vinegar.—The chief varieties of vinegar are as follows:—

(1.) *Malt-Vinegar.*—The great majority of commercial vinegars in this country are derived from the acetous fermentation of a wort, made from mixtures of malt and barley. Malt-vinegar is of a decided brown colour, in specific gravity varying from 1·017 to 1·019; it is of various degrees of strength, the manufacturers distinguishing different kinds as Nos. 18, 20, 22, and 24 respectively, the last being the strongest, and containing from 4·6 to 6·6 per cent. of acetic acid.

Malt vinegars contain from ·095 to ·12 per cent. of nitrogen. Vinegar made from sugar contains, according to Allen, about ·016 per cent. of nitrogen.

Malt vinegars contain phosphates in quantities varying from ·057 to ·093 per cent. calculated as phosphoric acid. They should only contain sulphates derived from the malt and water used in their manufacture; this should not amount to more than about ·01 to ·04 per cent. calculated as sulphuric acid. Malt vinegar contains no acid potassium tartrate.

(2.) *Wine-Vinegar* is the chief vinegar in Continental commerce. It is prepared from grape-juice and inferior new wines; that made from white wine is most esteemed. The wine-vinegars vary in colour from pale yellow to red; they have nearly always an alcoholic odour, and contain some alcohol; specific gravity from 1·014 to 1·022. A litre of Orleans vinegar (according to Chevallier's¹ analyses of actual samples) saturates from 6 to 7 grms. of dry carbonate of soda. The extract from pure wine-vinegar varies from 1·38 to 3·2 per cent., the average being 1·93 per cent., and always contains from ·25 to ·5 grm. of acid potassium tartrate (see Table LIV.).

Small quantities of reducing sugar (from ·007 to ·46), as well as some of the wine ethers, are usually present. The relation between the extract and acid does not, as a rule, exceed 3·5 per cent.

Cider Vinegar has a characteristic taste and odour of apples. It contains an average extract of 2·8 per cent., and from 3·2 to 7·6 per cent. of acid and ·31 to ·51 per cent. of ash.

¹ *Journ. d'Hyg.*, 1877, No. 45.

The solids consist of glycerol, albuminous matter, gums, malic, and other organic acids. It gives no optical rotation, and has no reducing power after clarification with lead acetate. The ash contains a large amount of potassium, but no sodium. The phosphates in the ash are high (42.1 to 17.8 per cent. of the ash), and differ from those contained in other vinegars in that at least $\frac{2}{3}$ of them are soluble in water. *Crab-Vinegar* is made from the crab apple, and is common in Wales.

Spirit Vinegar is made from alcohol, and differs from the others in having a small extract and only a trace of ash which has a very slight alkalinity. It contains only a trace of phosphates.

Grain and Sugar Vinegars.—These are made from various grains, such as maize and rice. They are similar to malt vinegars, but contain less nitrogen. They often contain much sulphate. Dextrin is usually present, and may be precipitated by alcohol.

Various Artificial Vinegars, such as diluted acetic and pyroligneous acids, coloured to resemble the dearer kinds of vinegar.

TABLE LIV.—THE COMPOSITION OF VARIOUS KINDS OF VINEGAR.

	Specific Gravity.	Extract.	Acetic Acid.	Calculated Original Solids. Acetic Acid \times 1.5 + Extract.	Relation between Acid and Extract. $\frac{a}{b}$	Ash.
Wine Vinegar—		p.c.	p.c.	p.c.		
Max.,	1.0213	3.19	7.58	14.26	2.3	0.68
Min.,	1.0129	1.38	4.44	8.04	3.2	0.16
Mean,	1.0175	1.93	6.33	11.42	3.2	0.32
Spirit Vinegar—						
Max.,	1.013	0.57	7.98	12.54	13.8	0.08
Min.,	1.008	0.16	4.98	7.63	30.0	Trace.
Mean,	1.0082	0.35	6.34	9.86	18.1	0.04
Date Vinegar—						
Max.,	1.0195	2.68	6.60	12.58	2.4	0.47
Min.,	1.0170	2.29	6.30	11.74	2.7	0.40
Mean,	1.0185	2.40	6.44	12.10	2.6	0.44
Malt Vinegar,	1.019	2.93	5.78	11.60	2.9	0.44
Malt Vinegar, which has also been made from sugar, }	1.015	2.01	5.52	10.24	2.8	0.34
Rice Vinegar,	1.017	2.53	5.64	10.99	2.2	0.34
Sugar Vinegar,	1.010	1.64	4.21	10.84	2.6	0.27
Vinegar certified to contain 70 p.c. of pyroligneous acid, }	1.007	0.21	4.70	7.26	22.2	0.04

The above table contains results reduced and collated from Sanglé Ferrière, Allen, Hehner, and others.

§ 335. *Adulterations*.—The adulterations of vinegar are—

(1.) Mineral acids, especially sulphuric, more rarely hydrochloric, and still more rarely nitric acids.

(2.) Metallic adulterations, or rather impurities; such as arsenic¹

¹ "The observations of M. Deschamps induced us to analyse a vinegar sold by a certain Sieur C. . . . The presence of arsenic in this vinegar was ascertained, and the Sieur C. was compelled to confess that the vinegar had been mixed with wood-vinegar. On resorting to the person who furnished the latter product, the whole of the wood-

(derived from sulphuric acid), copper,¹ lead, zinc, and tin, from the solvent action of the acid on any metallic surfaces with which it may have come in contact.

(3.) Pyroligneous acid.

(4.) Various organic substances, such as colouring agents, and capsicum.

§ 336. *Analysis of Vinegar.*—(1.) *Water.*—Good vinegar should contain from 5 to 7 per cent. of acetic acid ($C_2H_4O_2$). Three per cent. of acetic acid is, however, the standard adopted in this country by the Government Laboratory for the purposes of the Sale of Food and Drugs Act.

The amount of acetic acid may be estimated with fair accuracy by diluting 6 c.c. of the sample with 50 c.c. of water and titrating with d. n. sodium hydroxide. The percentage of acetic acid is one-tenth of the number of c.c.s of alkali required for neutralisation.

The acetic acid may also be estimated by distilling 110 c.c. until 100 c.c. have been drawn over, that is, ten-elevenths. The 100 c.c. will contain 80 per cent. of the whole acetic acid present in the 110 c.c., and may be titrated; or the specific gravity of the distillate may be taken, and the strength found from the following table:—

Per cent.	Sp. gr.	Per cent.	Sp. gr.	Per cent.	Sp. gr.
1	1·001	8	1·012	15	1·022
2	1·002	9	1·013	16	1·023
3	1·004	10	1·015	17	1·024
4	1·005	11	1·016	18	1·025
5	1·007	12	1·017	19	1·026
6	1·008	13	1·018	20	1·027
7	1·010	14	1·020		

Vinegar may also be distilled in a vacuum produced by a mercury or water pump; it should be distilled into caustic soda or potash of known strength, and then titrated back. By distilling thrice to dryness, adding a little water each time, the whole of the acetic acid comes over.

It will be necessary to test the distillate for the presence of hydrochloric acid, and also to take the acidity of the vinegar without distillation, so as to control the results.

If absolutely accurate determinations are required, it is best to add an excess of carefully weighed pure carbonate of lime to a known weight of the vinegar; the liquid is boiled, filtered, and the residual carbonate of lime filtered off, dissolved in slight excess of normal hydrochloric acid, and titrated back with caustic soda and cochineal solution. From the amount of carbonate thus found to have been unacted on by the vinegar, the total acidity is calculated. Carbonate of barium may with advantage replace the lime carbonate.

Total Solids or Extract.—5 c.c. of the vinegar should be evaporated to a constant weight in a platinum dish on the water-bath.

Apple pulp exhausted of malic acid is said to be added to vinegars in order to increase the solid residue. According to Dr. Frear² any residue containing more than 50 per cent. of reducing sugar in its total solids contains unfermented sugar material. If the sample is supposed to be

vinegar in his possession was found arsenical, and seized, in order to be employed only for industrial use."—"Le Vinaigre," Chevallier, *Journ. d'Hyg.*, No. 46, June, 1877.

¹ Seven out of twelve samples of vinegar sold in Paris, and analysed by Alfred Riche, contained copper varying from 5 to 15 mgrms. per litre. *Journ. Pharm. Chim.* [4], xxvj., 23-28.

² *Report of the Pennsylvania Department of Agriculture*, 1898, p. 138.

true wine vinegar, the solid residue should be tested for acid potassium tartrate.

Ash.—The residue from 10 c.c. should be burned at as low a temperature as possible. It should be tested for chlorides and sulphates; these, if excessive, should be estimated. The total phosphates and the soluble phosphates should be estimated.

If the ash of vinegar is less than 10 per cent. of the total solids, unfermented sugar material is probably present; while if it be less than 6 per cent. of the solids, it cannot be a genuine cider vinegar.¹

Nitrogen.—The amount of nitrogen should be estimated, by the Kjeldahl-Gunning method, in 50 c.c. of the sample.

Mineral Acids.—By the Vinegar Act of 1818, sulphuric acid was allowed to be added to vinegar, "in the proportion not exceeding one-thousandth part thereof by weight." This Act was, however, repealed in 1861, and it is now illegal to add any mineral acid to vinegar. Good vinegar should contain no mineral salts other than those contained in the brewing materials. There is, however, at present no legal decision preventing manufacturers from using water heavily charged with sulphates, or from adding calcium sulphate, or from introducing calcium sulphate by 'killing' the acid with lime in the manufacture of vinegar from grain.

Free Hydrochloric Acid is detected by the distillation already described, and the testing of the distillate with nitrate of silver.

Free Nitric Acid may (in the absence of other reducing agents) be detected by the rapid decoloration of a solution of indigo carmine added to the boiling vinegar, or by the diphenylamine test (see article on Water).

Free Sulphuric Acid cannot be detected by the usual chloride of barium test, for it fails to distinguish between free and combined sulphuric acid. The charring effect of the acid on paper, on sugar, or its action on starch (formerly taken as the basis of the older tests), is now replaced by more scientific methods, and need not be described here.

One of the most speedy tests for the presence of mineral acids is that proposed by A. Hilger²:—Two or three drops of a solution of methyl aniline violet (0.1 : 100) are added to 25 c.c. of vinegar; if pure, no colour is produced; but if .2 per cent. of any mineral acid is present, the colour is blue; or if .5 per cent., blue-green; and if 1 per cent., green.

A. Ashby's test is also a reliable and rapid one:—A drop of logwood extract in water (0.5 gramme to 100 c.c.) is dried on a porcelain plate, a drop of vinegar added, and again dried. Pure vinegar gives a yellow residue, but if free mineral acid be present the residue is red.

Another useful test is that of M. Strohl;³ it is based on the well-known fact, that oxalate of lime is insoluble in acetic, but soluble in mineral acids. The solutions requisite are—a solution of calcic chloride (15.1 grms. to the litre) and a solution of crystallised ammonic oxalate (28.4 grms. to the litre); $\frac{1}{2}$ c.c. of each of these liquids is added to 50 c.c. of the vinegar under examination, and if the turbidity which is at first produced does not disappear, the liquid contains less than—

1.70	gram.	per cent.	sulphuric acid	(specific gravity 1.843)	per litre.
2.85	"	"	hydrochloric acid	(" " 1.174)	"
4.40	"	"	nitric acid	(" " 1.174)	"

The test, without claim to great accuracy, is extremely useful; for if any

¹ Report of the Pennsylvania Department of Agriculture, 1898, p. 138.

² Archiv der Pharmacie, 1876, 193.

³ Arch. Pharm. [5], 4, 342-346.

suspicious indication be observed, the vinegar may be then submitted to a more elaborate examination for free acids.

As speedy as any of the foregoing, and at once more scientific and accurate, is the process introduced by Hehner. Its principle is based upon the fact that vinegar always contains potash and soda salts of the organic acid; hence, it is obvious that sulphuric or hydrochloric acids, if added in small quantity, merely decompose an equivalent quantity of acetate or tartrate, as the case may be, and as *free acids* immediately disappear; but if added in excess of the amount of acetates and tartrates, the excess remains as free acid. It thus follows, that if any undecomposed acetate or tartrate exists in the vinegar, it is impossible for a free mineral acid to be present; and since the acetates and tartrates are decomposed by ignition into carbonates, the readiest way to ascertain their existence is to examine the ash of the vinegar for carbonates. If that ash is *neutral*, free mineral acid is probably present; if *alkaline*, no free acid can be present, although, of course, a small quantity may originally have been added.

The qualitative test devised by Hehner is also made quantitative. If an accurately-estimated volume of d. n. soda solution is added to a known quantity of the vinegar, so as to neutralise slightly in excess the total amount of free mineral acid present, on ignition the alkalinity of the ash gives the measure of the quantity of free sulphuric or hydrochloric acid. The exact details of this operation, as practised by Hehner, are as follows:—50 c.c. of the vinegar are mixed with 25 c.c. of d. n. soda; the liquid is evaporated on a water-bath in a platinum basin, the residue dried at about 110° C., and carefully incinerated at the lowest possible temperature—the ash need not be burned white. 25 c.c. of a d. n. sulphuric acid solution are now added to the ash, the liquid heated to expel free CO_2 , and filtered. The filter is washed with hot water, litmus added,¹ and the acidity ascertained by d. n. soda. The volume of soda necessary for neutralisation directly gives the proportion of free mineral acid present in the vinegar, 100 c.c. of d. n. corresponding to 49 gm. of H_2SO_4 . If the amount of alkali originally added should have been insufficient, it is necessary to recommence the experiment. For this reason Allen and Bodmer made some experiments in which the preceding manipulation was modified by neutralising the *whole* of the acid, organic and inorganic, by soda solution. The results were satisfactory, but great care must be taken to titrate accurately.

Another very satisfactory way of separating and identifying the free mineral acids in vinegar is the following:—Saturate a known quantity with cinchonine, evaporate to dryness, take up the quinine salts with spirit, recover the spirit by distillation, dissolve the cinchonine salt in water, and precipitate by ammonia. The aqueous liquid will now contain the acetate of ammonia, together with the sulphate, chloride or nitrate; if any one, or all three, of the free acids were present, the acids may be determined in the usual way.

A method of separating free sulphuric acid from sulphates is to evaporate the vinegar to a syrup, precipitate the sulphates by alcohol, filter, wash the precipitated salts with alcohol, and determine the free sulphuric acid in the alcoholic solution. Provided sufficient alcohol be added, the separation of free from combined sulphuric acid is exact.

¹ Instead of litmus, cochineal may be used; the latter is unaffected by CO_2 , and therefore preferable.

Another method, the principle of which was proposed by Thresh, and which has been improved upon by W. C. Young, is to add to a known measure of vinegar an excess of BaCl_2 ; the chlorine in a portion of the liquid is now determined with great care, the rest is evaporated, ignited, and the chlorine of the ash determined. The difference represents the free mineral acid in terms of chlorine. The presence of free tartaric or citric acids quite invalidates the accuracy of the process, but, with these exceptions, it is generally applicable.

(3.) *Colouring Matter*.—Vinegar is often coloured with caramel; sometimes simply acetic acid thus coloured is sold as 'vinegar.' The vinegar should be well shaken up with a little Fuller's earth and filtered; if caramel is present the filtrate will be very much paler than the original liquid. The estimation and detection of caramel has been worked out by C. Amthor¹ as follows:—10 c.c. of the liquid are put into a flask with 30 to 50 c.c. of paraldehyde, and absolute alcohol added until the liquids have mixed thoroughly. The flask is corked and allowed to remain undisturbed for twenty-four hours; by that time the caramel will have precipitated. To still further identify it, the liquid is decanted, the precipitate washed with a little alcohol, dissolved in hot water, and reduced by evaporation until it measures a cubic centimetre. On now treating the solution with phenylhydrazine, an insoluble compound is obtained. This is distinguished from the osazones of sugar by its amorphous appearance.

Estimations of caramel are best made by imitating the colour of the paraldehyde precipitate; for this purpose the precipitate is dissolved in water, made up to 100, and a solution of known caramel content used as a comparison liquid. Possibly the phenylhydrazine compound could be weighed, but the limits of accuracy in this direction have not yet been defined, and the formula of caramel is not accurately known. A. Sabaneef and J. Antushevitch² believe, from cryoscopic researches, that its formula is $\text{C}_{125}\text{H}_{188}\text{O}_{80}$.

The colouring of vinegar with coal tar dyes does not seem to be practised at the present time, but it is probable that these dyes will be put to this purpose, and in special cases they should be sought for by the ordinary methods.

(4) *Metallic Adulterations*.—Metals of the first group in vinegar are detected by saturating the liquid with hydric sulphide (if the sample is not deeply coloured) or by specially testing the ash for copper, zinc, tin, and lead, by the usual methods. Arsenic may be tested for by Reinsch's, or Marsh's test (see pp. 436–441).

Zinc may be presumed to be absent if the nearly neutralised vinegar gives no precipitate with hydric sulphide.

From the foregoing it will be seen that it is possible to distinguish between the different vinegars by a consideration of the analytical figures obtained from them. The watering of a genuine vinegar will be indicated by the low solids and low acetic acid. Diluted acetic acid coloured with caramel or with strongly scorched malt, or diluted pyroligneous acid, will be found to have low solids and ash, and only a trace of phosphates, while the acetic acid is usually high. Pyroligneous acid may sometimes contain tarry products. It may be recognised, according to the French chemists, by the larger amount of furfural than is the case with the other vinegars. A vinegar made from glucose, or from rice converted into glucose by

¹ *Zeit. f. anal. Chemie*, xxiv., 30–33.

² *J. Russ. Chem. Soc.*, xxv., 23–31.

inverting with sulphuric acid, may show traces of its origin in excessive sulphates. Dextrin may often be precipitated and identified.

§ 337. *Interpretation of the Results of Vinegar Analyses.*—The chemists of the Municipal Laboratory, Paris, interpret the results of their analyses of wine-vinegar according to the following principles. The normal weight of extract and alcohol in wine is in the proportion of 4 : 1; a tenth per cent. of the extract is lost during acetification; on the other hand, 130 grms., theoretically, of acetic acid is produced from 100 grms. of alcohol; in practice this must be diminished 15 per cent. The following table gives a calculation on the above basis:—

Alcoholic Strength. Per cent. (vol.).	Acetic Acid. Per cent.	Extract of Vinegar. Per cent.
6	5·315	1·08
7	6·211	1·26
8	7·105	1·44
9	8·00	1·62
10	8·895	1·80
11	9·801	1·98
12	10·71	2·16

The relation existing between the acetic acid and the extract is equal to 4·9; if a vinegar has a relation higher than this, allowing at the most a tenth (that is, really 5·0), an addition of alcohol is indicated, whether an actual addition or whether from the use of a fortified wine. The table also indicates the maximum and minimum quantities of acid which can be furnished by wines of from 6 to 12 per cent. of alcohol; and should the proportion of acetic acid fall below 5·3 per cent., watering may be concluded, especially if the potassic tartrate, ash, and extract are diminished in the same proportions. The French standard for vinegar is, therefore, near the standard of our old pharmacopœia (5·41 per cent.). Vinegar is now omitted from the British Pharmacopœia.

It has been proposed by Hehner to calculate the various constituents on the original solids of the vinegar. As 60 parts of acetic acid are theoretically produced from 90 of glucose, the acetic acid is multiplied by 1·5, as representing theoretically the sugar from which the acetic acid was derived. To this figure is added the total extractive matter still contained in the wort-vinegar, and the number thus obtained represents the 'original solids' of the wort. Examples of this calculation are shown in Table LIV. This number obtained, the ash, acetic acid, and albuminoids may be calculated out in terms of the original solids, which is doubtless a better guide to composition than the usual way of stating them. For example, calculating a malt-, a 'spirit-', and a 'sugar'-vinegar in per cent. of the total solids, the results are as follows:—

	Malt- Vinegar.	Spirit- Vinegar.	Sugar- Vinegar.
Acetic acid,	49·8	63·2	50·0
Albuminoids,	5·9	0·8	1·15
Phosphoric acid,	0·8	0·0	0·18
Ash,	3·8	0·4	3·35

LEMON JUICE AND LIME JUICE.

§ 338. Lemon juice is the expressed juice of the *Citrus limonum*, and lime juice that of the *Citrus acida* and *Citrus limetta*. The Board of Trade standard for lemon juice is a density of 1·030 [when dealcoholised], and an acidity equivalent to 30 grains per ounce of citric acid. The British Pharmacopœia directs that lemon juice should have a specific gravity of 1·039, and should contain 32·5 grains of citric acid per ounce.

Lemon, lime, and bergamot juice are all similar in their composition, containing citric acid as the predominant free acid, and small quantities of acetic, formic, and other organic acids, together with albumen, sugar, mucilage, and extractive matters. The mineral matter is very small, and contains 54 per cent. of its weight of potash and 15 per cent. of phosphoric acid.

The juice is expressed in England, and also in Sicily, where the method of preparation is to mix one ounce of brandy with ten ounces of the juice, and over the surface of the liquid to pour a layer of olive oil. This crude process of preservation is effectual, but is now being supplanted by more modern methods.

The following table shows the specific gravity, free acid, and combined organic acid of the citric commercial juices, the acid being expressed in terms of crystallised citric acid [Allen]:—

	Specific Gravity.	Free Acid, Ozs. per gallon.	Combined Organic Acid. Ozs. per gallon.
Lemon Juice—			
Raw Sicilian,	6 to 9	0·85
„ English,	1·04 to 1·05	11 to 13	0·3
Concentrated,	1·20 to 1·25	57 to 72	6 to 8
Bergamot Juice—			
Concentrated,	1·22 to 1·25	47 to 55	7 to 8
Lime Juice—			
Raw,	1·035 to 1·040	10·6 to 13·5	0·4 to 0·7
Concentrated,	1·28 to 1·38	82 to 112	8·6

A. Bornträger gives the following as the percentage composition of lemon juice:—

	Ripe Fruit.	Unripe Fruit.
Citric acid,	7·25	7·70
Reducing sugar,	·75	·21
Sucrose,	·19	·78
Ash,	·39	·49
Total solids,	8·87	9·30

§ 339. *Adulterations and Analysis of Citric Juices.*—Lime juice has been rather extensively adulterated, and at the present time it is by no means uncommon to meet with a wholly fictitious article under this name.

The chief adulterants of citric juices are:—Tartaric acid, mineral acids, glucose, cane and invert sugar, coal tar dyes, and preservatives. *Tartaric acid* may be estimated as follows¹:—20 grammes of the juice are mixed with 5 grammes of KCl, the solution neutralised with potassium hydroxide,

¹ *Select Methods of Food Analysis*, 1901, p. 323. Leffmann and Beam.

and made up to 50 c.c. with water. 5 grammes of citric acid are added, the solution well stirred, and allowed to stand all night. The precipitated acid potassium tartrate is washed, first, with a saturated solution of the same substance, then twice with a 10 per cent. solution of potassium chloride, and titrated hot with standard alkali.

Mineral Acids.—The general principles of the detection of these acids in a free state are entirely the same as in vinegar, and the remarks with regard to the alkalinity of vinegar-ash when genuine apply equally to the ash from the citric juices. (See p. 476.) Good juice contains insignificant traces of sulphates and chlorides, so that the mere addition of silver nitrate or of barium chloride will at once show whether there has been any tampering with the liquid.

Nitric acid may be detected and estimated as described *post* in the sections on Water Analysis.

F. Scribani¹ adds to the suspected lemon juice an aqueous solution of ferrous chloride, strongly acidified with hydrochloric acid and free from ferric salt. The liquid is boiled for a few minutes, and then a little sulphocyanide is added. If nitric acid has been present, it will have oxidised the ferrous salt into a ferric salt, and a deep blood-red colour will be produced by the test.

Glucose and sugars generally will be detected and estimated by the methods already described, remembering that the juices naturally contain a small quantity of sucrose and reducing sugar.

Coal tar dyes and preservatives should be sought for by the ordinary methods.

Poisonous Metals.—The analyst should never neglect to look for these, paying special attention to the examination for lead.

§ 340. *The Analysis of Lime Juice*, with a view to ascertain its strength, is confined to the determination of the amount of citric acid and citrates.

The amount of citric acid is determined frequently for technical purposes by the aid of a special hydrometer, called 'a citrometer,' but this method is not exact enough for the purposes of the food-analyst. Nevertheless, it will always be well to take the specific gravity of the juice by a specific gravity bottle, or by any other reliable method, first boiling off any alcohol which the juice may contain, and making it up to the same bulk as before the dealcoholisation.

The amount of free acid may be estimated by means of decinormal soda. A known quantity of the juice is taken and titrated with d. n. soda, using phenol-phthalein as an indicator. About 10 c.c. of the ordinary raw juice may be taken and diluted to 50 c.c.; but the concentrated juice must be much diluted before titration.

The free acid known, the next step is to determine the amount of citrates and other organic acids combined with bases. For this purpose, the measured quantity of the juice which has already been neutralised by soda is evaporated down, charred, and the charred mass treated with a known volume of decinormal sulphuric acid, which must be sufficient to more than neutralise the carbonates.

The acid solution is filtered and neutralised by d. n. soda; this will give the necessary data from which to calculate the amount of sulphuric acid used by the carbonates produced by the action of heat on the organic acids. This amount is equivalent to the total amount of organic acid; if

¹ *Gaz. Chim. Ital.*, viii., 294.


expressed as citric acid, forty-nine parts of sulphuric are equal to seventy of $2\text{H}_3\text{C}_6\text{H}_5\text{O}_7\text{H}_2\text{O}$.

The amount of free acid already obtained is now subtracted from the total acid, the difference being that which is combined with bases.

To ascertain the amount of real citric acid present in the juice, it must be determined as citrate of lime, for it need hardly be said that the process given above does not distinguish between malates, meconates, or any other organic acids converted by heat into carbonates. To determine the real citric acid, Warrington¹ recommends the following process: from 15 to 20 c.c. of raw lime juice are exactly neutralised by d. n. soda, the whole made up to about 50 c.c., and heated to boiling.

While boiling, so much of a solution of chloride of calcium is added as is known to be rather more than equivalent to the total amount of organic acids present. The boiling must be continued for half an hour, and the precipitate collected and washed with hot water. The filtrate and washings are evaporated to a small bulk (not more than 15 c.c.), and a little ammonia added to exact neutralisation, if the liquid gives an acid reaction; a further precipitation takes place, and this second precipitate must be collected on a filter. The filters are dried and burnt up at a low heat, and their neutralising power with regard to acid is determined. For this purpose, the ash may be dissolved in standard hydrochloric acid, and titrated back; each c.c. of normal HCl neutralised is equivalent to .070 gm. of crystallised citric acid. If either oxalic or tartaric acids should be present, the results are, of course, inaccurate.

¹ *Journ. Chem. Sec.*, 1875, 934.



PART VIII.
MUSTARD, PEPPER, SPICES,
CONDIMENTS.

PART VIII.—MUSTARD, PEPPER, SPICES, CONDIMENTS.

MUSTARD.

§ 341. *Mustard* is made from the seeds, finely ground, of the *Sinapis nigra*, or black mustard, or from those of the *Sinapis alba*, or white mustard, or, again, from a mixture of both varieties. The manufacturer reduces the seeds to powder, and passes the product through a series of sieves. The portion in the first sieve is called the *dressings*; that which passes through is an impure *mustard flour*. The impure flour, on being passed through a second sieve, yields the pure flour of mustard and a second quantity of dressings. The dressings are utilised, by being submitted to pressure, for the sake of the fixed oil they contain.

Microscopical Structure of the Seed.—The white mustard seed is made up of the husk and the seed proper. The *seed proper* is simple in structure, consisting entirely of minute oil-bearing cells; their size averages $\cdot 00041$ inch in the finely powdered seed; and they look extremely like starch corpuscles, but neither polarise light nor strike a blue colour with iodine.

The complicated structure of the husk of the mustard seed, in part unravelled by Hassall, has more recently been fully investigated by the labours of Semplowski and v. Höhnels.¹ It is built up of no less than six layers (see fig. 67).

(1.) The most superficial is composed of almost quadratic thin-walled cells ($\cdot 05 - 1$ micro-millimetre in diameter) covered with a thin cuticle. The lumen is narrow, and the cells are almost entirely filled with a gelatinous substance. On the addition of water the cells swell up to several times their original volume, and the gelatinous substance escapes from some of the ruptured cells. If looked at from the surface these cells are hexagonal (see fig. 68), and the central lumen or cavity is seen surrounded by ring-like layers of the mucilage.

(2.) The second layer is formed of large, roundish, polygonal cells with thickened walls; their form is not so well seen on section as on the surface. The cells present differences in different species of mustard, the

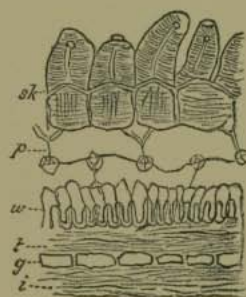


FIG. 67.—Section through the coats of the seed of *Sinapis alba*.—*sk*, The outer skin, filled with swollen mucus; *p*, the polygonal cells in section, their form better seen in fig. 68; *w*, the layer of wine-glass shaped cells; *t*, a thin-walled parenchyma; *g*, a layer of gluten-like cells; *i*, the inner seed coating. Potash preparation $\times 160$ (after MOELLER.)

¹ Moeller's *Mikroskopie*, p. 261.

white mustard has cells with two distinct layers, and with intercellular spaces; the black mustard has larger cells, with only a single homogeneous cell-wall, while the Russian mustard has likewise cells single-walled, but irregularly polyhedral in outline.

(3.) The third layer can only be seen properly in section; it consists of cells which have been likened to wine-glasses, though their outline rather suggests to the authors the soles of narrow-heeled shoes (see fig. 67). A striking peculiarity of these cells is that they vary in length or height; the surface of the seed is mapped out into small pits; in the middle of each of these small pits the cells are lowest, at the borders highest. The cells are colourless in *Sinapis alba*, but red-brown in *Sinapis nigra*.

(4.) The fourth layer is a thin-walled parenchyma filled with pigment in black and Russian mustard, but colourless in white mustard.

(5.) The fifth layer belongs morphologically rather to the substance of the seed than the husk. It is a thick-walled, single layer of polyhedral cells, containing a granulated substance.

(6.) The sixth layer is composed of an irregular parenchyma of trans-

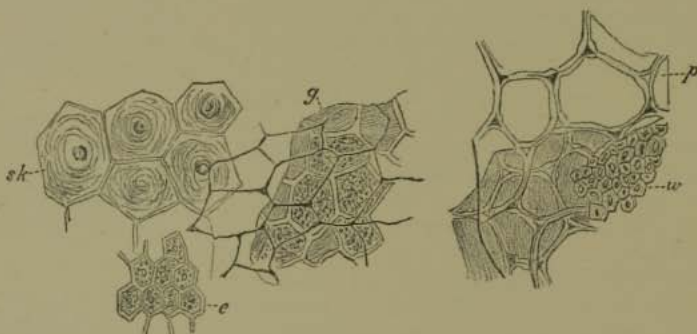


FIG. 68.—A superficial view of elements delineated in section in fig. 67; the same lettering has been followed—*e*, embryonal tissue. Potash preparation $\times 160$ (after MOELLER).

parent, colourless cells (see fig. 68), well seen from the surface, but indistinct on section (see fig. 67).

As a rule, none of the husk is found in commercial ground mustard, the main bulk being made up of the substance of the seed.

§ 342. The accompanying tables (LV., LVI.) give some careful analyses by C. H. Piesse and Lionel Stansell¹ of black and white mustard:—

§ 343. *The Chemistry of Mustard* is extremely interesting; both seeds, white and black, contain a fixed oil (from about 36 per cent.), and a sulphocyanate of sinapin and myrosin. Black mustard seeds contain, in addition to the foregoing, myronate of potash (about .5 to .6 per cent.). When the powdered black mustard seeds (or the mixed black and white) are moistened with water, the myronate of potash acts upon the myrosin, and produces *the volatile oil of mustard*. White mustard seeds, on the other hand, contain also a sulphur principle, *sinalbin*, not found in black.

Sinapin, $C_{10}H_{23}NO_5$.—Sinapin exists as a sulphocyanate, both in black and white mustard seeds, as well as in the seeds of *Turritis glabra*, L. It

¹ *Analyst*, 1880, p. 161.

was first prepared by Henry and Garot in 1825. The best process for extraction of the sulphocyanate on a small scale is (according to Von Babo) to exhaust the oil from the seeds by ether, then to treat with cold absolute alcohol, which only takes up a little of the sulphur compound, and lastly, to dissolve the sulphocyanate of sinapin out with 90 per cent. alcohol. The excess of alcohol is then separated by distillation, and the substance crystallises out, yielding about 1·1 per cent.

Sinapin itself cannot be obtained pure, but a watery solution may readily be prepared by decomposing a solution of the bisulphate with the

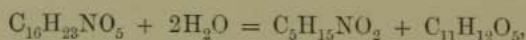
TABLE LV.

	WHITE MUSTARD.					BROWN MUSTARD.			
	Mustard, whole seeds.		Mustard farina.			Mustard, whole seeds.	Mustard farina.		
	Yorkshire.	Cambridge.	Superfine.	Fine.	Second.	Cambridge.	Superfine.	Fine.	Second.
Moisture,	9·32	8·00	6·30	5·78	6·06	8·52	4·35	4·52	5·63
Fat,	25·56	27·51	37·18	35·74	32·55	25·54	36·96	38·02	36·19
Cellulose,	10·52	8·87	3·90	4·15	9·34	9·01	3·09	2·06	3·26
Sulphur,	0·99	0·93	1·33	1·22	1·26	1·28	1·50	1·48	1·30
Nitrogen,	4·54	4·49	5·05	4·89	4·25	4·38	4·94	5·01	4·31
Albuminoids,	28·37	28·06	31·56	30·56	26·56	26·50	29·81	30·25	26·06
Myrosin and albumen,	5·24	4·58	7·32	6·67	6·11	5·214	6·46	6·78	6·14
Soluble matter,	27·38	26·29	36·31	36·60	33·90	24·22	31·14	32·78	31·41
Volatile oil,	0·06	0·08	0·03	0·04	0·03	·0473	1·437	1·500	1·381
Potassium myronate,	1·692	5·141	5·366	4·940
Ash,	4·57	4·70	4·22	4·31	4·30	4·98	5·04	4·84	4·91
Ash soluble,	0·55	0·75	0·44	0·55	0·33	1·11	1·01	0·98	0·77

TABLE LVI.—ANALYSES OF ASH OF MUSTARD SEED.

	WHITE SEEDS.		BROWN SEEDS.
	Yorkshire.	Cambridge.	Cambridge.
Potash,	21·29	18·88	21·41
Soda,	0·18	0·21	0·35
Lime,	13·46	9·34	13·57
Magnesia,	8·17	10·49	10·04
Iron oxide,	1·18	1·03	1·06
Sulphuric acid,	7·06	7·16	5·56
Chlorine,	0·11	0·12	0·15
Phosphoric acid,	32·74	35·00	37·20
Silica,	1·00	1·12	1·41
Sand,	1·82	1·95	1·38
Charcoal,	12·82	15·14	7·57
	99·83	100·44	99·70

proper quantity of baryta. After filtering away the sulphate of baryta, the filtrate is of a yellow colour and intensely alkaline reaction; it precipitates many metals from their solution, but on evaporation its colour changes through green and red into brown, and at last it leaves behind an uncrystallisable brown residue. On boiling a solution of sinapin with the alkalies or alkaline earths, the sinapin splits up into choline or sinkalin and sinapic acid,

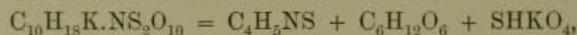


and similar treatment of the sulphocyanate of sinapin produces the same decomposition. To sulphocyanate of sinapin is ascribed the formula $C_{16}H_{23}NO_5CNHS$. It forms colourless, transparent, truncated prisms, in warty or starlike groups, without odour, but of a bitter taste, of neutral reaction, melting at 130° C. to a yellow fluid, solidifying again in an amorphous mass. The sulphocyanate is readily soluble in water; but ether, turpentine, and bisulphide of carbon do not dissolve it. If to a hot solution in alcohol concentrated sulphuric acid be added, bisulphate of sinapin, $C_{16}H_{23}NO_5, SH_2O_4 + 2OH_2$, separates on cooling in rectangular plates. From this salt the neutral sulphate may be obtained by solution in water, and precipitating half the sulphuric acid by baryta.

Sinalbin, $C_{30}H_{44}N_2S_2O_{16}$, a substance which exists only in white mustard, and may be supposed to take the place of myronate of potash. It splits up into sugar, bisulphate of sinapin, and sulphocyanide of acrinyl, C_8H_7NSO . The last, on treatment with alkalies, yields ammonia and the salt of an acid melting at 136° C., to which the formula $C_8H_8O_3$ is ascribed.

Myrosin, a substance analogous to emulsin, has not yet been obtained albumen- or lime-free; its solution froths on being shaken; it is coagulated by warming to 60° C., as well as by acids and alcohol.

Myronate of Potash, $C_{10}H_{18}KNS_2O_{10}$, crystallises out of spirit, in needles; out of water, in rhombic prisms. It is destitute of water of crystallisation, is of neutral reaction, and has no odour, but is of a bitter taste. It is easily soluble in water, with difficulty in diluted spirit, and scarcely at all in absolute alcohol, whilst it is quite insoluble in ether, chloroform, and benzole. If the concentrated aqueous solution of the salt be digested with tartaric acid and absolute alcohol, the tartrate of potash separated, and the filtered fluid evaporated with carbonate of baryta, the filtrate from the latter will yield easily soluble crystals of myronate of baryta ($C_{10}H_{18}BaNS_2O_{10}$), which soon become opaque on exposure to the air; if heated, it develops ethereal oil of mustard, leaving behind sulphate of baryta. A solution of myronate of potash gives with zinc and hydrochloric acid sulphuretted hydrogen, and then contains a salt of ammonia, sugar, and half of the sulphur as sulphuric acid. Boiling hydrochloric acid decomposes similarly. Concentrated potash-lye digested on the dry salt, and heated, develops volatile oil of mustard, cyanide of allyl, and ammonia. If to a watery solution of myronate of potash, myrosin is added, volatile oil of mustard, sugar, and bisulphate of potash are formed; thus,



the same reaction takes place if the freshly-prepared watery extract of the white or black mustard seeds be added.¹

¹ In the analysis of mustard, A. R. Leeds and E. Everhart (*Bied. Centr.*, 1884, 140) have proposed to dissolve out potassium myronate by treatment with aqueous alcohol,

§ 344. *The Fixed Oil of both Black and White Mustard appears to be identical.*—It is a yellow, non-drying oil of 0.915 to 0.920 specific gravity at 15° C., solidifying at from -12° C. to -16° C., saponification value 170 to 175, iodine value 92 to 106, and of a mild taste. It contains the glycerides of erucic acid, of stearic acid, and of oleic acid, which last Darby considers different from ordinary oleic acid.

Erucic Acid, $C_{22}H_{42}O_2$, was discovered by Darby in the fatty oil of the seeds of the white and black mustard in 1849, and the same acid has also been found in rape oil. It is easily obtained by saponifying the oil with litharge, treating the soap with ether, which dissolves out the erucate of lead, and decomposing the salt with hydrochloric acid. The erucic acid in solution is filtered from the chloride of lead, the filtrate evaporated in the water-bath, and the residue recrystallised from ether. Erucic acid forms slender, long, white, glittering needles, without odour or taste, melting at from 33° to 34° C., and coagulating again at 33° C.; it is insoluble in water, but dissolves easily in alcohol and ether. The acid, exposed to the air, gradually becomes coloured and rancid. If to water saturated with erucic acid bromine be added gradually, a crystalline compound can be obtained, $C_{22}H_{42}Br_2O_2$, which crystallises out of alcohol in small, white, warty masses, melting at 42° to 43° C. Again, if the solution be decomposed with hydrochloric acid and sodium amalgam, it can again be changed back to erucic acid. Erucic acid forms definite salts, of the formula $HC_{22}H_{41}O_2$.

The Volatile Oil of Mustard, C_4H_5NS , is mixed with cyanide of allyl, which may be separated by fractional distillation.¹ Ordinary distillation of black mustard seeds yields it in the proportion of 0.5 to 0.7 per cent. It is colourless or slightly yellow; has a boiling point of 148° C., and specific gravity 1.015 to 1.025; is somewhat soluble in water, dissolving easily in alcohol, ether, and petroleum ether. According to Hager (*Pharm. Centralb.*, x., 65), the commercial oil is much adulterated; he enumerates as fraudulent additions, alcohol, bisulphide of carbon, oil of gilliflowers, and castor-oil. The volatile oil of mustard prevents the coagulation of serum-albumen, as well as alcoholic fermentation. According to Mitscherlich, this is the most deadly of all the ethereal oils, 4 grms. killing a kitten in two hours, 15 grms. in a quarter of an hour. The post-mortem appearances were those of acute gastroenteritis, and the smell of the oil pervaded the blood, urine, and lungs. It has been used in medicine, chiefly externally, for its powerful rubefacient properties, blistering the skin when applied to it.

§ 345. *Adulterations.*—The adulteration most commonly met with is a dilution of ground mustard with wheat flour, and coloured by either turmeric or Martin's yellow dinitro-naphthol. Other substances usually enumerated as having been fraudulently mixed with mustard are—cayenne

after the oil has been removed by dry ether. The solution is evaporated, dried at 105°, weighed, and ignited, and the myronate is calculated from the potassium sulphate of the ash. Myrosin and cellulose are not dissolved by dilute alcohol; these are then treated with a half per cent. soda solution; the undissolved cellulose is weighed and ignited, whilst the solution of myrosin is neutralised and precipitated by Ritthausen's solution of copper sulphate.

¹ V. Dircks (*Landw. Versuchs-Stat.*) estimates the volatile oil as follows:—The powdered substance is mixed with ten times its weight of water, and allowed to stand for nine hours at 50° C.; it is then distilled into alkaline permanganate, and thus the sulphur is oxidised into sulphate. This sulphate is precipitated in the usual way by baric chloride.

pepper, ginger, gamboge, potato starch, pea flour, radish and rape seed, linseed meal, yellow ochre, chromate of potash, plaster of Paris, and clay, besides the ground seeds of the *Sinapis arvensis*.

A careful microscopical examination by both ordinary and polarised light will detect most organic adulterations. If on the addition of iodine to an infusion of the mustard in hot water, no blue colour is produced, it is certain that neither wheat nor any other starch is present.

The chemical examination of mustard, for the purposes of the food-analyst, mainly resolves itself into—

1. Testing for turmeric, naphthalene yellow, and other coal tar dyes.
2. Estimation of the total sulphur.
3. Estimation of the fat or oil.
4. Estimation of the ash.
5. Testing for gamboge.

1. *Testing for Turmeric and Naphthalene Yellow.*—The detection of turmeric by its microscopical appearance is usually satisfactory; there are, however, some good chemical tests.

A. Extract the mustard with two or three times its volume of methylic alcohol, filter, and evaporate to dryness. If turmeric be present, the addition of hydrochloric acid to the extract will produce a red-orange colour, turned by excess of an alkali to green and blue; or the extract may be dissolved in the least possible quantity of methyl alcohol, and evaporated to dryness in a porcelain capsule, in which there has been placed a small piece of filter-paper. When the evaporation is complete, the paper is moistened with a strong solution of boric acid, and then dried; if turmeric be present the paper will take a reddish colour; if it be then treated by an alkali, there is a play of colours, among which green and purple predominate.

B. Advantage may be taken of the fact that the colouring-matter of turmeric is strongly fluorescent; that of mustard, on the other hand, is devoid of fluorescence. The simplest method to detect the fluorescence of the yellow colouring-matter of turmeric, when mixed with mustard, is to pass a little castor-oil through the suspected sample on a filter; the oil, if turmeric be present, shows a very distinct green colour; this is a test of considerable delicacy. Or an alcoholic solution may be placed in a test-tube, and held vertically in water contained in a glass blackened internally; if the observer now slightly incline the top from the window, and look from above *outside* the test-tube, the green fluorescence, if present, will be readily observed.

Naphthalene Yellow or Dinitro-Naphthol, $C_{10}H_6(NO_2)_2O$, has been detected in mustard by E. Waller and E. W. Martin (*Analyst*, 1884). The colouring-matter is readily extracted by alcohol of 95 per cent., and dyes wool a bright yellow—its tinctorial power is very great.

2. *Estimation of the Total Sulphur.*—Most of the adulterants of mustards contain no sulphur, or at least no very appreciable amount, in the unoxidised state. Mustard, on the other hand, in common with a large number of cruciferous plants, contains sulphur-organic compounds; hence a great deficiency or excess of sulphur is indicative of adulteration, a normal amount no conclusive sign of purity. The percentage of sulphur should be between 1 and 1.6.

The total sulphur in mustard may be estimated by oxidation with fuming nitric acid, or, more conveniently, by alkaline permanganate. A

weighed quantity of mustard is boiled with excess of alkaline permanganate, and the resulting sulphate precipitated by barium chloride. The volatile sulphur compounds may be distilled by a current of steam into strong alkaline permanganate, and a similar determination of sulphates be made. A. Schicht¹ recommends to add alcohol to the permanganate after cooling, and then a solution of iodine in potassium iodide until a feeble yellow colour remains. V. Genin prefers to distil and oxidise the distillate by bromine water.

3. *Estimation of the Fat or Oil.*—This is particularly useful when wheat starch is the adulterating agent. Wheat flour does not contain more than 1.2 to 2.1 per cent. of oil; mustard, on the other hand, from 33.9 to 36.7 per cent. A weighed portion of the previously dried sample may be placed in the little apparatus figured at p. 50. As a rough guide the following formulæ may be used:—

x = per cent. of mustard in the mixture, y = per cent. of oil found.

$$32.7x + 120 = 100y$$

or

$$34.7x + 200 = 100y$$

and the mean values of the two equations may be taken.

4. *Estimation of the Ash.*—The ash is taken in the way already described (see p. 93). The total ash of dried mustard averages 5 per cent. The highest number the writers have obtained is 5.3 per cent., the lowest 5.088 per cent. Of this ash from .2 to 1.25 is soluble in water. It hence follows, that if found above 5.5 per cent., mineral matters of foreign origin are present; if below 4 per cent., it is an indication of some organic adulterant. The American Association of Official Agricultural Chemists take 4 to 7 per cent. as the limits for the ash of ground mustard seeds.

5. *Gamboge.*—Gamboge as an adulterant of mustard is somewhat apocryphal; if suspected of being present, an alcoholic extract of the mustard must be prepared; such an extract when treated with caustic soda becomes of a bright red colour, hydrochloric acid produces a yellow colour.

PEPPER.

§ 346. Black pepper is the dried *immature* fruit of *Piper nigrum*, one of the *Piperaceæ*, or pepperworts.

White pepper is the same berry decorticated, or deprived of its outer and black husk or covering.

The pepperworts are a well-defined natural order, confined to the hottest parts of the world, and delighting in low places, valleys, and the banks of rivers. Although neither the number of its genera nor of its species is great, yet the whole order is remarkable for a variety of active and useful plants—*e.g.*, the aromatic black and long peppers, the astringent matico, the intoxicating *Macropiper methysticum*, the different varieties of cubebs, useful in the treatment of inflamed mucous membranes, and several other plants possessing medicinal properties,² belong to the natural order of *Piperaceæ*.

¹ *Zeit. anal. Chem.*, xxx., 661–665.

² The *Artanthe eucalyptifolia* is used in Brazil in cases of colic; *Piper parthenium*, used in menstrual disturbances; *Chavica betle* and *Siriboa* cause salivation, and decrease

Black pepper itself is a climbing plant, attaining the height of from 8 to 12 feet; the berries— or, botanically speaking, ‘drupes’—are at first green, then red, and if left still longer ungathered, turn to black; but before this latter change takes place the berries are gathered by hand and dried in the sun, the result being an entire change of appearance; instead of a red, smooth berry, a black or reddish-black peppercorn, with the cortex contracted and shrivelled in such a manner as to form a veined network, is obtained. The plant is cultivated in various portions of the equatorial regions of the earth, the zone of cultivation being confined to the isotherms of 82° F. It would not, however, be strictly correct to say that this high mean annual temperature is essential, or even necessary; for the fact is, that it is produced principally in the cooler valleys, where the mean annual temperature does not, perhaps, exceed 70° Fahr.

The black pepper imported into this country principally comes from the islands of Malacca, Java, Borneo, and Sumatra. The commercial varieties are at least five—viz., Malabar, Penang, Sumatra, Trang, and Tellicherry, names indicating the localities whence they are derived. The differences which these different varieties of pepper present to the eye are evident enough when the several samples are at hand for comparison; but it takes a very practised observer to identify a solitary sample; and if samples of each of the kinds named were mixed together, it is doubtful whether an adept even could separate the berries again, identifying each sort with any correctness. The merchant, indeed, relies more upon the weight than the appearance; he takes a handful of peppercorns, and by long practice can tell in a moment whether it is a light or a heavy sample. Chevallier has determined the weight of what is technically called heavy, half-heavy, and light pepper. A litre of the first weighed 530 grms.; of the second, 512 grms.; of the third, 470 grms. That there is considerable difference in weight in the different berries is certain, for one of the writers carefully weighed 100 berries of each kind, with the following result:—

	Grammes.
100 peppercorns of Penang weighed	6·2496
100 " Malabar " 	6·0536
100 " Sumatra " 	5·1476
100 " Trang " 	4·5736
100 " Tellicherry " 	4·5076

If, then, quality is to be judged of by weight, Penang and Malabar may be bracketed together as standing first, Sumatra holding the second place, and Trang and Tellicherry bracketed together in the third. The general opinion of the trade is, that Malabar is really the heaviest, and possibly the samples of Penang examined were unusually fine. The whole of the ground peppers of commerce are mixtures of different kinds of pepper; there is no such thing to be found in the shops as a pure ground Malabar or a pure ground Penang. The principal varieties mixed for household purposes and retailed are Malabar, Penang, and Sumatra; the first of these is the dearest.

The usual mixture, according to Chevallier, is—

33	per cent.	of Malabar	to give weight,
33	" "	Penang	" strength, and
33	" "	Sumatra	" colour.

the function of the skin. Besides these, *Acrocarpidium hispidulum*, *Cocobryon capense*, *Artanthe adunca*, *Chavica adunca* and others, possess active and useful properties.

The pepper thus mixed is either ground by the aid of large mill stones, or in an apparatus perfectly analogous to a coffee-mill. The latter mode is far preferable to the former, as the friction of the stones develops considerable heat, and dissipates some of the aromatic principles. Pepper thus damaged by the heat of the mechanical operations is technically known as 'burnt.'

Microscopical Structure of Pepper.—The black pepper berry is composed of the seed enveloped by its pericarp, both being adherent. The microscopical structure will be readily understood on reference to figs. 69 and 70. There is first a rather thick cuticle (.005 mm.) containing a single layer of small square cells, filled with a brownish substance; beneath the epidermis is seen a closely-packed, yellow layer of radially-arranged cells, with thickened walls of about .05 mm. in diameter. The cavity of each cell is filled up with a lump of resin; beneath the layer of thick cells is the parenchyma of the pericarp, consisting of two sets of cells. The outer layer is composed of soft tangentially-extended cells containing numerous starch granules, drops of oil and lumps of resin; it is the shrinking of this soft loose layer that is the chief cause of the wrinkling of the berry; among these cells may be often seen bast fibres and spiral vessels. The inner layer of the parenchyma is large-celled, the cells containing many drops of ethereal oil. A layer of one or two rows of thick-walled, tangentially-arranged cells concludes the structures belonging to the pericarp proper; the rest of the berry is made up of the testa and albumen of the seed. The testa consists of an outer brown, and an inner transparent, membrane; the outer brown consists of two layers of cells exhibiting no very definite form on cross section; but when the seed is soaked in water and thin strips torn from the testa, the outer layer is then seen to be composed of regular, long, five-sided cells. The albumen of the seed is formed of angular radially-arranged, large-celled parenchyma; most of the cells are filled with extremely small starch granules (see page 144), each of which, when examined by the aid of a high power, shows a central nucleus. Here and there may be seen cells containing little masses of a yellow resin; such yellow masses kept under glycerin for some time develop crystals.

White Pepper is the ripe fruit of the same plant; the berries are soaked in water, and then most of the dark pericarp is rubbed off. All the structures belonging to the pericarp are not detached, the skin separating at the vascular zone. The microscopic structure of white pepper is, therefore, the same as that of black, minus the portions removed.

Pepper contains two alkaloids, piperin and piperidine, a volatile oil,

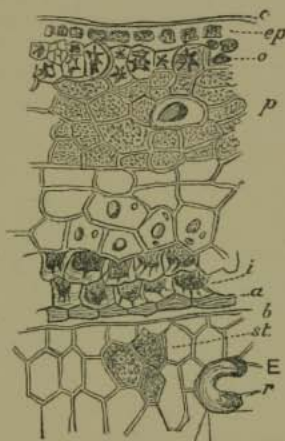


FIG. 69.—A section of black pepper $\times 160$.—*ep*, Epidermis covered with the cuticle, *c*; *o*, the outer layer of hard cells; *p*, thin-walled parenchyma, many of the cells having oily contents; *i*, the inner layer of hard cells, showing a peculiar one-sided thickening; *a*, *b*, coatings of the seed proper, *a* being the more external brown layer, and *b* the inner colourless layer; *E*, the endosperm, containing the starch cells, *st*, and here and there lumps of resin as at *r*.

and an acrid resin, besides gum, starch, vegetable albumen, salts, and other substances.

Oil of Pepper has a specific gravity of from 0.86 to 0.99, and a boiling point of 167° to 170° C. It is a clear fluid, possessing a mild taste, and corresponds to the formula $C_{10}H_{16}$. Both white and black pepper contain a little more than one per cent. of this oil.¹

§ 347. *Piperin*—($C_{17}H_{19}NO_3$)—was discovered by Oerstedt in 1819; it is found in white, long, and black pepper, in *Chavica officinarum*, in cubebs, in the berries of *Schinus mollis*, and in the bark of *Liriodendron tulipifera*. The quantity of piperin in peppers varies from 4 to 8 per cent. When pure, piperin crystallises in colourless, brilliant, four-sided prisms; it is almost tasteless, and presents no alkaline reaction. It melts to an oily mass at about 128° C., solidifying in a resinous form; is soluble in petroleum ether, alcohol, ether, the volatile oils, benzole, chloroform, and creosote. Concentrated nitric acid changes it into an orange-red resin;

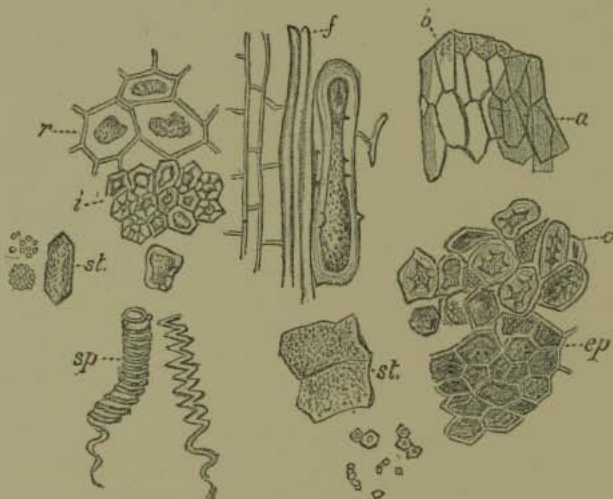


FIG. 70.—Ground pepper $\times 160$.—The lettering is the same as in the previous figure: *f*, bast fibres; *sp*, spiral vessels; *st*, the individual granules of starch $\times 600$.

if this be treated with a solution of caustic potash, a blood-red colour is produced, and on boiling piperidine is developed. Heating with alcoholic potash decomposes piperin into piperidine and piperinate of potash, and the same substance is quickly developed by heating with soda lime.

Piperidine, $C_5H_{11}N$ (Hexa hydro-pyridine), is a liquid boiling at 106° C. It is easily soluble in water and in alcohol. According to W. Johnstone² the amounts in black pepper range from 0.39 to 0.77 per cent., mean value 0.56 per cent.; white pepper contains smaller quantities, from 0.21 to 0.42 per cent.

Piperic Acid— $C_{12}H_{10}O_4$ —is obtained by boiling piperin with alcoholic potash, decomposing the piperate of potash by the addition of HCl, and

¹ Dumas, *Journ. Chem. Med.*, xi., 308.

² *Chem. News*, lviii., 235.

subsequent purification of the acid by crystallisation from alcohol. The acid is in the form of yellow hair-like needles, some of which may be sublimed undecomposed; they dissolve easily in boiling alcohol, but are scarcely soluble in water. The crystals melt at 217° C.

Bucheim has given the name of "Chavicin" to a substance which he separated as follows:—Black pepper, after being exhausted of all matters soluble either in alcohol or water, was treated with ether. The ethereal extract was shaken up with potash; on then separating and distilling the ether, *chavicin* is left behind, of the consistence of thick turpentine, and possessing a biting taste; it has not yet been obtained pure.

§ 348. *The Ash of Pepper.*—The following is an analysis of the ash of Tellicherry pepper:—

100 grms. of Ash.		100 grms. of Ash.	
Potash,	24·380	Phosphoric acid,	8·470
Soda,	3·226	Sulphuric acid,	9·613
Magnesia,	13·000	Chlorine,	7·570
Lime,	11·600	Carbonic acid,	14·000
Iron,	0·300	Sand,	6·530

The total ash of pure black pepper should not exceed 7·0 per cent., and of white pepper 3 per cent. No pepper ash should contain more than 2 per cent. of SiO_2 . The phosphoric acid averages 8·5 per cent. of the ash. Pepper also contains small quantities of carbonate; *i.e.*, from ·5 to 1·5 per cent. The ash itself will be found to contain from 30 to 60 per cent. of carbonates of the alkaline earths.

Nitrates and Nitrites in Pepper.—Comparatively few observations of the amount of nitrates and nitrites in organic substances are on record: it is a subject of some scientific interest, especially since it has been observed that nitrates and nitrites are decomposed in the presence of free oxalic acid. Whether the determination of nitric acid will be of service to the food-analyst or not is unknown; it certainly may be so, if it be found that a substance rich in nitrates is fraudulently mixed with one poor in nitrates.

100 grms. undried Penang pepper yield	Calculated as Nitric Acid. Grm.
" " Malabar "	0·04470
" " Tellicherry "	0·03858
" " Sumatra "	0·08860
" " Trang "	0·06560
" " " "	0·11870

§ 349. *General Composition of Pepper.*—A sample of Penang pepper analysed by one of the authors gave the following results:—

	Per cent.
Volatile oil,	1·04
Acrid resin,	1·77
Piperin,	5·17
Substances soluble in water, gum, starch, and other matters, subtracting ash,	14·74
Substances insoluble in alcohol and water,	67·75
Water,	9·23

The following table exhibits some analyses made in 1876 of genuine

black peppers, and may be compared with similar determinations of white and long peppers :—

TABLE LVII.—GENERAL COMPOSITION OF COMMERCIAL PEPPERS.

	Hygroscopic Moisture.	Piperin in Pepper dried at 100° C.	Resin in Pepper dried at 100° C.	Aqueous Extract in Pepper dried at 100° C.	Ash in Pepper dried at 100° C.	
					Soluble in water.	Total.
	Per cent.	Per cent.	Per cent.	Per cent.		
Penang,	9.53	5.57	2.08	18.33	2.21	4.18
Tellicherry,	12.90	4.675	1.70	16.5	3.38	5.77
Sumatra,	10.10	4.702	1.74	17.59	2.62	4.31
Malabar,	10.54	4.632	1.74	20.37	3.45	5.19
Trang,	11.66	4.600	1.70	18.17	2.53	4.77
White pepper, com- mercial, }	10.30	5.600	2.05	...	0.56	1.12
Long pepper,	1.800	0.80	16.82	4.47	8.30

§ 350. *Analysis of Pepper.*—The analyses (Table LVIII.) of genuine peppers and pepper adulterants, by A. L. Winton, A. W. Ogben, and W. L. Mitchell, will be found of great value. The ash and hygroscopic moisture are estimated in the usual manner. The piperin and resin are the most important to determine.

W. Johnstone recommends the conversion of piperin into piperidine. 10 grms. of the pepper are digested at 100° C. in a closed flask with 25 c.c. of water and 25 c.c. of alcoholic potash. After from four to five hours' digestion, the flask is cooled and the contents distilled and titrated until the distillate is no longer alkaline.

Piperin decomposes into piperidine, $C_5H_{11}N$, and piperic acid, $C_{12}H_{10}O_4$, which unites with potash; hence it follows that every molecule of piperidine found is equal to a molecule of piperin—*i.e.*, 1 c.c. of d. n. acid used by the distillate indicates 0.0285 of piperin.

An alcoholic extract of pepper practically consists of resin and piperin, so that if the alcoholic extract be determined, and then the piperin by Johnstone's method, this last value subtracted from the total weight of extract, *minus* any ash, should give approximately the resin.

Instead of the above methods an ether extract may be made and evaporated to dryness; this, according to A. L. Winton, should be not less than 6.5 per cent. Piperin may be isolated from this extract by dissolving the resin in sodium hydroxide, then treating the residue with alcohol, filtering and evaporating to dryness.

It has been proposed by Lenz to take as a basis for calculation of the amount of any adulterant the organic matters of the pepper. This is of course obtained by subtracting the hygroscopic moisture and ash from the total weight. The pepper-starch is then converted into sugar, and the sugar estimated; in this way very constant results are obtained, as shown in Lenz's experiments¹ and also in some determinations on the same lines by Heisch.²

¹ *Zeit. anal. Chem.*, xxiii., 501.

² *Analyst*, 1886, 186.

TABLE LVIII.

Description.	Moisture.	ASH.			ETHER EXTRACT.		Alcohol Extract.	Reducing matters by direct inversion calculated as Starch.	Starch by Diastase method.	Crude Fibre.	Total N. less N. in Non-volatile Ether Extract $\times \frac{6}{4}$.	NITROGEN.		Parts of N. in 100 parts of Non-volatile Ether Extract.
		Total.	Soluble in Water.	Insoluble in HCl.	Volatile.	Non-volatile.						Total.	In Non-volatile Ether Extract.	
<i>Black pepper:</i>														
Singapore,	12.00	3.49	2.10	0.12	1.06	7.73	8.89	42.80	38.51	11.04	12.71	2.36	0.31	3.99
Tallicherry,	11.56	4.21	2.75	0.01	0.83	6.94	8.80	41.67	37.01	12.20	11.79	2.15	0.27	3.89
Lampung,	11.40	5.69	2.18	0.83	1.17	8.86	9.72	39.25	35.50	12.14	10.93	2.09	0.34	3.83
Acheen,	12.31	5.87	3.04	0.86	1.34	9.54	10.64	32.99	28.13	15.78	11.94	2.29	0.38	4.00
<i>White pepper:</i>														
Decorticated,	12.89	1.06	0.47	0.01	0.56	7.24	7.83	64.85	63.16	0.60	11.03	2.08	0.32	4.42
Singapore,	13.47	1.33	0.33	0.09	0.92	7.89	8.45	56.71	53.89	4.10	11.06	2.11	0.34	4.33
Siam,	13.63	1.47	0.38	0.10	0.69	8.64	7.36	59.01	56.18	3.52	10.69	2.00	0.29	4.30
Penang,	13.68	2.84	0.65	0.15	0.76	6.32	7.32	57.17	54.01	3.80	10.88	2.01	0.27	4.23
<i>Pepper by-products and adulterants:</i>														
Pinhead, Siftings from } Acheen, }	10.95	8.25	2.34	1.74	1.26	8.24	9.66	29.01	25.03	17.51	11.25	2.10	0.30	3.65
Pepper shells (hulls only),	10.57	11.91	3.20	4.70	0.68	3.04	4.00	11.43	2.30	32.15	14.19	2.36	0.09	2.91
Pepper shells (hulls and dust),	10.52	10.30	2.28	2.88	1.06	4.77	5.71	21.69	15.30	23.61	12.94	2.21	0.14	3.01
Pepper shells (hulls and dust),	10.66	10.2	2.90	2.63	1.02	4.97	6.30	20.99	14.12	23.27	12.31	2.12	0.15	2.94
Long pepper,	9.47	5.93	4.20	0.22	1.55	6.61	8.67	42.88	39.55	5.76	12.25	2.18	0.22	3.34
Buckwheat hulls,	7.63	1.84	1.24	0.00	0.07	0.38	2.17	20.51	1.46	43.76	3.06	0.49

H. Rabourdin's¹ method has for its principle the estimation of the residue not capable of transformation into sugar and soluble matters by boiling with acid. A grm. of the sample is boiled continuously for an hour in 100 grms. of distilled water and 4 grms. of sulphuric acid; the boiling is best carried out in a flask adapted to a vertical condenser. The insoluble matters are washed, dried, and weighed in a tared filter. Rabourdin found that ground genuine black pepper gave, as an average, '35 per cent. insoluble residue; olive-stones, '745 per cent. A better method is to use the American method of estimating crude fibre:—2 grammes of the ether extracted substance are boiled for thirty minutes under an inverted condenser with 200 c.c. of 1·25 per cent. H_2SO_4 , then filtered and washed. The undissolved substance is then boiled for thirty minutes with 200 c.c. of 1·25 per cent. KOH, filtered, washed, dried, and weighed, burnt to an ash and the ash deducted from the weight found. A pure black pepper should not contain more than 16 per cent. of fibre, and a pure white pepper not more than 8 per cent.

§ 351. *Adulterations of Pepper.*—Pepper has been adulterated for at least two centuries and a half; for Pierre Pomet,² writing in 1614, says: "As the greatest part of pepper, white as well as black, is sold '*battu*' (that is to say, powdered), it should only be bought of honest merchants; because all the pepper the retailers sell is no other thing for the white than '*épices d'Auvergne blanchées*,' or rather black pepper whitened with ground rice; the black is only the dust either of the crust of bread, grey Auvergne spices, or manigette."

The list of the adulterations enumerated by authors is an extraordinary one. Linseed-meal, rice, pepper, leaves, mustard, wheat flour, sago, woody fibre, chillies, rape-seed, potato, spices, capsicum, manigette (otherwise known as Guinea pepper), chicory, rye, powdered leaves of the laurel, which had been previously used to wrap round extract of liquorice, the stones from olives, bone-dust, marine salt, and various mineral adulterations, are all said to have been detected.

However various may be the adulterations in France (where, Chevallier tells us, in Paris alone he was acquainted with a manufactory producing 1200 to 1500 kilogrammes annually of a mixture sold solely for the purpose of adulterating pepper), the only common adulterations of this country were until lately what are known in the trade as P.D., H.P.D., and W.P.D., abbreviations for pepper-dust, hot pepper-dust, and white pepper-dust. The first, or P.D., used to be principally composed of faded leaves, but linseed-meal is now preferred; H.P.D. is chiefly the husks of mustard, and W.P.D. is ground rice.

Maize has lately been used in England to adulterate pepper. Other starches, nut shells and bleached pepper hulls will all be detected by a careful microscopical examination. Pepper hulls will also have the effect of raising the ash, decreasing the ether extract and lowering the ratio of calcium carbonate to total ash. The amount of fibre is also considerably raised.

Cayenne pepper is said to be added to pepper to restore pungency. It may be detected by heating some of the separated red particles, when irritating vapours are given off.

Long pepper may be detected in black or white pepper by the strong and characteristic odour when warmed. The ash will be found to be high and the ethereal extract low.

¹ *Journal de Pharmacie* [5], ix. 289-287. ² Pomet: "*Hist. Gén. des Drogues*," 1735.

Olive-Stones, Poivrette Pepperette.—Towards the end of 1886, considerable consignments of pepper came into English commerce adulterated with poivrette or ground olive-stones.

The olive-stone has an extremely hard endocarp, difficult to grind, and by boiling in dilute sulphuric acid or by heating with potash, the particles become of a reddish colour, and may thus be picked out of mixtures. Microscopically, ground olive-stones will be found to consist mainly of hard prosenchymatous cells; the superficial layers are fibrous, and are in company with numerous vascular bundles; the more numerous cells are short cells of various forms, but all showing frequently pore-canals (see

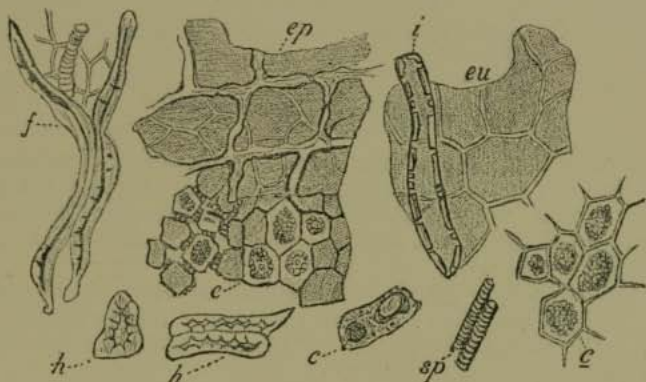


FIG. 71.—Tissues of the olive-stone, $\times 160$.—*f*, Thickened fibres with spiral vessels; *h*, hard cells from the middle layer of the stone; *i*, inner layer of hard cells, resting on the endothelium, *eu*; *sp*, spiral vessels; *ep*, epidermis, through which may be seen a brown parenchyma; *c*, embryonal tissue; *c*, soft cells from the olive fruit occasionally adhering to the olive-stone; *C*, tissue of the cotyledons.

fig. 71). These structures have but a slight similarity to the analogous structures of the pepper-berry.

The specific gravity of pepper is about 1.173, that of olive-stones higher. If glycerin and water be mixed so as to have a density of 1.173 at 15° C., the pepper either floats or is suspended, the olive-stones sink. In this way small percentages of olive-stones may be roughly separated.

Advantage has also been taken of the different effect of colouring-matters on pepper and olive-stones. Aniline acetate colours olive-stones yellow-brown, pure pepper remains of a greyish-white. Naphthylamine gives a yellow orange, thalline pure orange. The French chemists use the hydrochlorate of dimethylparaphenylenediamine.¹ The pepper is put in a porcelain dish, and a solution of the above-named salt poured direct on the powder; a little water is added, and the whole stirred; if olive-stones are present, they sink to the bottom and are stained a brilliant red colour.

D. Martilli digests for two or three days 1 gm. of phloroglucol in 50 to 60 c.c. of HCl, specific gravity 1.1; the clear solution is decanted. To about half a gramme of the pepper, enough of the reagent is added to just cover it, and the mixture is heated for a few minutes. Poivrette and, generally, similar substances give a reddish-violet

¹ This reagent may be made as follows:—In a porcelain dish 10 grms. of dimethyl-aniline are mixed with 20 grms. of pure hydrochloric acid; 100 grms. of crushed ice are added, and little by little 100 c.c. of a 7 per cent. solution of sodic nitrite. At the end of half an hour, 30 to 40 grms. more hydrochloric acid are added, and 20 grms. of leaf tin. The reduction is allowed to cool for an hour, then the tin is precipitated by granulated zinc. The liquid is filtered, and saturated with sodic carbonate until it becomes cloudy; it is cleared by acetic acid; 10 grms. of sodic bisulphite are added to prevent oxidation, and it is diluted to 2 litres.

colour. On adding water and decanting, the powder left mainly consists of the adulterant. The authors have tried this process, and it works fairly well. According to Gillet a 7 per cent. alcoholic solution of iodine imparts to pepper a brown stain and to poivrete a yellow one.

Sand is a common adulterant, whether derived from the sweepings of the shops, or added as sand, is by no means clear. The sand, of course, influences the weight of the ash, which should never exceed 7 per cent.

Dr. Hassall made some determinations of the ash of some fifteen or sixteen commercial samples of black pepper; of these only one was under 5 per cent., the percentages of the other fifteen being distributed as follows:—

One	gave between 5 and 6	per cent. of ash.
Three	“ 6 and 7	“ “
Three	“ 7 and 8	“ “
Seven	“ 9 and 10	“ “
One	“ 11 and 12	“ “

It is difficult to believe that more than 2 per cent. of unavoidable material dust can get into the pepper by grinding, etc., and the inference naturally is that most of the above samples were adulterated. The maximum percentage of ash from genuine pepper which the writers have obtained is 5.3 per cent.

The sand foreign to the pepper ash is best separated by the chloroform process as used for alum in flour (p. 158). Nearly all the foreign mineral matter sinks to the bottom of the tube, while the lighter starchy matters float.

Besides the formidable list of adulterations already mentioned, the berry itself is not free from manipulation; for, as the merchant judges by the weight of the sample, means are taken to render the lighter sorts equal in weight to the heavy Malabar and Penang, and in order to do this they are macerated in tubs of brine for twenty-four hours, and thus impregnated with salt and water find their way into the market as Malabar; but such samples are quickly recognised by the astute merchant; and the high chlorides, the high ash, the great amount of humidity, could hardly fail to reveal their nature to the analyst.

As coffee has been cleverly imitated by chicory pressed into the shape of the coffee-berry, so by pressing various pastes into the shape of the pepper-berry has pepper been imitated. Of this adulteration there is the most undoubted evidence. Accum noticed artificial peppercorns made of oilcake, common clay, and Cayenne pepper, and Chevallier states that in 1843 he was requested to examine a sample taken from forty bales, in which he found from 15 to 20 per cent. of artificial pepper, composed of pepper-dust, bran, and other matters.

CAYENNE PEPPER.

§ 352. Cayenne pepper consists of the powdered pods or seeds of the *Capsicum annuum*, a plant belonging to the natural order *Solanaceæ*. It is a native of America, but is also cultivated, to a slight extent, in the greenhouses of England and other European countries. It is sold entire under the name of *Chillies*. The microscopical structure of the capsules is somewhat peculiar: the epidermis is formed of cells, the walls of which are thick, flattened, tortuous, well defined, and punctated here and there;

frequent drops of a reddish-orange oil occur, especially in the parenchyma, which is formed of thin-walled, rounded cells. The envelope of the grain itself, when cut in thin vertical sections, presents a very singular appearance, that of radiating dentiform processes, the apex of each being apparently fixed in the outer membrane. The substance of the seed proper is composed of small angular cells, with black colourless walls, filled with granules and a yellow-orange oil, but without starch.

Cayenne pepper, as met with in commerce, is in the form of a somewhat coarse, brick-dust like powder, the least particle of which, if heated strongly, volatilises a very acrid vapour, causing intense irritation of the throat. This sensation can be produced by so minute a portion of cayenne, that any foreign substance mixed with it could in this way be detected; it would be only necessary to separate carefully, by the aid of the microscope and a camel's-hair brush, all particles of cayenne, and heat the portion suspected; if no acrid vapours were given off, the substance could not be cayenne. This intense acidity appears to be due to a body discovered by Dr. Thresh, and named by him Capsaicin.

§ 353. *Capsaicin*—($C_{18}H_{28}NO_3$), specific gravity 1.060—is in the form of minute crystals, which melt at $63-63.5^\circ C.$, volatilise unchanged at $115.6^\circ C.$ ($240^\circ F.$), and at $120^\circ C.$ ($248^\circ F.$) become brownish-black. The fruit yields about .03 per cent. Capsaicin contains a methoxy and a hydroxy group and has the properties of a weak phenol-like acid. It may be obtained by exhausting cayenne by petroleum, evaporating the petroleum, and treating the extract thus obtained by dilute solution of potash; on now saturating the solution with carbonic anhydride, it is precipitated in very small crystals. It dissolves slightly in cold, and more readily in boiling, water; is easily soluble in alcohol, proof spirit, ether, amylic alcohol, acetic ether, acetic acid, benzine, the fixed oils, and solutions of the alkalies. It dissolves slowly in turpentine and carbonic disulphide; when pure, petroleum does not dissolve it readily, but the presence of the red oil in the pepper increases its solvent powers; it is totally insoluble in solutions of the carbonates of the fixed alkalies, and in ammonia. Excess of platin chloride added to an alcoholic solution of capsaicin and allowed to evaporate, develops the odour of vanillin. The crystals moistened with strong sulphuric acid and a little sugar added become after some hours a fine blue.¹ Silver nitrate gives a precipitate with alcoholic solutions of capsaicin; it also yields white precipitates with barium and calcium chlorides. It is powerfully pungent, causing, if volatilised, severe fits of coughing.

It would appear that capsaicin is not contained in the substance of the seed; for if the pericarp be carefully separated, the seeds are entirely devoid of acrid taste.

Dr. Thresh has also described a conium-like alkaloid, obtained by exhausting the pericarp with benzine, evaporating, dissolving in ether, shaking the solution with dilute sulphuric acid, partially neutralising with barium carbonate, and evaporating to a small bulk. Some red fat now separates, and after the removal of this (upon adding an excess of alkali, shaking with ether, and evaporating) a brown residue is obtained, smelling like conium, and giving precipitates with Nessler reagent, iodine, and iodides of potassium and cadmium.

The acrid oil *Capsicol*, *Capsicin*, and other substances described by Bucholz, Braconnot, Buchheim, etc., are undoubtedly mixtures. The

¹ Karl Micko. *Zeit. Unters. Nahr. Genussm.*, II. 411.

general composition of cayenne pepper may, however, be gathered from the following analysis by C. Richardson :—

	Seed.	Pod.	Whole Fruit.
Water at 100° C.,	8·12	14·75	11·94
Albuminoids,	18·31	10·95	13·88
Ether extract,	28·54	5·48	15·26
Nitrogen-free matter by difference,	24·33	38·73	32·63
Crude fibre,	17·50	23·73	21·09
Ash,	3·20	6·62	5·20
Nitrogen,	2·93	1·71	2·22

The hygroscopic moisture ranges in different samples from 10 to 13 per cent. One of the authors analysed several samples of genuine cayenne, and the mean of these analyses was as follows :—

	Per cent.
Aqueous extract of dried cayenne,	32·1
Alcoholic extract,	25·79
Benzole extract,	20·00
Ethereal extract,	10·43
Ash,	5·693 (soluble, 3·32)
Total nitrogen in 100 grms.,	2·04

The ash, as a rule, will be found to be less than 6 per cent. and should not be more than 8 per cent. The ether extract should exceed 10 per cent., and the alcohol extract 25 per cent.

§ 354. *The Adulterations of Cayenne* usually enumerated are: all kinds of red mineral powders, from brick-dust to cinnabar, and a few starches. An artificial red containing barium is sometimes added, and lead has been found. It has been stated that if lead or barium salts are present in a soil they may be contained in the cayenne pepper grown upon it. George Gregory¹ has, however, proved that there is no truth in this statement. There does not appear, however, to have been any conviction recently for the adulteration of cayenne, and the numerous samples the senior writer has examined were all genuine. Most of these additions would be easily detected in the ash, or by the microscope. Cinnabar is highly improbable; for its detection, see "Mercury," in *Poisons*, by A. Wynter Blyth.

SPICES.

§ 355. The most commonly used spices are ginger, allspice, nutmeg, mace, cloves, and cinnamon. Those spices which are sold in a powdered form are especially liable to adulteration, various starches, stems, barks, sawdust, ground olive-stones, walnut shells, etc., all having been used from time to time. Microscopical examination² will, as a rule, show the presence of any coarse adulterant, but chemical examination will also assist in the detection of adulteration, and in the estimation of the

¹ *Zeit. Nahr. Genussm.*, 1900, iii., 450 471.

² It cannot be too strongly impressed upon the analyst that however useful diagrams and descriptions of microscopical preparations may be, in teaching him what to look for, the only possible way to learn the microscopical structures of tissues, etc., is to prepare and examine specimens for himself.

TABLE LIX.—SPICES AND SPICE ADULTERANTS.

Description.	Moisture.	ASH.			ETHER EXTRACT.		Alcohol Extract.	Reducing matters by direct inversion calculated as Starch.	Starch by Diastase method.	Crude Fibre.	Nitrogen × 64.	Total Nitrogen.	Cold Water Extract.	Oxygen absorbed by Aqueous Extract.	Quercitannic Acid equivalent to O absorbed.	Lime (CaO).
		Total.	Soluble in Water.	Insoluble in HCl.	Volatile.	Non-volatile.										
<i>Ginger</i> : Jamaica, bleached (limed),	10.56	8.31	2.63	0.02	1.27	3.12	4.70	57.31	54.78	2.37	9.34	1.49	15.89	2.72
Jamaica, unbleached,	11.22	4.17	2.96	0.22	1.79	3.91	5.15	57.59	56.09	3.72	7.85	1.24	15.69	0.26
Cochin,	10.43	3.86	2.40	0.10	2.09	3.70	4.62	59.08	57.33	3.62	8.15	1.30	12.42	0.48
"	9.31	5.22	2.78	0.45	2.80	5.11	5.77	53.70	49.45	5.30	7.84	1.25	12.22	0.55
Japan (limed),	11.39	6.19	1.81	0.71	0.96	3.94	5.02	60.55	57.05	2.73	5.40	0.86	11.82	1.66
African,	9.97	4.00	2.52	0.11	2.73	5.35	6.34	56.74	52.68	4.66	7.92	1.27	13.10	0.25
<i>Ginger by-products</i> : Rough, scraggy ginger, } rejections from Cochín D,	4.99	8.05	4.03	0.89	6.05	9.55	11.60	31.38	19.35	13.18	7.00	1.12	14.65	0.61
Ginger cuttings,	3.19	9.20	3.90	1.81	7.06	2.76	9.20	40.23	31.14	8.69	8.69	1.39	17.72	1.06
Exhausted ginger from English ginger-ale works, } Exhausted ginger from extract works,	10.61	2.12	0.59	0.18	1.61	3.86	4.88	59.86	54.57	5.17	6.94	1.11	16.42
<i>Claves</i> : Penang,	7.99	5.29	3.37	0.03	19.02	6.43	15.22	9.16	2.65	7.90	6.75	1.08	...	2.39	18.64	...
Ambonyna,	7.89	6.18	3.62	0.04	20.47	6.56	15.20	9.44	2.73	8.62	6.02	0.96	...	2.13	16.94	...
Zanzibar,	7.62	6.07	3.63	0.10	18.00	6.45	14.31	9.44	2.73	8.62	6.02	0.96	...	2.49	18.44	...
<i>Clove stems</i> (adulterant),	8.74	7.99	4.26	0.60	5.00	3.83	6.79	14.13	2.17	18.71	5.88	0.94	...	2.40	18.79	...
<i>Nutmegs</i> : Singapore,	3.63	2.28	0.86	0.00	3.02	36.70	10.77	25.56	23.72	2.51	6.73	1.08
Macassar (limed),	5.24	3.32	1.25	0.00	4.70	32.88	16.79	29.47	29.25	2.07	6.95	1.11
Damaged (worm eaten),	17.23	6.37	4.33	0.12	9.76	11.32	14.71	5.71	1.63	7.95	9.73	1.49
<i>Mace</i> ,	11.05	2.01	1.13	0.07	7.58	22.48	23.11	31.73	27.37	3.20	6.47	1.03
Bombay (adulterant),	0.32	1.98	1.37	0.07	4.65	59.81	44.27	16.20	14.51	3.21	5.06	0.51
<i>Cinnamon</i> : Ceylon,	8.63	4.82	1.87	0.13	1.39	1.44	12.21	19.30	...	36.20	3.70	0.59
<i>Cassia</i> : Saigon,	7.64	5.23	2.06	0.37	4.04	2.87	6.60	21.83	...	23.88	4.18	0.67
Batavia,	9.33	4.58	1.72	0.04	1.99	1.43	13.50	21.55	...	30.19	4.99	0.80
China,	11.27	4.21	1.14	1.32	1.31	1.80	5.32	27.08	...	24.51	3.89	0.62
<i>Cassia buds</i> ,	7.93	4.64	2.88	0.27	3.88	5.96	10.88	10.71	3.04	19.35	7.53	1.20	...	1.24	9.71	...
<i>Allspice</i> ,	4.47	2.47	0.00	4.05	5.84	11.79	18.03	...	22.39	5.75	0.92	...	0.53	2.08	...
English walnut shells,	7.69	1.40	0.07	0.00	0.12	0.55	1.84	19.30	1.01	56.58	1.69	0.27	...	0.33	1.30	...
Brazil nut shells,	9.08	1.59	1.06	0.17	0.07	0.57	1.01	12.96	0.73	50.98	4.19	0.67	...	0.40	1.66	...
Almond shells,	7.80	2.86	2.39	0.05	0.16	0.64	5.16	22.72	0.84	49.89	1.75	0.28	...	0.47	1.82	...
Cocoonut shells,	7.36	0.54	0.50	0.00	0.00	0.25	1.12	20.88	0.73	56.19	1.13	0.18	...	0.61	2.34	...
Date stones,	8.24	1.24	0.76	0.04	0.36	3.38	16.72	20.88	2.19	5.72	5.31	0.85	...	0.80	1.17	...
Spruce sawdust,	8.77	0.23	0.16	0.00	0.07	0.77	1.50	15.48	1.13	64.03	0.56	0.09	...	3.13	12.22	...
Oak sawdust,	5.73	1.22	0.32	0.02	0.07	0.84	6.25	17.10	1.68	47.79	1.63	0.26	...	1.00	3.90	...
Lanseed meal,	8.71	5.72	1.74	0.55	0.04	6.58	9.46	21.15	14.06	8.30	31.81	5.09	...	1.26	4.94	...
Cocoa shells,	10.44	8.40	4.66	0.83	1.00	2.99	4.77	8.68	3.15	14.12	16.19	2.59	...	0.59	2.29	...
Red sandalwood,	4.42	0.70	0.28	0.07	1.21	11.47	19.37	6.79	1.12	52.30	3.06	0.49

adulterant. The most important determinations to make in the majority of cases are:—ash, fibre, starch, volatile oil, fixed ether extract, and total nitrogen; certain other determinations will be found useful in the case of individual spices. The careful determinations of Messrs. A. L. Winton, A. W. Ogben, and W. L. Mitchell at the Connecticut Experimental Station, given in Table LIX., will be found of great assistance to the analyst.

§ 356. *Ginger* is the dried rhizome of *Zingiber officinale*. It is sold both with and without the outer integument and also in the form of powder. The characteristics of ginger starch have been described on page 142.

As well as admixture with such substances as have been mentioned above, ginger is very often mixed with exhausted ginger; this is usually indicated by a low cold water extract, and low soluble ash (see Table LIX.).

§ 357. *Allspice* or *Pimento* is the dried unripe fruit of *Eugenia pimenta*. For analytical figures see Table LIX.

§ 358. *Nutmeg and Mace*.—*Nutmeg* is the dried seed of *Myristica fragrans* stripped of its hard shell. This hard shell or arillus constitutes the *mace* of commerce. Nutmeg contains from 3 to 5 per cent. of a volatile oil having a specific gravity which varies from .880 to .95. It boils at 160° C., is soluble in alcohol, and turns a ray of polarised light to the right.

Artificial whole nutmegs are sometimes made from exhausted or damaged nutmeg powder. These are mixed with genuine nutmegs. According to J. Vanderplauten¹ they may be detected by cutting the nut in half, when all vegetable structure will be noticed to be absent. Treated with boiling water for a few minutes and pressed, they fall to powder. The ash is exceedingly high, *i.e.*, from 11 to 18 per cent. F. Rauwez gives the following analysis of some artificial nuts, which may be compared with the analyses of nutmegs in Table LIX.

	Per cent.
Moisture,	11.09
Ash,	11.34
Ash insoluble in HCl,	3.90
Ether extract,	15.42
Essential oil,	1.76
Cellulose (? crude fibre),	8.24

§ 359. *Mace*.—In addition to the ordinary adulterants, mace is often mixed with Bombay mace, a variety which contains only a small proportion of volatile oil, *i.e.*, about 3 per cent. The fixed ether extract is, however, higher than in the case of true mace (see Table LIX.). The volatile oil in true mace should, according to König, be at least 3 per cent. Wild mace (*Myristica fatua*) is sometimes mixed with or substituted for true mace; it may be recognised by means of the microscope and also by Hefelman's test, which is as follows:—A piece of filter paper is saturated with an alcoholic solution of the mace; the excess of liquid is removed by pressing between more filter paper. It is then treated with a drop of potash solution. A red coloration is produced in the presence of wild mace.

Cinnamon and Cassia (see Table LIX.).—True cinnamon or Ceylon cinnamon is the bark of *Cinnamomum zeylanicum*, but the commercial substance is more often derived from *cassia* barks which are sold as true

¹ *British Food Journal*, March 1900.

cinnamon. They are said to contain more manganese oxide than cinnamon.

§ 360. *Cloves* are the dried unexpanded flower-buds of *Caryophyllus aromaticus*. They contain about 16 per cent. of a volatile oil. This oil has a sp. gr. of from 1.034 to 1.061; it is colourless when freshly distilled and becomes reddish brown with keeping. It is soluble in alcohol, in ether, and in acetic acid. In addition to the ordinary spice adulterants, cloves are often mixed with stems and clove fruit. Clove stems will be readily detected by the microscope.

THE SWEET AND BITTER ALMOND.

§ 361. The sweet almond, the seed of *Amygdalus communis*, and the bitter almond, the seed of *Amygdalus communis*, var. *amara*, enter either in whole or in part into so many articles of food (such as sweetmeats and pastry, and as a flavouring ingredient into certain drinks), that it is absolutely necessary to be acquainted with their chemical composition. Both varieties of almond agree in containing about 50 per cent. of a bland fixed oil (consisting chiefly of olein, and liable to become rancid), as well as an albuminous principle, emulsin, sugar, gum, and woody fibre; but only in the bitter almond is found, in addition to the foregoing, *amygdalin*.

§ 362. *The Oil of Almonds* is a thin fluid oil, of a clear yellow colour, specific gravity 0.914 to .920, not coagulated by a cold of -10° C.; at -16° C. it becomes cloudy, and at -22° C. it solidifies to a white butter. Oil of almonds appears to be rather frequently adulterated with other oils.

2-5 parts of the oil, shaken with an equal bulk of nitric acid (specific gravity 1.20) and bisulphide of carbon, should not show any colour after standing a few minutes; if it becomes within half an hour yellow, or reddish-yellow, the change indicates oil from *cherry* or *apricot kernels*.

The following test will detect drying oils:—Dissolve one part of starch in 3 parts of warm nitric acid, of 1.20 specific gravity, and warm in a capacious vessel over the water-bath with 10 parts of almond oil, until all evolution of gas ceases. The oil after cooling is within two days changed into a warty, crystalline, greasy mass of elaidin. Should it, however, contain a drying oil (*poppy*, for example), it either remains quite fluid or semi-fluid, according to the proportion of adulterant present. The colour of the elaidin is also a guide; that produced by the sweet almond is pure white, by the bitter, yellowish-white, and by the small or inferior kinds of almonds, brownish-yellow; if the elaidin should be red, it denotes adulteration of some foreign oil, especially of *sesamé*.

Pure almond oil dissolves in 25 parts of cold and 6 of hot alcohol. The above tests, and in addition the low temperature required for congelation, should detect all ordinary adulterations.

§ 363. *Amygdalin* ($C_{20}H_{27}NO_{11} + 3H_2O$), is a glucoside, discovered in 1830 by Robiquet and Boutron-Charlard. It may be extracted from almond-cake by boiling alcohol of 95 per cent., and then precipitated from the somewhat concentrated alcoholic solution by ether. *Amygdalin*¹

¹ Lehmann, in an elaborate research, found the method of Liebig and Wohler the best for obtaining *crystalline amygdalin*. The process consists in boiling the substance with strong alcohol (of 94 to 95 per cent.) twice successively, after having first removed the fixed oil by petroleum benzine, concentrating to about one-half or one-sixth of its

crystallises from 80 per cent. alcohol in colourless glittering scales, containing two molecules of water: it can also be obtained in crystals. Amorphous amygdalin of the before-mentioned cherry-laurel leaves and buckthorn bark is best obtained by the following method:—The dried buckthorn bark is boiled with absolute alcohol, agitated with lead oxide, and evaporated to dryness. Dried in a vacuum over SO_2H_2 it forms a brittle, yellow, transparent, resin-like mass, which, when heated to 100°C ., becomes dark-brown; it can be dissolved by boiling alcohol and by water, but is insoluble in ether. Although amorphous, it is a crystalloid with three molecules of water, as proved by dialysis from water or weak spirit, but in such a case it loses one molecule if dried over sulphuric acid. At 100° to 120°C . it may be obtained anhydrous.

Amygdalin possesses no smell; it has a slightly bitter taste; its reaction is neutral, and it polarises to the left $[\alpha]_D = -35.57$. It dissolves in all proportions in boiling water, and in 12 parts of cold at 10°C .; requires 148 parts of alcohol, specific gravity 0.939, 904 parts of alcohol, specific gravity 0.819, if cold—but if boiling, 11 parts of the first and 12 of the last; it is insoluble in ether. It melts at 120°C ., and begins to carbonise at 160°C ., when it develops a caramel smell, and is at length fully destroyed.

Amygdalin, by the action of dilute hydrochloric acid, splits up into glucose and mandelic acid, volatile oil of almonds, and formic acid. If boiled with solutions of potash or baryta it forms ammonia and amygdalic acid. The most interesting decomposition is, however, that which takes place by the action of emulsin; it then breaks up into volatile oil of almonds, hydrocyanic acid, and formic acid.

Volatile Oil, or Essence of Almonds, does not exist as such in the bitter almond; it is, as above explained, the result of the decomposition of the amygdalin. The oil of almonds, when properly purified from prussic acid, is identical with the hydride of benzole, $\text{C}_6\text{H}_5\text{OH}$. It is colourless, thin, turning a ray of polarised light to the right, of a peculiar, pleasant odour, and a burning aromatic taste. Its specific gravity is 1.043 to 1.07, usually 1.06 (*Hirsch*). Its boiling point is 180°C . By the action of light and air it is gradually oxidised into benzoic acid. It is soluble in equal parts of alcohol, 0.830 specific gravity, and in about 30 parts of water. The ethereal or volatile oil is officinal in the French, Swiss, and Norwegian pharmacopœias. The ethereal oil is much adulterated. The analyst will specially look for alcohol, prussic acid, nitro-benzine, and ethereal oils.

If *alcohol-free*, the addition of an equal weight of fuming nitric acid produces no effervescence, and after two or three days the mass becomes emerald green, and crystals of benzoic acid appear. On the other hand, if it contain alcohol from 0.08 per cent. upwards, there is immediately a strong effervescence. Some of the tests given for alcohol at p. 382 *et seq.* will be of service.

volume; and then adding ether, which precipitates the amygdalin, and removes any of the remaining fixed oil. Lehmann obtained from

Bitter almonds,	.	.	2.5	per cent. crystallised amygdalin.
Cherry-kernels,	.	.	0.82	" " "
Plum-kernels,	.	.	0.96	" " "
Apple-seeds,	.	.	0.60	" " "
Peach-kernels,	.	.	2.35	" " "
Cherry-laurel leaves,	.	.	1.38	per cent. amorphous amygdalin.
Bark of <i>Rhamnus frangula</i> ,	.	.	0.7	" " "

Cherry laurel leaves and Rhamnus bark contain hydrocyanic acid ready formed.

The detection and estimation of *prussic acid* in the essence is carried out on the principles detailed in the article on *Prussic Acid* in *Poisons*, by A. Wynter Blyth.

Nitrobenzine is indicated when the essence is not entirely soluble in a solution of bisulphide of potash, and the specific gravity is higher than 1·07, the specific gravity of nitrobenzine being 1·20 to 1·29; the boiling point will also be raised. In such a case nitrobenzine should be specially tested for, by changing it into aniline by reducing agents. For this purpose 10 parts of dilute sulphuric acid (specific gravity 1·117) may be added to 10 of granulated zinc and 1 part of essence. At the end of two hours (after frequent agitation) the fluid is passed through a moistened filter, and a crystal of chlorate of potash added to the filtrate with a drop of concentrated sulphuric acid. If a violet or red colour is produced, it is due to the presence of an aniline salt, produced from nitrobenzine; but if there is no coloration, nitrobenzine must have been absent.

To estimate the amount of nitrobenzine in an adulterated sample 5 c.c. of the essence are shaken up in a separating tube with 35 c.c. of a concentrated solution of sodic bisulphite at 25° and made up to 50 c.c. by addition of water. The nitrobenzine is now taken up with ether, the ether separated and dried by fused calcic chloride and allowed to evaporate spontaneously, and the nitrobenzine weighed.

Another special method used for the detection of nitrobenzine was proposed by Maisch:—1 grm. of the essence is dissolved in twelve times its volume of alcohol, .75 of caustic melted potash is added, and the whole heated until the liquid is diminished to about one-third. The pure essence, on cooling, is of a light brown colour, and dissolves entirely in water; but if nitrobenzine is present, the residue is brown, crystalline, and insoluble in water.

The action of sodium on the essence may also be utilised as a test:—Pure almond essence, when treated with sodium, gives white flocks; if nitrobenzine should be present, the sodium is immediately covered with yellow or brown flakes, according to the amount of adulteration; if the percentage rises as high as 0·30 to 0·50, the whole liquid after a minute becomes thick and opaque. (*Dragendorff*.)

However, the action of potash alone on a sample adulterated with nitrobenzine is tolerably conclusive. If one grm. of the essence is treated in a test-tube with half its weight of pure caustic potash, a yellow coloration is produced, should the essence be pure; but if nitrobenzine be present, the tint soon becomes yellowish-red, and at the end of a minute green. On the addition of a little water, the mixture separates into two layers, of which the lower is yellow and the upper green, the latter changing in the course of a day into red. Most foreign *etheral oils* may be detected by the bisulphite of soda test:—If a little of the pure essence be dropped into a warm solution of this salt, of from 1·24 to 1·26 specific gravity, shaken, and then diluted with hot water, it is fully dissolved; other essences, on the contrary, are insoluble.

ANNATTO.

§ 364. Annatto is a colouring-matter obtained from the seeds of the *Bixa orellana*, chiefly prepared in Brazil and Cayenne. Although not used itself as a food, it enters into several articles of consumption, and has been employed to colour milk, butter, and cheese.

Microscopical Characters.—When annatto is examined by the microscope, the outer red portion presents an almost homogeneous appearance, and the surface of the seed proper consists of narrow or elongated cells or fibres vertically disposed, while the inner white portion consists of cells filled with starch corpuscles, well defined, of medium size, and resembling in the elongated and stellate hilum the starch granules of the pea and bean.

In commercial unadulterated annatto, but little structure is met with. Portions of the outer cells may be seen; and in those specimens, which in the course of their preparation have not been subjected to the action of boiling water, a few starch granules may be noticed.

Since this is the case with annatto itself, we can the more easily detect the presence of most foreign vegetable substances, such as turmeric powder, the starch of wheat, rye, barley and sago flours. The salt and alkali present in the annatto generally greatly alter the appearance of the turmeric. Most of the colouring-matter of the cells is discharged, so that the starch corpuscles contained within them become visible. Loose starch granules of turmeric may also be frequently seen, and in consequence of the action of the alkali much enlarged.

§ 365. *Chemical Composition of Annatto.*—Dr. John found the pulp surrounding the fresh seed to consist of 28 parts of colouring resinous matter, 26.5 of vegetable gluten, 20 of ligneous fibre, 20 of colouring extractive matter, 4 formed of matters analogous to vegetable extractive, and a trace of spicy and acid matters. The colouring-matter consists of a red substance—*bixin*, associated with a yellow, *orellin*; the latter has been as yet but little studied.

Bixin, $C_{15}H_{18}O_4$, when pure, is an amorphous, resinous, cinnabar-red substance. It is scarcely soluble in water and bisulphide of carbon, soluble in about 89 parts of cold, and in 25 of hot, alcohol; in 345 parts of ether, in 93 of chloroform, and also in the caustic and carbonated alkalies. The alcoholic solution is coloured orange-red by lead acetate, brownish-yellow by chloride of mercury and acetate of copper, brown-red by chloride of iron, and it is precipitated yellow by stannous or stannic chloride; concentrated sulphuric acid produces a deep blue. A solution of *bixin* in an alkaline liquid, on neutralisation with an acid, gives a precipitate of the resin, and in this way it may be purified.

§ 366. *Adulterations.*—Annatto is one of the most adulterated substances met with in commerce, the adulterants being both organic and inorganic. The organic substances used are—turmeric, rye, barley, and wheat flours, as well as aniline dyes. These latter should always be looked for in the solution of annatto in oil or alkalies so extensively used for colouring dairy products. The inorganic—sulphate of lime, carbonate of lime, salt, alkali, an oily substance (probably soap), red ferruginous earths (mostly Venetian red), red lead, and copper.

When large quantities of flour and lime are used, the colour of the annatto is so reduced that it becomes necessary to use salt, alkalies, and the red earths, to restore it to its original standard. Salt heightens the intensity of vegetable reds, hence its use. Lead is probably introduced into the annatto through the Venetian red used. Copper is added to prevent the annatto becoming attacked by fungi.

The following is an analysis by one of the writers of a fair commercial sample.

No. 1. The sample was in the form of a paste, colour deep red, odour peculiar, but not disagreeable:—

Water,	24.2
Resin,	28.8
Ash,	22.5
Starch and extractive matter,	24.5
	100.0

The following is an analysis of an adulterated sample. It was in the form of a hard brown cake, texture hard and leathery, odour disagreeable:—

Water,	13.4
Resin,	11.0
Ash—consisting of iron, silica, chalk, alumina, and salt,	48.3
Extractive matter,	27.3
	100.0

§ 367. The *Analysis of Annatto*, as may be gathered from the preceding description, principally resolves itself into a determination of the ash and an estimation of the resin. The former is determined in the usual way, the latter by exhausting the sample by boiling alcohol, getting rid of the spirit by evaporation, and then redissolving the extract thus obtained in an alkaline solution, and finally precipitating the nearly pure resin by careful neutralisation with an acid.

OLIVE OIL.

§ 368. Olive oil is derived from the fruits of the olive tree, *Olea Europæa sativa*, of which there are many varieties; in Italy alone, three hundred.

The constants of olive oil, as compared with others, are set forth in the following table:—

TABLE LX.—CHEMICAL AND PHYSICAL CONSTANTS OF OLIVE OIL AND SOME OILS USED FOR THE ADULTERATION OF OLIVE OIL.

	Specific Gravity, Water=1.		Solidifying Point.		Insoluble Acids, Hehner Value.	Saponification, Mgms. KOH.	Iodine Value.	Mauméné Thermal Test.
	At 15° C.	At 100° C.	Fat.	Fatty Acids.				
Olive oil,914 to .917	.862	-2° C.	21° to 22° C.	94 to 96	186 to 196	79 to 88 generally 81 to 85	34° to 35° C., mean value 35°
Cotton-seed oil,922 to .930	.867 to .868	0° to -1°	31° to 36°	96.0	191 to 196	100 to 117	75° to 90°
Arachis oil,916 to .922	.864 to .869	-3° to -7°	24° to 30°	98.0	190 to 196	87 to 100	44° to 67°
Rape oil,914 to .917	.856 to .864	-2° to -10°	17° to 18°	95.0	175 to 179	97 to 102	49° to 51°
Sesame oil,922 to .924	.868 to .871	-4° to -6°	18° to 22°	96.0	188 to 191	103 to 112	63° to 68°
Maize oil,921 to .922	.868 to .869	-10° to -15°	14° to 16°	96.0	189 to 190	111 to 119	79° to 86° cocoa-nut olein.
Cocoa-nut oil,923	.873	16° to 20°	16° to 20°	88.0	246 to 268	8 to 9	26° to 27° (Allen).
Poppy oil,923 to .924	.873	-18°	15° to 16°	95.4	190 to 195	133 to 138	86° to 88°
Castor oil,9600 to .9736	..	-10° to -18°	176 to 182	82.4 to 86	46° to 47°
Curcas oil,911 to .920	..	-8°	..	87.9	210 to 230	100 to 127	..

N.B.—None of the above (save cocoa-nut oil) give more than sufficient volatile acid to neutralise 1 c.c. of d. n. soda when submitted to the Reichert-Wolny test. Cocoa-nut oil, however, gives a definite Reichert-Wolny value of about 7 c.c., and castor oil about 3 c.c. of d. n. soda per 5 grammes of fat.

The composition varies within certain limits according to the species and locality, and as to whether the oil has been expressed or extracted by solvents. The finest edible oils are known under the names of 'Virgin oil,' 'Provence oil,' 'Aix oil'; and a second quality is sold under the name of 'finest Tuscan cream.'

Olive oil contains about 28 per cent. of solid glycerides consisting of palmitin, stearin, and a small quantity of arachin. The liquid portion of the oil contains olein, linoleic acid, and unsaponifiable matter (cholesterol) in the following proportions:—Olein, 66 per cent.; linoleic acid, 5 per cent.; unsaponifiable matter, 1 per cent.

Free fatty acid is always present in quantities varying from 1 per cent. to 25 per cent.

Most specimens of olive oil when examined by the spectroscope show a chlorophyll spectrum.

TABLE LXI.—MIXED FATTY ACIDS.

	Melting Point.	Saponification Value, Mgrms. KHO.	Iodine Number.
Olive oil,	24° to 27°	192	85 to 90
Sesamé oil,	24° to 26°	199	109 to 112
Rape oil,	16° to 19°	170 to 177	99·8 to 103
Cotton oil,	30° to 36°	204 to 208	111 to 113
Arachis oil,	28° to 31°	199·7	96·5 to 103
Poppy oil,	20° to 21°	199	96 to 103

§ 369. *Adulterations of Olive Oil.*—Olive oil commands a good price, and is therefore extensively adulterated with other oils, especially with cotton and arachis oils, but sesamé, rape, poppy-seed, lard, curcas and castor oils have been used. Green oils should always be examined for copper.

Refraction of Olive Oil.—Jean's refractometer may be applied for the detection of certain adulterants. Oliveri gives the following values:—

	Deviation.
Olive oil,	0° to 2°
Sesamé oil,	15°·5
Arachis oil,	7°·5
Cotton-seed oil,	18°
Colza oil,	26°·5
Poppy-seed oil,	28°·5
Castor oil,	41°–44°

Zeiss' refractometer may be used with advantage for the detection of cotton-seed oil. The table (LXII., p. 511) gives the normal readings of olive and cotton-seed oils at various temperatures.

J. H. Long¹ has examined a number of oils and determined the specific gravity at 24° C. as compared with water at 4° C., the oils being weighed in a vacuum. He has also determined the refraction at 20° C., using the sodium light; the following are his chief results:—

	Specific Gravity.	Refractive Index.
Olive oil,	0·9130	1·4608
Cotton-seed oil,	0·9191	1·4732
Sesamé oil,	0·9191	1·4740
Mustard oil,	0·9121	1·4742
Castor oil,	0·9589	1·4791
Lard oil,	0·9122	1·4686
Peanut oil,	0·9173	1·4717

¹ *Amer. Chem. Journ.*, x. 392–405.

The variations caused by temperature he found to be 0·00068 specific gravity, and 0·0004 refraction, for each rise of 1 degree C. in temperature.

TABLE LXII.—READINGS ON ZEISS REFRACTOMETER OF OLIVE AND COTTON-SEED OILS.

Temperature (Centigrade).	SCALE READING—		Temperature (Centigrade).	SCALE READING—	
	Olive Oil.	Cotton-seed Oil.		Olive Oil.	Cotton-seed Oil.
35·0	57·0	61·8	25·5	62·4	67·5
34·5	57·2	62·1	25·0	63·0	67·9
34·0	57·4	62·3	24·5	63·3	68·2
33·5	57·7	62·5	24·0	63·6	68·5
33·0	58·0	62·8	23·5	63·9	68·8
32·5	58·3	63·0	23·0	64·2	69·1
32·0	58·5	63·2	22·5	64·5	69·4
31·5	59·0	63·6	22·0	64·8	69·7
31·0	59·2	64·0	21·5	65·1	70·0
30·5	59·4	64·2	21·0	65·4	70·3
30·0	59·9	64·5	20·5	65·7	70·6
29·5	60·1	64·9	20·0	66·0	70·9
29·0	60·3	65·1	19·5	66·3	71·2
28·5	60·6	65·3	19·0	66·6	71·5
28·0	60·9	65·7	18·5	66·9	71·8
27·5	61·1	66·0	18·0	67·2	72·1
27·0	61·5	66·5	17·5	67·5	72·4
26·5	62·0	67·0	17·0	67·8	72·7
26·0	62·2	67·3	16·5	68·1	73·0

The following are the chief special tests for foreign oils :—

Sesamé Oil.—20 c.c. of the oil are mixed in a test-tube with 10 c.c. of hydrochloric acid (specific gravity 1·19), which contains in solution 0·1 gm. of sugar; on shaking and allowing to stand a minute, should sesamé oil be present, there will be a more or less crimson colour developed.

The red colour in some olive oils adulterated with sesamé appears in the oil rather than in the aqueous layer.

According to V. Villavecchia and G. Fabris¹ sesamé oil may be thus detected: 0·1 c.c. of a 2 per cent. solution of furfural is placed in a test-tube and 10 c.c. of the oil to be tested added, followed by 10 c.c. of HCl, specific gravity 1·19. The tube is shaken for half a minute; a red colour denotes sesamé oil.

Sesamé, if present, will also alter the 'constants' (see Table LX., p. 509), its presence tending to raise the specific gravity, the iodine number, and the thermal value; but a small percentage of sesamé will only affect these slightly.

Arachis Oil.—Arachis oil is so similar to olive oil in its general reactions that the only way in which it can be demonstrated with certainty is the isolation and estimation of arachidic acid. It is true that olive oil also contains arachidic acid, but in so small a quantity as not to be estimable from such quantities as 10 grms. of oil; whereas it is in about the proportion of 5 per cent. in arachis oil. To isolate arachidic acid the method of Renard is in use. 10 grms. of the oil are saponified, the soap decomposed by hydrochloric acid, the fatty acids dissolved in 90 per cent. alcohol, and

¹ *Zeit. f. angewandte Chem.*, 1893, 505, 506.

precipitated by lead acetate. (Lewkowitsch shortens the process by neutralising the soap by acetic acid, and then precipitating direct with lead acetate.) The lead salts are extracted with ether, which does not dissolve lead palmitate and lead arachidate. These latter are warmed with hydrochloric acid, the fatty acids allowed to solidify, and separated from lead chloride. The solid fatty acids are dissolved in 50 c.c. of hot 90 per cent. alcohol. If arachidic acid be present, crystals of the acid are formed on cooling; they have a definite form, and melting-point of 71° to 72° C., they may also be identified by dissolving known arachidic acid in a similar quantity in hot alcohol, and comparing with them the melting-point and microscopical characters of the crystals extracted from the oil. The crystals should be weighed, and the weight corrected by adding a number representing the arachidic acid still held in solution. 100 c.c. of 90 per cent. alcohol dissolve 22 mgrms. at 15° C. and 45 mgrms. at 29° C. The quantity of arachis oil present is found approximately by multiplying the weight of the crystals by 20, calculating that arachis oil contains about 5 per cent.

De Negri and Fabris have examined different mixtures of olive and arachis oils, and have obtained the following results (the arachis oil used containing apparently 4.78 per cent. of arachidic acid):—

TABLE LXIII.

Sample containing		Arachidic Acid found.			Arachis Oil.
Olive Oil.	Arachis Oil.	Weight of separated Crystals.	Crystals in Solution.	Total.	
		Grm.	Grm.	Grm.	Per cent.
70	30	0.107	0.0315	0.1385	29.08
80	20	0.0605	0.0315	0.0920	20.24
85	15	0.0385	0.0315	0.070	14.00
90	10	0.0200	0.0315	0.0515	10.30

In two other experiments with 10 per cent. arachis oil and 90 per cent. olive oil only unweighable crystals were obtained, and in a third a quantity equal to 9.54 per cent. arachis oil; hence, when operating on 10 grms. the limit of detection appears to be 10 per cent. Should, therefore, qualitative evidence be obtained of small quantities of arachidic acid, from 25 to 50 grms. of the oil must be saponified to get quantitative results.

Cotton-seed Oil.—This is detected by the alteration it produces in the specific gravity, in the refractometer reading by the higher iodine number, by the higher melting-point of the fatty acids, by Becchi's nitrate of silver test, and by Halphen's test.

For Becchi's test two solutions are required—viz., an alcoholic solution of silver nitrate, AgNO₃, 1 grm.; alcohol (98 per cent. by volume), 200.0 c.c.; ether, 40 c.c.; nitric acid, 0.1 grm., and a solution of colza oil, 15 parts, in 100 parts of amylic alcohol. The colza oil should be 'cold drawn,' and only slightly coloured. To apply the test, 10 c.c. of the oil to be examined are mixed with 1 c.c. of the silver nitrate solution, and then from 8 to 10 c.c. of amylic alcohol solution of colza oil are added; the mixture is shaken up, and heated in a water-bath for five or ten minutes.

If cotton-seed oil is present, there is produced a brownish colour or turbidity of varying grade from light maroon to black. A small quantity of cotton-seed may give no reaction.

Halphen's Test.—Carbon bisulphide containing about 1 per cent. of sulphur in solution is mixed with an equal volume of amylic alcohol. 3 c.c. of the sample and 3 c.c. of the reagent are mixed in a test-tube and heated for fifteen minutes in a bath of boiling brine. If no reaction takes place add another 1 c.c. of the reagent and heat for ten minutes; if there is no reaction add another 1 c.c. and heat again. A red or orange coloration indicates cotton-seed oil.

The unsaponifiable matter in olive oil is cholesterol; in other oils, such as cotton-seed oil, phytosterol; hence Salkowski's process, already described in the article on *Lard*, is applicable to the detection of foreign oils, especially cotton-seed oil in olive oil.

Rape Oil.—This may be detected by the character of the 'constants,' especially by the melting and solidifying points of the fatty acids, the lower saponification and the higher iodine numbers.

Poppy-seed oil is chiefly indicated by the higher specific gravity and the high iodine number.

Castor Oil is soluble in every proportion in glacial acetic acid and in absolute alcohol. Olive oil is only slightly soluble in absolute alcohol, 3.6 in 100.

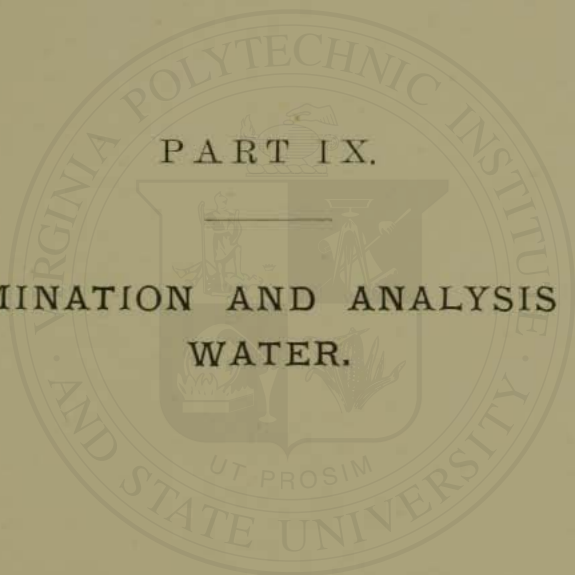
Curcas Oil will be indicated by a high iodine value and a high saponification value. It differs from castor oil in being insoluble in acetic acid. Treated with nitric acid and copper, an intense reddish brown colour is produced.

Lard Oil may be detected by the odour of lard on warming and by the high melting-point of the fatty acids.

It may be finally stated that experience has shown that, save a few special tests (such as Becchi's for cotton oil and the colour test for sesamé), reliance must be mainly placed on a careful determination of the specific gravity, of the melting and solidifying points of the fatty acids, the refraction, the iodine number, and the saponification values for the detection of adulterations of olive oil.

PART IX.

EXAMINATION AND ANALYSIS OF
WATER.



PART IX.—EXAMINATION AND ANALYSIS OF WATER.

§ 370. Ordinary drinking water is water containing dissolved gases, mineral salts and organic matter in solution or suspension, and usually various living organisms. The researches of the analysts in the examination of water have for their object the unveiling and determining the nature, and when possible, the origin of these substances. The nature of the foreign matter contained in water depends upon the purity of the air through which the water has fallen as rain, and upon the nature of the various soils or surfaces with which the water has come in contact. On this account it is important that the analyst should obtain, as far as possible, a full history of the sample to be examined, *i.e.* whether it is surface water, shallow well water, or deep well water, and the nature of the soil from which the supply is obtained. This latter information is of especial importance in enabling the analyst to draw inferences from the nature and quantity of the mineral matter present; thus, water which has simply flowed over *Metamorphic* or *Igneous rocks*, as for instance the water of Loch Katrine, differs but little from rain water. The total solids are very low and they contain little or no organic matter. Water from the *chalk*, if taken from deep wells, contains, as a rule, very little organic matter, but is specially characterised by its great temporary hardness, due to calcium carbonate held in solution by carbonic acid.

Water from Magnesian Limestone is as organically pure as that from chalk, but is not so suitable as a drinking water or for domestic and manufacturing purposes, on account of its high total hardness and the large amount of magnesium sulphate it contains rendering it permanently hard.

Water from Oolitic Limestone differs but little from water from chalk, except that this limestone is not such an efficient filtering material, and owing to its liability to become cracked or fissured, there is, especially with shallow wells, more chance of its contamination.

Water from Sandstones varies very considerably in organic purity and in the quantity of dissolved mineral matter. It is, as a rule, fairly hard, containing carbonate and sulphate of calcium. A considerable quantity of sodium chloride is also usually present.

Water from Clay is usually organically impure, small in quantity, and of great permanent hardness.

Water from Peat contains a large amount of organic matter, but need not, on that account, be dangerous for drinking purposes. It contains but little dissolved mineral matters.

It must be remembered that the water from deep wells, as often as not passes through a number of different geological strata, and so takes up various substances from each, its final composition being the result of a complicated series of changes.

The routine examination of ordinary samples of water comprises the following:—

- I. EXAMINATION BY THE SENSES.
- II. CHEMICAL METHODS.
- III. BIOLOGICAL METHODS.

I. EXAMINATION BY THE SENSES.

Water that is evidently turbid, that possesses an odour and an unpleasant taste, *requires no analytical processes to condemn it* as unfit for drinking in its unaltered condition; such waters may, however, be often greatly improved by filtration through sand, or other treatment. A water that even possesses any one of the enumerated bad qualities will, as a rule, be found to hold in solution sufficient impurities to make it decidedly objectionable. Most drinking waters, when looked at, or tasted, or smelt, without special aids to the senses, have neither colour nor odour; on the other hand, all water, if viewed through a sufficiently deep stratum, possesses colour.

Colour.—To ascertain the colour of water, it is usual for analysts to be provided with a colourless glass tube, at least 2 feet in length, having the ends closed with plate glass, and a small opening in the side of the tube through which to pour the water. The purest waters have the slightest tinge of blue; the next in order of purity have a just distinguishable shade of green. Decided green tints, London fog hues, amber yellow, and brown tints are those possessed by waters tinged with peat, containing suspended matters, of second-class composition, or those of considerable impurity.

Messrs. Crookes, Odling, and Tidy, in their report on the London water-supply for 1881, describe an ingenious 'colour meter,' consisting of two hollow wedges filled, one with a brown and the other with a blue solution. Any desired combination of green and blue may be made by sliding the wedges across each other in front of a circular aperture in a sheet of metal, and thus imitating the tint of water under examination; each prism is graduated from 1 to 50, the figures representing the thickness in millimetres at that particular part of the prism.

A far more scientific method is to fix two right-angled prisms, the reflecting surface of each being in opposite directions, in front of the divided slit, and then to fill a 400-c.c. tube, such as is used for saccharimetry, with the water under examination, and a second 400-c.c. tube with distilled water, and by means of two sources of light of equal luminosity, examine by the methods described (p. 60, *et seq.*) the different parts of the spectrum, and tabulate out the absorption of the water as compared with that of distilled water.

Smell.—Half a litre or more of the water is warmed in a large corked or stoppered flask to 38° C. [100° F.]; a long glass tube three-quarters of an inch in diameter is now inserted, and the water sucked up once or twice so as to wet the side of the tube thoroughly; then, without taking the tube out of the flask, one nostril is applied to the orifice of the tube, the other closed by the finger, and deep inspirations or 'sniffs' taken.

Another simpler plan is to warm a quantity of the water, without removing the stopper, up to the temperature given, then shake, remove the stopper, and smell; a putrid odour denotes decomposing animal or vegetable matter. If the sample is much polluted by fresh sewage, a

urinous odour is not unfrequently distinct. But, again, it may be specially noted that water quite unfit to drink may have no odour, hence the usefulness of the test is limited. A positive smell teaches volumes—a negative result is of little value.

Taste.—A few waters, and a few only, have a decided taste. It is scarcely to be recommended that analysts should taste samples derived from fever-stricken localities; but, on the other hand, when there is no suspicion of the samples having been the cause of any illness, the palate may detect some not unimportant peculiarity.

II. CHEMICAL METHODS.

A.—Preliminary Qualitative Chemical Examination.

§ 371. The qualitative examination of drinking water is not of much value save when applied to a water considerably polluted. It may be restricted to the direct addition of Nessler solution, when a water containing a considerable amount of free ammonia will give an amber colour or even a precipitate, and to the testing for nitrites, nitrates, and metals.

Detection of Nitrites.—The best tests for nitrites are—(1.) The meta-phenylenediamine test; (2.) Meldola's test; (3.) The naphthylamine test; (4.) The zinc iodide starch test.

(1.) *The Meta-phenylenediamine Test*; the solution is made by dissolving 8 grammes of the hydrochloride in 1 litre of distilled water, and adding a little hydrochloric acid. A c.c. of the solution added to 50 c.c. of water acidified with sulphuric acid (1 : 3 made very faintly pink with permanganate of potash), strikes a pale straw-yellow to a deep orange-red, according to the quantity of nitrite present. By comparing with known dilute solutions of nitrite this method gives fair quantitative results. The limit of the reaction is, according to R. Warington, 1 part of nitrite in ten millions of solution.

(2.) *Meldola's Test* is a solution of para-amido-benzene-azo-dimethylaniline in water, acidified with hydrochloric acid, strength about .02 per cent. The reagent is added to the water to be tested, and the whole is acidified with sulphuric acid, warmed from ten to fifteen minutes on the water-bath, and then alkalisied by ammonia. If no nitrites are present, the tint is first a pale citron-yellow, changing on the addition of the acid to a rose-pink, and reconverted by ammonia or alkalies to citron-yellow. If, however, nitrites are present, the acid-liquid becomes of a salmon colour, and the final tint on the addition of ammonia is that of a sap green with small quantities of nitrite, and of a sapphire blue with larger quantities; on acidifying the liquid it changes back to a salmon colour. The tints are not permanent, but soon fade. The reaction, according to Warington, succeeds in a dilution of 1 part in one hundred millions.

(3.) *The Naphthylamine Test.*—Reagents:—Solution I.: 1 grm. sulphanic acid, 14.7 c.c. glacial acetic acid, 285 c.c. distilled water.

Solution II.:—2 grm. α -naphthylamine, 14.7 c.c. glacial acetic acid, 325 water. Keep the solutions separate and mix equal volumes before using. To 50 c.c. of the sample add 2 c.c. of the mixed solution; a minute trace of nitrite strikes a pale pink; but, if much nitrite be present, a deep ruby colour is produced, and the solution becomes turbid from the precipitation of colouring-matter. The reaction may be made to give good quantitative results. Warington found the reaction distinct with a

dilution of 1 part of nitrite in five hundred millions of water, but in these great dilutions the reaction requires to go on for an hour or two before the colour is developed.

(4.) *The Zinc Iodide Starch Test.*—To 100 c.c. of the water are added 2 c.c. of strong sulphuric acid, and a little zinc iodide and starch solution (see *Appendix*); in the presence of nitrites a blue colour appears.

Detection of Nitrates.—Since no natural water is absolutely free from nitrate, the quantitative estimation of nitrates is alone of importance. Most tests are common to nitrites and nitrates. Nitrates, in the absence of nitrites, can be readily tested for by acidifying, adding a little zinc, and then testing with the zinc iodide starch test. (For the brucine and other tests, see pp. 523–525.)

Detection of Poisonous Metals.—The poisonous metals most frequently found in ordinary waters are lead or copper, and the most sensitive test for these is to acidify and add either ammon. sulphide or sulphuretted hydrogen water; a dark tint or precipitate denotes either lead or copper, or both; by adding potassic cyanide solution, if the dark hue be due to copper sulphide only, the solution clears; if to lead sulphide only, it remains dark; if to a mixture, it partially clears. To confirm a copper reaction, test with potassic ferrocyanide; this produces a brownish colour or precipitate, according to the quantity of copper present.

A convenient reagent is the senior author's cochineal test. A solution of cochineal in spirit strikes with a neutral or alkaline solution containing dissolved lead or copper, a deep mauve-blue to a red with a faint blue tinge, according to the amount present. The test will indicate $\frac{1}{10}$ of a grain of lead per gallon in ordinary drinking-water, and by the aid of comparison lead or copper-free solutions, smaller quantities of these metals may be detected.

F. P. Venable¹ has found 4.48 grains of zinc carbonate per gallon in a spring water. Heaton²—in a Welsh spring, after flowing through half a mile of galvanised iron pipe—found 6.41 grains per gallon of $ZnCO_3$. Dr. Frankland has recorded a case of poisoning from a zinc-polluted well-water.

Potassic ferrocyanide, added to the filtered and acidulated water containing zinc, gives either a light white cloud or heavy precipitate, according to the amount present.

B.—Quantitative Analysis.

§ 372. A complete examination by chemical processes embraces the following determinations:—

1. Total solid residue, and solids in solution and suspension.
2. Estimation of the halogens, chlorine, and occasionally iodine, and in a few cases bromine.
3. Phosphates.
4. Nitrates and Nitrites.
5. Estimation of dissolved oxygen.
6. Sulphates.
7. Oxygen absorbed from permanganate.
8. Free and albuminoid ammonia.
9. Hardness.
10. Alkalinity.
11. Organic Analysis—Estimation of organic carbon and nitrogen.
12. Mineral analysis of water.

¹ *Chem. News*, vol. li. 18.

² *Ib.*, xlix. 85.

The ordinary analyses, sufficient in most cases to pronounce an opinion as to the fitness of a water for drinking purposes, embrace only 1, 2, 3, 4, 7, 8, and 9.

1. *Total Solid Residue.*—By the total solid residue of a water is meant the substances in solution, as determined by drying up a measured portion, and weighing the dried residue; if the water contain suspended matters, it should first be filtered, and a portion of the clear filtered liquid taken. The amount suitable for this determination depends upon the characters of the water. The soft Devon waters yield a very insignificant residue from 100 c.c., and to obtain trustworthy results, at least a quarter of a litre is required; while, on the other hand, with calcareous waters, good results may be always obtained from 100 c.c. With waters the characters of which are unknown, it will be best to operate on a quarter of a litre, or (if working with English measures) one-twentieth of a gallon. The water may be placed in a platinum dish, and evaporated down to a small quantity over a ring burner, taking care that the liquid in no case boils or even simmers; the last drops are driven off on the water-bath. It is recommended by the Society of Analysts to heat the residue up to 104° C. (220° F.) in the air-bath, and then to cool under a desiccator; but with waters of unknown composition, it will be best to weigh the residue, which has not been exposed to a greater heat than 100° C., for it is always open to the chemist to expose the residue thus obtained to higher temperatures. The examination of the solid matters by the eye will often reveal much. Iron gives a coppery lusture to the dish, manganese a green to the ash, and very pure waters leave a residue almost white. The dish with its contents is next heated to a low redness, by the aid of a good Bunsen's burner, furnished with a rose, and then cooled and weighed. Note should be taken of any blackening or scintillation. The loss of weight is returned as loss on ignition, and this final residue is dissolved in the manner to be described, and used for the qualitative determination of the phosphates. The suspended solids are best estimated by evaporating to dryness, and weighing the residues of known quantities of the water unfiltered and of the water filtered through a hard filter, the difference in the weights equalling suspended solids.

2. *Estimation of the Halogens.*—The estimation of chlorine is an essential part of the ordinary scheme of water analysis; that of iodine is rarely (perhaps too rarely) performed, while so few waters contain an estimable amount of bromine, that it need not be here described.

Chlorine.—Chlorine exists in ordinary waters in the form of sodic chloride; occasionally a small portion of the total chlorine is combined with potassium, and sometimes quite a quantity of calcium chloride is present. It may be estimated volumetrically by a standard solution of silver nitrate (*see Appendix*), using as an indicator neutral potassic chromate. Nitrate of silver in presence of potassic chromate and alkaline chlorides (when the solution is neutral) first uses up or decomposes all the chlorides, and then attacks the chromates. Chloride of silver being white, and chromate of silver being red, the formation of silver chromate is indicated immediately by a red colour. At least 100 c.c. of ordinary water (or, if grains are worked with, 140 grains) are to be taken for the determination of chlorine. With much-polluted waters, with those near the seashore or other places in which the ground is impregnated with salt, such a quantity may be inconvenient, and it will be necessary then to dilute with distilled water, taking of the diluted liquid a known quantity. In any

case, the water is put into either a white porcelain dish or a beaker standing on a white slab. 1 c.c. of the chromate solution (or 15 grains) is added to the water, and the standard solution run in from a graduated burette or pipette.

The exact termination of the process is best observed through a glass cell, in which a little pale chromate solution has been placed. Since the eye, looking thus through yellow light, is very sensitive to the red rays, it may be necessary—especially where great accuracy is required—to repeat the determination in the following way:—The water from which the red colour of the silver chromate cannot be discharged by stirring, is rendered again whitish-yellow by the cautious addition of a very dilute solution of common salt. A fresh portion of water is titrated in a fresh dish or beaker, side by side with the former; in this way the first permanent difference of colour can be observed. The results may be expressed in chlorine as chlorides, or it may be returned as common salt; for the latter purpose multiply the chlorine by the factor 1.648, or more exactly by 1.64788. The following short table may facilitate calculation:—

Chlorine.	Sodium Chloride.	Chlorine.	Sodium Chloride.
1.	1.648	6.	9.888
2.	3.296	7.	11.536
3.	4.944	8.	13.184
4.	6.592	9.	14.832
5.	8.240	10.	16.480

In a general laboratory it may be more convenient to estimate chlorine by Volhard's method,¹ because the solutions are in that case applicable to the determination of the halogens in acid solutions, in which they have hitherto been estimated by weight.

For ordinary purposes the solution of silver nitrate should be decinormal (that is, 17 grms. per litre), but for water analysis the strength given in the Appendix is most convenient.

Beside the silver solution, are required—(1) A solution of ammonium sulphocyanide; (2) strong nitric acid which has been boiled; (3) a saturated solution of ferric alum. The sulphocyanide solution is diluted so that 10 c.c. shall exactly equal 10 c.c. of the silver.

This is easily effected as follows:—10 c.c. of the silver solution, acidified with nitric acid, are run into a beaker, and a drop of the iron-alum solution added; the sulphocyanide solution is now run in from a burette until a single drop gives a red colour; the number of c.c.s. used is noted, and the sulphocyanide diluted accordingly.

The titration of the chlorides in water is done on similar principles; an excess of silver solution is added to 100 c.c. of water, the mixture well shaken, and a few drops of alum solution added, and then the excess of silver solution determined by carefully running in the sulphocyanide until the red colour denotes the end of the reaction.

Iodine.—M. Chatin² has upheld the theory, that goitre is caused by waters insufficiently iodised—a proposition which cannot be considered proved. However, although M. Chatin has failed to convince the scientific world of the truth of his theory, he has done good service in showing how easy the detection and estimation of iodine in water really is, and in demonstrating the fact that most waters contain it in appreciable

¹ "On the Estimation of the Halogens," by A. Percy Smith. *Analyst*, Jan., 1886. *Liebig's Annalen*, etc. 24.

² *Compt. Rend.*, t. xxxv., xxxix.

quantity. The process which M. Chatin used in his researches was: To evaporate one or two litres of the water to dryness with pure potassic carbonate, to calcine very moderately this dry residue, and then to extract with strong alcohol of 94 per cent. This alcoholic solution is again evaporated to dryness, and moderately calcined; the last residue is dissolved in a very little water, and will show all the reactions of potassic iodide. It is colorimetrically estimated by palladium. A solution of chloride of palladium gives a distinct colour with an infinitesimal quantity of iodine; hence it is only necessary to have a standard solution of potassic iodide, containing say 1 milligramme in 100 c.c., and to estimate it precisely on the same principles as detailed (*post*) for ammonia. Mr. Marchand,¹ pursuing the same line of researches, has preferred to precipitate from ten to twenty litres with nitrate of silver, collect the precipitate which may contain the chloride, iodide and bromide of silver, and dissolve it in sodic hypsulphite. The silver is now thrown out of this solution by sulphuretted hydrogen, and the solution, when freed from silver sulphide, evaporated to dryness with a little hydro-potassic carbonate. In this way he obtains the chloride, bromide, and iodide of potassium. When the residue is perfectly dry, it is extracted with strong alcohol of 85 per cent.; the alcoholic liquid is evaporated to dryness at a temperature not exceeding 75° C. This last residue is again taken up by alcohol, and treated similarly to the potassic iodide obtained by Chatin's method.

3. *Phosphates*.—The residue after ignition is treated with a very little nitric acid, and evaporated to dryness; this treatment renders the silica insoluble. It is now again dissolved in a few drops of nitric acid, some water added, and filtered through an exhausted filter. If the filtrate is more than 5 c.c., it should be concentrated to a smaller bulk, and its own volume of the molybdic solution (see *Appendix*) added. The solution thus treated, and gently warmed, gives a more or less deep colour or a decided precipitate, according to the amount of phosphoric acid present. It may be estimated colorimetrically by a known solution of sodic phosphate, but this with no great accuracy. To make a gravimetric estimation of phosphates, save in polluted waters, may require several litres, and will seldom repay the trouble. Hence phosphates may be returned in a qualitative manner as '*traces*,' with a feeble colour; '*decided evidence*' with a darker colour, and as '*estimable amount*' if there should be a precipitate. The Analyst Committee have adopted '*traces*,' '*heavy traces*,' and '*very heavy traces*,' as expressing three degrees of phosphate contamination. Such phrases are convenient, though somewhat paradoxical, and the authors therefore prefer the more logical form of expression given above.

4. *Estimation of Nitrates and Nitrites*.—The several methods in use for the estimation of nitrates and nitrites (see also *ante*, p. 519) may be arranged under the following heads:—

- (1.) Colorimetric methods of estimation.
- (2.) Estimation of nitrates by conversion of the nitrate into ammonia.
- (3.) Estimation of nitrates by decomposing the nitrate into nitric oxide, and measuring the gas.
- (4.) Indirect estimation by means of indigo.
- (5.) Estimation by the deficiency of hydrogen evolved from the action of sulphuric acid on iron powder.

(1.) *Colorimetric Methods*—(a.) *The Brucine Method*.²—1 grm. of brucine is dissolved in 100 c.c. of alcohol; 10 c.c. of the water are evaporated to dryness; and from 0.5 to 2 c.c. of the brucine solution added to the residue and six drops of a saturated solution of oxalic acid. The colour must be a bright red; if it be brown, an insufficient quantity of brucine has been added; if pink, too large a quantity has been added. So it is best (in waters of unknown nitric content) to put three or four

¹ *Compt. Rend.*, xxxv.

² See a paper by J. West Knight, *Analyst*, 1881, 56-58.

separate 10 c.c. in porcelain dishes and to evaporate them down with various quantities of the brucine solution (such as 0.5 c.c.; 1.0 c.c.; 1.5 c.c.; and 2 c.c.), and to make any quantitative determination on that which has the proper red colour. The colour is imitated by a standard test made by evaporating 10 c.c. of a solution containing 72.1 mgrms. of potassic nitrate per 100 c.c. with 3 c.c. of brucine solution and six drops of the oxalic acid solution, and diluting up to 100. This test solution is diluted, if necessary. The original water treated in the same manner may, after evaporating and diluting, require filtration.

G. Lunge and A. Lwoff¹ do not evaporate to dryness, but apply the test to the water direct; they acidify with sulphuric acid, and the contents are heated to between 70° and 80° C. until the liquid assumes a permanent greenish-yellow; then this colour is imitated by a standard solution. They find that nitrites do not interfere.

(b.) *The Diphenylamine Method.*—A solution of diphenylamine sulphate strongly acidified with sulphuric acid strikes a blue colour with nitrates. This colour can be imitated by a standard solution of nitrate, and the nitrates be thus estimated on colorimetric principles.

This test is also well suited for quantitative spectroscopy.

Hans Settegärt² has worked out the absorption factors of diphenylamine as applied to small quantities of nitrates. Reducing his notation to wave-lengths, the following are his chief results:—

TABLE LXIV.

Wave-lengths.	Absorption Coefficients.	
	0.120 to 0.500 Light Strength.	0.500 to 0.780 Light Strength.
616.6 to 543.2	.000002822	.000002913
543.2 to 532	.000003162	.000003266
532 to 521.6	.000003606	.000003687
521.6 to 513.6	.000003758	.000003811
513.6 to 505.7	.000004226	.000004290

The values are correct for solutions containing nitric acid from 4.0 to 0.1 per 10,000. The diphenylamine must be in 0.1 per cent. solution, and, of course, the sulphuric acid must be free from nitric or nitrous acids.

(c.) *The Carbazol Test.*—Samuel C. Hooker³ has proposed carbazol as a test for nitrates; it gives a green colour with oxidising agents. The objections to its use are that chlorine and iron have to be first removed, and that the presence of much organic matter makes the results too low. Assuming that the water has been freed from chlorine by silver sulphate and contains no iron, then two c.c. of a sulphuric acid solution of carbazol are added, and the resulting green colour imitated by means of a solution of potassic nitrate.

(d.) *Phenol and Resorcinol.*—D. Lindo⁴ has experimented with phenol in alcohol as well as with resorcinol. A .0005 per cent. solution of N₂O₃.

¹ *Zeit. angew. Chem.*, 1894, 345-350.

² Beiträge zur quantitativen Spectral-analyse. *Annalen der Physik. u. Chemie.* Bd. vii., 1879.

³ *Analyst*, Sept., 1889.

⁴ *Chem. News*, lviii, 1-3, 15-17, 28-29.

yields a faint pink band, green below, with a 10 per cent. solution of phenol in weak alcohol; if 0.5 c.c. of the solution be mixed with a single drop of the test in a test-tube, and then 2 c.c. of sulphuric acid run down the tube so as to form bands of colour, N_2O_3 , in the proportion of one 2,000th, gives very intense green and red bands.

Phenol is also a good test for nitrates in the presence of free hydrochloric acid.

Resorcinol, in 5 per cent. solution, is a good test for nitrites, so small a quantity as 1 in 500,000 giving (after standing four hours) a pink colour if acidified with sulphuric acid. One in 10,000 gives a pink colour at once. Iodides, bromides, and very large quantities of chlorides interfere with these tests, and must be removed by silver sulphate.

Both the above are adapted for quantitative colour estimation.

(2.) *Estimation as Ammonia.*—A convenient method of obtaining the nitrogen of nitrites and nitrates in the form of ammonia is decidedly by the aid of the 'copper-zinc couple.' The results are, however, of no value unless all the processes are carried out with extreme care. This method was first proposed by Gladstone and Tribe, and afterwards worked out in detail by M. Whitley Williams.¹ It appears that the copper-zinc couple decomposes nitrates first into nitrites, and then the nitrites into ammonia; nitrites are present to the last, and when all the nitrites have disappeared, it is certain the conversion into ammonia is complete. A low temperature, alkalies, alkaline earths and their carbonates, retard the reaction, while carbon dioxide, mineral acids, oxalic, phosphoric and generally all acids and common salt, as well as elevation of temperature, increase the reaction. In practice, a temperature of 24° C. is recommended as easily attainable.

Manufacture of the Copper-Zinc Couple.—Pieces of clean zinc-foil, about 3 inches by 2 inches, are immersed in a 3 per cent. solution of cupric sulphate; the zinc rapidly becomes coated with metallic copper. When a sufficient coating is obtained, the solution is poured off, and the couple well washed with water, finally drained, and the water for analysis poured on to the couple. It is best to do these processes in one and the same stoppered bottle. The water may nearly fill the bottle, and the stopper may be inserted, for there will be no gas evolved until the nitrates are entirely decomposed. The water thus treated is put in a warm place, and if the action is allowed to go on all night, the ammonia will be ready for estimation in the morning. The quantity to be taken for the estimation of nitrates according to this plan may be a quarter of a litre, or, if English measures are used, say 5 ounces. To very hard waters the addition of a little oxalic acid is recommended. In any case where there is doubt whether the conversion into ammonia is complete, one of the tests (page 519), should be used; and if there is evidence of nitrous acid, the water must be left for a longer period. If the water possesses colour interfering with, or matters precipitated by, Nessler's reagent, it must be distilled in the ordinary way, and the ammonia estimated in a fractional part of the much-diluted distillate. In most cases this is unnecessary, and by taking a measured quantity of the water and diluting it considerably, a fairly correct colorimetric estimation can be made by the direct addition of the Nessler reagent to the water thus diluted. It will be necessary to subtract from the amount of ammonia found, that which has been determined to exist in the water as ammonia. The ammonia derived from nitrates and nitrites must be expressed either as nitrogen or as nitric acid.

¹ *Journal of Chemical Society*, March, 1881.

The Aluminium Process.—The metal aluminium, when acted on by a caustic alkaline solution, decomposes nitrates into ammonia. A solution of

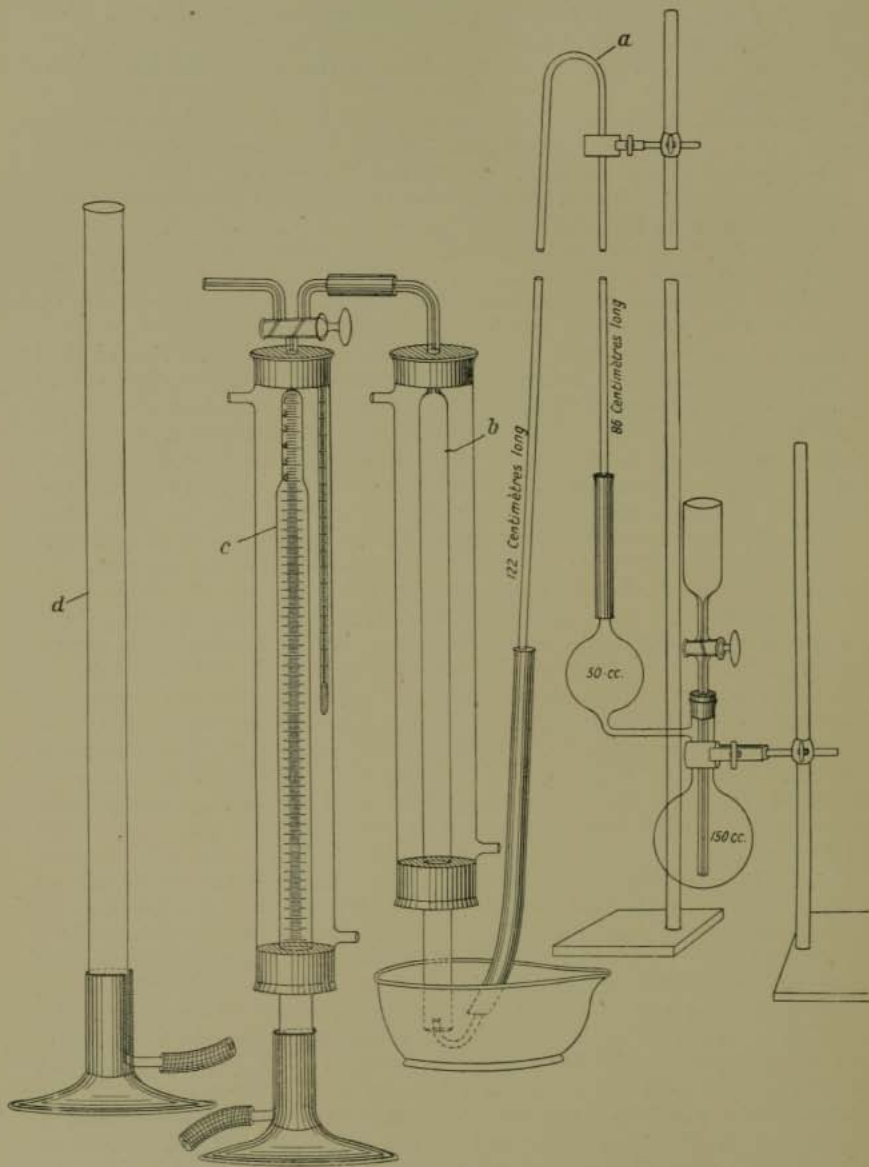


FIG. 72.

soda of about 10 per cent. is prepared perfectly free from nitrates, by dissolving bit by bit metallic sodium in water. Any convenient quantity (such, *e.g.*, as 100 c.c., or 2,000 grains) of the water is placed in a suitable

retort, which is fitted in an air-tight manner to a condenser, terminating in a flask as in the arrangement figured at page 387. An equal quantity of the soda solution is added, and the whole boiled until free from ammonia; the retort is cooled, and the aluminium-foil dropped into the liquid; the whole is left over-night, and in the morning heat is applied to the retort, and the ammonia distilled over, and estimated in the usual way.

*Ulsch's Method of Estimation of Nitric Acid by Reduction to Ammonia.*¹
—Half a litre of the water is concentrated down to 15 c.c.; this is transferred to a flask of about 300 c.c. capacity, 5 grms. of reduced iron (*Ferrum redactum*) added, and 10 c.c. of diluted sulphuric acid, specific gravity 1.35. The liquid is heated to gentle boiling for some five or ten minutes, diluted with 100 c.c. of distilled water, alkalised by from 20 to 25 c.c. of soda lye (specific gravity 1.35), and distilled into 25 to 30 c.c. of d. n. acid; the difference in the titre of the d. n. acid, before and after the distillation, gives the data for measuring the nitric acid converted into ammonia. A correction by blank experiments should be made for impurities in the reagents.

Example.—Half a litre of water was treated in the way described; and distilled, after being made alkaline, into 30 c.c. of d. n. acid; at the end of the operation, the acid used, instead of 30, 14 c.c. of d. n. soda, $14 - 30 = 16$, which 16 of d. n. soda is exactly equivalent to 16 c.c. of d. n. sulphuric acid which have been neutralised by ammonia. Since 1 c.c. of d. n. acid is equal to 5.4 mgrms. of nitric anhydride (N_2O_5); $16 \times 5.4 = 86.4$ mgrms.; hence the litre contained twice that amount, or 172.8 mgrms. [12.1 grains per gallon].

(3.) *Estimation of Nitrates and Nitrites as Nitric Oxide—Crum Process.*
—Strong sulphuric acid acting on nitrates or nitrites in the presence of mercury, decomposes the nitrates or nitrites, and the whole of the nitrogen is evolved in the form of nitric oxide.

Half a litre of the water is evaporated to dryness, the nitrates extracted by hot water, the hot-water extract evaporated down to 1 c.c., and the liquid transferred to the decomposition tube, which is a short tube about 3 inches long, constricted at one end, and furnished with a cup and stopcock; open at the other, and having a bore easily closed with the thumb.

This tube is filled with mercury, inverted, and clamped in a mercury trough with the cup uppermost; it is now easy to transfer the solution of nitrates by pouring the solution into the cup, and cautiously opening the stopcock. The vessel in which the filtrate has been concentrated is then rinsed into the cup with pure strong sulphuric acid, and ultimately one and a half times the volume of the concentrated nitrate solution of strong sulphuric acid is worked into the tube by carefully opening the stopcock. No air must be allowed to gain admittance. Should gas be immediately evolved, it is carbonic dioxide, and must be got rid of, for nitric oxide is not at once evolved. On the mixture of sulphuric acid and nitrate having been transferred into the tube, the lower end is closed by the thumb, and the tube shaken so as to mix up the acid and the mercury, when the gas in a short time begins to come off, and considerable pressure may have to be exerted.

When the reaction is complete, the contents are transferred to any gas apparatus and measured, or as the tube itself is now graduated, the simplest method is to plunge the tube into a vessel of distilled water, cool it to the temperature of the water, adjust it so that the level of the water inside and outside the tube is the same, and measure it direct. The small absorption of the nitric oxide by water requires no correction. Of course, the value of the divisions on the tube must be ascertained with accuracy.

Every two volumes of nitric oxide equals one volume of nitrogen. The weight of the nitrogen is obtained from Table LXIX.

(4.) *Schulze-Tiemann Method.*—The method is based on the fact that a strong solution of ferrous chloride in the presence of hydrochloric acid liberates nitric oxide. The reaction is not interfered with by the presence

¹ *Zeit. f. analyt. Chem.*, xxx. 175; xxxi. 3 2. Tiemann-Gartner's *Handbuch*, Braunschweig, 1895.

of organic matter. The apparatus hitherto used has neither been convenient nor capable of giving results of the highest accuracy, but by adopting the improvements suggested by one of the authors, the process is now one of the best for the estimation of nitrates.

A quarter of a litre of ordinary water is evaporated nearly to dryness, the insoluble carbonates filtered off, and the residue washed with hot water. The filtrate and washings now contain all the nitrates and nitrites originally present in the water in a fairly concentrated form. This liquid is transferred to a tubulated flask (see fig. 72), closed by a perforated caoutchouc stopper, carrying a thistle head funnel, the tubule being connected with a long delivery tube partly of glass and partly of thick walled rubber. The glass tube is carried up well above the barometric height to which mercury ascends and thus makes an efficient valve. The tube delivers any gas into a pneumatic trough, and from thence into a suitable measuring apparatus.

The liquid is first just acidified with hydrochloric acid and boiled to concentrate it still farther and to get rid of any carbonates, the steam being allowed to escape into the air; after boiling a little time the nozzle of the delivery tube is inserted under the tube previously filled with mercury and the boiling continued until air or gas ceases to be evolved. It is best to take away the flame for a minute or so and thus allow condensation and the creation of a vacuum in the flask; on now applying heat any trace of air or carbon dioxide remaining is effectually got rid of. When this is the case a hot acid solution of ferrous chloride (made by dissolving 15 grms. of ferrous chloride in 100 c.c. of hydrochloric acid) is added through the funnel, care being taken to allow no bubbles of air to enter the flask.

The boiling is now continued. Should gas be at once evolved, it is not nitric oxide derived from nitrates, but from nitrites, and when this first evolution of gas has ceased, the gas should be transferred to the measuring-tube *c*, for the purpose of estimation; by lowering the pressure tube *d*, and opening the two-way stopcock so as to allow a communication between *b* and *c*, the gas can be drawn over without the water, which has condensed from the steam and has collected at the top of *b*. The nitric oxide is not evolved from nitrates until the solution is quite concentrated. The last trace of the gas is obtained by allowing the flask to cool for a few minutes, thus producing a strong vacuum. On reboiling, the gas is obtained without difficulty, and, ultimately, the steam issuing from the flask makes the peculiar metallic click which is a sign of freedom from gas.

The measurement of the gas is made in the usual way. After correction to normal temperature and pressure, 4.1 c.c. of NO gas equals 10 mgrms. of nitric anhydride, N_2O_5 .

(5.) *Indigo Process*.—This process is based on the decolorisation of indigo when nitrates or nitrites are decomposed by strong sulphuric acid.

When certain kinds of organic matter are present, the results are entirely without value. On the other hand, with careful working the test is correct with the great majority of waters, and as a means of rapidly determining the nitrates in unknown samples, with a view to their determination by other more exact processes, it is very useful. Four mgrms. of sublimed indigotin are digested for some hours with five times their weight of Nordhausen oil of vitriol; the liquid is diluted with water, filtered, and brought to the volume of 2 litres.¹ A normal nitre solution is made by

¹ See Warington's excellent paper, *Journ. Chemical Society*, Sept. 1879, p. 579; also, Frankland's *Water Analysis*, p. 31.

dissolving 1.011 gm. of pure potassic nitrate in 1 litre of water. From this solution, solutions of $\frac{1}{4}$, $\frac{1}{8}$, $\frac{1}{16}$, $\frac{1}{32}$, and $\frac{1}{64}$ normal are prepared. An assay is now made by mixing, say 20 c.c. of the nitre solution with any amount of the indigo solution deemed sufficient, in a wide-mouthed flask of 150 c.c. capacity. Oil of vitriol is run into a test-tube, the volume being equal to the united volumes of the indigo and nitre. The contents of the test-tube are then suddenly tipped into the flask, and the flask transferred to a chloride of calcium bath maintained at 140° C. If the solution of indigo is insufficient, the liquid will be suddenly decolorised; if it is too much, no bleaching will take place, the liquid still retaining its blue colour. In either case a fresh determination will be requisite, and by doubling or halving the amount of indigo for the next experiment, as the case may be, the operator will soon find the limits, and five or six experiments will standardise the solution. In every instance a quantity of sulphuric acid, equal to the united volumes of indigo and water, must be used; the indigo solution should be diluted so as to be about equal to the nitre solution. As it is found that the quantity of indigo consumed is not precisely in proportion to the nitric acid present, but diminishes as the nitrate solution becomes more dilute, the further standardising of the indigo solution by the more dilute solutions of nitre already alluded to is necessary. The results may be thrown into a table as follows. (See Table LXV.)

TABLE LXV.—VALUE OF INDIGO, IN NITROGEN, FOR DIFFERENT STRENGTHS OF NITRE SOLUTION.

Strength of Nitre Solution Used.	Indigo required.	Difference between Amounts of Indigo.	Nitrogen Corresponding to 1 c.c. of Indigo.	Difference between the Nitrogen Values.	Difference in the Nitrogen Values for a Difference of 1 c.c. in the Amount of Indigo.
	c.c.	c.c.			
$\frac{8}{64}$ normal, .	10.00000035000
$\frac{7}{64}$ " .	8.71	1.29	.000035161	*000000161	*000000125
$\frac{6}{64}$ " .	7.43	1.28	.000035330	*000000169	*000000132
$\frac{5}{64}$ " .	6.14	1.29	.000035627	*000000298	*000000231
$\frac{4}{64}$ " .	4.86	1.28	.000036008	*000000381	*000000298
$\frac{3}{64}$ " .	3.57	1.29	.000036764	*000000756	*000000586
$\frac{2}{64}$ " .	2.29	1.28	.000038209	*000001445	*000001129
$\frac{1}{64}$ " .	1.00	1.29	.000043750	*000005541	*000004295

The method of using the table is sufficiently obvious to those who are accustomed to calculations of the kind; supposing, for example, 20 c.c. of the water used up 6.64 c.c. of the indigo; this is .5 c.c. above the nearest number in the table, viz., 6.14. Now, taking the extreme right-hand column, the difference for the nitrogen values of 1 c.c. will be found; and as there is in this case a difference of only half that quantity, halving the number gives us .000000115; this number has to be subtracted from the unit value of nitrogen found in the first column, thus:—

$$.000035627 - .000000115 = .000035512,$$

which is the nitrogen-value of each c.c. of the indigo. Hence, as we have supposed that 20 c.c. of the water decolorised 6.64 c.c. of indigo, the

nitrogen as nitric acid in parts per 100,000 is 1.179, or in grains per gallon .82 grain. If the indigo estimation of nitric acid is only a preliminary step to a further and more exact determination of the Crum method, these refinements are not necessary. The indigo solution is standardised once for all by the normal solution of nitre, and if the nitrates are either very large or very small, an allowance is made.

(6.) *Ulsch's Method of Estimating Nitric Acid by Measuring the Deficiency of Hydrogen evolved on Reduction.*—One of Ulsch's methods has been already detailed. This method is in its principle an indirect one, for what is measured is not the ammonia produced, but the deficiency in the evolution of hydrogen.

A quarter of a litre of water is evaporated down to 15 c.c. and, while still hot, filtered. The filter is washed with a little boiling water into a small measuring flask, the size depending on the amount of nitrate present, which can be approximately ascertained by a brucine or other colour test; 50 c.c. is the capacity for most waters—viz., those containing under 150 mgrms. of N_2O_5 per litre; larger flasks are used for larger quantities. The collective fluid with washings should not be more, for a 50 c.c. flask, than 40 c.c. The flask is cooled to the temperature of the room, and sufficient normal sulphuric acid added to make the contents equal to $\frac{1}{2}$ normal when water is added to fill the flask to the mark on the neck. This fluid is the 'testing fluid,' of which 10 c.c. are taken for the estimation of nitric acid. In a thin walled flask, *a* (fig. 73) the bulb of which is equal to about 30 c.c., 3 grms. of iron powder (*Ferrum pulveratum*) are placed by means of a dry funnel. The flask is closed by a caoutchouc stopper containing three holes; one of which carries the twice bent 3 mm. wide tube, *b*, the one end of which goes almost to the bottom of the flask, the other end is connected by means of a bit of rubber tubing to a short piece of glass tubing—the rubber is supplied with a screw clip. The middle hole of the caoutchouc stopper carries the funnel, *c*, which is provided with a stopcock; the end of the stem of the funnel is bent slightly, so that fluids will run down the neck of the flask. The third hole carries a right-angled tube, connected by narrow rubber tubing, to the gas-measuring apparatus. This consists of a burette, *f*, divided into tenths of a c.c., the upper end of which is provided by a T-tube, *e*, and the lower end connected by rubber tubing to the spherical funnel, *g*.

The first operation is to fill the whole system with hydrogen, water is poured into *g*, the stopcock at *e* opened, and the funnel raised until the whole of *f* is filled up to the stopcock. Into the funnel, *c*, are poured 12 c.c. of dilute sulphuric acid (33 c.c. strong sulphuric acid to a litre of water) and some 10 c.c. of it allowed to flow on to the iron, by opening the stopcock; an evolution of gas commences, and in about two minutes ceases. Before it actually ceases the stopcock, *e*, is closed, and the clip at *b*. The funnel, *g*, is lowered as much as possible. Now the rest of the acid is allowed to flow into the flask, and the funnel washed twice with a 2 per cent. copper solution, which is also allowed to flow into the flask, care being taken not to allow the admittance of air. The flask is now warmed in a beaker of water at 60° C. for two minutes without shaking; then the flask is shaken without interruption for another two minutes; after which the flask is cooled as quickly as possible to the temperature of the air of the room. The funnel, *g*, is now raised, and the clip on *b* cautiously opened. The fluid contents of the flask, *a*, are under the pressure of the hydrogen forced through *b*, and may be received in a beaker.

If the twice right-angled tube, *b*, is properly adjusted, none of the iron will escape; the clip is closed while there is still a small column of liquid in the tube. After thus emptying the flask, its temperature is adjusted to that of the air, and the excess of gas is got rid of by carefully opening the stopcock at *e*; while the water level of *f* and *g* is adjusted to the zero point of the burette, *f*.

The apparatus is now ready for the purpose of ascertaining once for all how much hydrogen is developed by 10 c.c. of $\frac{1}{2}$ normal sulphuric acid acting on the iron in the *absence* of nitric acid.

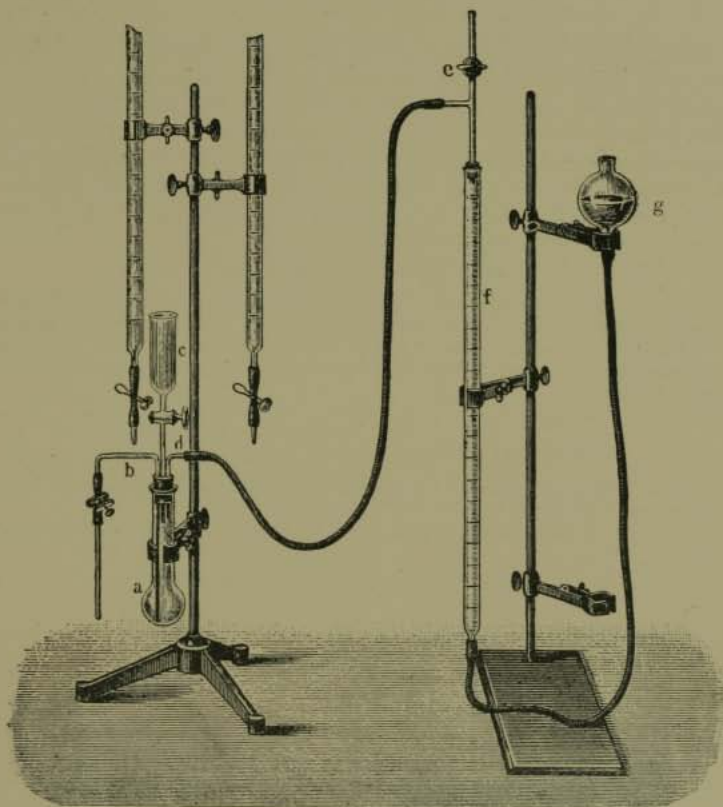
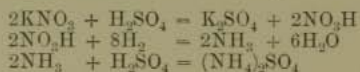


FIG. 73.

From a burette exactly 10 c.c. of the acid are allowed to flow into the funnel attached to the small flask, *a*, while *g* is lowered to the level of the table; then the acid is allowed to flow slowly into the flask. From a second burette 10 c.c. of 2 per cent. copper sulphate are added to the flask, always avoiding entrance of air. The flask is warmed to 60° C., shaken, and finally cooled exactly as detailed previously. The amount of gas, on adjusting the water level of *g* to the water level of *f*, is read, the temperature of the water in *g* being taken, and also the height of the barometer, and the volume of gas reduced to

normal temperature and pressure—always subtracting 20 c.c. from the known volume of the flask, that being occupied by the 20 c.c. of liquid. The flask may now be emptied as before, and a second or third determination made, the mean being taken as the standard.

To estimate nitric acid the process is precisely the same, save that 10 c.c. of the testing fluid are taken instead of 10 c.c. of $\frac{1}{2}$ normal acid. In the presence of nitrates there will be a deficiency in hydrogen, from which the nitric acid can be calculated—the following reactions taking place:—



or 8 molecules of hydrogen are required for the reduction of one molecule of nitric acid, besides which, for the decomposition of an equal molecule of nitric acid from its combination with a base, and the combination of the ammonia formed with a farther molecule of sulphuric acid, 2 molecules of hydrogen are required. A molecule of saltpetre then will in respect of a normal hydrogen volume show a deficit of 10 molecules of hydrogen. A deficit of 1 c.c. of hydrogen will, therefore, be equal to 0.90416 mgrm. KNO_3 or 0.4834 mgrm. nitric anhydride (N_2O_5).

An example of this method may here be given.

50 c.c. of test fluid derived from a quarter of a litre of spring water were prepared according to the method described in the test. 10 c.c. of the latter yielded as a mean of three estimations (subtracting the 20 c.c. of fluid in the flask) 20.2 c.c. of hydrogen, the barometer being 741 mm. (temp. $18^{\circ}5$), and the corresponding tension 15.9 mm. This, reduced as follows:—

$$\frac{20.2 \times (741 - 15.9)}{(1 + 0.0030 \times 18.5) \times 760} = 18.05 \text{ c.c. of hydrogen.}$$

The normal hydrogen volume in the absence of a nitrate was 21.62 c.c.; the hydrogen deficit is, therefore, $21.62 - 18.05 = 3.57$ c.c.

$3.57 \times 0.4833 = 1.725$ mgrm. of nitric acid in 50 c.c. of water; therefore (a litre) contains

$$\frac{1.725 \times 1000}{50} = 34.5 \text{ mgrms. } (\text{N}_2\text{O}_5).$$

The process is made inaccurate by the presence of iron or carbonates. The carbonates of the earths as well as any iron are, however, by evaporating to a small volume as described, fully separated. Should alkaline carbonates be present a little gypsum must be added to the water before evaporation.

Small quantities of nitrites introduce no material error; in the presence of much nitrite advantage is taken of the fact that the iron copper couple reduces nitrites in the cold. The test fluid is, therefore, freed from nitrites by digesting the test fluid with copper solution and iron in the cold. A correction may also be made by estimating the nitrites by one of the colour tests for nitrites already described.¹

(7.) *Estimation of the Dissolved Oxygen in Water.*—The amount of air dissolved in water is dependent on temperature and pressure, the amount of oxygen in the air of pure water having a mean value of 34.91 per cent. The following table gives the absorption coefficient of air in water; and also the amount in c.c. of oxygen that a litre of water will dissolve. The oxygen, in what may be called the water atmosphere, is diminished by the

¹ Nitrites may also be estimated by Piccini's method, which depends upon the fact that if a solution of nitrite is treated with urea, and thoroughly acidified with sulphuric acid and gently heated, all the nitrogen of the nitrite is evolved as gas. The acidification must take place in a vacuum, or, at all events, atmospheric air must be excluded, otherwise the reaction is not quantitative, some of the nitrite passing into nitrate. Nitrites may be also estimated by titration with potassic permanganate.

activity of micro-organisms; hence its determination is valuable. It is especially useful in the investigation of the polluted water of a river, particularly if determinations of oxygen are made on the spot. On the other hand, water sent from a distance to an analyst, and probably one or two days on the road, cannot have its dissolved oxygen directly determined with advantage. The senior writer, in such a case, proceeds as follows:—Half a litre of the water is shaken up in a large Winchester quart until saturated with air; the dissolved oxygen is determined in half of this—that is, $\frac{1}{2}$ of a litre; the other $\frac{1}{2}$ of the litre is put on one side, a layer of xylene having been poured on the surface of the water to exclude air, and again titrated for oxygen at the end of forty-eight hours; the difference will have a direct relation to the organic matter and micro-organisms in the water. This method may be called “the differential method of estimating oxygen in water.”

TABLE LXVI. — SHOWING THE ABSORPTION COEFFICIENT OF AIR DISSOLVED IN WATER AND THE C.C.'S OF OXYGEN WHICH MAY BE OBTAINED FROM 1 LITRE OF WATER (AFTER BUNSEN).¹

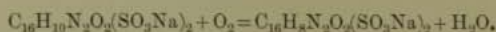
Temp. C.	Absorption Coefficient of Air dissolved in Water (c.c. of Air dissolved in 1 c.c. of Water).	The Number of c.c. of Oxygen which 1 Litre of Water will dissolve.
0	0·02471	8·63
1	0·02406	8·34
2	0·02345	8·19
3	0·02287	7·98
4	0·02237	7·80
5	0·02179	7·60
6	0·02128	7·43
7	0·02080	7·26
8	0·02034	7·10
9	0·01992	6·95
10	0·01953	6·81
11	0·01916	6·69
12	0·01882	6·57
13	0·01851	6·46
14	0·01822	6·36
15	0·01795	6·26
16	0·01771	6·18
17	0·01750	6·11
18	0·01732	6·05
19	0·01717	5·99
20	0·01704	5·95

Preusse and Tiemann have arranged a convenient apparatus, by means of which the dissolved air is boiled out of water into soda lye, which has been completely deprived of its gases by boiling. The oxygen in the collected gas is determined either by exploding with hydrogen or by absorbing it with alkaline pyrogallate.

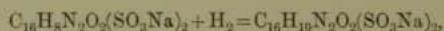
Schutzenberger and Risler cause the dissolved oxygen to act on sodium

¹ *Gasometrische Methoden*, 2nd Edit., p. 387. Winkler has also published values for the amount of oxygen dissolved in water; his numbers closely correspond to those of Bunsen's for temperatures 16° to 20° C., but are more than 1 c.c. higher for lower temperatures.

indigo-white-disulphonate in excess, and change a portion into sodium indigo-blue-disulphonate, thus—



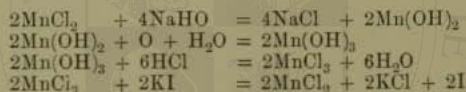
The salt is changed back again by a titrated solution of sodium hydro-sulphite, according to the equation—



the end of the reaction being sharply indicated by the change of colour from blue to yellow.

Both the above processes and their various modifications demand more or less complicated apparatus, and are not adapted for estimations on the banks of streams or anywhere else, save in a laboratory; the writers, therefore, prefer a modification of Winkler's method; it has great accuracy, and may be performed almost anywhere.

The essential fact on which Winkler's method is based is that manganese hydrate absorbs oxygen; a third hydroxyl being added in the presence of oxygen and water to the two hydroxyls in manganese hydrate—that is to say, $Mn(OH)_2$ become $Mn(OH)_3$. This last hydrate, by excess of HCl , is transformed into manganese trichloride, two molecules of which react with two molecules of potassium iodide, setting free two atoms of iodine; so that for every 16 parts of oxygen 254 parts of iodine are set free; the iodine set free is, therefore, titrated and converted by calculation into oxygen. The equations setting forth the general reactions are as follows:—



The following solutions are required:—

- (1.) 10 grms. potassium iodide dissolved in 100 grms. of 33 per cent. pure soda solution.
- (2.) Solution of 80 per cent. manganous chloride (free from iron).¹
- (3.) Hydrochloric acid (specific gravity 1·16 to 1·18).
- (4.) Starch solution.
- (5.) Solution of thiosulphate—the iodine value of which is known; each c.c. should be equivalent to a centinormal iodine solution—that is to say, 0·000127 l.

The temperature of the water and the height of the barometer being first ascertained, 250 c.c. of the water are covered with xylene to the depth of an inch, and 1 c.c. of the alkaline iodide solution is allowed to flow in through a pipette, the end of the pipette being held below the xylene; next 1 c.c. of the manganous chloride solution is allowed to flow in. The mixture is stirred with a glass rod until the contents are completely mixed and the flask put on one side for a little time. Then 3 to 5 c.c. of hydrochloric acid are added; the precipitate dissolves, and the iodine is set free; this is titrated by the thiosulphate, using as an indicator starch. If the thiosulphate is equal to centinormal iodine solution, each c.c. equals 0·0000798 grm. oxygen, or 0·0558325 c.c. oxygen; if the c.c.s. of water

¹ The solution may be freed from iron by boiling and precipitating with soda; the filtrate is acidified with HCl , evaporated to a syrup, and then crystallised.

used = V , the c.c. of thiosulphate = n , then the content of oxygen in a litre of water is obtained by the following calculation:—

$$\frac{0.055825 \ n \times 1000}{V}$$

The number thus obtained must, of course, be reduced to standard pressure and temperature. A reference to the table on p. 533 will show whether (at the temperature of the experiment) the water is below the standard or not.

A correction is usually necessary for most waters, especially those that contain nitrates, organic matter, and other impurities. For this purpose 1 c.c. of the manganous chloride solution is mixed with half a litre of distilled water, alkalisied with 1 c.c. of a 33 per cent. solution, shaken, and the brown precipitate collected on a small filter. The precipitate is dissolved in hydrochloric acid, and the solution diluted to half a litre. Two separate portions (each 100 c.c.) of this solution are taken, and mixed, the one with 100 c.c. of distilled water, the other with the water to be tested. After a few minutes, a few crystals of potassium iodide are added to both mixtures, and the iodine separated from each estimated by titration. The difference of the values equals the amount to be added. In waters of this kind the titration of oxygen is best done by adding the manganese chloride solution and simply 33 per cent. soda lye, the potassium iodide being finally added in crystals; the following is an example:—

250 c.c. of the water of the river Exe just below Exeter, at 15° C. and 760 mm., treated in the method stated, set free iodine equal to 12 c.c. of centinormal thiosulphate.

The correction was obtained by mixing 100 c.c. of the manganous dichloride solution with 100 c.c. of distilled water, and by treating 100 c.c. of the river water in the same way, adding to each potassium iodide, and titrating the iodine set free. The distilled water gave 7 c.c., the river water 4 c.c.; the difference is, therefore, 3 c.c. Hence, the content of oxygen at 15° C. of a litre of Exe water is

$$\frac{(12 + 3) \times 0.055825 \times 1000}{250} = 3.35 \text{ c.c. O.}$$

For most practical purposes it is not necessary to reduce the c.c. of oxygen thus found to normal temperature and pressure; hence, in such a case as the above, it would suffice to report that the water contained 3.35 c.c. oxygen at 15° C., or about half the normal quantity.

Rideal and Stewart¹ have recently proposed to destroy nitrites and organic matter by adding enough $\frac{N}{10}$ acid permanganate to the water to give a faint pink colour after standing 10 minutes. .1 c.c. more than the right amount of permanganate ascertained by experiment is introduced into the bottle together with 1 c.c. of strong H_2SO_4 ; the water is then added, and the excess of permanganate removed by adding $\frac{1}{2}$ c.c. of a 2 per cent. solution of neutral potassium oxalate; the estimation is then made in the ordinary way. A larger proportion of soda is required to balance the sulphuric acid added.

Ramsay and Homfray² have devised the following process:—The water (in a tube) is introduced into a special form of colorimeter,³ contact with the air being prevented by a layer of paraffin oil. The comparison tube is filled with water saturated with oxygen at a known temperature. Both tubes are treated with the same quantity of cuprous chloride dissolved in HCl just before use; an excess of ammonia is then added. After a short time the colours are compared and the usual calculations made.

(8.) *Sulphates*.—Any convenient quantity of the water, carefully meas-

¹ *Analyst*, June, 1901.

² *J. Soc. Chem. Ind.*, 1901, 20.

³ This colorimeter can be obtained from Messrs. Townson and Mercer, Bishopgate Street.

ured, is acidified with hydrochloric acid, and heated nearly to boiling; while hot, some solution of chloride of barium is added, so as to be in slight excess, and the solution kept near the boiling point for some time. The sulphate of barium is allowed to settle, collected on a filter, dried, ignited, and weighed: one part of baric sulphate equals $\cdot 134333$ of sulphuric acid.

An estimation of sulphates in water can also be made on colorimetric principles. To 100 c.c. of water, barium chloride is added in slight excess and then the water is acidified by hydrochloric acid, the turbidity produced is now imitated by a dilute solution of sodic sulphate tested with the same reagents. It is best to observe the turbidity by looking through the colorimeter at a black porcelain plate.

(9.) *The Forchammer, Oxygen or Permanganate Process.*—The principle of this process is the abstraction of oxygen by the organic elements of the water, and the estimation of the oxygen thus abstracted. $\cdot 395$ grm. of potassic permanganate is dissolved in a litre of water, which gives a solution containing 1 mgrm. in every 10 c.c., or, if working in grains and septems, 2 grains of permanganate in 1,000 septems of water, equalling $\cdot 01$ grain of available oxygen in 20 septems. This is the standard solution.

The determination is now usually made, as recommended by the Society of Analysts, in two stages, on two equal quantities of water, viz.—(1) The amount of oxygen absorbed in fifteen minutes, and commonly due to nitrites, or, at all events, substances very readily oxidisable; and, (2) the amount of oxygen absorbed in four hours. The time for this last determination used to be given as three hours, but the four-hours' period is preferable; and even then it is easy of proof that, if the water be allowed to stand, there still remain matters capable of being oxidised. The temperature is an important factor, for numerous experiments have shown that the amount of oxygen consumed varies greatly at different temperatures. The Analysts' Society have adopted $26^{\circ}\cdot 6$ C. (80° Fahr.), and in order to ensure uniformity this temperature is here recommended. It is, however, probable that better and more uniform results would be attained by boiling the water and permanganate for an hour. In some interesting experiments by Messrs. Wigner and Harland,¹ river water, to which a known quantity of pure sugar had been added, was found to have absorbed more oxygen at the end of two hours, at $37^{\circ}\cdot 7$ C. (100° Fahr.), than during six hours at $15^{\circ}\cdot 5$ C. (60° Fahr.), and almost as much as during six hours at $26^{\circ}\cdot 6$ C. (80° Fahr.). Similarly, river water contaminated by a known quantity of urine used up equal quantities of oxygen when acted upon by permanganate for six hours at $26^{\circ}\cdot 6$ C. (80° Fahr.), as it did when the process was accomplished in two hours at $37^{\circ}\cdot 7$ C. (100° Fahr.). The actual operation is as follows:—

Two stoppered flasks are taken, and a quarter of a litre of the water put in each [or 3,500 grains]. The bottles, with their contents, are immersed in an air-bath until the temperature rises to $26^{\circ}\cdot 6$ C. (80° Fahr.), then 10 c.c. [or 100 grains] of dilute sulphuric acid [1 : 3] are added, and the same quantity of the standard permanganate. One of the bottles is taken out at the end of a quarter of an hour, and two or three drops of potassium iodide solution added to remove the pink colour. After thorough admixture, there is run into it from a burette a solution of sodium hypo-

¹ "On the Action of Permanganate on Potable Waters at Different Temperatures." *Analyst*, March, 1881, p. 39.

sulphite, until the yellow colour is nearly destroyed; then a few drops of starch water are added, and the hyposulphite added until the blue colour is just discharged. The value of the sodium hyposulphite must be determined by titrating in a similar manner 10 c.c. of the standard permanganate in distilled water. At the end of four hours the other bottle is removed and titrated in exactly the same way. Should the pink colour diminish very rapidly during the four hours, another measured quantity of permanganate must be added.

If A be taken to express the amount of hyposulphite used for a blank experiment with pure distilled water, B the water under examination, and a the amount of available oxygen in the quantity of permanganate originally added: then, the oxygen consumed by the quantity of water operated on would be

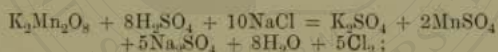
$$\frac{(A - B) a}{A}$$

or, in actual figures, 10 c.c. of a permanganate solution, equivalent to 1 mgrm. of oxygen, were added to a quarter of a litre of distilled water, and to the same quantity of a sample under analysis. The distilled water used 40 c.c. of hyposulphite, the water 15 c.c. at the end of four hours. Then the oxygen consumed by the quarter litre was 0.625 mgrm., according to the equation

$$\frac{40 - 15 \times 1.0}{40} = 0.625 \text{ mgrm.}$$

or per litre, 2.5 mgrm. [175 grain per gallon].

Blair prefers to use the oxygen process at 100° C., and has published an elaborate series of experiments proving that permanganate and sulphuric acid in the absence of organic matter or reducing agents can be boiled for two hours without change; a serious objection is that a boiling permanganate of potash solution will decompose chlorides, setting chlorine free, thus—



but with waters containing up to 8 grains of chlorine per gallon, it appears that this reaction has absolutely no effect; with larger quantities of chlorine, a control may be run containing an equal amount of chlorine in the form of common salt. A great number of organic substances, when treated with permanganate at 100° C., absorb the quantity of oxygen theoretically necessary to convert the carbon into carbon dioxide, and the hydrogen into water; 100 mgrms. of cane sugar in two hours absorbed 111.2 mgrms. O, as against 111.9; strychnine, brucine, morphine, all gave the theoretical amounts; but starch came out low, *i.e.*, 86.4 instead of 118.5. The writers consider that with regard to ordinary waters the oxygen process at 100° C. is by far the most reliable, and have, for many years, used it exclusively. We are also able to confirm Kruss, that the strength of a permanganate solution may be estimated by the spectroscope by the use of the divided slit (see p. 62); those who have the necessary appliances may make use of this method in preference to titration.

The German chemists generally estimate oxygen consumed at 100° C., but only boil for ten minutes. The method of Kubel is the oxidation by a centinormal solution of permanganate in ten minutes in acid solution: that of Schulze is, first, oxidation by alkaline permanganate; then the

solution is acidified by sulphuric acid and again boiled; in each case the boiling is for ten minutes. Probably the method of Schulze is the better of the two. In either case the oxygen consumed (or, in other words, the permanganate used) is estimated by oxalic acid as follows:—

The permanganate strength is first ascertained by centinormal oxalic acid. The water which has been boiled with permanganate has its colour discharged by means of centinormal oxalic acid added in known volume and in slight excess, and then the solution of permanganate is dropped in until a weak red colour is permanent. The calculation is obvious, but it may be useful to give an example.

A centinormal solution of permanganate exactly equivalent to a centinormal solution of oxalic acid was used, and 15 c.c. of such a solution added to 100 c.c. of water; after boiling for ten minutes and then cooling, 10 c.c. of the oxalic acid solution were run in; and then to the colourless fluid it was found that 5 c.c. of the permanganate solution was necessary to just redden the colourless liquid. In all, the solution contained, therefore, 20 c.c. of permanganate, of which 5 have been used; 1 c.c. of centinormal potassic permanganate is equal to 0.08 mgrm. of oxygen; hence the 100 c.c. used 0.4 mgrm. of oxygen, equivalent to 4 parts per million.

The Oxygen Consumed Applied to the Indirect Estimation of Volatile Organic Matter.—Preusse and Tiemann¹ have submitted various waters to distillation, and have estimated the amount of oxygen consumed in the distillate. They have come to the conclusion that the products of putrefaction may be, in this way, detected, and that the process is a valuable aid to the judgment of drinking waters. Those waters which are good, reducing but small quantities of permanganate; those that are impure, large quantities.

An example of one of their experiments may be given. Half a litre of water derived from the Pankö, a dirty brook running through the north-west of Berlin, was distilled, 400 c.c. of distillate being collected in four successive fractions and boiled, after Kubel's method, ten minutes as before described. Half a litre was first distilled from the neutral water; another half litre was acidified; a third was alkalisied. In all three cases volatile organic matter was present.

	Mgrms. of Oxygen consumed by 100 c.c. of Distillate from		
	(a) Neutral Water.	(b) Acid Water.	(c) Alkalisied Water.
1	1.28	1.14	1.08
2	0.65	0.69	0.48
3	0.39	0.45	0.27
4	0.27	0.40	0.26

(10.) *Ammonia, Free and Albuminoid.*—The estimation of ammonia depends on the principle that it admits of ready distillation when it exists in the water as ammonia, provided that the water is alkaline. Since, therefore, nearly every natural water is alkaline, distillation of

¹ Tiemann-Gartner's *Handbuch der Untersuchung u. Beurtheilung der Wasser*. Braunschweig, 1895.

water is alone sufficient to expel the ammonia. If a water, by testing with cochineal, is found to be acid, then it will be necessary to add a little recently ignited carbonate of soda (or, perhaps better, a little recently burnt magnesia), until an alkaline reaction is obtained. The apparatus required for the estimation of free and albuminoid ammonia, is—

- (1.) A good large stoppered retort, fitting into a full-sized Liebig's condenser, through which a constant stream of water is running.
- (2.) Measuring-flasks, either in septems or litres.
- (3.) Cylinders made of clear glass, "Nessler cylinders," or a colorimeter.
- (4.) One or two pipettes.
- (5.) Nessler reagent (*see Appendix*).
- (6.) Standard solutions of ammonium chloride (*see Appendix*).
- (7.) Solution of alkaline permanganate (*see Appendix*).

The water is first tested with a little of the Nessler reagent; if it shows any decided colour it may be necessary to distil a very small portion, say a quarter of a litre, diluted with a sufficient quantity of pure, ammonia-free water. But if, on the other hand, there is no colour, or a doubtful one, a litre of the water should be distilled, or a fifth of a gallon. On distillation, 100 c.c., or 1,400 grains, are collected in one of the glass cylinders, and 5 c.c., or one-twentieth of its volume, of clear straw-coloured Nessler solution added. If there is any ammonia the distillate thus tested will be tinted or coloured, the colour varying from a very pale straw up to a dark amber. If the colour should be very deep, it is impossible to estimate the ammonia with even an approach to accuracy, unless the dark solution is very much diluted and made up to a definite volume, of which definite fractional parts are taken. The next step is to estimate the ammonia by imitating the colour. This is done by running into some distilled water one, two, or more cubic centimetres of the standard ammonium chloride solution, and adding exactly the same amount of Nessler solution as had been added to the distillate. The solution is now made up to precisely the same bulk as the distillate, and the liquids, thus in equal columns, compared by looking down through them on to a glass plate or white porcelain tile or slab.

Accurate estimations may be made by the special colorimeters described on pp. 60-62, but the accuracy greatly depends upon the practice of the observer, and the sensitiveness of his eye for differences of colour. There are many persons who, from some physical peculiarity of sight, can only distinguish a few shades, and even with the greatest care can make no very accurate colorimetric observation.

By graduated Nessler glasses, having taps near the bottom in order to run off a portion, as well as by colorimeters, such as Mill's and other like contrivances, "Nesslerising" is much expedited and facilitated.

Returning to the actual estimation of free ammonia, the water must be distilled in successive fractions, until no more free ammonia is detected in the distillate. This occurs generally when 150 c.c. or 200 c.c. (that is, one-fifth of the entire quantity) of the water taken has come over, then the water is ammonia-free. The next step is to estimate in the same water the albuminoid ammonia.

Albuminoid Ammonia. — When Mr. Wanklyn first published the

albuminoid process it was very generally adopted, and it may be considered as yielding quickly certain data, assisting in the final verdict of an analyst. To the water left in the retort from the estimation of free ammonia about one-tenth of its original volume of the alkaline permanganate (see *Appendix*) is added; the water is again distilled; successive fractions of the distillate are tested with Nessler, and the ammonia therein contained determined in the same way as in the free ammonia estimation. Here the analyst may occasionally have difficulty, from the circumstance that the alkaline permanganate sets free certain compound ammonias, which strike a tint with the Nessler re-agent entirely different from that given by pure ammonia. In certain cases it may, indeed, be necessary to estimate the ammonia by titrating with a feeble and very dilute acid. The free ammonia is usually returned as ammonia; the albuminoid should properly be returned as "*nitrogen as albuminoid ammonia.*" It is scarcely necessary to remind the operator that all retorts, condensers, etc., used for these estimations must be ammonia-free, and that ammonia from any analytical operation must not be allowed to contaminate the laboratory atmosphere. The most ready way to render it certain that there is no ammonia in the condenser is to acidify a little water with sulphuric acid, and then distil until the distillate is ammonia-free.

Estimation in Water of Proteid Ammonia. — L. W. Winkler has proposed a new method of estimation of proteid ammonia, and has suggested that this method may be used advantageously as a substitute for the ordinary albuminoid ammonia process. It is, however, obvious that it may also be used as an addition to the albuminoid process, any want of fair agreement between the albuminoid ammonia and the proteid ammonia as estimated by Winkler's process sometimes giving valuable insight into the nature of the nitrogenous substance present; urea, for example, yields more than ten times; hippuric acid about double, and uric acid more than double, the weight of ammonia by Winkler's process than can be obtained by distillation with permanganate. The process is simply this:—Proteid substances in solution are more or less oxidised to ammonia by persulphate of potash slightly acidulated with sulphuric acid and submitted to a heat of 100° C. for a quarter of an hour.

The following table gives the amount of ammonia obtained by Winkler, using his process as compared with the ammonia evolved by alkaline permanganate from various nitrogenous substances:—

	Proteid Ammonia per cent.	Albuminoid Ammonia per cent.	Ammonia calculated per cent.
Urea,	27·0	2·2	56·67
Hippuric acid,	7·4	3·1	9·45
Uric acid,	10·6	4·3	40·17
Theine,	15·2	15·0	26·29
Leucine,	10·2	11·3	12·98
Tyrosine,	7·9	8·0	10·30
Asparaginic acid,	11·2	11·8	12·81
Gelatine (18·3 p.c. N),	9·1	8·2	22·23
Albumen,	7·1	6·7	19·06

In each case 10 mgrms. of the substance were dissolved in 1 litre of water.

The following are the details of the process :—

Solutions required—

(1.) *Potassic Persulphate Solution.*—1 gm. of commercial persulphate is dissolved in 100 c.c. of water. Since the commercial persulphate is mostly contaminated with ammonia it is purified as follows: 15 grms. of potassic persulphate, finely powdered, are dissolved in 100 grms. of warm water (50–60° C.) and 1.5 gm. of potash added. The solution is filtered through a small wad of asbestos and allowed to stand for a few hours in a cold place. Pure crystals separate out; these are collected on a glass filter and dried in the air at ordinary temperatures. The solution rapidly changes, so that only small quantities are prepared at a time. The freshly prepared pure solution gives no turbidity with barium chloride and its reaction is neutral. A slight turbidity with barium chloride may, however, be neglected.

(2.) Approximately one-fifth normal sulphuric acid. It is sufficient to dilute 6 c.c. of pure concentrated acid up to a litre with water.

The ammonia is not distilled off, but estimated by a colorimetric method with the same water, for which purpose the following solutions are required.

(3.) *Ammonium chloride solution*, 315 mgrms. to the litre. 1 c.c. = 0.1 mgrm. NH_3 .

(4.) *Nessler's reagent.*—This must not be prepared with mercuric chloride, but with mercuric iodide, the proportions being as follows:—

Mercuric iodide,	10 grms.
Potassic iodide,	5 grms.
Sodium hydroxide,	20 grms.
Distilled water,	100 c.c.

The mercuric iodide is rubbed to a powder in a small porcelain mortar with some water washed into a flask and the potassic iodide added. The soda is dissolved in the rest of the water and only added when quite cold. This solution must be kept in the dark. It must not be used for some days, in order that the excess of mercuric iodide may crystallise out and the liquid become clear. When properly made the liquid is pale yellow, caustic, quite clear, and of sp. gr. 1.28.

(5.) *Solution of potassium sodium tartrate.*—50 grms. of the salt are dissolved in 100 c.c. of warm water, filtered, 5 c.c. of Nessler solution added, and kept in the dark. This solution is also allowed to stand a few days before use.

For the estimation of ammonia equal parts of the tartrate and of the Nessler solution are mixed.

The process itself as applied to drinking water is as follows:—100 c.c. of the water are placed in a suitable flask, 5 c.c. of the sulphuric acid and 5 c.c. of the persulphate solution added. Should the fluid not react acid, 5 c.c. more of sulphuric acid are added. The flask is now sunk in the water bath and heated at 100° C. for a quarter of an hour. At the end of that time it is removed from the bath, put in a glass cylinder, the mixed Nessler solution added (5 c.c.), and a comparison made with a second 100 c.c. of the same water by adding to it in the cold the same quantities of acid persulphate and mixed Nessler solution,—the colour being equalised by dropping from a burette a sufficiency of the chloride of ammonium solution into this last which, if any proteid ammonia exists in the water at all, will naturally be of a less deep colour than the water which has been

heated with the persulphate. The amount of ammonium chloride solution added gives the necessary data from which to calculate the proteid ammonia.

The process is not easy of application to dirty waters, to sewage or to any coloured liquid; but it is suitable for ordinary potable and for polluted waters without turbidity or marked colour.

The very best spring waters yield no proteid ammonia; polluted waters yield from .1 to more than .7 mgrm. per litre.

(11.) *Hardness*.—A. *Before Boiling*.—In a corked or stoppered bottle 1000 grains, or 100 c.c., of the water to be tested are placed. The standard soap solution (*see Appendix*) is run in, 10 grains, or 1 c.c., at a time, and after each addition the cork or stopper is replaced, and the bottle shaken violently, and observed as to whether a permanent lather forms or not. If not, then another measured quantity is run in, and so on until the desired effect is produced. Waters containing but little magnesia give a good lather, and the reaction is fairly sharp. With magnesian waters the reaction is slow, and not so easy to observe. When a lather has been obtained, it is well to repeat the experiment, and in this second assay to run in within half a division the whole of the amount of soap solution thought to be necessary; then a further portion of the soap solution is run in very gradually in tenths of a c.c., or single grains, until the lather is permanent. The hardness is expressed in degrees. However, when the hardness is more than 16°, it is not possible to estimate it in this way with accuracy, and the water under examination must be diluted with distilled water to double its bulk, and then the same quantity as above recommended taken for the estimation: in this case the number of degrees found must, of course, be multiplied by 2.

B. *Hardness after Boiling*.—A quantity of water, precisely the same in bulk as in the former experiment, is boiled briskly for half-an-hour, filtered, cooled, and made up to the original bulk with distilled water; and treated with soap solution, exactly in the same way as in the previous case.

A different method of determining hardness has been proposed by *Hehner (Analyst, May, 1883)*. 20 c.c. of normal sulphuric acid are made up to 1000 c.c., and a solution of sodic carbonate (1.06 : 1000) is prepared, an equal volume of which exactly neutralises the acid. 1 c.c. of the acid neutralises 10 mgrms. of calcic carbonate. 100 c.c. of the water are tinted with cochineal, phenacetoline, or methylorange, and titrated in the usual way with the acid. Each c.c. used indicates one degree of temporary hardness. To another 100 c.c. a measured quantity of the sodic carbonate solution is added, more than enough to decompose the whole of the soluble lime and magnesian salts. The mixed solutions are evaporated in a platinum vessel to dryness. The residue is extracted with a little hot water, filtered and titrated hot with the standard acid. The alkali added, *minus* the acid used, indicates the permanent hardness as CaCO_3 .

(12.) *Alkalinity*.—The alkalinity of water is best taken by using as an indicator an alcoholic solution of cochineal, which is not affected by carbonic acid, and strikes a beautiful crimson purple colour with a trace of alkali, a reddish yellow with acids. 100 c.c. or more are placed in a tall cylinder of colourless glass, and a decinormal hydrochloric acid is run in, drop by drop, from a burette until the colour changes to a yellowish hue. The result is expressed in terms of carbonate of lime, each c.c. of decinormal acid equalling 5 mgrms. of carbonate of lime.

(13.) *Organic Analysis of Water: Estimation of Organic Carbon and*

Nitrogen.—(1.) *Carbon.*—There are four main ways in which the carbon in a water residue is estimated—(1.) as gas; (2.) gravimetrically; (3.) nephelometrically; (4.) indirect methods.

(1.) *Carbon as gas.*—The first method (which consists in burning up the carbon into carbon dioxide, and estimating both it and the nitrogen in a suitable gas-apparatus) we owe to Dr. Frankland, who proposed and practised it as early as 1867.

Frankland's Combustion Process.—A quantity of water, varying from 100 c.c. to a litre, according to the amount of impurity suspected from other determinations [especially of the free ammonia] is evaporated to dryness with special precautions.

These precautions are mainly two—(1.) The protection of the sample from dust during the evaporating process, and (2.) the destruction of carbonates, nitrates, and nitrites, which, it is scarcely necessary to say, would greatly interfere with the results, and indeed render them valueless. Small quantities of water, such as sewage and the like, can be evaporated under any improvised cover, but for larger quantities Frankland recommended a self-filling circular water-bath, on the top of which rests a flanged copper capsule, serving as support to a thin glass dish, in which the evaporation of the water takes place. The dish is protected from dust by being covered by a tall glass shade, such as is used for statuettes. The bulk of the water for evaporation is contained in a flask, to the neck of which is a ground glass tube bent appropriately. The flask, when filled with the water and connected with this tube, is by a quick movement inverted, so that the end of the tube rests on the glass dish. A little above the end of the tube there is a short side tube bent at right angles, of smaller diameter than the tube itself, the effect of which is that directly the water in the dish falls below the little angle of this tube, air bubbles up into the flask, and more water runs into the dish. In this way the evaporating dish is kept at a constant level until the whole of the water is used up. The steam condenses on the inside of the glass shade, and collects in the copper capsule underneath the glass dish, and is finally conducted away by a piece of tape which passes over the copper lip of the bath. The evaporation of a litre of water takes about twenty-six hours, but with proper arrangements it is continuous, and when once started requires no supervision. The evaporating time is really a small matter, for the analyst can begin it one morning, and it will be ready the next. Before the water is submitted to evaporation it is boiled briskly with 20 c.c. of sulphurous acid; or if previous estimations have shown that there is a larger quantity of nitrogen as nitrates and nitrites than 5 per 100,000, a larger amount of sulphurous acid must be added. To ensure the destruction of nitrates, a drop of ferrous chloride is added to the first dishful of water. Lastly, in dealing with waters deficient in carbonates (in which case the sulphurous acid, when oxidised to sulphuric, might not offer sufficient base for combination, and therefore there might be some destruction of the organic matter), 1 or 2 c.c. of a saturated solution of hydric sodic sulphite are added, which will give any sulphuric acid, otherwise free, sufficient base for combination.

When the evaporation is complete, the next step is to remove the residue from the dish and burn it up in a vacuum with oxide of copper. To avoid this removal, Dr. Dupré has proposed and used a collapsible silver dish: the water in this dish is evaporated down in the usual way, and then the dish can be rolled up and thrust into a combustion tube. If the analyst does not use the silver dish, the residue must be removed by the aid of a flexible spatula, and mixed with copper oxide. The combustion tube, 18 inches long and of rather narrow bore, is cleansed and dried, and charged by the aid of a small metallic scoop, first with a little coarse oxide, after which the residue is mixed with oxide; lastly some more oxide is added, and in front of this is placed a roll of copper gauze which has first been oxidised in air and then reduced in hydrogen.

The usual precautions in filling a combustion tube for organic analysis are, of course, to be strictly observed. The tube is now placed in a combustion furnace, exhausted of all air by a Sprengel pump, the tube made gradually red-hot, and the gas finally transferred to a gas apparatus.

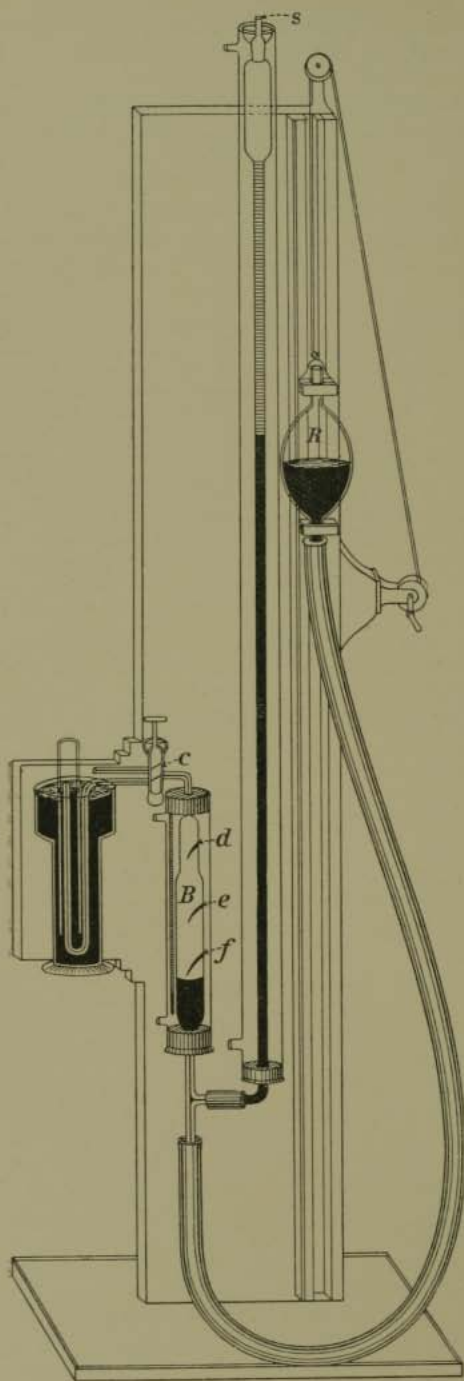


FIG. 74.

The writers use the mercury pump described at page 52. The combustion tube is attached by a short piece of pressure tubing direct to the tube from the pump, and made perfectly vacuum.

The gas is pumped out by the mercury pump into any convenient test-tube previously filled with mercury and inverted in the pneumatic trough. The gas thus collected is transferred to a measuring apparatus, the one the authors use being of the type devised by C. N. Huntley (fig. 74). The method of transference is obvious from the diagram. On lowering the reservoir *R* there is a vacuum produced in the burette *B*, and on turning the two-way cock *C* so as to place the burette and the interior of the test-tube in communication, the gas is aspirated into the burette.

The burette has no divisions, but there are three opaque glass points, *d*, *e*, *f*. The mercury, by careful first lowering then raising the reservoir, can be brought exactly in contact with one or other of these points, coincidence being easily adjusted by observing that the point coincides with its image reflected on the mercury mirror. The capacity of the spaces above the mercury for each point is ascertained by weighing the volumes of mercury or water for each division. It is also necessary to ascertain the exact divisions of the barometer tube which are on the same level with each of the three points. The barometer tube has no stopcock, but simply a ground glass stopper (*s*). The stopper is surrounded by a little mercury in the

mercury cup, so that leakage is impossible; both burette and barometer tube are jacketed and maintained at the same temperature by a stream of water.

If the gas consists of nitrogen and carbon dioxide, after measuring the total volume the gas is retransferred to the test-tube and a little bit of moist solid potash on a wire carried up through the mercury into the tube. After all absorption has ceased, the gas is transferred back again and remeasured, provided the temperature of the water-jacket is the same as before and the same division on the burette is used. The barometric height will be of course different, and the heights of the first to the second observation will be proportional to the gas that has disappeared or been absorbed.

Instead of absorption by solid potash, absorption pipettes may be used.

The pipette (fig. 75) consists of two bulbs, A and B. C is a thick-walled glass tube, with a capillary bore; at X is the India-rubber connection.

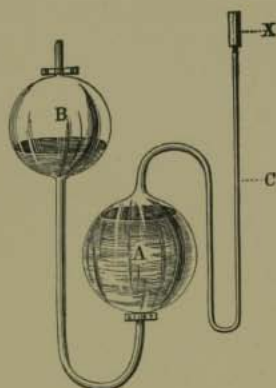


FIG. 75.

The pipette may now be disconnected and well shaken without any loss of gas, and absorption is far more rapid than when liquid reagents are applied in the ordinary manner. An analysis of a sample of air, and one of a gas consisting of carbon dioxide, nitric oxide and nitrogen, may be cited as an example.

Air.—A sample of air brought to the lowest division of the measuring pipette, barometer tube reading 900 mm., but the barometer reading corresponding to the lowest opaque glass point, is 325 mm. Therefore this has to be subtracted: $900 - 325 = 575$ mm., which is the pressure of the total gas. On absorbing by a stick of potash after the manner described, and again reading at the same division, the barometer reading is now 899.8 mm.; this subtracted from 900 gives, as the tension of the carbon dioxide, .2 mm. Lastly, on impelling the gas into the pipette (fig. 75), and submitting it to the action of alkaline pyrogallate for two hours with frequent shaking, and measuring at the same division, the barometer reading is 780.9, which, subtracted from the former reading, gives as the pressure of the oxygen 119 mm. Hence

Pressure of carbon dioxide,2
,, oxygen,	119.0
,, nitrogen,	455.8
Total pressure,	575.0

Since the temperature was constant throughout, if the volume in percentages only is required, the calculation is as follows:—

$$\begin{aligned}
 (1.) \quad & 575 : .2 :: 100 = \text{carbon dioxide} \\
 & \text{or } \frac{20.0}{575} = .03 \\
 (2.) \quad & 575 : 119.0 :: 100 = \text{oxygen} \\
 & \text{or } \frac{11900}{575} = 20.69.
 \end{aligned}$$

That is, the air contained .03 per cent. of carbon dioxide, and 20.69 per cent. of oxygen.

In a determination of the carbon and nitrogen of a water residue (from a litre), the following is an example of the method and of the numbers obtained:—The gas was pumped out by the mercury-pump direct into a thick-walled test-tube, and sucked into the measuring pipette and the mercury exactly brought down to one of the glass points; the pressure of the mercury as read on the barometer tube was 850 mm.; but since the division itself equalled or corresponded to 284, the total pressure of the gas was $850 - 284 = 566$ mm. On now absorbing by a stick of potash, the pressure was found to be 584, therefore the tension of the carbon dioxide was $850 - 584 = 266$. Two bubbles of pure oxygen were now added, and the gas, which immediately became of a red colour, submitted to the action of alkaline pyrogallate; after this operation the barometer reading was 574, and therefore the pressure of the nitric peroxide was $584 - 574 = 10$ mm. We have, therefore, the following determinations:—

Division 4 = 23.5 c.c. Temp. = 11° 5 C.		mm.
Tension of carbon dioxide,		266
,, nitric oxide,		10
,, nitrogen,		290
,, the three mixed gases,		566

These operations have furnished three uncorrected volumes of gases—

- A. Volume of the three mixed gases.
- B. Volume of nitric oxide and nitrogen.
- C. Volume of nitrogen.

The volumes must, therefore, all be reduced by the usual calculations to 0° C. temp. and 760 mm. pressure.

From these corrected volumes of nitrogen and carbonic dioxide, the weights of carbon and nitrogen can be obtained by calculation or by tables. There is a simple means of arriving at the desired result by the aid of the following data:—

1. The weights of carbon and nitrogen contained in equal volumes of carbon dioxide and nitrogen gases, measured at the standard temperature and pressure, are to each other as 3 : 7.

2. The weights of nitrogen contained in equal volumes of nitrogen and nitric oxide are as 2 : 1.

Hence, if we assume that for the purpose of calculation the gaseous mixture consists entirely of nitrogen, and that two successive portions of the nitrogen are removed from it by the reagents; then, if A be the weight of the total gas calculated as nitrogen, B the weight after absorption of the first portion (CO_2), and C the weight after the absorption of the second portion (N_2O_3); further, if x and y represent respectively the weights of carbon and nitrogen contained in the gaseous mixture, then the following simple equations express the values of x and y :—

$$x = \frac{3(A - B)}{7} \qquad y = \frac{C + B}{2}$$

In the example given the calculation is as follows:—

A.

Log. of 23.5 c.c., the capacity of the pipette at the point measured,	=	1.37106	
Log. of 566,	=	2.75281	
Log. from Table LXIX., corresponding to 11° 5 C.,	=	-6.20029	
			= .02109
			-2.32416

B.

Log. 23.5 c.c.,	=	1.37106	
Log. of 300,	=	2.47712	
Log. from Table LXIX., corresponding to 11° 5 C.,	=	-6.20029	
			= .01187
			-2.04847

C.

Log. 23.5 c.c.,	=	1.37106	
Log. of 200,	=	2.46239	
Log. from Table LXIX., corresponding to 11° 5 C.,	=	-6.20029	
			= .01080
			-2.03374
Carbon,	=	3(.02109 - .01187)	= .00438
			$\frac{7}{}$
Nitrogen,	=	.01187 + .01080	= .01133
			$\frac{2}{}$

Or the water contains in 100,000 parts .438 carbon, 1.133 nitrogen.

Blair's Method of Moist Combustion.—250 c.c. of water are acidified with 2 c.c. of pure, strong sulphuric acid when the water has been concentrated to 100 c.c.; all CO₂ from carbonates is then boiled off by concentrating to 50 c.c. A globular receiver with two necks, one of which can be adapted airtight by a rubber connection to the retort, is now charged with 10 c.c. of permanganate and 100 c.c. of water. The receiver is connected with the retort, and to the second tubular portion is adapted an indiarubber tube with clip. The liquid in the retort and that in the receiver are each made to boil by means of two Bunsen burners; after the steam has issued from the exit tube for a few minutes, the exit tube is clipped, and at the same moment the flames removed. The condensation of the steam produces a vacuum, as shown by the flattening of the rubber tube. By tilting the apparatus a little, about 30 c.c. of the permanganate solution are allowed to flow into the retort from the receiver, and the retort heated gently; distillation from the retort into the cooler receiver at once commences, and ultimately the acid, becoming concentrated, oxidises all the organic matter; the resulting solution should be colourless, but, if it is not so, a little more permanganate should be transferred as before, and the operation repeated until a colourless liquid results. The gases now in the apparatus are carbonic acid, oxygen, and chlorine, and it is obvious that, by means of a Sprengel or mercury pump, they could be transferred to a suitable gas measuring apparatus, and the quantity of CO₂ ascertained after absorption of the chlorine by direct measurement; Blair, however, prefers to estimate the CO₂ by titration. 10 c.c. of a 10 per cent. solution of ferrous sulphate are passed into the receiver by adjusting the charged pipette to the exit rubber tube, and carefully loosening the clip, so as to allow no air to enter. The contents of the receiver decolorised by the ferrous sulphate are allowed to flow into the retort, the liquid heated nearly to boiling and then poured backwards and forwards from retort to receiver. In a few minutes all the

chlorine is absorbed. A 6-oz. flask is prepared, having an accurately fitting rubber cork, through which passes a glass tube fitted with a rubber connection; a little water is placed in this flask, the water boiled to expel air, and the rubber tube clipped while the water is boiling; the flame is removed, and the flask cooled. Into the vacuous flask 10 c.c. of barium hydrate solution, 0.315 per cent., tinted with phenolphthalein are run in through the indiarubber tube by placing the end of the charged pipette in the end of the rubber tube and carefully loosening the clip; the glass tube is then pushed down until the lower end nearly reaches the bottom of the flask. The flask is now connected with the receiver by the indiarubber connections, the clips loosened, and the liquids in receiver and retort boiled, the flask being cooled in water; if the rubber tube remains flat, this is a sign that no air has leaked in, and that the steam has not sufficient tension to do any damage. Boiling from five to seven minutes expels all CO_2 ; the flames are then removed, the rubber tubes clipped, the flask finally disconnected, and the contents titrated with oxalic acid. The exact amount of c. n. oxalic acid required to neutralise 10 c.c. of the baryta water deducted from that required to neutralise the same quantity in the flask, gives the requisite data for calculating the CO_2 . 10 c.c. of c. n. oxalic acid = 1.2 mgrm. of carbon.

A. Wyster Blyth's Method of Moist Combustion.—Half a litre of the water to be examined is first saturated with SO_2 , and transferred through the thistle-headed funnel (fig. 76) of A into the flask, A. This is a strong Florence flask of about 600 c.c. capacity, having a side tube, the side tube being connected with a water pump and a mercury apparatus as follows:—A smaller flask, capacity about 70 c.c., with a T-side tube, is connected with the side-tube of A by means of a rubber cork. There is a Bunsen valve at *v*, so that no gas or liquid can run back into A; one limb of the T-piece is connected with the water pump, and the other with a gas burette graduated in cubic centimetres and tenths of cubic centimetres charged with mercury, and provided with a pressure tube and a stopcock.

There are also suitable clamps on the rubber parts. The measuring burette is first filled with mercury by raising the pressure tube, P, and, when filled, shutting off by means of the stopcock at *s*; the clamps, *t* and *g*, remain open. The flask, A, is placed on a sand-bath, the water pump set going, and the water boiled so as to get rid of free SO_2 ; this only takes a few minutes. Next, about 20 c.c. of clear saturated baryta water are transferred by means of the thistle funnel into A, without letting any air in, and this is followed by 10 c.c. of a recently boiled 1 per cent. permanganate. The alkaline solution is boiled down in a vacuum almost to dryness. The flask is then raised, cooled slightly, and 5 c.c. of organically pure sulphuric acid diluted with about 20 c.c. of water are transferred through the funnel in the same way. The clip, *t*, on the tube leading to the water pump is screwed down, and the apparatus put in connection with the gas burette by opening *s*. The flask, A, is now lowered on to the sand-bath and allowed to boil; nearly all the water condenses in B, which, at the beginning of this part of the operation, is almost empty, the condensed vapour from the former operations having passed away to a receiving flask connected with the water pump. The liquid is distilled to dryness and allowed to fume for about an hour. With ordinary waters the acid fumes are arrested in B. At the end of an hour the flask is removed from the sand-bath, allowed to cool slightly, and then recently boiled solution of ferrous sulphate run in through the thistle funnel.

This is boiled for a few minutes, and lastly, heat is applied for a few minutes to B. All the carbon dioxide is now in the gas burette, *s* is closed, the flame removed, and the gas burette detached from the apparatus; it is allowed to cool, and the gas carefully measured by bringing the meniscus of the pressure tube, *P*, on an exact level with that in the measuring burette and reading the number of cubic centimetres. The height of the barometer and the temperature is also ascertained. The gas is now transferred to the bulb pipette charged with potash, figured at p. 545, the bulb pipette is shaken, the unabsorbed gas drawn again into the measuring burette, and the gas again measured. The contraction will be equal to the amount of CO_2 . The whole readings must be reduced to normal temperature and pressure, and the volume of CO_2 translated into parts by weight of carbon.

If, instead of measuring the carbon dioxide as a gas, the analyst prefers to absorb

it in baryta water, the burette is replaced by an absorption apparatus consisting, first, of a bulb containing potassic permanganate solution, and then of absorption vessels charged with clear baryta, the last vessel being connected with the water pump; the carbonate of baryta can be filtered off under xylene, washed with hot water, dissolved in hydrochloric acid, evaporated to dryness, and converted into barium sulphate by adding a little sulphuric acid to the dry residue and driving the excess of acid off by heat.

The advantage of this method of making a moist combustion is obvious. The permanganate, by acting first in alkaline solution in a vacuum, at once fixes those volatile organic matters which in boiling down would otherwise be lost; while (except for a few minutes) the water is constantly under the influence of an oxidising medium, which is not the case with other processes.

Gravimetric Estimation of Minute Quantities of Carbon.—Drs. Dupré and Hake in 1879¹ published a method of gravimetrically estimating minute quantities of carbon by burning up in a current of pure oxygen, and then absorbing the CO_2 thus produced in baryta water, and converting the baryta carbonate into baric sulphate. A product was thus obtained, which weighs 19.4 times as much as the carbon originally present.

The details of the process are as follows:—A combustion tube, open at both ends, and about 24 inches long, is drawn out and bent downwards at one end at an angle of 120° , so that it may be conveniently attached

¹ *Journal of Chemical Society*, March, 1879.

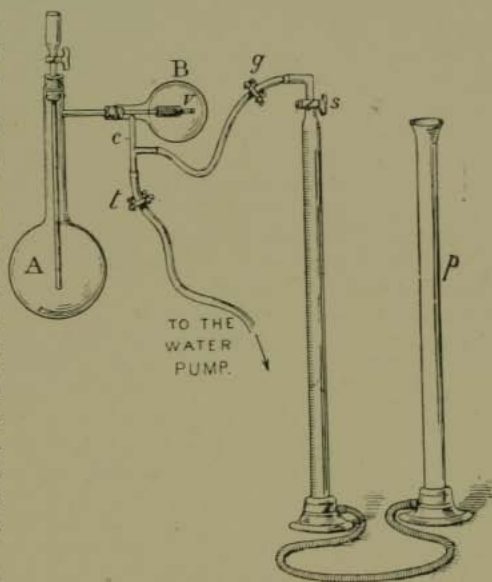


FIG. 76.

to a Pettenkofer's absorption tube, the other end being connected, by means of a caoutchouc stopper and glass tubing, with an oxygen reservoir. This combustion tube is filled half way from the bent end with granulated cupric oxide, which may conveniently be held in position either by plugs of asbestos or by platinum wire gauze, or by a combination of both. The connection with the oxygen reservoir being then made, the greater part of the tube is heated to redness, with the ordinary precautions, and a stream of oxygen (which is first conducted through a long tube containing caustic potash) is passed over the glowing oxide of copper until the issuing gas ceases, after long bubbling, to cause any turbidity in the bright baryta water. As soon as this point is reached, the portion of the combustion tube preceding the layer of cupric oxide is allowed to cool somewhat, and the tube is now ready to be connected with the absorption apparatus. The clean absorption tube is carefully rinsed with water, and is clamped in front of the furnace, in such a manner that its bulb end is somewhat higher than the end to be connected to the combustion tube. Both ends must be provided with convenient stoppers, consisting of short pieces of caoutchouc tubing closed with a small piece of glass rod. The stoppers being removed, air, which is first caused to pass through a tube containing caustic potash, is pumped through the tube for about two minutes, and it is then filled with baryta water as follows:—The baryta water (of strength 1.5 per cent.) is kept in a sufficiently large stock bottle, provided with a caoutchouc stopper, through which pass two bent glass tubes, the long one for syphoning, the shorter, to which a potash tube is attached, being connected with a small hand-bellows. In filling the absorption apparatus, the longer syphon tube is connected with it by means of flexible tubing, and the baryta water is forced over by gentle pressure of the bellows, the bulb end of the absorption apparatus being provided with a potash tube. As soon as the absorption apparatus is half filled, the flow of baryta water is arrested; the ends of the Pettenkofer tube are immediately closed by its stoppers, and is now ready for use. By these means the tube is filled with perfectly clear and bright baryta water. The absorption apparatus is now connected with the combustion tube, and the combustion proceeded with. The silver dish containing the water residue having been inserted just behind the copper oxide, it is burnt in a slow current of oxygen, and the carbon dioxide is absorbed and converted into baric carbonate in the absorption tube. In order to filter off and convert the baric carbonate, a funnel and filter are arranged to stand over a beaker containing a layer of caustic potash solution at the bottom, the whole being covered by a bell jar, which itself stands in a layer of caustic potash solution. The mouth of the bell jar, which is immediately over the funnel, is closed by a thick caoutchouc cap with two narrow openings, one of which is provided with a caustic potash tube. (Soda lime apparently answers equally well.) The other, which is temporarily stoppered, contains a straight glass tube, placed immediately over the filter so that, after the whole arrangement has been left some time to itself, in order that all enclosed air may be free from CO_2 , direct connection may be made with the Pettenkofer tube by means of flexible tubing sufficiently long to admit of some slight freedom of action. Filtration may thus be carried on without danger of CO_2 being introduced from the atmosphere, the additional precaution being taken of compelling all air which passes through the Pettenkofer tube during this process of filtration, to pass through a tube containing caustic potash attached to the

tube itself. The washing of the precipitate in the tube and on the filter is effected almost entirely with boiling water, which has been previously saturated with carbonate of barium [solubility 1 in 15,000], but finally with a small quantity of boiling distilled water. After complete washing the tube is disconnected, and the filter ultimately rinsed round, while still under the bell jar, by means of the long tube already mentioned, and which, when not clamped, may be moved freely in all directions. The bell jar is then removed, and the precipitate is rapidly washed together into the bottom of the filter.

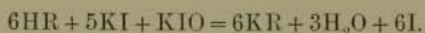
The Pettenkofer tube, which may contain minute particles of baric carbonate not removed by the washing, is rinsed twice with small quantities of dilute pure hydrochloric acid (about 1 in 50), and finally with distilled water: the rinsings are poured on to the filter on which the greater mass of baric carbonate is already collected. The filter is further washed with dilute hydrochloric acid, and finally with distilled water: and the whole of the solution of baric chloride so formed is carefully collected in a small beaker. The quantity of such solution need not exceed 50 c.c. This solution of chloride of barium has next to be evaporated, which is best done in a platinum vessel on the water-bath. It is then transferred, when greatly decreased in bulk, to a much smaller platinum dish, weighing about 5 grms., and finally evaporated to dryness after the addition of a few drops of pure sulphuric acid. The dish and its contents have then to be ignited, the residue moistened with a drop of nitric acid and redried, and the whole re-ignited and weighed to conclude the operation. The amount of carbon present is obtained by dividing the weight of the baric sulphate by 19.4.

Nephalometric Method.—This ingenious method we also owe to Dupré and Hake. The carbonic acid resulting from the combustion of an organic residue is passed into perfectly pure clear solution of basic lead acetate, and the turbidity produced is imitated by known weights of CO_2 ; in fact, the operation is a colour method conducted on the same principle as 'Nesslerising,' with this important difference, that no success will be obtained unless there are special precautions taken to prevent the contamination of the solutions by the breath and air, etc.

§ 373. *The Estimation of Organic Nitrogen after Kjeldahl's Method.*—Half a litre of the water is placed in a retort and the free ammonia distilled off. Then 5 to 10 c.c. of diluted sulphuric acid are added, and the water concentrated down on a sand-bath until the acid fumes. The acid is allowed to fume for about half an hour, or until it is almost colourless. The acid solution is then cooled, diluted, alkalisied with pure soda lye, and the liquid distilled, the alkaline distillate being neutralised with decinormal sulphuric acid, each c.c. of which is equal to 1.4 mgrm. of nitrogen. This simple process is applicable to most pure waters containing but little organic matter and feeble nitrates. On the other hand, it will not give accurate results with waters containing much nitrate or much organic matter. In such a case the following is the best method:¹—Half a litre, as before, is taken. The water (after getting rid of the free ammonia) is saturated with SO_2 , and a drop of iron chloride solution added; it is then gently heated for about twenty minutes; and is next boiled down to about 20 c.c. To this residue is added 20 c.c. of sulphuric acid containing 4 grms. of phosphorus pentoxide and then 0.12 gm. of anhydrous copper sulphate and 5 drops of platin chloride solution. The contents of the flask, closed by a glass marble, are heated gradually to a gentle boil, and the heating continued

¹ Ulsch, *Zeitschrift f. analyt. Chemie*, xxv. 579.

until the fluid remains of a green colour. After cooling, the acid fluid is diluted, alkalisied by ammonia-free soda solution, a little granulated zinc added, and the whole distilled into a measured volume of decinormal sulphuric acid. The decinormal acid arrests any ammonia, and on titrating the distillate with d. n. soda, there will be a loss of acidity proportionate to the ammonia, from which (as before) the amount of nitrogen can be calculated. There has been found a slight practical difficulty in thus titrating ammonia with the greatest accuracy, and Kjeldahl¹ has therefore recommended the utilisation of the reaction which takes place between a free acid and iodide and iodate. This reaction (denoting free acid by HR, takes place as follows :—



To the acid distillate is added 0.4 grm. of potassium iodide and 0.1 grm. of potassic iodate; after standing two hours the iodine set free is estimated by means of a decinormal thiosulphate solution, the strength of which has been checked and adjusted by the aid of a decinormal iodine solution.

An example will make the calculation clear.

The distillate from half a litre of water was submitted to Kjeldahl's process, and was received in 30 c.c. decinormal sulphuric acid. Potassic iodide and iodate were added thereto (as above described), and the mixture allowed to stand two hours; at the end of that time the free iodine was estimated by means of a decinormal thiosulphate solution, using starch as an indicator; 25.9 c.c. of thiosulphate were used. Since 30 c.c. of thio-sulphate are equivalent to 30 c.c. of decinormal acid, it is clear that 30 - 25.9—that is, 4.1 c.c. of free acid—have been saturated with ammonia; hence the distillate contains 4.1 c.c. \times 1.4 mgrm. = 5.74 mgrms. nitrogen, and the water contains 11.48 mgrms. per litre or 11.48 parts per million of organic nitrogen.

12. *Mineral Analysis of Water.*²—Ordinary drinking water holds dissolved but few saline matters, and when an analyst has determined chlorine, nitrates, sulphates, phosphates, and carbonates, and also lime and magnesia and alkalies, he will usually find, on adding the several amounts together, that he gets numbers very nearly equal to the solid saline residue.

An excellent method of approximately estimating the various saline constituents of a water is to evaporate down to dryness a known quantity, then to treat the residue with a

¹ Kjeldahl, *ib.*, xxii. 366.

² A method of determining calcium and magnesium has been described by Professor C. L. Bloxam (*Chem. News*, 1886); it depends upon the precipitation of calcium and magnesium as ammonia-arsenates, and is specially applicable to their separation from strontium salts. The determination of calcium as ammonia-arsenate in ordinary drinking waters has some advantage—the precipitate is as nearly insoluble in water as calcium oxalate, and being highly crystalline is not liable to run through the filter—the formula of the precipitate dried at 100° C. is $Ca_3NH_4H_2(AsO_4)_6 \cdot 3H_2O$, and 100 parts are equal to 20 of calcium or 50 calcic-carbonate. If a rapid determination be desired, arsenic acid is added to $\frac{1}{4}$ of a litre in water, which is then strongly alkalisied by ammonia. The mixture is well stirred, and allowed to stand for ten minutes, the precipitate is then collected on a weighed filter, washed with ammonia water (8.5 per cent.), and dried at 100° C. The gain in weight represents the united magnesian and calcic ammonia-arsenates. The precipitates are now dissolved off the filter by acetic acid, and the calcium precipitated as oxalate by ammonium oxalate, the solution boiled and filtered. On adding to the filtrate ammonia, the ammonia-magnesium arsenate is in this way reprecipitated, and may be collected on a weighed filter, washed with ammonia water, and dried at 100° C.; its composition is $(MgNH_4AsO_4 \cdot \frac{1}{2}H_2O)$; 100 parts are equal to 44.2 of magnesium carbonate.

little hot water, which will dissolve all the soluble salts out, but leave insoluble carbonates of lime and magnesia, and silica. In the soluble portion, the soluble salts of the alkaline earths and the alkalis are determined; the chlorides, sulphates, and nitrates are estimated in the unconcentrated water by the processes already detailed. It is also always open to make the analysis in the old-fashioned way—that is, to evaporate down a large quantity of water, to separate the silica by treatment of the ash or residue with hydrochloric acid, and after separation of the silica to divide the solution into three or four quantities, in which sulphuric acid, lime, magnesia, etc., are determined by the ordinary methods.

W. W. Fisher¹ has suggested a very good method of checking the mineral analyses of water, which is also applicable to the indirect determination of alkalis. He calculates the molecular proportions of the bases and acids present; if the analysis is correct, they should very nearly correspond. The following example explains itself.

TOTAL SOLIDS DRIED AT 100° C. = 243·60; DRIED AT 240° C. = 230·00.

BASES.				ACIDS.			
	Grains per gallon.	Mol. weight.	Mol. propor- tions.		Grains per gallon.	Mol. weight.	Mol. propor- tions.
Soda, Na ₂ O . . .	53·14	÷ 62	= 0·8571	Cl ₂	18·00	÷ 71	= 0·2535
Magnesia, MgO, . . .	11·99	÷ 40	= 0·299	SO ₃	103·88	÷ 80	= 1·2985
Lime, CaO, . . .	35·28	÷ 56	= 0·630	CO ₂	9·55	÷ 44	= 0·2170
				N ₂ O ₅	0·84	÷ 108	= 0·0077
Total, . . .	100·41		1·7861	Total, . . .	132·27		1·7767
				Deduct O = Cl ₂ = 4·07			
					128·20		

Thus total salts = 228·61, and the difference will be due to silica and oxide of iron. It is obvious that if the soda be unknown it may be easily calculated if we assume the molecular proportions of the total bases = 1·7767.

III. BIOLOGICAL METHODS.

§ 374. *Microscopical Appearances.*—To make a microscopical examination of water, it is necessary to collect the sediment or deposit which falls to the bottom of the vessel in which the water stands. A convenient way of doing this is to use the tube (fig. 77), which holds a little more than a litre. The little glass cell, C, is adjusted to the pipette-like end, the rod is removed, and after introduction of the water the tube is covered and set aside for twenty-four hours. At the end of that time any deposit will have collected in the glass cap. On now carefully inserting the rod-like stopper, the cap or cell can be removed with great ease, and its contents submitted to microscopical examination. With very pure waters merely a little sand or formless *débris* collects in the cap, and there is no life. If, however, in the first place eight or ten gallons are allowed to deposit in a capacious vessel, most of the water run off, and then the last litre rinsed into the tube, in nearly every case there may be a few life-forms and sufficient matter collected to give definite results. It need scarcely be said that an opinion must not be formed upon a microscopical examination without taking into account the amount of water from which the sediment has been collected, and a definite quantity should be generally agreed on by analysts.

*The Sedgwick-Rafter Method.*²—This ingenious method of obtaining a quantitative estimation of organisms and objects in water is in use by the

¹ *Analyst*, May, 1902.

² *Massachusetts State Board of Health Report*. Boston, 1890.

Massachusetts biologists. A brass gauze stop is put in the mouth of a funnel, and on this stop is packed a layer of sharp quartz sand; the sand grains should pass through a sieve 80 meshes to the inch, but not through a sieve 100 to the inch. Whipple¹ prefers pure ground quartz. On to this sand is poured a convenient quantity of the water (say 500 c.c.), well shaken, so as to get a fair sample. The sand and organisms, when filtration is complete, but before the sand has become dry, are washed into a test-tube, with from 3 to 5 c.c. of distilled water. The tube is shaken thoroughly, the sand allowed to settle for a moment, and the water quickly decanted. This procedure leaves the sand, but transfers most of the organisms. 1 c.c. of the liquid is placed on a counting-plate, consisting of a shallow cell with a brass border, the cell holding exactly 1 c.c. A fractional part of the field is now observed, and the organisms counted therein; in order to do this, the slide is ruled into millimetre squares, or a metal disc is fitted into the eye-piece with a square hole cut in its centre, the area of the square hole being such that with the powers used it just covers a square millimetre.



The Massachusetts biologists have by this method examined an enormous number of waters; some having been examined on several days in each month in the year. The method of recording and tabulating the results are shown in Table LXVII., giving the biological results of the water supply of Nantucket. This water supply, owing to the increase of *Anabaena* (one of the blue-green algæ), in the months of August, September, and October smells and tastes offensively.

Dibdin's Process.—A better method than the Sedgwick-Rafter process is that which has been invented by the late chemist to the London County Council (Mr. Dibdin). A litre of the water (or less, should the water contain much suspended matter) is filtered through hard filter paper. The deposit is washed off the filter paper into what is called a "micro-filter." The micro-filter is prepared as follows :—A piece of clean combustion tubing is drawn out into a capillary tube of a diameter of 2 mm. The open small end is plugged by a paste made of equal parts of air-dried clay and kieselguhr. This plugged end is dried in the Bunsen flame, and ultimately heated to redness. The residue from the water is now placed in this micro-filter, and the micro-filter is fitted into any convenient flask or bottle by means of an india-rubber plug attached by a side tube to a good water-pump, and the superfluous liquid drawn through, until only about 1 to 1.5 c.c. remain. The sediment and suspended matter are thus collected in the form of a compact cylinder just above the porous filtering substance, and are now carefully measured, the results being expressed in millimetres per litre. The cylinder of deposit is removed by scratching the tube with a sharp file about half an inch from the filter plug and breaking it off. A platinum wire is pushed in so as to loosen the deposit from the porous plug. Upon inverting the tube, open end downwards, the deposit falls away from the plug towards the open end. The tube is then cut close to the plug and the plug removed. On holding the little tube, now open at both ends, and giving it a downward shake, the whole of the deposit can be transferred in about a drop of water to a microscope slide and examined in the ordinary way; afterwards portions of

¹ *The Microscopy of Drinking Water*, by G. C. Whipple, 1899.

TABLE LXVII.—MICROSCOPICAL EXAMINATION OF WATER FROM WANNACOMET POND, NANTUCKET. NUMBER OF ORGANISMS PER CUBIC CENTIMETRE.

(25th Annual Report of State Board of Health, Massachusetts, 233.)

	1893.												
	16th Aug.	24th Aug.	31st Aug.	9th Sept.	15th Sept.	22nd Sept.	30th Sept.	6th Oct.	13th Oct.	21st Oct.	27th Oct.	11th Nov.	8th Dec.
PLANTS.													
Diatomaceæ,	0	0	0	0	0	0	0	0	0	0	2	pr.	400
Cyanophyceæ,	2224	1100	467	336	925	508	176	968	1748	800	449	5	0
Anabæna,	2160	760	200	228	860	356	176	968	1748	744	440	5	0
Anabæna spores,	64	340	260	108	65	152	0	0	0	56	9	0	0
Microcystis,	0	0	7	0	0	0	0	0	0	0	0	0	0
Algæ,	16	2	9	1	25	10	0	0	0	1	9	36	14
Pandorina,	10	0	1	1	0	0	0	0	0	0	7	0	0
Protococcus,	0	0	3	0	25	10	0	0	0	0	0	1	12
Scenedesmus,	4	2	2	0	0	0	0	0	0	0	2	34	2
Staurastrum,	2	0	3	0	0	0	0	0	0	1	0	1	0
Fungi, Crenothrix,	56	0	0	0	0	0	2	2	0	0	0	1	0
ANIMALS.													
Infusoria,	8	9	20	2	0	0	0	2	10	3	21	354	16
Cryptomonas,	0	0	0	0	0	0	0	0	0	0	0	0	7
Dinobryon,	0	0	0	0	0	0	0	0	0	0	10	60	0
Dinobryon cases,	0	0	0	0	0	0	0	0	0	0	0	260	4
Euglena,	2	0	0	0	0	0	0	0	0	0	0	pr.	0
Glenodinium,	0	0	0	0	0	0	0	0	0	0	0	0	2
Monas,	4	0	0	0	0	0	0	0	8	0	0	0	0
Peridinium,	2	9	20	2	0	0	0	2	2	3	6	34	3
Vorticella,	0	0	0	0	0	0	0	0	0	0	5	0	0
Vermes,	8	4	9	4	0	0	0	4	0	7	16	6	3
Anurea,	4	1	5	3	0	0	0	2	0	6	15	5	0
Monocerca,	0	0	0	0	0	0	0	2	0	0	0	0	0
Polyarthra,	2	2	0	1	0	0	0	0	0	0	0	1	3
Rotatorian ova,	2	0	1	0	0	0	0	0	0	1	1	0	0
Rotifer,	0	1	3	0	0	0	0	0	0	0	0	0	0
Crustacea,	·08	0	·02	·01	·10	·07	0	0	0	0	0	·01	·04
Cyclops,	0	0	0	0	0	·02	0	0	0	0	0	·01	·04
Daphnia,	·08	0	·02	·01	·10	·05	0	0	0	0	0	0	0
Miscellaneous,	144	76	200	0	·10	0	208	136	72	6	0	2	5
Acarina,	0	·01	0	0	·10	0	0	·04	·04	0	0	0	0
Zoogloea,	144	76	200	0	0	0	208	136	72	6	0	2	5
Total,	2456	1191	705	343	950	578	386	1112	1830	817	497	404	438

the deposit may be stained with aniline dyes, cultivated and otherwise investigated.¹

1. Lifeless Forms.

1. *Mineral Matters*, especially sand, clay, and not unfrequently fine spicula of glass derived from the glass pipette, etc.

2. *Vegetable Matters*.—In shallow pools, in rivers, reservoirs, and, in fact, all open waters, the microscopist seldom fails to find vegetable *débris* in the shape of dotted ducts, spiral vessels, parenchymatous cells, bits of cuticle with the hair still adhering, the down of seeds, roots of duckweed, bits of chara, etc. It depends on the amount as to what conclusions are to be drawn; but this is certain, that a water showing these matters is not likely to be from a deep spring, but one over which the atmosphere more or less freely plays.

3. *Dead Animal Matters*—(a.) *Purely Animal*, such as hairs from domestic or wild animals, striped muscular tissue, the scales of moths, butterflies, or other lepidoptera, eggs of euzoza (which, of course, may, for aught we know, be living).

(b.) *Human Débris*.—Human hair, human epithelium.

(c.) *Manufactured Matters*.—Wool, silk, etc. All animal matters, whether derived from insect, human, or domestic animal life cannot be considered a favourable indication; and even the presence of cotton, silk, hemp, and the like, though innocuous in themselves, yet afford evidence that the water is in such a position as to be liable to accidental contamination.

2. Living Forms.

Microscopic organisms. These may be conveniently considered as (a) Vegetable, (b) Animal, although a division is sometimes difficult.

(a) Vegetable.—Putting aside for the moment Bacteria or Schizomycetes the most common living vegetable forms are the following:—*Beggiatoa*, *Crenothrix*, *Cladothrix*—then there are confervæ, oscillatoria, volvocinæ, desmids, diatoms, etc. To these may be added the green, or sometimes red, cells of palmellæ, and the moving reproductive spores of confervoids, charæ,² etc. Those forms which possess cells holding 'chlorophyll' denote water which is exposed to daylight.

Beggiatoa.—There are several species of *Beggiatoa*; the commonest being *Beggiatoa alba*. *Beggiatoa* forms long flexible threads, 1 to 5 mm. in diameter, containing a number of round bodies. It grows on dead vegetable and animal matters, sometimes in enormous quantities. The senior author has seen the bottom of a polluted brook lined with it for the extent of half a mile. The attached part of the thread is thinner and shows divisions into segments with but few particles, but towards the free end the segments become less obvious and bright round coloured particles more numerous. These particles consist mainly of sulphur. The free ends are occasionally liberated, and give rise to a swarm of spiral threads which (under the name of *Ophidomonas*) were formerly classed as a distinct genus.

¹ *Analyst*, 1895.

² For a full account of all the organisms (except bacteria) to be found in drinking waters from all sources the reader is referred to the excellent work by G. C. Whipple, *The Microscopy of Drinking Water*.

The growth of *Beggiatoa* is favoured by organic matter and sulphur compounds; hence its presence usually denotes sewage pollution.

Leptothrix is another thread form met with in waters. A few botanists consider it to be simply a stage in the life of *Crenothrix* and *Cladothrix*; but the majority consider it a distinct genus.

Leptothrix ochracea forms thin threads stained yellow or yellow-red by a deposition of iron oxide in the sheath.

Crenothrix forms a brown slimy precipitate consisting of fine threads from 1.5 mm. to 5 mm. in diameter. The threads are composed of oblong cells, the cell divisions being evident. Old cells are often coloured by iron oxide. Some of the cells develop spores which form masses of brownish scum. These grow to fresh threads. Sometimes the spores grow within the cell to new threads, and then the new threads break through the cell, forming a bundle of fine filaments. Old threads may show no sign of division into cells.

Cladothrix is similar to *Crenothrix*, but the threads are of less diameter. It may always be, however, distinguished from *Crenothrix* by the threads dividing dichotomously, hence the name *Cladothrix dichotoma*.

Desmids, beautiful microscopic algae, consisting always of two symmetrical cells, are in colour remarkably green: one of the most common is a species of *Closterium* (fig. 78). Desmids have been referred to by Dr. Macdonald as rather indicating a good water.

It is true that desmids occurring by themselves, with no other indication of animal and vegetable life, could in no way be pronounced injurious. The fact, however, remains that, as a result of many years' experience in the almost daily analysis of water, the writers have never found desmids except in more or less surface supplies of water abounding with impurity. *The diatoms*, which are composed of a siliceous skeleton clothed by a sarcode substance, consist, like the desmids, of two exactly similar parts; they possess no chlorophyll, and are now considered either as a class or order of the Algae or as an independent group. Fig. 79 is *Diatoma vulgare*, very frequently found, and by itself certainly affording no indication of a bad water. In fact, diatoms generally are of little importance.

§ 375. *Bacteria*.—The simplest and lowest forms of plant-life are called bacteria or *Schizomycetes*. They are for the most part exceedingly small cells, the width being generally about 0.5μ and the lengths about 1μ (the μ is $\frac{1}{1000}$ mm. or $\frac{1}{25,000}$ in.), but the smaller proportion may range beyond the sizes given, or from 0.15μ to 5μ , or in the case of the sulphur bacteria to 20μ or more in width, while the length in some exceptional cases may exceed 100μ . The cells are in many cases single; in some instances they are in pairs (as in *Diplococcus*), or in chains of two, three or more (as in *Streptococcus*), and in yet other instances they form masses. The form of the cells is spherical, oval-shaped or spiral in the vegetative state, and nail-headed, clubbed, fusiform or somewhat dumb-bell shaped in the sporing conditions. When actively growing, the multiplication is by fission; but in some species, when growth is checked by unfavourable conditions, spores are formed. These spores correspond to the seed of higher plants, and are endowed with extraordinary powers of resisting extremes of cold and heat, desiccation and other influences which are fatal to the vegetating individuals. With few exceptions, they are devoid

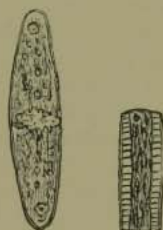


FIG. 78. FIG. 79.

of chlorophyll, a feature which distinguishes them and the fungi proper from green plants, and they live mainly on organic matter, in which respect they agree with fungi and differ from higher plants generally. Some live on decaying organic matter, and are termed *Saprophytes*; others occur in living organic tissues as parasites; while a few live solely on mineral matter.

The essential function of bacteria is to reduce diseased and decaying organic matter to simpler chemical forms by oxidising it and using the energy thereby liberated in building up their own bodies.

The living home of most bacteria is wherever lifeless organic matter has accumulated; generally on the surface soil and to a few feet within, on the bottom of oceans, lakes, pools, and in the organic matter suspended in flowing waters or exposed to moist air. If the organic matter becomes dried up, the bacteria form spores and these spores are carried with the dust into the air, the winds carrying them to moist places where they find favourable conditions for growth. The main factors regulating their distribution are, first, a supply of suitable food, and, secondly, currents of air or water, and carriage by active organisms and inanimate motors.

The rate of multiplication of the vegetative form is exceedingly high, but such multiplication is limited by the amount of available food. Hence, in soils and in flowing waters the organic matter is ever undergoing a conversion into harmless products, and, if there is no fresh accession of organic matter, the soil or the stream becomes purified at a rate proportioned to the accessibility of oxygen for the bacteria, etc. Each organism has its own limited sphere of action, so that the co-operation of many species are required for the complete decomposition of the organic matter. The first stages of the decomposition are effected by species which can only obtain the requisite oxygen from easily deoxidised organic compounds; these are termed *anaërobic*; others can only obtain their supply from the oxygen of the air or from the oxygen dissolved in water; these are *aërobic*; while some can obtain it in either way.

Most bacteria are so sensitive to physical and chemical conditions more or less special to themselves that their absolute and relative numbers are liable to great changes within very short periods of time. Hence, the bacteriological examination of a water should be made as speedily as possible, and, if delay is unavoidable, growth should be checked by placing the samples in tubes, and the tubes in special boxes kept cool by ice, and surrounded by some substance which will prevent the ready accession of heat.

It has been proved that the purification of water takes place chiefly while it is in contact with the layers of soil at and near the surface. Not only is it here that the greatest amount of organic matter is reduced to ammonia and nitrates, but pathogenic organisms are actually killed. Dr Sidney Martin¹ has shown that the micro-organisms common to soil, prove rapidly fatal to *Bacillus typhosus*, Gärtner's bacillus and *Bacillus coli communis*. Dr Houston² has further shown that the same thing takes place when sewage is allowed to remain in contact with superficial soil. The soil bacteria rapidly gain a mastery over the majority of the sewage bacteria, streptococci, *B. coli*, and *Bacillus enteritidis sporogenes*. The streptococci, especially, are killed in a very short space of time. Dr Houston found that sewage organisms persist for a very much longer period in deep soil than they do in surface soil.

¹ Appendix B. Supplement to 30th Annual Report, L. G. B.

² *Ibid.*

It is thus evident that the changes that water may undergo are rapid, and it is necessary to exercise great care and speed in their examination and to place but little reliance upon the absolute number of bacteria present. For although the number of bacteria may, and usually does if excessive, indicate that a large amount of organic matter is present, it gives no indication as to the origin of this organic matter, or whether the bacteria present are likely to be harmful. An estimation of the number of bacteria present is, however, of great value if constant examinations of the same supply are being made, or when comparing different portions of the same supply or when testing the efficacy of filters, etc. Of more value, as an indication of pollution, is an estimation of the different varieties of organisms present. It is often useful to also estimate the number of spores of bacteria, the number of gelatin-liquefying bacteria, and the number of bacteria capable of multiplying at blood heat, a temperature unsuitable to the majority of harmless water organisms.

But in the present state of our knowledge, since no convenient and certain method has been devised for isolating *B. typhosus* from the number of more hardy organisms which invariably accompany it, chief reliance has to be placed upon the isolation of organisms of the *Bacillus coli communis* group. These organisms are contained in large numbers in all animal excreta, and the work of Theobald Smith, of Klein and others has demonstrated beyond doubt that the presence of *Bacillus coli* and its near allies in drinking water, in anything but the smallest numbers, is conclusive evidence of sewage contamination. Since, however, these organisms are widely distributed in dust, etc., a few may be found in good waters, if they have been exposed to the air. The cause of the presence of *B. coli* should always be sought, and a water persistently found to contain this organism should not be declared absolutely safe for use as a drinking water until it is known that the organisms are not derived from sewage pollution. In undoubtedly polluted waters, from four organisms of the *Coli* group to many thousands may be found in 100 c.c. of the water, and they will always be found associated with a great variety of other organisms. *Bacillus (Proteus) vulgaris* and *Bacillus sporogenes enteritidis* (Klein) should also be looked for, as they are always present in sewage in large numbers.

Streptococci and *Staphylococci* may be sometimes looked for; as Houston has recently pointed out, their presence is very strong evidence as to recent contamination.

Only in very special cases need Cholera vibrios and the spores of Anthrax be sought for; fortunately the isolation of these organisms presents no great difficulty.

Only those organisms associated with sewage contamination will be described here. The reader is referred to the special works on the bacteriology of water (as, for instance, P. G. and C. Frankland's *Micro-organisms in Water*, 1894) for a full description of the many organisms to be met with, and to any of the many works on practical bacteriology for the methods of culture, staining, etc.

Total Numbers of Organisms growing at 22° C. — The organisms contained in a definite volume of water capable of growing on standard nutrient gelatin are usually spoken of as total numbers of organisms, although this number does not represent all the organisms contained in the water. In estimating the total numbers, the following precautions should be observed.

(1) The sample should be examined as soon as possible after taking ; at the time of taking if possible.

(2) The nutrient gelatin used should be of a definite alkalinity and be always prepared in the same manner.

(3) A definite incubation temperature (22° C) should be used ; on no account should plates be allowed to incubate at the temperature of the room or any other indefinite temperature.

(4) Not more than 50 or 100 organisms should be allowed to grow upon one ordinary Petri dish ; since if organisms are crowded together many do not grow.

The actual operation of estimating total numbers is simple. Six standard nutrient gelatin tubes (see *infra*) are taken, and the gelatin gently melted at 40° C. The water to be tested is shaken up, and 1 c.c., 0.5 c.c. or .25 c.c. injected into each of the gelatin tubes by means of a sterile pipette ; after mixing, the contents of the tube are at once poured into a sterile Petri dish, and allowed to set in a cold place. The used tube is replugged with its scorched cotton wool, and held under a stream of cold water, while it is slowly rolled so as to cause the gelatin to set evenly round the sides of the tube. This tube and the Petri dish, which must be inverted when the gelatin has set, are placed in the incubator at 22° C., and examined every day, the colonies being dotted off with an ink spot on the base of the Petri dish as they appear. If the water is suspected of being very rich in organisms, 10 c.c. of the original water are injected into 100 c.c. of cold sterile tap water (always use tap water, as distilled water is inimical to many organisms), and of this dilution inject 1 c.c., 0.5 c.c., and .25 c.c., respectively, into gelatin tubes, and treat as before. This method is far better than taking very small quantities of the original water, as such quantities cannot be measured with any degree of accuracy.

The estimation of total numbers is very important when testing the efficacy of filters, or if regular examinations of a supply are being made, but is of little value when applied to a single sample.

Total Organisms growing at 38° C.—Agar-agar tubes are treated in the same manner as gelatin tubes, and then placed in the warm incubator, and examined after twenty-four hours and on subsequent days.

Total Aerobic Spores.—1000 c.c. of the water are filtered through a sterile Pasteur-Chamberland candle, and the surface of the candle brushed into 10 c.c. of sterile tap water contained in a sterile dish, by means of a small sterile brush. 1 c.c. or less, according to the suspected purity of the water, is injected into melted gelatin. After heating in a water bath maintained at a temperature of 80° C. for ten minutes, the gelatin is poured into a Petri dish, and incubated at 22° C.

Care should be taken to heat the upper portion of the test tube, or else the estimation is liable to be too high.

Standard Nutrient Gelatin.—In order to have uniformity in the composition of the nutrient gelatin used, we have adopted the mode of preparation suggested by Pakes, which is as follows :—

One pound of finely-chopped beef, as free from fat and connective tissue as possible, is incorporated in a litre of water. The whole is then raised slowly to 60° C., preferably in a water-bath, kept at this temperature for about ten minutes, and then quickly heated to boiling-point. This is now filtered into a clean flask, and made up with water to one litre again. Distilled water should be used rather than ordinary tap-water, because the

character of the water affects the growing power of the gelatin. This meat infusion now contains the salts and extractives of the beef, and a little acid albumen.

Ten grammes of Witte's peptone and 5 grammes of common salt are carefully weighed, and placed in a clean dish. The mixture is rubbed up very carefully with small quantities of the meat infusion in order to dissolve the peptone. If this is not done, dry masses of peptone may be found even after boiling. If by this time the infusion is cold, it should be warmed in a water-bath, and 100 grammes of gelatin ('Best French') added. The flask is now placed in the steam steriliser, in order that the gelatin may be thoroughly dissolved; it should be taken out every few minutes, and well shaken, in order to prevent the gelatin settling as a sticky layer at the bottom. When the gelatin is quite dissolved, the flask is removed, and at once neutralised with $\frac{N}{1}$ NaOH, using litmus as the indicator. When it has been neutralised, 4 c.c. of the NaOH are added, the contents of the flask are cooled to 50° or 55° C., and the white of an egg is added and well incorporated by vigorous shaking. The flask is now placed in the steam steriliser for half an hour, by which time the egg has coagulated in large flakes, leaving the internatant fluid clear. It is then filtered through 'papier Chardin' into a clean flask, and run through a filtering apparatus (delivering 10 c.c.) into the sterile test-tubes. The gelatin tubes are at once placed in the steam steriliser for twenty minutes, and treated in a similar manner on the two succeeding days.

THE ISOLATION OF BACILLUS COLI COMMUNIS.

§ 376.¹ *The Neutral Red method.*

W. G. Savage's method is as follows:—*The medium.*—To ordinary broth made from Liebig's extract, add peptone and sodium chloride, and make faintly alkaline to litmus, then add 5 per cent. of glucose and 12 c.c. per litre of $\frac{1}{2}$ per cent. watery solution of Grubler's neutral red. Steam and filter into tubes and sterilise in the steamer for half an hour on three successive days. Make also a stock of broth containing twice the above quantity of neutral red, and four times the quantity of the other constituents except water. Make neutral red agar by adding 10 c.c. per litre of $\frac{1}{2}$ per cent. solution of neutral red and 0.5 per cent. glucose to the ordinary agar medium just before filtration.

Process.—Shake the water, and to a series of broth tubes add $\frac{1}{2}$, 2, 10 c.c. or further quantities of the water. To about 40 c.c. of the water add one tube of four times strength broth, and if the water is suspected to be very pure, one or two litres may be treated in the same manner. All the tubes and bottles are incubated at 37° to 38° C., and examined daily. In from 2 to 3 days, or at the latest a week, if *B. coli* is present, a complete reaction (fluid quite yellow, with marked fluorescence) or a partial reaction (orange, with more or less fluorescence) will be obtained.

If a reaction is obtained, select the tube containing the smallest quantity of water giving the reaction, take one loop of the mixture and add it to 10 c.c. of sterile water, mix thoroughly, brush a little in the ordinary way over two agar plates, incubate at 37° C. for 24 hours, examine and subcultivate in the ordinary way.

¹ Makgill, *Journ. of Hygiene*, 1901, p. 430. Savage, *ibid.*, p. 437. Savage, *Public Health*, May 1902, p. 485.

From an examination of the tubes it is possible to make a good comparative estimation of the number of organisms of the Coli group present. Thus if a positive reaction is obtained with 2 c.c. of water and not with 1 c.c., at least 50 organisms are present in 100 c.c. of water.

W. C. C. Pakes' method.—Two litres of the water are filtered through a special apparatus described as follows:¹

“A small bougie is used and the filtration is from within outwards. As this is very slow, it is necessary to exert pressure as well as suction. The apparatus for this purpose consists (see fig. 80) of a brass cylinder with two shoulders. The neck at each end is provided with an opening with a screw thread. Into the top screw fits a nut carrying a manometer; into the bottom a brass box carrying the small bougie. Leakage is prevented by means of the india-rubber washers shown in the diagram. Into the top shoulder a tube is inserted with a tap. The whole of the brass work is tinned both inside and outside.



FIG. 80.

“In order to sterilise the apparatus the bougie is put in place, and the box is screwed on loosely. The manometer is removed and the whole apparatus heated in the autoclave at 120° C. for twenty minutes. It is then taken out and allowed to cool. The box carrying the bougie is now screwed tight and the water poured into it. In order to do this, the neck of the Winchester quart is well flamed and inserted into the neck of the cylinder. The Winchester quart is then inverted and the water allowed to run in. As the water is running in the air is forced out, and no accidental contamination is likely to take place. When the bottle is empty the nut carrying the manometer, having been held in boiling water for a short time, is screwed in. The tube is connected with a glass cylinder, and the tap is opened to allow the compressed air to enter. The pressure is kept at forty-five pounds. The tube at the bottom of the box carrying the bougie is connected with pressure tubing to an aspirator. Two litres take rather less than twenty minutes to come through.

“When all the water has run through, the bougie is removed and the mouth plugged with sterile cotton-wool; 10 c.c. of sterile water or broth are now poured into the bougie, and the bacteria are brushed into the broth with a special sterile test-tube brush. When this is completed the brush is removed and the cotton-wool plug reinserted. The organisms originally present in 2,000 c.c. are now contained in 10, *i.e.*, the water has been concentrated 200 times. Any fixed volume of this emulsion will therefore represent 200 times its volume of the original water; 0.1 c.c. will represent 20 c.c., and so on.”

1 c.c. and 5 c.c. of the original water and from 1 to 2 c.c. or more of the

¹ *Public Health*, March 1901, 423.

emulsion are injected into glucose formate broth¹ and incubated at 42° C. in Buchner tubes, from 18 to 24 hours.

The tubes are then examined, and those which show no growth are replaced for another twenty-four hours, and if at the end of that time there is still no growth, for a further twenty-four hours. Those tubes which show growth are subcultivated and further examined in the ordinary way.

The number of *B. coli* present is computed from the smallest quantity of water yielding the organism, remembering that every 1 c.c. of the emulsion = 200 c.c. of original water.

Phenol Gelatin method.—Klein² recommends this method “for the detection of *Bacillus coli*, bacillus of typhoid, and a certain variety of *Proteus Zenkeri*, a normal inhabitant of sewage.” Phenolated broth and phenolated gelatin are prepared by adding 1 c.c. of a 5 per cent. phenol solution to every 100 c.c. of broth or of gelatin.

1000 c.c. of the water to be examined is filtered through a sterilised Berkefeld or Pasteur pressure filter. Klein uses a special apparatus for holding the filter; we prefer the simpler apparatus figured in the diagram. (See fig. 81.)

A is a piece of stout glass tubing about two feet long. The Berkefeld filter candle B is fitted into this by means of the tightly-fitting rubber ring C. The nozzle of the candle is directly connected by means of a short piece of pressure tubing with a glass tube passing into the pressure flask E, which is in its turn connected with a filter pump. A litre flask of the water to be examined is inverted into the tube A, round the top of which sterile cotton wool is packed; as the level of the water in the tube falls, bubbles of air pass up into the flask, and so keep the level of the water in the tube constant until nearly all the water is filtered.

The inverted flask and cotton wool prevent the water from becoming contaminated with air organisms during filtration. All the parts of this apparatus are easily sterilised.

After filtering the water through the sterilised apparatus, the candle is taken out, “and 10 c.c. of sterile water are measured into a sterile glass dish; the whole surface of the candle is well brushed into these 10 c.c. of water by means of a thoroughly clean sterile nail-brush.”

“Of the above distribution then, to each phenol gelatin or phenol broth tube 0.5 c.c. or 1 c.c. is added, the gelatin is shaken and poured out into a plate, and after setting kept at 20° C., the phenol broth is incubated at 37° C. From the phenol broth incubated for twenty-four hours, plates in phenolated gelatin are then made. If in the original water the *Bacillus coli communis*, or the sewage variety of *Proteus Zenkeri* or the typhoid

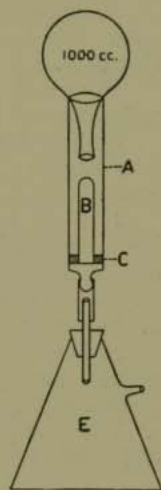


FIG. 81.

¹ The glucose formate broth is made by adding to ordinary meat infusion 1 per cent. peptone, .5 per cent. NaCl, 2 per cent. glucose and 0.4 per cent. sodium formate. When these are dissolved by heating, the medium is neutralised and then 2 c.c. of $\frac{N}{1}$ NaOH added. The broth is then boiled in the steam steriliser for 20 minutes, filtered, poured into the test-tubes, and sterilised for 20 minutes on three succeeding days.

² *Micro-organisms and Disease.*

bacillus be present, the phenol broth cultivation will be found uniformly turbid after twenty-four hours' incubation."

"The phenol gelatin plates when ready, after two to four days' incubation at 20° C., must be carefully examined, and all the surface colonies which resemble in aspect the above organisms have to be tested by fresh preparation, by flagella staining, and by subcultures."

It is also advisable to make at the same time surface cultures on gelatin, as typical colonies of *Bacillus coli communis* or typhoid are more easily recognised when growing on the surface of gelatin than when in the depth. The typical colonies must always be identified by subcultivation and microscopical examination.

Microscopical and Cultural Characters of B. coli communis and B. typhosus.

Microscopical appearance.—*B. coli communis* is, as a rule, a short fat bacillus, having almost the appearance of a coccus. Little or no movement. Stained for flagella (*see* any modern practical Bacteriology). It will be seen, as a rule, to have only three or four. If, however, the staining is done with great care, as many as eight may be seen.

B. typhosus.—In fresh broth cultivations many long threads will be seen. The bacilli themselves are short rods having a length of about twice that of *B. coli*.

Movement in fresh broth culture is very rapid, the chains and bacilli wriggling and darting about.

Stained for flagella, each bacillus will be seen to have a number of long flagella; if stained very carefully as many as sixteen may be seen. Seven to ten is a more usual number to find.

It must be remembered that the Coli group of organisms is a very large one, and that many organisms belonging to this group differ in certain cultural characters from the typical *Bacillus coli communis*; but for all practical purposes, since the near allies of the typical bacillus are as important to the analyst as is the organism itself, any organism which has the microscopical characters already described, does not liquefy gelatin, and produces gas in gelatin shake culture may be concluded to be a variety of *Bacillus coli*.

The following table gives the chief cultural characters of typical *Bacillus coli communis* and *Bacillus typhosus*, according to Delépine.¹

Media.	<i>Bacillus coli communis.</i>	<i>Bacillus typhosus.</i>
1. Alkaline peptone, bouillon gelatin. Alk. = carbonate sodium, 2: 1000, 22° C.	Growth rapid — superficial colonies often thick and more coarsely granular than in B.t. (24 to 48 hours).	Growth distinctly slower, superficial colonies usually very thin and surface marked by wavy bundles (48 hours).
2. Slightly acid potato gelatine, 22° C.	Do., do.	Growth very slow; colonies remain small and transparent.
3. Slightly acid potato, 37° C.	Rapid abundant growth or dark-brownish yellow (24 to 48 hours).	Transparent, very thin glaze-like growth (24 to 48 hours).
4. Glucose gelatine, stab or diffuse culture, 22° C.	Abundant production of gas bubbles (24 to 48 hours).	No production of gas.
5. Alkaline peptone bouillon, 37° C.	Production of indol, variable in green tints (48 hours).	No production of indol.

¹ *Med. Chronicle*, Nov. 1897.

Media.	<i>Bacillus coli communis</i> .	<i>Bacillus typhosus</i> .
6. Feebly alkaline litmus lactose agar peptone bouillon, 37° C.	Production of acid (litmus turns red, 24 to 48 hours).	Litmus never turns red.
7. Separated sterilised milk, 37°.	Coagulated (24 to 48 hours).	Never coagulated.
8. Recent (20 hours) bouillon culture and 1 part in 10 of blood, or blood serum from typhoid patient.	Usually no stoppage of motion and no agglomeration within 1 hour.	Variable amount of immobilisation and agglomeration usually well marked within 5 minutes.
1. Size.	Variable, especially in cultures. 0.3 to 0.6 μ .	Variable; there are always some larger and thicker bacilli among the others; 0.7 to 0.9 μ .
2. Number of cilia.	Maximum 6. ¹ Not easy to stain.	10 to 12. Easy to stain by Van Ermengem's method.
3. Staining reaction.	Not well stained by Gram's method. Stains well by alkaline methylene blue (<i>Loeffler</i>), <i>id.</i> , carbol fuch sine (<i>Ziehl</i>).	

Widal's Reaction.—No bacillus should be concluded to be the typhoid bacillus unless it gives a positive Widal reaction.

A broth culture is taken of the suspected organism and nine loopfuls of this culture are placed on a slide round a drop of serum obtained from a patient suffering from typhoid fever. An emulsion is then made by rubbing the serum and broth together.

Bacillus typhosus usually shows decidedly less mobility, and marked clumping in less than 5 minutes, whereas, as a rule, *Bacillus coli communis* shows no stoppage of motion and no clumping in the first 60 minutes, when Prof. Delépine's rapid film method is used.

Control experiments should always be made at the same time with known typhoid cultures, and with known *coli* cultures.

§ 377. *Proteus Vulgaris*.—*Gelatin Plates*.—Appears as small greyish dots after 15 to 20 hours' incubation at 20° C. The colonies are irregular in outline, the irregularities consisting of mobile bacilli which rapidly increase and cause neighbouring colonies to coalesce. Liquefaction of the gelatin is rapid.

Gelatin Stab.—Liquefaction commences in a few hours in the upper portion, forming in a few days a funnel-shaped liquefied mass along the line of inoculation.

Agar-agar Plates.—Moist, sticky grey growth.

Broth.—Turbid in twenty-four hours if incubated at 37° C. After some days a pellicle forms on the surface.

Microscopically, the bacilli from a liquefied gelatin culture will be seen to be very mobile. The bacilli are protean in form, cocci, short oval, dumb bell, cylindrical and filamentous.

§ 378. *Streptococci and Staphylococci*.—Houston employs the following method:—"Surface agar cultures (at 37° C.) should be made of the sample of water, and all minute colonies resembling streptococci subcultured in broth; afterwards resorting to further subcultures if a microscopic examination of these broth tubes yielded seemingly positive results. As regards the amount of the samples to be used in making these agar

¹ The number of cilia or flagella found depends upon the method and care with which the staining is done (see page 564).

cultivations, it was indicated that in certain cases it is necessary to filter large quantities of the water through a sterile Pasteur filter, and to use the 'filter brushings' of the candle; in others, the water may be examined directly; in other cases, again, the water may require to be first greatly diluted with a definite volume of sterile water."

There are a number of species of *streptococci*; for the purpose of water examination it is, however, unnecessary to distinguish between them.

Broth.—In twelve hours the broth is turbid. After a few days the growth sinks to the bottom of the test-tube, leaving the broth quite clear.

Microscopically the organism, which should be stained by *Gram's method* will be seen to consist of chains of cocci of varying length as well as single cocci.

There are also several species of *staphylococci*.

Agar-agar Surface Culture.—The colonies appear as yellowish or white dots in twenty-four hours. The organism should be subcultured in broth and stained by *Gram's method*, when the cocci will be seen arranged in chains or triangular masses.

Bacillus enteritidis sporogenes.—The water, unless very foul, must be passed through a Pasteur-Chamberland filter in exactly the same manner as in the search for *B. coli* by Klein's method. Various quantities of the brushings from the filter are then injected into sterile milk tubes. (It has been found necessary to heat the milk tubes just before using, in order to drive off the dissolved oxygen, and allow them to cool to below 80° C.)

The tubes are then heated to 80° C., and kept at this temperature for ten minutes, care being taken that the upper portion of each tube is heated, or the upper portion of each tube may be passed through a Bunsen burner flame. The tubes are then at once incubated anaerobically at 37° C. by Buchner's method. This method consists in placing the tubes in a wider tube containing pyrogallic acid, and pouring on the pyrogallic acid a 5 per cent. solution of caustic potash, and at once sealing the outer tube with a well-fitting rubber cork. About one gramme of pyrogallic acid should be allowed for every 10 c.c. of potash solution. After twenty-four hours' incubation, the casein of the milk will have coagulated and separated from the turbid whey. A large amount of gas is produced at the same time, and the layer of casein is usually blown up the test-tube. If a small quantity of the milk whey (1 c.c.), thus containing products of the *B. enteritidis* culture, be injected into a guinea-pig, the animal will die in about forty-eight hours; the peritoneal cavity will be found to be much inflamed, and to contain a large amount of fluid. This fluid should be subcultured upon blood serum, and in milk under anaerobic conditions; a portion should also be sealed up in a small glass pipette and placed in the cool incubator for some days, in order that spores may form.

Rod-shaped and cylindrical bacilli, which stain well by *Gram's method*, will be found in the milk and serum cultures, and in the peritoneal fluid. Spores will be found in the serum cultures, and in the sealed-up peritoneal fluid.

Klein gives the following dimensions for the bacilli and spores:—

Bacillus—length, 1.6 to 4.8 μ ; thickness, .8 μ . Free spores—length, 1.6 μ ; thickness, .8 to 1 μ .

Bacillus anthracis.—Frankland, in *Micro-organisms in Water*, gives the following experiment:—From 1 to 3 c.c. of the water containing anthrax spores were mixed with 1 c.c. of sterile broth, and heated for periods of from two to five minutes to 50° C., to 70° C., or to 90° C., after

which they were submitted to ordinary plate cultivation. After heating to 50° C. for five minutes, the 100,000 water bacteria previously contained in 1 c.c. of the contaminated water were reduced to from 35 to 39 colonies per c.c., and several of these were recognisable as colonies of *B. anthracis*. Heating to 70° C. for two minutes reduced the number from 30 to 10 colonies per c.c., in which from 4 to 10 were recognised as those of anthrax bacilli. Heating to 90° C. for two minutes resulted in the appearance of only 7 to 10 colonies, of which 3 to 6 were found to be those of anthrax.

Cultivations.—

Gelatin Surface.—Colonies appear as grey irregular dots after twenty-four hours' incubation. After from two to three days the margin, when viewed with a lens, may be seen to be made up of twisted fibres. At the same time, commencing liquefaction of the gelatin causes the centre of the colony to sink.

Agar-agar Plate Cultivations.—Colonies appear in twenty-four hours as round, oval, or heart-shaped colonies. Under a lens the surface of each colony is seen to be marked with fine lines. In older colonies, twisted fibres, like platts of hair, may be seen.

Microscopically, the bacilli are seen to be short, rod-shaped organisms, with square ends, varying from 2 to 10 μ in length and 1 to 1.5 μ in breadth. Chains of these bacilli are usually found. The bacilli stain by Gram's method. Spore formation is so well marked that it is not, as a rule, necessary to stain specially for spores in order to observe them. *In impression specimens*, made from agar-agar plates, the hair-like appearance will be seen to be due to chains of bacilli, and the spores will be seen as highly refractory vacuole-like bodies.

Spirillum cholerae asiaticae.—

Cultivation gelatin surface.—In about eighteen hours it begins to appear as minute points of growth of an opaque white. The gelatin after a few days is liquefied.

Agar-agar surface.—Moist greyish-white growth.

Potato.—Light yellow growth, difficult to see.

The vibrios under the microscope are seen to be slender rods, curved so as to form commas. And two are often joined together forming S-shaped bodies. Several may also be joined to form a spirillum.

Water may be examined for the cholera vibrio in the following manner:—Place 100 c.c. in each of five or six sterile conical flasks, plugged with cotton wool; add to each flask 1 gramme of peptone and 0.5 gramme of sodium chloride. Incubate at 37° C. for from eight to twelve hours. From the surface of one or all of the liquids prepare films and stain them with aqueous gentian-violet or aqueous fuchsine. If any comma bacilli are seen, some of the surface liquid should be diluted with sterile water and sub-cultures made on agar-agar, on which the organism will be visible after ten hours' incubation; pure cultures may in this way be obtained.

Nitroso-indol or Cholera Red Reaction.—Test one of the flasks containing the water and peptone, by adding to it a few drops of pure sulphuric acid; if cholera is present a distinct crimson hue will be obtained after a very few hours' incubation.

Durham-Gruber Reaction.—Dilute the serum of an animal immunised against cholera with ten times its volume of distilled water. Mix this with an emulsion of the suspected vibrios. Incubate in a narrow test-tube for one hour at 38° C. If the vibrios are those of cholera, they will collect together as a white deposit at the bottom of the tube, the

supernatant fluid being clear. The vibrios under the microscope will be found to be motionless and aggregated into clumps.

§ 379. *Animal forms*.—Without taking into consideration various animal water organisms which can be seen with the naked eye (*e.g.*, water fleas), but confining the attention solely to the microscopic forms of life, there is such an inexhaustible variety of the latter that it takes a special study to

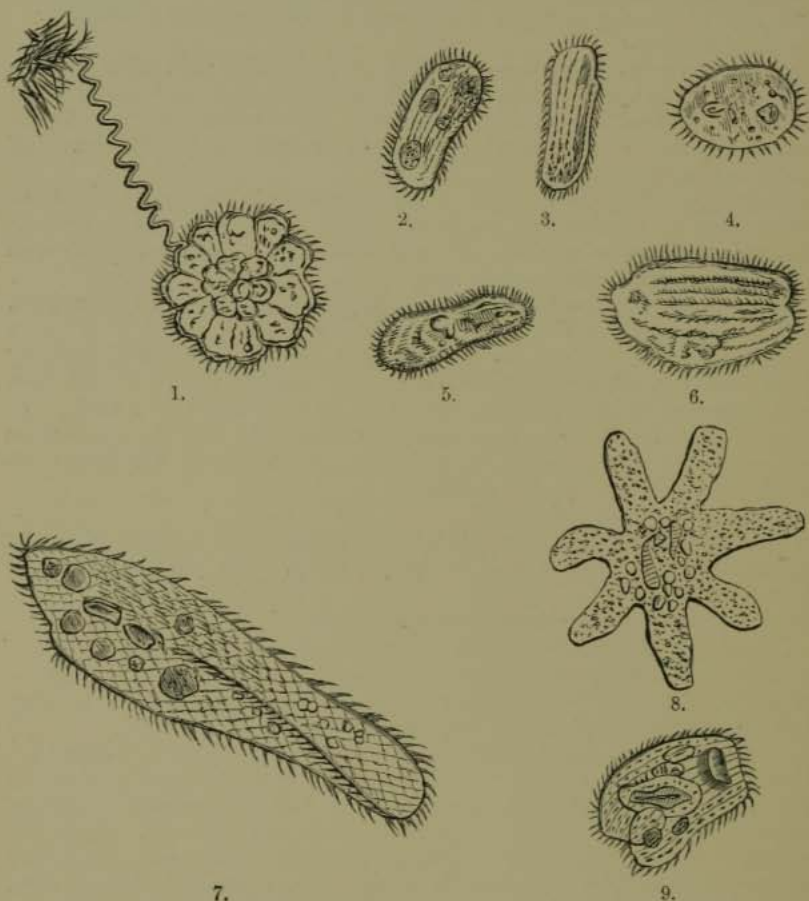


FIG. 82.

1. Colony of Vorticella. 2. Oxytricha lingua. 3. Pellionella. 4. Glaucocoma scintillans. 5. Glaucocoma gibba. 6. Paramecium aurelia. 7. Paramecium caudatum. 8. Amoeba. 9. Chilodon.

ascribe each form to its particular species; fortunately, this to the analyst is not necessary, and it may at once be laid down that if the deposit from a reasonable quantity of water (for example, a gallon) exhibits forms of infusorial life, the water cannot be pure. For although from all natural water, if a sufficient bulk be taken, it is possible to extract life-forms, yet all good drinking-water is devoid of such when moderate quantities are taken for the search. There are, however, certain animalcules (types

of which are figured) that specially point to sewage contamination, such as *Amœba* (fig. 8), a soft, colourless granular mass, which puts out finger-like processes. *Euglena*, which lives on decaying vegetable matter, is fusiform of a red or green colour, very flexible, with a single flagellum. *Vorticella* (fig. 1), an oval animalcule attached by a contractile spring-like pedicle. *Paramœcium aurelia* (fig. 6), colourless and finely ciliated; it moves swiftly. Contractile vesicles and nucleus conspicuous. In addition to these, *Cercomonas*, *Colomonas*, and many others, will often be observed. Of the larger organisms the most common are *Cyclops*, (belonging to the Crustacea), *Anguillula*, a small colourless worm, and *Nais*, a worm with bristles.

Fresh-water sponges occur as a thin incrustation on objects; portions often die, putrefy, and impart a disagreeable odour to water. The spiculæ, in the form of fine needles, are most indestructible and easy of recognition. The presence of sponge in water mains often occurs, and always is a misfortune. The examination of a water pipe for sponge is best performed by splitting the pipe and scraping the upper, not the lower surface, for the fresh-water sponge is always found with its crater-like openings downwards.

A genus of colony-building infusoria, not unlike *Volvox* in general appearance, has been discovered in the waters of Massachusetts, and is described and figured in *23rd Annual Report of the Massachusetts State Board of Health* under the name of *Uroglena*. It is important, because it imparts to water a most offensive odour. This odour is apparently not a sign of the decomposition of *Uroglena*, but is evolved when the several united colonies disintegrate.

It will not be necessary to use any special colouring-agents to see infusoria, for they are quite sufficiently visible without any special reagent. M. A. Certes has, however, found out a colouring-matter which concentrates itself in certain parts of living infusoria, and may, therefore, be of some utility; this reagent is quinoleine or cyanine, and it appears chiefly to colour the fatty parts of the protoplasm. He considers it a reagent for living fat.¹

§ 380. *Standards of Purity*.—It is now generally admitted that no hard and fast standards can be fixed for drinking waters either from a chemical or bacterial standpoint. Every water should be judged with a full knowledge as to the surroundings of the supply and the geological formation from which it is drawn.

When waters are kept under constant observation it is often possible to draw valuable inferences from slight variations. Especially is this so in the case of nitrates, which in unpolluted waters will rarely show any great variations at different times.

With regard to organic carbon and nitrogen, Frankland considers that some importance should be attached, not only to the gross amount of carbon and nitrogen, but also to the relative proportion they bear to one another, and this especially with regard to surface waters. In deep springs the limit of organic carbon should be about $\cdot 1$ in 100,000, the amount actually found ranging from $\cdot 02$ to $\cdot 1$ in 100,000 of water. In spring water the ratio of C : N varies from 2 : 1 to 6 : 1, with an average of about 2 : 1. Surface water derived from cultivated land, containing C : N below the proportion of 6 : 1, and having more than $\cdot 3$ part of the carbon in 100,000, with these data supported by other indications, should be condemned as

¹ *Comptes Rendus*, xcii. 425.

unfit for dietetic purposes. In surface water, if carbon is to nitrogen as 3 : 1, the organic matter is of animal origin; if it be as high as 8 : 1, it is chiefly, if not exclusively, of vegetable origin. The content of organic elements in sewage, of course, varies according to its state of dilution; but it usually averages from four to five parts of carbon per 100,000, and from one to two parts of nitrogen per 100,000. Frankland divides waters into two sections, according to the results of a combustion.

SECTION I.—UPLAND SURFACE WATER.

CLASS I.—Water of great organic purity, containing a portion of organic elements (organic carbon and organic nitrogen), not exceeding 0·2 part in 100,000 parts of water.

CLASS II.—Water of medium purity, containing from 0·2 to 0·4 part of organic elements in 100,000.

CLASS III.—Water of doubtful purity, containing from 0·4 to 0·6 part of organic elements in 100,000.

CLASS IV.—Impure water, containing more than 0·6 part of organic elements in 100,000.

SECTION II.—WATER OTHER THAN UPLAND SURFACE.

CLASS I.—Water of great organic purity, containing a proportion of organic elements not exceeding 0·1 part in 100,000.

CLASS II.—Water of medium purity, containing from 0·1 to 0·2 part of organic elements in 100,000.

CLASS III.—Water of doubtful purity, containing from 0·2 to 0·4 part of organic elements in 100,000.

CLASS IV.—Impure water, containing upwards of 0·4 part of organic elements in 100,000.

Wanklyn's standards :—

	Parts per million.	Grain per gallon.	
Albuminoid ammonia,	0·05	0·0035	Great purity.
" "	0·10	0·0070	Organically safe.
" More than,	0·10	0·0070	More or less impure.

Professor Tidy's oxygen process :—

	Parts per million.	Grain per gallon.	
Oxygen used in 3 hours,	0·5	0·035	Great purity.
" "	0·5 to 1·5	0·035 to 0·105	Medium purity.
" "	1·5 to 2·1	0·105 to 0·147	Doubtful.

Blair's oxygen process at 100° C. :—

	Parts per million.	Grain per gallon.	
Oxygen absorbed less than,	2	0·14	Great purity.
" between,	2 and 4	0·14 to 0·28	Medium purity.
" "	4 and 6	0·28 to 0·42	Suspicious (unless peaty).
" over,	6	0·42	Impure.

Blair's classification according to organic nitrogen as estimated by Kjeldahl's process :—

	Parts per million.	Grain per gallon.	
Organic nitrogen less than,	0·06	0·0042	Great purity.
„ between, 0·06 and 0·12		0·0042 to 0·0084	Medium purity.
„ over,	0·12	0·0084	Suspicious unless peaty (peaty waters may contain anything under 0·25 part per million, 0·0175 grain per gallon, and yet be potable).
„ „	0·32	0·224	To be condemned, even if peaty.

APPENDIX TO WATER ANALYSIS.

STANDARD SOLUTIONS AND REAGENTS, ETC., ALPHABETICALLY ARRANGED.

Parts by Weight: Liquids by Measure.

§ 381. *Ammonium Chloride*.—(1) Ammonium chloride, 3146 part; pure water, 1000 parts; dissolve, 1 c.c. contains 0·001 gm. of ammonia; (2) 100 c.c. solution (1) diluted to 1000 c.c. with pure water. 1 c.c. = 0·0001 gm. of ammonia.

Calcic Chloride Solution.—Iceland spar, or other pure form of calcic carbonate, 1 part. The calcic carbonate is converted into chloride by evaporating to dryness with pure dilute hydrochloric acid, and the calcic chloride thus obtained is dissolved in water and made up to 1000 parts.

Copper Sulphate.—Sulphate of copper, 30 parts; pure water, 1000 parts; dissolve.

Diphenylamine Test for Nitrates.—1 gm. of diphenylamine is dissolved in 10 to 30 c.c. of absolute alcohol and diluted with 150 c.c. of sulphuric acid. A c.c. of this solution, added direct to the water, strikes a blue colour in the presence of nitrates, which colour, on dilution, changes to a yellow-green. It gives the same colour with nitrites.

Ferrous Chloride Solution.—A solution of pure ferrous sulphate is precipitated with sodic hydrate, and washed thoroughly with pure water, and then dissolved in the smallest possible quantity of pure hydrochloric acid.

Griess' Reagent for Nitrous Acid or Nitrites.—(A.) 1 gm. sulphanilic acid, 14·7 grms. glacial acetic acid, 285 c.c. water.

(B.) 0·2 gm. *a*-naphthylamine, 14·7 grms. glacial acetic acid, 325 c.c. water. Mix equal parts just before use.

Meta-phenylenediamine.—(A.) 8 grms. meta-phenylenediamine hydrochloride, 1000 c.c. water.

(B.) Sulphuric acid (1 : 3).

Meta-phosphoric Acid.—Meta-phosphoric acid 100 parts, made up to 1000 with distilled water; 10 parts should contain no appreciable amount of ammonia.

Molybdic Solution.—Molybdic acid, 1 part; solution of ammonia, specific gravity 0·960, 4 parts; nitric acid, 1·20 specific gravity, 15 parts.

The molybdic acid is dissolved in the ammonia, filtered, and poured with constant stirring into the nitric acid. It should be kept in the dark, and freed by decantation from any precipitate which may form.

Nessler Solution.—35 parts of potassium iodide are dissolved in 100 parts of water; 17 parts of mercuric chloride are boiled in 300 parts of water, and then cooled. The mercuric solution is added to the potassium iodide, little by little, until a permanent precipitate is produced. The liquid is now made up to 1000 parts with a solution of sodic hydrate of 20 per cent. strength. Lastly, the reagent is made more sensitive by the final addition of a little more of the mercuric chloride solution, until a permanent precipitate begins to form. The solution is put on one side to deposit, and the clear liquid decanted for use. It is best to keep that intended for use in a small bottle, while the larger stock is carefully stoppered down.

Palladium Solution.—Palladium, 1 part is dissolved in nitro-hydrochloric acid, and evaporated to dryness at 100° C., 50 parts of hydrochloric acid are added, and 2000 of water, ultimately making the bulk up to 2370 parts.

Potassium Iodide Solution.—(A.) Potassium iodide 1 part in 10 of water, for use in the oxygen or Forchhammer process.

(B.) Potassium iodide 1, water 100,000, for use in the volumetrical estimation of iodine.

Potassium Monochromate.—Potassium monochromate 50 parts, dissolved in 1000 parts of distilled water. To ensure absence of chlorides it is recommended to add a little nitrate of silver until a permanent red precipitate forms, which is allowed to settle, and the clear solution used.

Potassium Permanganate.—(A.) *Alkaline*—Potassium permanganate 8 parts; potassium hydrate 200 parts; distilled water 1100 parts. The solution is boiled rapidly down to 1000 parts, and kept in properly stoppered bottles.

(B.) *Standard Volumetric Solution for Oxygen Process.*—395 part of potassic permanganate is dissolved in 1000 of water. Each c.c. contains '0001 gramme of available oxygen, and each one grain contains '0001 grain.

Silver Nitrate, Standard Solution of.—4.7887 parts of silver nitrate are dissolved in pure water, and made up to 1000 parts. The solution may be standardised, if necessary, by the sodium chloride solution, 1 c.c. = '001 gm. chlorine; 10 grains = '01 grain chlorine.

Soap, Standard Solution of.—150 parts of lead plaster are triturated in a mortar with 40 parts of dry potassic carbonate, and made into a cream with the addition of absolute alcohol; when dissolved, filter, and by the addition of water reduce the alcoholic strength to that of proof spirit. The solution of soap is then reduced to the proper strength by proof spirit. If working in c.c. and grms. it should be of such a strength that 14.25 c.c. are required to form a permanent lather with 50 c.c. of the calcic chloride solution; if working in grains, then it will be most convenient to make it of that strength that it just forms a permanent lather when 180 grain measures are shaken with 1000 grain measures of the standard calcic solution, equalling water of 8°.

Sodium Chloride Solution.—1.648 parts of pure sodium chloride are dissolved in water, and the solution made up to 1000 parts. Pure sodium chloride can be obtained by passing hydrochloric acid gas through a saturated solution of commercial sodic chloride; a precipitate of pure sodic chloride falls, which may be collected, and dried in the hot air oven at 260°

to 300°. Each c.c. contains .001 grm. chlorine, or each grain contains .001 grain chlorine.

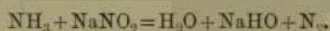
Sodium Hydrate, Solution for Estimation of Nitrates.—56 parts of metallic sodium are dissolved little by little in 1000 parts of water.

Sodic Hyposulphite.—One part of crystallised sodium hyposulphite in 1000 parts of water.

Sodic Nitrite Solution.—406 part of silver nitrite is dissolved in a little distilled water, and pure sodic chloride is added until no more argentic chloride is thrown down. Dilute with water to 1000 parts. Allow to settle. Then of the clear solution take 100 parts, and dilute to 1000; 1 c.c. equals .00001 grm. of N_2O_3 ; similarly each 10 grains equals .0001 grain.

Starch Solution.—1 part of starch is rubbed up with 20 parts of boiling water. The liquid is filtered, boiled, and after being allowed to stand for twenty-four hours, the clear liquid is syphoned off.

Sulphuric Acid (Pure).—Pure sulphuric acid is obtained by heating the strong acid with a few crystals of potassic permanganate until the green colour is permanent. It may still contain nitrogen as ammonia. Blair purifies the acid from ammonia as follows:—5 c.c. of the purified acid are neutralised with ammonia-free soda, the mixture distilled, and the ammonia estimated in the distillate; then the amount of sodium nitrite necessary to destroy the ammonia is calculated from the equation—



Each part of ammonia will require four times its weight of sodic, or five times its weight of potassic nitrite to satisfy the equation. A slight excess of this quantity is added to the acid and the acid heated on a sand-bath, and the drops distilled, tested, from time to time, for nitrous acid by means of starch and potassium iodide; when the distillate is free from nitrous acid, the operation is finished, and the acid is (for the purposes of water analysis) pure—that is, free from carbon and free from nitrogen.

Zinc Iodide Starch Solution.—4 grms. of starch are triturated in a mortar with a little water and transferred little by little to a boiling solution of 20 per cent. zinc chloride. The heating is continued, replacing the water from time to time, until the starch is dissolved and the fluid is almost colourless; the liquid is then diluted, and, after adding 2 grms. of zinc iodide, filtered; the filtration is slow, but a clear filtrate can ultimately be obtained, which, kept in closed bottles in the dark, preserves its properties for an indefinite time. On acidifying with sulphuric acid, if properly made, there should be no blue colour produced.

TABLE LXVIII.—REDUCTION OF CUBIC CENTIMETRES OF NITROGEN TO GRAMMES.

$$\log \frac{0.0012562}{(1 + 0.00367)} 760 \text{ for each tenth of a degree from } 0^\circ \text{ to } 30^\circ \text{ C.}$$

c. c.	00	01	02	03	04	05	06	07	08	09
T										
0	$\bar{6}$.21824	808	793	777	761	745	729	713	697	681
1	665	649	633	617	601	586	570	554	538	522
2	507	491	475	459	443	427	412	396	380	364
3	349	333	318	302	286	270	255	239	223	208
4	192	177	161	145	130	114	098	083	067	051
5	035	020	004	*989	*973	*957	*942	*926	*911	*895
6	$\bar{6}$.20879	864	848	833	817	801	786	770	755	739
7	723	708	692	676	661	645	629	614	598	583
8	567	552	536	521	505	490	474	459	443	428
9	413	397	382	366	351	335	320	304	289	274
10	259	244	228	213	198	182	167	151	136	121
11	106	090	075	060	045	029	014	*999	*984	*969
12	$\bar{6}$.19953	938	923	907	892	877	862	846	831	816
13	800	785	770	755	740	724	709	694	679	664
14	648	633	618	603	588	573	558	543	528	513
15	497	482	467	452	437	422	407	392	377	362
16	346	331	316	301	286	271	256	241	226	211
17	196	181	166	151	136	121	106	091	076	061
18	046	031	016	001	*986	*971	*956	*941	*926	*911
19	$\bar{6}$.18897	882	867	852	837	822	807	792	777	762
20	748	733	718	703	688	673	659	644	629	614
21	600	585	570	555	540	526	511	496	481	466
22	452	437	422	408	393	378	363	349	334	319
23	305	290	275	261	246	231	216	202	187	172
24	158	143	128	114	099	084	070	055	041	026
25	012	*997	*982	*968	*953	*938	*924	*909	*895	*880
26	$\bar{6}$.17866	851	837	822	808	793	779	764	750	735
27	721	706	692	677	663	648	634	619	605	590
28	576	561	547	532	518	503	489	475	460	446
29	432	417	403	388	374	360	345	331	316	302

TABLE LXIX.—FOR THE CALCULATION OF TOTAL SOLIDS FROM FAT AND SPECIFIC GRAVITY (RICHMOND).

Specific Gravity.	Fat per cent.									
	1	2	3	4	5	6	7	8	9	10
	Total Solids per cent.									
22.0	5.77	5.89	6.01	6.13	6.25	6.37	6.49	6.61	6.73	6.85
22.5	5.90	6.02	6.14	6.26	6.38	6.50	6.62	6.74	6.86	6.98
23.0	6.02	6.14	6.26	6.38	6.50	6.62	6.74	6.86	6.98	7.10
23.5	6.15	6.27	6.39	6.51	6.63	6.75	6.87	6.99	7.11	7.23
24.0	6.27	6.39	6.51	6.63	6.75	6.87	6.99	7.11	7.23	7.35
24.5	6.40	6.52	6.64	6.76	6.88	7.00	7.12	7.24	7.36	7.48
25.0	6.52	6.64	6.76	6.88	7.00	7.12	7.24	7.36	7.48	7.60
25.5	6.65	6.77	6.89	7.01	7.13	7.25	7.37	7.49	7.61	7.73
26.0	6.77	6.89	7.01	7.13	7.25	7.37	7.49	7.61	7.73	7.85
26.5	6.90	7.02	7.14	7.26	7.38	7.50	7.62	7.74	7.86	7.98
27.0	7.02	7.14	7.26	7.38	7.50	7.62	7.74	7.86	7.98	8.10
27.5	7.15	7.27	7.39	7.51	7.63	7.75	7.87	7.99	8.11	8.23
28.0	7.27	7.39	7.51	7.63	7.75	7.87	7.99	8.11	8.23	8.35
28.5	7.39	7.51	7.63	7.75	7.87	7.99	8.11	8.23	8.35	8.47
29.0	7.52	7.64	7.76	7.88	8.00	8.12	8.24	8.36	8.48	8.60
29.5	7.64	7.76	7.88	8.00	8.12	8.24	8.36	8.48	8.60	8.72
30.0	7.77	7.89	8.01	8.13	8.25	8.37	8.49	8.61	8.73	8.85
30.5	7.89	8.01	8.13	8.25	8.37	8.49	8.61	8.73	8.85	8.97
31.0	8.02	8.14	8.26	8.38	8.50	8.62	8.74	8.86	8.98	9.10
31.5	8.14	8.26	8.38	8.50	8.62	8.74	8.86	8.98	9.10	9.22
32.0	8.26	8.38	8.50	8.62	8.74	8.86	8.98	9.10	9.22	9.34
32.5	8.38	8.50	8.62	8.74	8.86	8.98	9.10	9.22	9.34	9.46
33.0	8.51	8.63	8.75	8.87	8.99	9.11	9.23	9.35	9.47	9.59
33.5	8.63	8.75	8.87	8.99	9.11	9.23	9.35	9.47	9.59	9.71
34.0	8.75	8.87	8.99	9.11	9.23	9.35	9.47	9.59	9.71	9.83
34.5	8.88	9.00	9.12	9.24	9.36	9.48	9.60	9.72	9.84	9.96
35.0	9.00	9.12	9.24	9.36	9.48	9.60	9.72	9.84	9.96	10.08
35.5	9.12	9.24	9.36	9.48	9.60	9.72	9.84	9.96	10.08	10.20
36.0	9.24	9.36	9.48	9.60	9.72	9.84	9.96	10.08	10.20	10.32
36.5	9.36	9.48	9.60	9.72	9.84	9.96	10.08	10.20	10.32	10.44
37.0	9.49	9.61	9.73	9.85	9.97	10.09	10.21	10.33	10.45	10.57
37.5	9.61	9.73	9.85	9.97	10.09	10.21	10.33	10.45	10.57	10.69

TABLE LXIX.—FOR THE CALCULATION OF TOTAL SOLIDS FROM FAT AND SPECIFIC GRAVITY (RICHMOND)—*continued.*

Specific Gravity.	Fat per cent.									
	1·1	1·2	1·3	1·4	1·5	1·6	1·7	1·8	1·9	2·0
	Total Solids per cent.									
22·0	6·97	7·09	7·21	7·33	7·45	7·57	7·69	7·81	7·93	8·05
22·5	7·10	7·22	7·34	7·46	7·58	7·70	7·82	7·94	8·06	8·18
23·0	7·22	7·34	7·46	7·58	7·70	7·82	7·94	8·06	8·18	8·30
23·5	7·35	7·47	7·59	7·71	7·83	7·95	8·07	8·19	8·31	8·43
24·0	7·47	7·59	7·71	7·83	7·95	8·07	8·19	8·31	8·43	8·55
24·5	7·60	7·72	7·84	7·96	8·08	8·20	8·32	8·44	8·56	8·68
25·0	7·72	7·84	7·96	8·08	8·20	8·32	8·44	8·56	8·68	8·80
25·5	7·85	7·97	8·09	8·21	8·33	8·45	8·57	8·69	8·81	8·93
26·0	7·97	8·09	8·21	8·33	8·45	8·57	8·69	8·81	8·93	9·05
26·5	8·10	8·22	8·34	8·46	8·58	8·70	8·82	8·94	9·06	9·18
27·0	8·22	8·34	8·46	8·58	8·70	8·82	8·94	9·06	9·18	9·30
27·5	8·35	8·47	8·59	8·71	8·83	8·95	9·07	9·19	9·31	9·43
28·0	8·47	8·59	8·71	8·83	8·95	9·07	9·19	9·31	9·43	9·55
28·5	8·59	8·71	8·83	8·95	9·07	9·19	9·31	9·43	9·55	9·67
29·0	8·72	8·84	8·96	9·08	9·20	9·32	9·44	9·56	9·68	9·80
29·5	8·84	8·96	9·08	9·20	9·32	9·44	9·56	9·68	9·80	9·92
30·0	8·97	9·09	9·21	9·33	9·45	9·57	9·69	9·81	9·93	10·05
30·5	9·09	9·21	9·33	9·45	9·57	9·69	9·81	9·93	10·05	10·17
31·0	9·22	9·34	9·46	9·58	9·70	9·82	9·94	10·06	10·18	10·30
31·5	9·34	9·46	9·58	9·70	9·82	9·94	10·06	10·18	10·30	10·42
32·0	9·46	9·58	9·70	9·82	9·94	10·06	10·18	10·30	10·42	10·54
32·5	9·58	9·70	9·82	9·94	10·06	10·18	10·30	10·42	10·54	10·66
33·0	9·71	9·83	9·95	10·07	10·19	10·31	10·43	10·55	10·67	10·79
33·5	9·83	9·95	10·07	10·19	10·31	10·43	10·55	10·67	10·79	10·91
34·0	9·95	10·07	10·19	10·31	10·43	10·55	10·67	10·79	10·91	11·03
34·5	10·08	10·20	10·32	10·44	10·56	10·68	10·80	10·92	11·04	11·16
35·0	10·20	10·32	10·44	10·56	10·68	10·80	10·92	11·04	11·16	11·28
35·5	10·32	10·44	10·56	10·68	10·80	10·92	11·04	11·16	11·28	11·40
36·0	10·44	10·56	10·68	10·80	10·92	11·04	11·16	11·28	11·40	11·52
36·5	10·56	10·68	10·80	10·92	11·04	11·16	11·28	11·40	11·52	11·64
37·0	10·69	10·81	10·93	11·05	11·17	11·29	11·41	11·53	11·65	11·77
37·5	10·81	10·93	11·05	11·17	11·29	11·41	11·53	11·65	11·77	11·89

TABLE LXIX.—FOR THE CALCULATION OF TOTAL SOLIDS FROM FAT AND SPECIFIC GRAVITY (RICHMOND)—*continued.*

Specific Gravity.	Fat per cent.									
	21	22	23	24	25	26	27	28	29	30
	Total Solids per cent.									
22.0	8.17	8.29	8.41	8.53	8.65	8.77	8.89	9.01	9.13	9.25
22.5	8.30	8.42	8.54	8.66	8.78	8.90	9.02	9.14	9.26	9.38
23.0	8.42	8.54	8.66	8.78	8.90	9.02	9.14	9.26	9.38	9.50
23.5	8.55	8.67	8.79	8.91	9.03	9.15	9.27	9.39	9.51	9.63
24.0	8.67	8.79	8.91	9.03	9.15	9.27	9.39	9.51	9.63	9.75
24.5	8.80	8.92	9.04	9.16	9.28	9.40	9.52	9.64	9.76	9.88
25.0	8.92	9.04	9.16	9.28	9.40	9.52	9.64	9.76	9.88	10.00
25.5	9.05	9.17	9.29	9.41	9.53	9.65	9.77	9.89	10.01	10.13
26.0	9.17	9.29	9.41	9.53	9.65	9.77	9.89	10.01	10.13	10.25
26.5	9.30	9.42	9.54	9.66	9.78	9.90	10.02	10.14	10.26	10.38
27.0	9.42	9.54	9.66	9.78	9.90	10.02	10.14	10.26	10.38	10.50
27.5	9.55	9.67	9.79	9.91	10.03	10.15	10.27	10.39	10.51	10.63
28.0	9.67	9.79	9.91	10.03	10.15	10.27	10.39	10.51	10.63	10.75
28.5	9.79	9.91	10.03	10.15	10.27	10.39	10.51	10.63	10.75	10.87
29.0	9.92	10.04	10.16	10.28	10.40	10.52	10.64	10.76	10.88	11.00
29.5	10.04	10.16	10.28	10.40	10.52	10.64	10.76	10.88	11.00	11.12
30.0	10.17	10.29	10.41	10.53	10.65	10.77	10.89	11.01	11.13	11.25
30.5	10.29	10.41	10.53	10.65	10.77	10.89	11.01	11.13	11.25	11.37
31.0	10.42	10.54	10.66	10.78	10.90	11.02	11.14	11.26	11.38	11.50
31.5	10.54	10.66	10.78	10.90	11.02	11.14	11.26	11.38	11.50	11.62
32.0	10.66	10.78	10.90	11.02	11.14	11.26	11.38	11.50	11.62	11.74
32.5	10.78	10.90	11.02	11.14	11.26	11.38	11.50	11.62	11.74	11.86
33.0	10.91	11.03	11.15	11.27	11.39	11.51	11.63	11.75	11.87	11.99
33.5	11.03	11.15	11.27	11.39	11.51	11.63	11.75	11.87	11.99	12.11
34.0	11.15	11.27	11.39	11.51	11.63	11.75	11.87	11.99	12.11	12.23
34.5	11.28	11.40	11.52	11.64	11.76	11.88	12.00	12.12	12.24	12.36
35.0	11.40	11.52	11.64	11.76	11.88	12.00	12.12	12.24	12.36	12.48
35.5	11.52	11.64	11.76	11.88	12.00	12.12	12.24	12.36	12.48	12.60
36.0	11.64	11.76	11.88	12.00	12.12	12.24	12.36	12.48	12.60	12.72
36.5	11.76	11.88	12.00	12.12	12.24	12.36	12.48	12.60	12.72	12.84
37.0	11.89	12.01	12.13	12.25	12.37	12.49	12.61	12.73	12.85	12.97
37.5	12.01	12.13	12.25	12.37	12.49	12.61	12.73	12.85	12.97	13.09

TABLE LXIX.—FOR THE CALCULATION OF TOTAL SOLIDS FROM FAT AND SPECIFIC GRAVITY (RICHMOND)—*continued*.

Specific Gravity.	Fat per cent.									
	3·1	3·2	3·3	3·4	3·5	3·6	3·7	3·8	3·9	4·0
	Total Solids per cent.									
22·0	9·37	9·49	9·61	9·73	9·85	9·97	10·09	10·21	10·33	10·45
22·5	9·50	9·62	9·74	9·86	9·98	10·10	10·22	10·34	10·46	10·58
23·0	9·62	9·74	9·86	9·98	10·10	10·22	10·34	10·46	10·58	10·70
23·5	9·75	9·87	9·99	10·11	10·23	10·35	10·47	10·59	10·71	10·83
24·0	9·87	9·99	10·11	10·23	10·35	10·47	10·59	10·71	10·83	10·95
24·5	10·00	10·12	10·24	10·36	10·48	10·60	10·72	10·84	10·96	11·08
25·0	10·12	10·24	10·36	10·48	10·60	10·72	10·84	10·96	11·08	11·20
25·5	10·25	10·37	10·49	10·61	10·73	10·85	10·97	11·09	11·21	11·33
26·0	10·37	10·49	10·61	10·73	10·85	10·97	11·09	11·21	11·33	11·45
26·5	10·50	10·62	10·74	10·86	10·98	11·10	11·22	11·34	11·46	11·58
27·0	10·62	10·74	10·86	10·98	11·10	11·22	11·34	11·46	11·58	11·70
27·5	10·75	10·87	10·99	11·11	11·23	11·35	11·47	11·59	11·71	11·83
28·0	10·87	10·99	11·11	11·23	11·35	11·47	11·59	11·71	11·83	11·95
28·5	10·99	11·11	11·23	11·35	11·47	11·59	11·71	11·83	11·95	12·07
29·0	11·12	11·24	11·36	11·48	11·60	11·72	11·84	11·96	12·08	12·20
29·5	11·24	11·36	11·48	11·60	11·72	11·84	11·96	12·08	12·20	12·32
30·0	11·37	11·49	11·61	11·73	11·85	11·97	12·09	12·21	12·33	12·45
30·5	11·49	11·61	11·73	11·85	11·97	12·09	12·21	12·33	12·45	12·57
31·0	11·62	11·74	11·86	11·98	12·10	12·22	12·34	12·46	12·58	12·70
31·5	11·74	11·86	11·98	12·10	12·22	12·34	12·46	12·58	12·70	12·82
32·0	11·86	11·98	12·10	12·22	12·34	12·46	12·58	12·70	12·82	12·94
32·5	11·98	12·10	12·22	12·34	12·46	12·58	12·70	12·82	12·94	13·06
33·0	12·11	12·23	12·35	12·47	12·59	12·71	12·83	12·95	13·07	13·19
33·5	12·23	12·35	12·47	12·59	12·71	12·83	12·95	13·07	13·19	13·31
34·0	12·35	12·47	12·59	12·71	12·83	12·95	13·07	13·19	13·31	13·43
34·5	12·48	12·60	12·72	12·84	12·96	13·08	13·20	13·32	13·44	13·56
35·0	12·60	12·72	12·84	12·96	13·08	13·20	13·32	13·44	13·56	13·68
35·5	12·72	12·84	12·96	13·08	13·20	13·32	13·44	13·56	13·68	13·80
36·0	12·84	12·96	13·08	13·20	13·32	13·44	13·56	13·68	13·80	13·92
36·5	12·96	13·08	13·20	13·32	13·44	13·56	13·68	13·80	13·92	14·04
37·0	13·09	13·21	13·33	13·45	13·57	13·69	13·81	13·93	14·05	14·17
37·5	13·21	13·33	13·45	13·57	13·69	13·81	13·93	14·05	14·17	14·29

TABLE LXIX.—FOR THE CALCULATION OF TOTAL SOLIDS FROM FAT AND SPECIFIC GRAVITY (RICHMOND)—*continued*.

Specific Gravity.	Fat per cent.									
	4.1	4.2	4.3	4.4	4.5	4.6	4.7	4.8	4.9	5.0
	Total Solids per cent.									
22.0	10.57	10.69	10.81	10.93	11.05	11.17	11.29	11.41	11.53	11.65
22.5	10.70	10.82	10.94	11.06	11.18	11.30	11.42	11.54	11.66	11.78
23.0	10.82	10.94	11.06	11.18	11.30	11.42	11.54	11.66	11.78	11.90
23.5	10.95	11.07	11.19	11.31	11.43	11.55	11.67	11.79	11.91	12.03
24.0	11.07	11.19	11.31	11.43	11.55	11.67	11.79	11.91	12.03	12.15
24.5	11.20	11.32	11.44	11.56	11.68	11.80	11.92	12.04	12.16	12.28
25.0	11.32	11.44	11.56	11.68	11.80	11.92	12.04	12.16	12.28	12.40
25.5	11.45	11.57	11.69	11.81	11.93	12.05	12.17	12.29	12.41	12.53
26.0	11.57	11.69	11.81	11.93	12.05	12.17	12.29	12.41	12.53	12.65
26.5	11.70	11.82	11.94	12.06	12.18	12.30	12.42	12.54	12.66	12.78
27.0	11.82	11.94	12.06	12.18	12.30	12.42	12.54	12.66	12.78	12.90
27.5	11.95	12.07	12.19	12.31	12.43	12.55	12.67	12.79	12.91	13.03
28.0	12.07	12.19	12.31	12.43	12.55	12.67	12.79	12.91	13.03	13.15
28.5	12.19	12.31	12.43	12.55	12.67	12.79	12.91	13.03	13.15	13.27
29.0	12.32	12.44	12.56	12.68	12.80	12.92	13.04	13.16	13.28	13.40
29.5	12.44	12.56	12.68	12.80	12.92	13.04	13.16	13.28	13.40	13.52
30.0	12.57	12.69	12.81	12.93	13.05	13.17	13.29	13.41	13.53	13.65
30.5	12.69	12.81	12.93	13.05	13.17	13.29	13.41	13.53	13.65	13.77
31.0	12.82	12.94	13.06	13.18	13.30	13.42	13.54	13.66	13.78	13.90
31.5	12.94	13.06	13.18	13.30	13.42	13.54	13.66	13.78	13.90	14.02
32.0	13.06	13.18	13.30	13.42	13.54	13.66	13.78	13.90	14.02	14.14
32.5	13.18	13.30	13.42	13.54	13.66	13.78	13.90	14.02	14.14	14.26
33.0	13.31	13.43	13.55	13.67	13.79	13.91	14.03	14.15	14.27	14.39
33.5	13.43	13.55	13.67	13.79	13.91	14.03	14.15	14.27	14.39	14.51
34.0	13.55	13.67	13.79	13.91	14.03	14.15	14.27	14.39	14.51	14.63
34.5	13.68	13.80	13.92	14.04	14.16	14.28	14.40	14.52	14.64	14.76
35.0	13.80	13.92	14.04	14.16	14.28	14.40	14.52	14.64	14.76	14.88
35.5	13.92	14.04	14.16	14.28	14.40	14.52	14.64	14.76	14.88	15.00
36.0	14.04	14.16	14.28	14.40	14.52	14.64	14.76	14.88	15.00	15.12
36.5	14.16	14.28	14.40	14.52	14.64	14.76	14.88	15.00	15.12	15.24
37.0	14.29	14.41	14.53	14.65	14.77	14.89	15.01	15.13	15.25	15.37
37.5	14.41	14.53	14.65	14.77	14.89	15.01	15.13	15.25	15.37	15.49

TABLE LXIX.—FOR THE CALCULATION OF TOTAL SOLIDS FROM FAT AND SPECIFIC GRAVITY (RICHMOND)—*continued.*

Specific Gravity.	Fat per cent.									
	5.1	5.2	5.3	5.4	5.5	5.6	5.7	5.8	5.9	6.0
	Total Solids per cent.									
22.0	11.77	11.89	12.01	12.13	12.25	12.37	12.49	12.61	12.73	12.85
22.5	11.90	12.02	12.14	12.26	12.38	12.50	12.62	12.74	12.86	12.98
23.0	12.02	12.14	12.26	12.38	12.50	12.62	12.74	12.86	12.98	13.10
23.5	12.15	12.27	12.39	12.51	12.63	12.75	12.87	12.99	13.11	13.23
24.0	12.27	12.39	12.51	12.63	12.75	12.87	12.99	13.11	13.23	13.35
24.5	12.40	12.52	12.64	12.76	12.88	13.00	13.12	13.24	13.36	13.48
25.0	12.52	12.64	12.76	12.88	13.00	13.12	13.24	13.36	13.48	13.60
25.5	12.65	12.77	12.89	13.01	13.13	13.25	13.37	13.49	13.61	13.73
26.0	12.77	12.89	13.01	13.13	13.25	13.37	13.49	13.61	13.73	13.85
26.5	12.90	13.02	13.14	13.26	13.38	13.50	13.62	13.74	13.86	13.98
27.0	13.02	13.14	13.26	13.38	13.50	13.62	13.74	13.86	13.98	14.10
27.5	13.15	13.27	13.39	13.51	13.63	13.75	13.87	13.99	14.11	14.23
28.0	13.27	13.39	13.51	13.63	13.75	13.87	13.99	14.11	14.23	14.35
28.5	13.39	13.51	13.63	13.75	13.87	13.99	14.11	14.23	14.35	14.47
29.0	13.52	13.64	13.76	13.88	14.00	14.12	14.24	14.36	14.48	14.60
29.5	13.64	13.76	13.88	14.00	14.12	14.24	14.36	14.48	14.60	14.72
30.0	13.77	13.89	14.01	14.13	14.25	14.37	14.49	14.61	14.73	14.85
30.5	13.89	14.01	14.13	14.25	14.37	14.49	14.61	14.73	14.85	14.97
31.0	14.02	14.14	14.26	14.38	14.50	14.62	14.74	14.86	14.98	15.10
31.5	14.14	14.26	14.38	14.50	14.62	14.74	14.86	14.98	15.10	15.22
32.0	14.26	14.38	14.50	14.62	14.74	14.86	14.98	15.10	15.22	15.34
32.5	14.38	14.50	14.62	14.74	14.86	14.98	15.10	15.22	15.34	15.46
33.0	14.51	14.63	14.75	14.87	14.99	15.11	15.23	15.35	15.47	15.59
33.5	14.63	14.75	14.87	14.99	15.11	15.23	15.35	15.47	15.59	15.71
34.0	14.75	14.87	14.99	15.11	15.23	15.35	15.47	15.59	15.71	15.83
34.5	14.88	15.00	15.12	15.24	15.36	15.48	15.60	15.72	15.84	15.96
35.0	15.00	15.12	15.24	15.36	15.48	15.60	15.72	15.84	15.96	16.08
35.5	15.12	15.24	15.36	15.48	15.60	15.72	15.84	15.96	16.08	16.20
36.0	15.24	15.36	15.48	15.60	15.72	15.84	15.96	16.08	16.20	16.32
36.5	15.36	15.48	15.60	15.72	15.84	15.96	16.08	16.20	16.32	16.44
37.0	15.49	15.61	15.73	15.85	15.97	16.09	16.21	16.33	16.45	16.57
37.5	15.61	15.73	15.85	15.97	16.09	16.21	16.33	16.45	16.57	16.69

TABLE LXIX.—FOR THE CALCULATION OF TOTAL SOLIDS FROM FAT AND SPECIFIC GRAVITY (RICHMOND)—*continued.*

Specific Gravity.	Fat per cent.					Specific Gravity.	Fat per cent.				
	6.1	6.2	6.3	6.4	6.5		6.1	6.2	6.3	6.4	6.5
	Total Solids per cent.						Total Solids per cent.				
22.0	12.97	13.09	13.21	13.33	13.45	30.0	14.97	15.09	15.21	15.33	15.45
22.5	13.10	13.22	13.34	13.46	13.58	30.5	15.09	15.21	15.33	15.45	15.57
23.0	13.22	13.34	13.46	13.58	13.70	31.0	15.22	15.34	15.46	15.58	15.70
23.5	13.35	13.47	13.59	13.71	13.83	31.5	15.34	15.46	15.58	15.70	15.82
24.0	13.47	13.59	13.71	13.83	13.95	32.0	15.46	15.58	15.70	15.82	15.94
24.5	13.60	13.72	13.84	13.96	14.08	32.5	15.58	15.70	15.82	15.94	16.06
25.0	13.72	13.84	13.96	14.08	14.20	33.0	15.71	15.83	15.95	16.07	16.19
25.5	13.85	13.97	14.09	14.21	14.33	33.5	15.83	15.95	16.07	16.19	16.31
26.0	13.97	14.09	14.21	14.33	14.45	34.0	15.95	16.07	16.19	16.31	16.43
26.5	14.10	14.22	14.34	14.46	14.58	34.5	16.08	16.20	16.32	16.44	16.56
27.0	14.22	14.34	14.46	14.58	14.70	35.0	16.20	16.32	16.44	16.56	16.68
27.5	14.35	14.47	14.59	14.71	14.83	35.5	16.32	16.44	16.56	16.68	16.80
28.0	14.47	14.59	14.71	14.83	14.95	36.0	16.44	16.56	16.68	16.80	16.92
28.5	14.59	14.71	14.83	14.95	15.07	36.5	16.56	16.68	16.80	16.92	17.04
29.0	14.72	14.84	14.96	15.08	15.20	37.0	16.69	16.81	16.93	17.05	17.17
29.5	14.84	14.96	15.08	15.20	15.32	37.5	16.81	16.93	17.05	17.17	17.29

APPENDIX II.

THE SALE OF FOOD AND DRUGS ACT, 1875.

AN ACT TO REPEAL THE ADULTERATION OF FOOD ACTS, AND TO MAKE BETTER PROVISION FOR THE SALE OF FOOD AND DRUGS IN A PURE STATE. [38 & 39 VICT., ch. 63.]

WHEREAS it is desirable that the Acts now in force relating to the adulteration of food should be repealed, and that the law regarding the sale of food and drugs in a pure and genuine condition should be amended :

Be it therefore enacted by the Queen's most Excellent Majesty, by and with the advice and consent of the Lords Spiritual and Temporal, and Commons, in this present Parliament assembled, and by the authority of the same, as follows :

Repeal of 23 & 24 Vict. c. 84, & 31 & 32 Vict. c. 121 s. 24, 33 & 34 Vict. c. 26 s. 3, & 35 & 36 Vict. c. 74.

1. From the commencement of this Act, the statutes of the twenty-third and twenty-fourth of Victoria, chapter eighty-four, of the thirty-first and thirty-second of Victoria, chapter one hundred and twenty-one, section twenty-four, of the thirty-third and thirty-fourth of Victoria, chapter twenty six, section three, and of the thirty-fifth and thirty-sixth of Victoria, chapter seventy-four, shall be repealed, except in regard to any appointment made under them and not then determined, and in regard to any offence committed against them or any prosecution or other act commenced and not concluded or completed, and any payment of money then due in respect of any provision thereof.

Definitions.

2. The term "food" shall include every article used for food or drink by man, other than drugs or water :

The term "drug" shall include medicine for internal or external use :

The term "county" shall include every county, riding, and division, as well as every county of a city or town not being a borough :

The term "justices" shall include any police or stipendiary magistrate invested with the powers of a justice of the peace in England, and any divisional justices in Ireland.

Description of Offences.

Mixing, colouring, staining so as to injure health.

3. No person shall mix, colour, stain, or powder, or order or permit any other person to mix, colour, stain, or powder, any article of food with any ingredient or material so as to render the article injurious to health, with intent that the same may be sold in that state, and no person shall sell any such article so mixed, coloured, stained, or powdered, under a penalty in each case not exceeding fifty pounds for the first offence ; every offence, after a conviction for a first offence, shall be a misdemeanour, for which the person, on conviction, shall be imprisoned for a period not exceeding six months with hard labour.

No drug to be coloured, etc., so as to injure its quality.

4. No person shall, except for the purpose of compounding as hereinafter described, mix, colour, stain, or powder, or order or permit any other person to mix, colour, stain, or powder, any drug with any ingredient or material so as to affect injuriously the quality or potency of such drug, with intent that the same may be sold in that state, and no person shall sell any such drug so mixed, coloured, stained, or powdered, under the same penalty in each case respectively as in the preceding section for a first and subsequent offence.

Guilty knowledge essential to the proof of the offence.

5. Provided that no person shall be liable to be convicted under either of the two last foregoing sections of this Act in respect of the sale of any article of food or of any drug, if he shows to the satisfaction of the justice or court before whom he is charged that he did not know of the article of food or drug sold by him being so mixed, coloured, stained, or powdered, as in either of those sections mentioned, and that he could not with reasonable diligence have obtained that knowledge.

No person to sell foods or

6. No person shall sell to the prejudice of the purchaser any article of food or any drug which is not of the nature, substance, and quality of the article demanded by such

purchaser, under a penalty not exceeding twenty pounds; provided that an offence shall not be deemed to be committed under this section in the following cases; that is to say,

drugs to the prejudice of purchaser.
Exceptions.

- (1.) Where any matter or ingredient not injurious to health has been added to the food or drug because the same is required for the production or preparation thereof as an article of commerce, in a state fit for carriage or consumption, and not fraudulently to increase the bulk, weight, or measure of the food or drug, or conceal the inferior quality thereof;
- (2.) Where the drug or food is a proprietary medicine, or is the subject of a patent in force, and is supplied in the state required by the specification of the patent;
- (3.) Where the food or drug is compounded as in this Act mentioned;
- (4.) Where the food or drug is unavoidably mixed with some extraneous matter in the process of collection or preparation.

7. No person shall sell any compound article of food or compounded drug which is not composed of ingredients in accordance with the demand of the purchaser, under a penalty not exceeding twenty pounds.

Compound foods or drugs must be in accordance with the demands of purchaser.
A legible descriptive notice exempts seller.

8. Provided that no person shall be guilty of any such offence as aforesaid in respect of the sale of an article of food or a drug mixed with any matter or ingredient not injurious to health, and not intended fraudulently to increase its bulk, weight, or measure, or conceal its inferior quality, if at the time of delivering such article or drug he shall supply to the person receiving the same a notice, by a label distinctly and legibly written or printed on or with the article or drug, to the effect that the same is mixed.

9. No person shall, with the intent that the same may be sold in its altered state without notice, abstract from an article of food any part of it so as to affect injuriously its quality, substance, or nature, and no person shall sell any article so altered without making disclosure of the alteration, under a penalty in each case not exceeding twenty pounds.

Abstraction of constituents of food.

Appointment and Duties of Analysts, and Proceedings to obtain the appointment of Analysts.

10. In the city of London and the liberties thereof the Commissioners of Sewers of the City of London and the liberties thereof, and in all other parts of the metropolis, the vestries and district boards acting in execution of the Act for the better local management of the metropolis, the court of quarter sessions of every county, and the town council of every borough having a separate court of quarter sessions, or having under any general or local Act of Parliament or otherwise a separate police establishment, may, as soon as convenient after the passing of this Act, where no appointment has been hitherto made, and in all cases as and when vacancies in the office occur, or when required so to do by the Local Government Board, shall for their respective city, districts, counties, or boroughs, appoint one or more persons possessing competent knowledge, skill, and experience, as analysts of all articles of food and drugs sold within the said city, metropolitan districts, counties, or boroughs, and shall pay to such analysts such remuneration as shall be mutually agreed upon, and may remove him or them as they shall deem proper; but such appointments and removals shall at all times be subject to the approval of the Local Government Board, who may require satisfactory proof of competency to be supplied to them, and may give their approval absolutely or with modifications as to the period of the appointment and removal, or otherwise; provided that no person shall hereafter be appointed an analyst for any place under this section who shall be engaged directly or indirectly in any trade or business connected with the sale of food or drugs in such place.

The local bodies by whom analysts may be, or are to be, appointed in England.

In Scotland the like powers shall be conferred and the like duties shall be imposed upon the commissioners of supply at their ordinary meetings for counties, and the commissioners or boards of police, or where there are no such commissioners or boards, upon the town councils for burghs within their several jurisdictions; provided that one of Her Majesty's Principal Secretaries of State in Scotland shall be substituted for the Local Government Board of England.

Appointments in Scotland.

In Ireland the like powers and duties shall be conferred and imposed respectively upon the grand jury of every county and town council of every borough; provided that the Local Government Board of Ireland shall be substituted for the Local Government Board of England.

In Ireland.

11. The town council of any borough may agree that the analyst appointed by any neighbouring borough or for the county in which the borough is situated, shall act for their borough during such time as the said council shall think proper, and shall make due provision for the payment of his remuneration, and if such analyst shall consent, he shall during such time be the analyst for such borough for the purposes of the Act.

Town Councils may agree with analyst of neighbouring county or borough.

Any purchaser of foods or drugs is entitled, on payment of a fee, to have an analysis and certificate.

Medical officers of health, inspectors, and certain other officials may procure samples for analysis.

The person purchasing must notify his intention of having the article analysed by the public analyst, and divide it into three parts.

Articles may be forwarded through the post.

Penalty for refusing to sell.

Certificate.

Analysts must report quarterly.

Proceedings after receipt of certificate.

Recovery of penalties.

12. Any purchaser of any article of food or of a drug in any place being a district, county, city, or borough where there is any analyst appointed under this or any Act hereby repealed shall be entitled, on payment to such analyst of a sum not exceeding ten shillings and sixpence, or if there be no such analyst then acting for such place, to the analyst of another place, of such sum as may be agreed upon between such person and the analyst, to have such article analysed by such analyst, and to receive from him a certificate of the result of his analysis.

13. Any medical officer of health, inspector of nuisances, or inspector of weights and measures, or any inspector of a market, or any police constable under the direction and at the cost of the local authority appointing such officer, inspector, or constable, or charged with the execution of this Act, may procure any sample of food or drugs, and if he suspect the same to have been sold to him contrary to any provision of this Act, shall submit the same to be analysed by the analyst of the district or place for which he acts, or if there be no such analyst then acting for such place to the analyst of another place, and such analyst shall, upon receiving payment as is provided in the last section, with all convenient speed analyse the same and give a certificate to such officer, wherein he shall specify the result of the analysis.

14. The person purchasing any article with the intention of submitting the same to analysis shall, after the purchase shall have been completed, forthwith notify to the seller or his agent selling the article his intention to have the same analysed by the public analyst, and shall [offer to] divide the article into three parts to be then and there separated, and each part to be marked and sealed or fastened up in such manner as its nature will permit, and shall, if required to do so [proceed accordingly, and shall], deliver one of the parts to the seller or his agent.¹

He shall afterwards retain one of the said parts for future comparison, and submit the third part, if he deems it right to have the article analysed, to the analyst.

[15. Repealed by Sale of Food and Drugs Act, 1899.]

16. If the analyst do not reside within two miles of the residence of the person requiring the article to be analysed, such article may be forwarded to the analyst through the post office as a registered "letter,"² subject to any regulations which the Postmaster-General may make in reference to the carrying and delivery of such an article, and the charge for the postage of such article shall be deemed one of the charges of this Act, or of the prosecution, as the case may be.

17. If any such officer, inspector, or constable, as above described, shall apply to purchase any article of food or any drug exposed to sale, or on sale by retail on any premises, or in any shop or stores, and shall tender the price for the quantity which he shall require for the purpose of analysis, not being more than shall be reasonably requisite, and the person exposing the same for sale shall refuse to sell the same to such officer, inspector, or constable, such person shall be liable to a penalty not exceeding ten pounds.

18. The certificate of the analysis shall be in the form set forth in the schedule hereto, or the like effect.

19. Every analyst appointed under any Act hereby repealed or this Act shall report quarterly to the authority appointing him the number of articles analysed by him under this Act during the foregoing quarter, and shall specify the result of each analysis and the sum paid to him in respect thereof, and such report shall be presented at the next meeting of the authority appointing such analyst, and every such authority shall annually transmit to the Local Government Board, at such time and in such form as the Board shall direct, a certified copy of such quarterly report.

Proceedings against Offenders.

20. When the analyst having analysed any article shall have given his certificate of the result, from which it may appear that an offence against some one of the provisions of this Act has been committed, the person causing the analysis to be made may take proceedings for the recovery of the penalty herein imposed for such offence, before any justices in petty sessions assembled having jurisdiction in the place where the article or drug sold was actually delivered to the purchaser, in a summary manner.

Every penalty imposed by this Act shall be recovered in England in the manner prescribed by the eleventh and twelfth of Victoria, chapter forty-three. In Ireland such penalties and proceedings shall be recoverable, and may be taken with respect to the police district of Dublin metropolis, subject and according to the provisions of any Act regulating the powers and duties of justices of the peace for such district, or of the police of such district; and with respect to other parts of Ireland, before a justice or justices of

¹ The words in brackets have been repealed by section 13 Sale of Food and Drugs Act, 1899, p. 594.

² For "letter," the word "parcel" is to be substituted by section 15 Sale of Food and Drugs Act, 1899.

the peace sitting in petty sessions, subject and according to the provisions of "The Petty Sessions (Ireland) Act, 1851," and any Act amending the same.

Every penalty herein imposed may be reduced or mitigated according to the judgment of the justices.

21. At the hearing of the information in such proceeding the production of the certificate of the analyst shall be sufficient evidence of the facts therein stated, unless the defendant shall require that the analyst shall be called as a witness, and the parts of the articles retained by the person who purchased the article shall be produced, and the defendant may, if he think fit, tender himself and his wife to be examined on his behalf, and he or she shall, if he so desire, be examined accordingly.

22. The justices before whom any complaint may be made, or the court before whom any appeal may be heard, under this Act may, upon the request of either party in their discretion, cause any article of food or drug to be sent to the Commissioners of Inland Revenue, who shall thereupon direct the chemical officers of their department at Somerset House to make the analysis, and give a certificate to such justices of the result of the analysis; and the expense of such analysis shall be paid by the complainant or the defendant as the justices may by order direct.

23. Any person who has been convicted of any offence punishable by any Act hereby repealed or by this Act by any justices may appeal in England to the next general or quarter sessions of the peace which shall be held for the city, county, town, or place wherein such conviction shall have been made, provided that such person enter into a recognisance within three days next after such conviction, with two sufficient sureties, conditioned to try such appeal, and to be forthcoming to abide the judgment and determination of the court at such general or quarter sessions, and to pay such costs as shall be by such court awarded; and the justices before whom such conviction shall be had are hereby empowered and required to take such recognisance; and the court at such general or quarter sessions are hereby required to hear and determine the matter of such appeal, and may award such costs to the party appealing or appealed against as they or he shall think proper.

In Ireland any person who has been convicted of any offence punishable by this Act may appeal to the next court of quarter sessions to be held in the same division of the county where the conviction shall be made by any justice or justices in any petty sessions district, or to the recorder at his next sessions where the conviction shall be made by the divisional justices in the police district of Dublin metropolis, or to the recorder of any corporate or borough town when the conviction shall be made by any justice or justices in such corporate or borough town (unless when any such sessions shall commence within ten days from the date of any such conviction, in which case, if the appellant sees fit, the appeal may be made to the next succeeding sessions to be held for such division or town), and it shall be lawful for such court of quarter sessions or recorder (as the case may be), to decide such appeal, if made in such form and manner and with such notices as are required by the said Petty Sessions Acts respectively hereinbefore mentioned as to appeals against orders made by justices at petty sessions, and all the provisions of the said Petty Sessions Acts respectively as to making appeals and as to executing the orders made on appeal, or the original orders where the appeals shall not be duly prosecuted, and shall also apply to any appeal made under this Act.

24. In any prosecution under this Act, where the fact of an article having been sold in a mixed state has been proved, if the defendant shall desire to rely upon any exception or provision contained in this Act, it shall be incumbent upon him to prove the same.

25. If the defendant in any prosecution under this Act prove to the satisfaction of the justices or court that he had purchased the article in question as the same in nature, substance, and quality as that demanded of him by the prosecutor, and with a written warranty to that effect, that he had no reason to believe at the time when he sold it that the article was otherwise, and that he sold it in the same state as when he purchased it, he shall be discharged from the prosecution, but shall be liable to pay the costs incurred by the prosecutor, unless he shall have given due notice to him that he will rely on the above defence.

26. Every penalty imposed and recovered under this Act shall be paid in the case of a prosecution by any officer, inspector, or constable, of the authority who shall have appointed an analyst or agreed to the acting of an analyst within their district, to such officer, inspector, or constable, and shall be by him paid to the authority for whom he acts, and be applied towards the expense of executing this Act, any Statute to the contrary notwithstanding; but in the case of any other prosecution the same shall be paid and applied in England according to the law regulating the application of penalties for offences punishable in a summary manner, and in Ireland in the manner directed by the Fines Act, Ireland, 1851, and the Acts amending the same.

Production of certificate is evidence, but analyst must attend if required. The defendant or his wife may, if they choose, be examined. Justices may send sample to Somerset House.

Appeal to quarter sessions.

Appeals in Ireland.

Incumbent on defendant to prove that he comes within any exception with regard to mixed matters.

Written warranty a good defence

Penalties in England to go to the authority and be applied to expenses of Act; in Ireland in manner directed by Fines Act.

Forged
certificates.

27. Any person who shall forge, or shall utter, knowing it to be forged for the purposes of this Act, any certificate or any writing purporting to contain a warranty, shall be guilty of a misdemeanor and be punishable on conviction by imprisonment for a term of not exceeding two years with hard labour ;

Every person who shall wilfully apply to an article of food, or a drug, in any proceedings under this Act, a certificate or warranty given in relation to any other article or drug, shall be guilty of an offence under this Act, and be liable to a penalty not exceeding twenty pounds ;

Every person who shall give a false warranty in writing to any purchaser in respect of an article of food or a drug sold by him as principal or agent, shall be guilty of an offence under this Act, and be liable to a penalty not exceeding twenty pounds ;

False labels.

And every person who shall wilfully give a label with any article sold by him which shall falsely describe the article sold, shall be guilty of an offence under this Act, and be liable to a penalty not exceeding twenty pounds.

Indictment.

28. Nothing in this Act contained shall affect the power of proceeding by indictment, or take away any other remedy against any offender under this Act, or in any way interfere with contracts and bargains between individuals, and the rights and remedies belonging thereto.

Breach of
contract.

Provided that in any action brought by any person for a breach of contract on the sale of any article of food or of any drug, such person may recover alone or in addition to any other damages recoverable by him the amount of any penalty in which he may have been convicted under this Act, together with the costs paid by him upon such conviction and those incurred by him in and about his defence thereto, if he prove that the article or drug the subject of such conviction was sold to him as and for an article or drug of the same nature, substance, and quality as that which was demanded of him, and that he purchased it not knowing it to be otherwise, and afterwards sold it in the same state in which he purchased it ; the defendant in such action being nevertheless at liberty to prove that the conviction was wrongful, or that the amount of costs awarded or claimed was unreasonable.

Expenses of Executing the Act.

Expenses in
England.

29. The expenses of executing this Act shall be borne, in the city of London and the liberties thereof, by the consolidated rates raised by the Commissioners of Sewers of the city of London and the liberties thereof, and in the rest of the metropolis by any rates or funds applicable to the purposes of the Act for the better local management of the metropolis, and otherwise as regards England, in counties by the county rate, and in boroughs by the borough fund or rate ;

In Ireland.

And as regards Ireland, in counties by the grand jury cess, and in boroughs by the borough fund or rate ; all such expenses payable in any county out of grand jury cess shall be paid by the treasurer of such county ; and

The grand jury of any such county shall, at any assizes at which it is proved that any such expenses have been incurred or paid without previous application to presentment sessions, present to be raised off and paid by such county the moneys required to defray the same.

Special Provision as to Tea.

Special pro-
vision as
to tea.

30. From and after the first day of January one thousand eight hundred and seventy-six all tea imported as merchandise into and landed at any port in Great Britain or Ireland shall be subject to examination by persons to be appointed by the Commissioners of Customs, subject to the approval of the Treasury, for the inspection and analysis thereof, for which purpose samples may, when deemed necessary by such inspectors, be taken, and with all convenient speed be examined by the analysts to be so appointed ; and if upon such analysis the same shall be found to be mixed with other substances or exhausted tea, the same shall not be delivered unless with the sanction of the said commissioners, and on such terms and conditions as they shall see fit to direct, either for home consumption or for use as ships' stores or for exportation ; but if on such inspection and analysis it shall appear that such tea is in the opinion of the analyst unfit for human food, the same shall be forfeited and destroyed or otherwise disposed of in such manner as the said commissioners may direct.

31. Tea to which the term "exhausted" is applied in this Act shall mean and include any tea which has been deprived of its proper quality, strength, or virtue by steeping, infusion, decoction, or other means.

Cinque ports.

32. For the purposes of this Act every liberty of a cinque port not comprised within the jurisdiction of a borough shall be part of the county in which it is situated, and subject to the jurisdiction of the justices of such county.

33. In the application of this Act to Scotland the following provisions shall have effect:
1. The term "misdemeanor" shall mean "a crime or offence."
 2. The term "defendant" shall mean "defender," and include "respondent."
 3. The term "information" shall include "complaint."
 4. This Act shall be read and construed as if for the term "justices," wherever it occurs therein, the term "sheriff" were substituted:
 5. The term "sheriff" shall include "sheriff-substitute."
 6. The term "borough" shall mean any royal burgh and any burgh returning or contributing to return a member to Parliament:
 7. The expenses of executing this Act shall be borne in Scotland, in counties, by the county general assessment, and in burghs, by the police assessment:
 8. This Act shall be read and construed as if for the expression "the Local Government Board," wherever it occurs therein, the expression "one of Her Majesty's Principal Secretaries of State" were substituted:
 9. All penalties provided by this Act to be recovered in a summary manner shall be recovered before the sheriff of the county in the sheriff court, or at the option of the person seeking to recover the same in the police court, in any place where a sheriff officiates as a police magistrate, under the provisions of "The Summary Procedure Act, 1864," or of the Police Act in force for the time in any place in which a sheriff officiates as aforesaid, and all the jurisdiction, powers, and authorities necessary for this purpose are hereby conferred on sheriffs:

Every such penalty may be recovered at the instance of the procurator-fiscal of the jurisdiction, or of the person who caused the analyses to be made from which it appeared that an offence had been committed against some one of the provisions of this Act.

Every penalty imposed and recovered under this Act shall be paid to the clerk of court, and by him shall be accounted for and paid to the treasurer of the county general assessment, or the police assessment of the burgh, as the sheriff shall direct.
 10. Every penalty imposed by this Act may be reduced or mitigated according to the judgment of the sheriff:
 11. It shall be competent to any person aggrieved by any conviction by a sheriff in any summary proceeding under this Act to appeal against the same to the next circuit court, or where there are no circuit courts to the High Court of Justiciary at Edinburgh, in the manner prescribed by such of the provisions of the Act of the twentieth year of the reign of King George the Second, chapter forty-three, and any Acts amending the same, as relate to appeals in matters criminal, and by and under the rules, limitations, conditions and restrictions contained in the said provisions.
 34. In the application of this Act to Ireland,—

The term "borough" shall mean any borough subject to the Act of the session of the third and fourth years of the reign of Her present Majesty, chapter one hundred and eight, intituled "An Act for the Regulation of Municipal Corporations in Ireland."

The term "county" shall include a county of a city and a county of a town not being a borough.

The term "assizes" shall, with respect to the county of Dublin, mean "presenting term."

The term "treasurer of the county" shall include any person or persons or bank in any county performing duties analogous to those of the treasurer of the county in counties, and, with respect to the county of Dublin, it shall mean the finance committee:

The term "police constable" shall mean, with respect to the police district of Dublin metropolis, constable of the Dublin Metropolitan Police, and with respect to any other part of Ireland, constable of the Royal Irish Constabulary:
 35. This Act shall commence on the first day of October one thousand eight hundred and seventy-five.
 36. This Act may be cited as "The Sale of Food and Drugs Act, 1875."

Special applications to Scotland.

Definitions of borough.

Expenses.

Local Government Board.

Penalties

Special applications to Ireland. Definitions. Borough.

County.

Assizes.

Treasurer.

Police constable.

Commencement of Act. Title.

Schedule.

SCHEDULE.

FORM OF CERTIFICATE.

To¹

Certificate.

I, the undersigned, public analyst for the _____, do hereby certify that I received on the _____ day of _____, 18____, from² _____ a sample of _____, for analysis (which then weighed³ _____), and have analysed the same, and declare the result of my analysis to be as follows:—

I am of opinion that the same is a sample of genuine _____

or,

I am of opinion that the said sample contained the parts as under, or the percentages of foreign ingredients as under.

Observations.⁴

As witness my hand this _____

day of _____
A. B.
at _____

SALE OF FOOD AND DRUGS ACT AMENDMENT ACT, 1879 [42 and 43 Vict. c. 30].

38 & 39 Vict.
63.

Whereas conflicting decisions have been given in England and in Scotland in regard to the meaning and effect of section six of the Sale of Food and Drugs Act, 1875, in this Act referred to as the principal Act, and it is expedient, in this respect and otherwise, to amend the said Act: Be it enacted by the Queen's most Excellent Majesty, by and with the advice and consent of the Lords Spiritual and Temporal, and Commons, in this present Parliament assembled, and by the authority of the same, as follows:

Short title.

1. This Act may be cited for all purposes as the Sale of Food and Drugs Act Amendment Act, 1879.

In sale of adulterated articles, no defence to allege purchase for analysis.

2. In any prosecution under the provisions of the principal Act for selling to the prejudice of the purchaser any article of food or any drug which is not of the nature, substance, and quality of the article demanded by such purchaser, it shall be no defence to any such prosecution to allege that the purchaser, having bought only for analysis, was not prejudiced by such sale. Neither shall it be a good defence to prove that the article of food or drug in question, though defective in nature or in substance or in quality, was not defective in all three respects.

Officer, inspector, or constable may obtain a sample of milk at the place of delivery to submit to analyst.

3. Any medical officer of health, inspector of nuisances, or inspector of weights and measures, or any inspector of a market, or any police constable under the direction and at the cost of the local authority appointing such officer, inspector, or constable, or charged with the execution of this Act, may procure at the place of delivery any sample of any milk in course of delivery to the purchaser or consignee in pursuance of any contract for the sale to such purchaser or consignee of such milk; and such officer, inspector, or constable, if he suspect the same to have been sold contrary to any of the provisions of the principal Act, shall submit the same to be analysed, and the same shall be analysed, and proceedings shall be taken, and penalties on conviction be enforced in like manner in all respects as if such officer, inspector, or constable had

¹ Here insert the name of the person submitting the article for analysis.

² Here insert the name of the person delivering the sample.

³ When the article cannot be conveniently weighed, this passage may be erased, or the blank may be left unfilled.

⁴ Here the analyst may insert at his discretion his opinion as to whether the mixture (if any) was for the purpose of rendering the article portable or palatable, or of preserving it, or of improving the appearance, or was unavoidable, and may state whether in excess of what is ordinary, or otherwise, and whether the ingredients or materials mixed are or are not injurious to health.

In the case of a certificate regarding milk, butter, or any article liable to decomposition, the analyst shall specially report whether any change had taken place in the constitution of the article that would interfere with the analysis.

purchased the same from the seller or consignor under section thirteen of the principal Act.¹

4. The seller or consignor or any person or persons entrusted by him for the time being with the charge of such milk, if he shall refuse to allow such officer, inspector, or constable to take the quantity which such officer, inspector or constable shall require for the purpose of analysis, shall be liable to a penalty not exceeding ten pounds.

Penalty for refusal to give milk for analysis.

5. Any street or open place of public resort shall be held to come within the meaning of section seventeen of the principal Act.

Extension of Act as to sale in streets, etc.

6. In determining whether an offence has been committed under section six of the said Act by selling, to the prejudice of the purchaser, spirits not adulterated otherwise than by the admixture of water, it shall be a good defence to prove that such admixture has not reduced the spirit more than twenty-five degrees under proof for brandy, whisky, or rum, or thirty-five degrees under proof for gin.

Reduction allowed to the extent of 25 degrees under proof for brandy, whisky, or rum, and 35 degrees for gin.

7. Every liberty having a separate court of quarter sessions, except a liberty of a cinque port, shall be deemed to be a county within the meaning of the said Act.

8. The town council of any borough having a separate court of quarter sessions shall be exempt from contributing towards the expenses incurred in the execution of the principal Act in respect of the county within which such borough is situate, and the treasurer of the county shall exclude the expenses so incurred from the account required by section one hundred and seventeen of the Municipal Corporation Act, 1835, to be sent by him to such town council.

Extension of meaning of "county."

9. The town council of any borough having under any general or local Act of Parliament, or otherwise, a separate police establishment, and being liable to be assessed to the county rate of the county within which the borough is situate, shall be paid by the justices of such county the proportionate amount contributed towards the expenses incurred by the county in the execution of the principal Act by the several parishes and parts of parishes within such borough in respect of the rateable value of the property assessable therein, as ascertained by the valuation lists for the time being in force.

Quarter session boroughs not to contribute to county analyst.

5 & 6 W. IV. c. 76.

Provision for boroughs with separate police.

[10. Repealed by Sale of Food and Drugs Act, 1899.]

MARGARINE ACT, 1887.

AN ACT FOR THE BETTER PREVENTION OF THE FRAUDULENT SALE OF MARGARINE.
[50 & 51 VICT., ch. 29.]

WHEREAS it is expedient that further provision should be made for protecting the public against the sale as butter of substances made in imitation of butter, as well as of butter mixed with any such substances:

Be it therefore enacted by the Queen's most Excellent Majesty, by and with the advice and consent of the Lords Spiritual and Temporal, and Commons, in this present Parliament assembled, and by the authority of the same, as follows:

1. This Act may be cited as the Margarine Act, 1887.

2. This Act shall come into operation on the first day of January one thousand eight hundred and eighty-eight.

Short title. Commencement of Act.

3. The word "butter" shall mean the substance usually known as butter, made exclusively from milk or cream, or both, with or without salt or other preservative, and with or without the addition of colouring matter.

Definition.

The word "margarine" shall mean all substances, whether compounds or otherwise, prepared in imitation of butter, and whether mixed with butter or not, and no such substance shall be lawfully sold, except under the name of margarine, and under the conditions set forth in this Act.

4. Every person dealing in margarine, whether wholesale or retail, whether a manufacturer, importer, or as consignor or consignee, or as commission agent or otherwise, who is found guilty of an offence under this Act, shall be liable on summary conviction for the first offence to a fine not exceeding twenty pounds, and for the second offence to a fine not exceeding fifty pounds, and for the third or any subsequent offence to a fine not exceeding one hundred pounds.

Penalty.

5. Where an employer is charged with an offence against this Act he shall be entitled, upon information duly laid by him, to have any other person whom he charges as the actual offender brought before the Court at the time appointed for hearing the charge, and if, after the commission of the offence has been proved, the employer proves to the

Exemption from penalty.

¹ The leading case of *M'Nair v. Cave*, K.E.D., heard before the Lord Chief Justice and Wills and Campbell, J.J. (*Times*, Oct. 30, 1902), lays down clearly the law that an inspector can only take samples under section three in his own district and must submit such samples to the analyst appointed for such district. Milk, for example, consigned to St Pancras Station for distribution by retail in Westminster can only be sampled and analysed by the St Pancras inspector and the St Pancras analyst.

satisfaction of the Court that he had used due diligence to enforce the execution of this Act, and that the said other person had committed the offence in question without his knowledge, consent, or connivance, the said other person shall be summarily convicted of such offence, and the employer shall be exempt from any penalty.

Marking of cases.

6. Every person dealing in margarine in the manner described in the preceding section shall conform to the following regulations :

Every package, whether open or closed, and containing margarine, shall be branded or durably marked "Margarine" on the top, bottom, and sides, in printed capital letters, not less than three-quarters of an inch square ; and if such margarine be exposed for sale, by retail, there shall be attached to each parcel thereof so exposed, and in such manner as to be clearly visible to the purchaser, a label marked in printed capital letters not less than one and a half inches square, "Margarine" ; and every person selling margarine by retail, save in a package duly branded or durably marked as aforesaid, shall in every case deliver the same to the purchaser in [or with] a paper wrapper, on which shall be printed in capital letters, not less than a quarter of an inch square, "Margarine."¹

Presumption against vendor.

7. Every person dealing with, selling, or exposing, or offering for sale, or having in his possession for the purpose of sale, any quantity of margarine contrary to the provisions of this Act, shall be liable to conviction for an offence against this Act, unless he shows to the satisfaction of the court before whom he is charged that he purchased the article in question as butter, and with a written warranty or invoice to that effect, that he had no reason to believe at the time when he sold it that the article was other than butter, and that he sold it in the same state as when he purchased it, and in such case he shall be discharged from the prosecution, but shall be liable to pay the costs incurred by the prosecutor unless he shall have given due notice to him that he will rely upon the above defence.

Margarine imported or manufactured.

8. All margarine imported into the United Kingdom of Great Britain and Ireland, and all margarine, whether imported or manufactured within the United Kingdom of Great Britain and Ireland, shall, whenever forwarded by any public conveyance, be duly consigned as margarine ; and it shall be lawful for any officer of Her Majesty's Customs or Inland Revenue, or any medical officer of health, inspector of nuisances, or police constable, authorised under section thirteen of the Sale of Food and Drugs Act, 1875, to procure samples for analysis if he shall have reason to believe that the provisions of this Act are infringed on this behalf, to examine and take samples from any package, and ascertain, if necessary, by submitting the same to be analysed, whether an offence against this Act has been committed.

38 & 39 Vict. c. 63.

Registration of manufactory.

9. Every manufactory of margarine within the United Kingdom of Great Britain and Ireland shall be registered by the owner or occupier thereof with the local authority from time to time in such manner as the Local Government Boards of England and Ireland and the Secretary for Scotland respectively may direct, and every such owner or occupier carrying on such manufacture in a manufactory not duly registered shall be guilty of an offence under this Act.

Power to inspectors to take samples without purchase.

10. Any officer authorised to take samples under the Sale of Food and Drugs Acts, 1875, may, without going through the form of purchase provided by that Act, but otherwise acting in all respects in accordance with the provisions of the said Act as to dealing with samples, take for the purposes of analysis samples of any butter, or substances purporting to be butter, which are exposed for sale, and are not marked Margarine, as provided by this Act ; and any such substance not being so marked shall be presumed to be exposed for sale as butter.

Appropriation of penalties.

11. Any part of any penalty recovered under this Act may, if the court shall so direct, be paid to the person who proceeds for the same, to reimburse him for the legal costs of obtaining the analysis, and any other reasonable expenses to which the Court shall consider him entitled.

Proceedings.

12. All proceedings under this Act shall, save as expressly varied by this Act, be the same as prescribed by sections twelve to twenty-eight inclusive of the Sale of Food and Drugs Act, 1875, and all officers employed under that Act are hereby empowered and required to carry out the provisions of this Act.

Definition of local authority.

13. The expression "local authority" shall mean any local authority authorised to appoint a public analyst under the Sale of Food and Drugs Act, 1875.

¹ The words in brackets are repealed by section 6 (3) Sale of Food and Drugs Act, 1899, p. 593.

SALE OF HORSEFLESH, &c. REGULATION ACT, 1889.

AN ACT TO REGULATE THE SALE OF HORSEFLESH FOR HUMAN FOOD.
[52 & 53 VICT., ch. 11.]

WHEREAS it is desirable to make regulations with respect to the sale of horseflesh for human food :

Be it therefore enacted by the Queen's most Excellent Majesty, by and with the advice and consent of the Lords Spiritual and Temporal, and Commons, in this present Parliament assembled, and by the authority of the same, as follows :

1. No person shall sell, offer, expose, or keep for sale any horseflesh for human food, elsewhere than in a shop, stall, or place over or upon which there shall be at all times painted, posted, or placed in legible characters of not less than four inches in length, and in a conspicuous position, and so as to be visible throughout the whole time, whether by night or day, during which such horseflesh is being offered or exposed for sale, words indicating that horseflesh is sold there. Signs on horseflesh shops.

2. No person shall supply horseflesh for human food to any purchaser who has asked to be supplied with some meat other than horseflesh, or with some compound article of food which is not ordinarily made of horseflesh. Horseflesh not to be sold as other meat.

3. Any medical officer of health or inspector of nuisances or other officer of a local authority acting on the instructions of such authority or appointed by such authority for the purposes of this Act may at all reasonable times inspect and examine any meat which he has reason to believe to be horseflesh, exposed for sale or deposited for the purpose of sale, or of preparation for sale, and intended for human food, in any place other than such shop, stall, or place as aforesaid, and if such meat appears to him to be horseflesh he may seize and carry away or cause to be seized and carried away the same, in order to have the same dealt with by a justice as hereinafter provided. Power of medical officer of health to inspect meat, etc.

4. On complaint made on oath by a medical officer of health or inspector of nuisances, or other officer of a local authority, any justice may grant a warrant to any such officer to enter any building, or part of a building other than such shop, stall or place as aforesaid, in which such officer has reason for believing that there is kept or concealed any horseflesh which is intended for sale, or for preparation for sale for human food, contrary to the provisions of this Act ; and to search for, seize, and carry away or cause to be seized and carried away any meat that appears to such officer to be such horseflesh, in order to have the same dealt with by a justice as hereinafter provided. Power of justice to grant warrant for search.

Any person who shall obstruct any such officer in the performance of his duty under this Act shall be deemed to have committed an offence under this Act.

5. If it appears to any justice that any meat seized under the foregoing provisions of this Act is such horseflesh as aforesaid, he may make such order with regard to the disposal thereof as he may think desirable ; and the person in whose possession or on whose premises the meat was found shall be deemed to have committed an offence under this Act, unless he proves that such meat was not intended for human food contrary to the provisions of this Act. Power of justice with reference to disposal of horseflesh.

6. Any person offending against any of the provisions of this Act, for every such offence shall be liable to a penalty not exceeding twenty pounds, to be recovered in a summary manner ; and if any horseflesh is proved to have been exposed for sale to the public in any shop, stall, or eating-house other than such shop, stall, or place as in the first section mentioned, without anything to show that it was not intended for sale for human food, the onus of proving that it was not so intended shall rest upon the person exposing it for sale. Penalty.

7. For the purposes of this Act "horseflesh" shall include the flesh of asses and mules, and shall mean horseflesh, cooked or uncooked, alone or accompanied by or mixed with any other substance. Definition of "horseflesh."

8. For the purposes of this Act the local authorities shall be, in the City of London and the liberties thereof, the Commissioners of Sewers, and in the other parts of the county of London the vestries and district boards acting in the execution of the Metropolis Local Management Acts, and in other parts of England the urban and rural sanitary authorities, and in Ireland the urban and rural sanitary authorities under the Public Health (Ireland) Act, 1878. Local authorities for purposes of Act.

9. In the application of this Act to Scotland the expression "justice" shall include sheriff and sheriff-substitute, and the expressions "local authority" shall mean any local authority authorised to appoint a public analyst under the Sale of Food and Drugs Act, 1879, and the procedure for the enforcement of this Act shall be in the manner provided in the thirty-third section of the said Sale of Food and Drugs Act, 1875. 41 & 42 Vict. c. 52. Application to Scotland, 38 & 39 Vict. c. 63.

10. This Act may be cited as the Sale of Horseflesh, &c. Regulation Act, 1889.

11. This Act shall come into operation on the twenty-ninth day of September one thousand eight hundred and eighty-nine. Short title. Commencement of Act.

SALE OF FOOD AND DRUGS ACT, 1899.

AN ACT TO AMEND THE LAW RELATING TO THE SALE OF FOOD AND DRUGS.
[62 & 63 VICT., ch. 51.]

BE it enacted by the Queen's most Excellent Majesty, by and with the advice and consent of the Lords Spiritual and Temporal, and Commons, in this present Parliament assembled, and by the authority of the same, as follows:—

Precautions against importation of agricultural and other produce insufficiently marked.

1.—(1.) If there is imported into the United Kingdom any of the following articles, namely:—

- (a) margarine or margarine-cheese, except in packages conspicuously marked "Margarine" or "Margarine-cheese," as the case may require; or
- (b) adulterated or impoverished butter (other than margarine) or adulterated or impoverished milk or cream, except in packages or cans conspicuously marked with a name or description indicating that the butter or milk or cream has been so treated; or
- (c) condensed separated or skimmed milk, except in tins or other receptacles which bear a label whereon the words "Machine-skimmed Milk" or "Skimmed Milk," as the case may require, are printed in large and legible type; or
- (d) any adulterated or impoverished article of food to which Her Majesty may by Order in Council direct that this section shall be applied, unless the same be imported in packages or receptacles conspicuously marked with a name or description indicating that the article has been so treated;

the importer shall be liable, on summary conviction, for the first offence to a fine not exceeding twenty pounds, for the second offence to a fine not exceeding fifty pounds, and for any subsequent offence to a fine not exceeding one hundred pounds.

(2.) The word "importer" shall include any person who, whether as owner, consignee, or consignee, agent, or broker, is in possession of, or in anywise entitled to the custody or control of, the article; prosecutions for offences under this section shall be undertaken by the Commissioners of Customs; and subject to the provisions of this Act this section shall have effect as if it were part of the Customs Consolidation Act, 1876.

(3.) The Commissioners of Customs shall, in accordance with directions given by the Treasury after consultation with the Board of Agriculture, take such samples of consignments of imported articles of food as may be necessary for the enforcement of the foregoing provisions of this section.

(4.) Where the Commissioners of Customs take a sample of any consignment in pursuance of such directions they shall divide it into not less than three parts, and send one part to the importer and one part to the principal chemist of the Government laboratories, and retain one part.

(5.) In any proceeding under this section the certificate of the principal chemist of the result of the analysis shall be sufficient evidence of the facts therein stated, unless the defendant require that the person who made the analysis be called as a witness.

(6.) If, in any case, the Commissioners of Customs are of opinion that an offence against this section has been committed, they shall communicate to the Board of Agriculture for their information the name of the importer and such other facts as they possess or may obtain as to the destination of the consignment.

(7.) For the purposes of this section an article of food shall be deemed to be adulterated or impoverished if it has been mixed with any other substance, or if any part of it has been abstracted so as in either case to affect injuriously its quality, substance, or nature.

Provided that an article of food shall not be deemed to be adulterated by reason only of the addition of any preservative or colouring matter of such a nature and in such quantity as not to render the article injurious to health.

Power for Local Government Board or Board of Agriculture to sample articles of food.

2.—(1.) The Local Government Board may, in relation to any matter appearing to that Board to affect the general interest of the consumer, and the Board of Agriculture may, in relation to any matter appearing to that Board to affect the general interests of agriculture in the United Kingdom, direct an officer of the Board to procure for analysis samples of any article of food, and thereupon the officer shall have all the powers of procuring samples conferred by the Sale of Food and Drugs Acts, and those Acts shall apply as if the officer were an officer authorised to procure samples under the Sale of Food and Drugs Act, 1875, except that—

- (a) the officer procuring the sample shall divide the same into four parts, and shall deal with three of such parts in the manner directed by section fourteen of the Sale of Food and Drugs Act, 1875, as amended by this Act, and shall send the fourth part to the Board, and

(b) the fee for analysis shall be payable to the analyst by the local authority of the place where the sample is procured.

(2.) The Board shall communicate the result of the analysis of any such sample to the local authority, and thereupon there shall be the like duty and power on the part of the local authority to cause proceedings to be taken as if the local authority had caused the analysis to be made.

3.—(1.) It shall be the duty of every local authority entrusted with the execution of the laws relating to the sale of food and drugs to appoint a public analyst, and put in force from time to time, as occasion may arise, the powers with which they are invested, so as to provide proper securities for the sale of food and drugs in a pure and genuine condition, and in particular to direct their officers to take samples for analysis.

Power for Local Government Board or Board of Agriculture to act in default of local authority.

(2.) If the Local Government Board or Board of Agriculture, after communication with a local authority, are of opinion that the local authority have failed to execute or enforce any of the provisions of the Sale of Food and Drugs Acts in relation to any article of food, and that their failure affects the general interest of the consumer or the general interests of agriculture in the United Kingdom, as the case may be, the Board concerned, may, by order, empower an officer of the Board to execute and enforce those provisions or to procure the execution and enforcement thereof in relation to any article of food mentioned in the order.

(3.) The expenses incurred by the Board or their officer under any such order shall be treated as expenses incurred by the local authority in the execution of the said Acts, and shall be paid by the local authority to the Board on demand, and in default the Board may recover the amount of the expenses with costs from the local authority.

(4.) For the purposes of this section an order of the Board shall be conclusive in respect of any default, amount of expenses, or other matter therein stated or appearing.

(5.) Any public analyst appointed under the Sale of Food and Drugs Acts shall furnish such proof of competency as may from time to time be required by regulation framed by the Local Government Board.

4.—(1.) The Board of Agriculture may, after such inquiry as they deem necessary, make regulations¹ for determining what deficiency in any of the normal constituents of genuine milk, cream, butter, or cheese, or what addition of extraneous matter or proportion of water, in any sample of milk (including condensed milk), cream, butter, or cheese, shall for the purposes of the Sale of Food and Drugs Acts raise a presumption, until the contrary is proved, that the milk, cream, butter, or cheese is not genuine or is injurious to health, and an analyst shall have regard to such regulations in certifying the result of an analysis under those Acts.

Power for Board of Agriculture to make regulations as to analysis of milk, cream, butter, or cheese.

(2.) Any regulations made under this section shall be notified in the London and Edinburgh Gazettes, and shall also be made known in such other manner as the Board of Agriculture may direct.

5. The provisions of the Margarine Act, 1887, as amended by this Act, shall extend to margarine-cheese, and shall apply accordingly, with the substitution of "margarine-cheese" and "cheese" for "margarine" and "butter," and provided that all margarine-cheese sold or dealt in otherwise than by retail shall either be inclosed in packages marked in accordance with the Margarine Act, 1887, as amended by this Act, or be itself conspicuously branded with the words "margarine-cheese."

Extension of Margarine Act, 1887, to margarine-cheese. 50 & 51 Vict. c. 29.

6.—(1.) Where under this Act or the Margarine Act, 1887, it is required that any package containing margarine or margarine-cheese shall be branded or marked, the brand or mark shall be on the package itself and not solely on a label, ticket, or other thing attached thereto.

Marking of margarine and margarine-cheese.

(2.) The letters required to be printed on the paper wrapper in which margarine or margarine-cheese is sold shall be capital block letters not less than half an inch long and distinctly legible, and no other printed matter shall appear on the wrapper.

(3.) The words "or with" in section six of the Margarine Act, 1887, shall be repealed.

7.—(1.) Every occupier of a manufactory of margarine or margarine-cheese, and every wholesale dealer in such substances, shall keep a register showing the quantity and destination of each consignment of such substances sent out from his manufactory or place of business, and this register shall be open to the inspection of any officer of the Board of Agriculture.

Provisions as to manufacturers of and dealers in margarine and margarine-cheese.

(2.) Any officer of the Board of Agriculture shall have power to enter at all reasonable times any manufactory of margarine or margarine-cheese, and to inspect any process of manufacture therein, and to take samples for analysis.

(3.) If any such occupier or dealer—

(a) fails to keep such a register, or

(b) refuses to produce the register when required to do so by an officer of the Board of Agriculture, or

¹ The regulations made under this section will be found at pp. 45, 46.

- (c) fails to keep the register posted up to date, or
 (d) wilfully makes any entry in the register which is false in any particular, or
 (e) fraudulently omits to enter any particular which ought to be entered in the register,

he shall be liable on summary conviction for the first offence to a fine not exceeding ten pounds, and for any subsequent offence to a fine not exceeding fifty pounds.

50 & 51 Vict.
c. 29.

(4.) The provisions of section nine of the Margarine Act, 1887, relating to registration of manufactories shall extend to any premises wherein the business of a wholesale dealer in margarine or margarine-cheese is carried on.

(5.) The registration of a manufactory or other premises shall be forthwith notified by the local authority to the Board of Agriculture.

Restriction on amount of butter fat in margarine.

8. It shall be unlawful to manufacture, sell, expose for sale, or import any margarine, the fat of which contains more than ten per cent. of butter fat, and every person who manufactures, sells, exposes for sale, or imports any margarine which contains more than that percentage, shall be guilty of an offence under the Margarine Act, 1887, and any defence which would be a defence under section seven of that Act shall be a defence under this section, and the provisions of the former section shall apply accordingly.

Provided that nothing in this section shall apply to any margarine manufactured or imported in fulfilment of any contract made before the twentieth day of July one thousand eight hundred and ninety-nine.

Provision as to name and address of person selling milk or cream in a public place.

9. Every person who, himself or by his servant, in any highway or place of public resort, sells milk or cream from a vehicle or from a can or other receptacle shall have conspicuously inscribed on the vehicle or receptacle his name and address, and in default shall be liable on summary conviction to a fine not exceeding two pounds.

Division of samples taken in course of delivery or transit.

10. In the case of a sample taken of milk in course of delivery, or of margarine or margarine-cheese forwarded by a public conveyance, the person taking the sample shall forward by registered parcel or otherwise a portion of the sample marked, and sealed, and fastened up, to the consignor if his name and address appear on the can or package containing the article sampled.

Provisions as to condensed separated or skimmed milk.

11. Every tin or other receptacle containing condensed separated or skimmed milk must bear a label clearly visible to the purchaser on which the words "Machine-skimmed Milk," or "Skimmed Milk," as the case may require, are printed in large and legible type, and if any person sells or exposes or offers for sale condensed separated or skimmed milk in contravention of this section he shall be liable on summary conviction to a fine not exceeding ten pounds.

Notice of mixtures.
38 & 39 Vict.
c. 63.

12. The label referred to in section eight of the Sale of Food and Drugs Act, 1875, shall not be deemed to be distinctly and legibly written or printed within the meaning of that section unless it is so written or printed that the notice of mixture given by the label is not obscured by other matter on the label: Provided that nothing in this enactment shall hinder or affect the use of any registered trade mark, or of any label which has been continuously in use for at least seven years before the commencement of this Act; but the Comptroller-General of Patents, Designs, and Trade Marks shall not register any trade mark purporting to describe a mixture unless it complies with the requirements of this enactment.

Amendment of 38 & 39 Vict. c. 63 as to samples.

13. In section fourteen of the Sale of Food and Drugs Act, 1875, the words "offer to" and the words "proceed accordingly and shall" shall be repealed.

Taking samples in course of delivery.
42 & 43 Vict.
c. 30.

14. The provisions of section three and section four of the Sale of Food and Drugs Act Amendment Act, 1879 (relating to the taking of samples of milk in course of delivery), shall apply to every other article of food: Provided that no samples shall be taken under this section except upon the request or with the consent of the purchaser or consignee.

Amendment of 38 & 39 Vict. c. 63 as to registered parcels.

15. In section sixteen of the Sale of Food and Drugs Act, 1875, the words "registered parcel" shall be substituted for the words "registered letter."

Obstruction of officer in discharge of his duties.

16. Any person who wilfully obstructs or impedes any inspector or other officer in the course of his duties under the Sale of Food and Drugs Acts, or by any gratuity, bribe, promise, or other inducement prevents, or attempts to prevent, the due execution by such inspector or officer of his duty under those Acts, shall be liable, on summary conviction, for the first offence to a fine not exceeding twenty pounds, for the second offence to a fine not exceeding fifty pounds, and for any subsequent offence to a fine not exceeding one hundred pounds.

Penalties for offences under the Sale of Food and Drugs Acts.

17.—(1.) Where, under any provision of the Sale of Food and Drugs Act, 1875, a person guilty of an offence is liable to a fine which may extend to twenty pounds as a maximum, he shall be liable for a second offence under the same provision to a fine not exceeding fifty pounds, and for any subsequent offence to a fine not exceeding one hundred pounds.

(2.) Where under any provision of the Sale of Food and Drugs Acts, a person guilty of

an offence is liable to a fine exceeding fifty pounds, and the offence, in the opinion of the court, was committed by the personal act, default, or culpable negligence of the person accused, that person shall be liable (if the court is of opinion that a fine will not meet the circumstances of the case) to imprisonment, with or without hard labour, for a period not exceeding three months.

18. Notwithstanding anything in section seventeen of the Sale of Food and Drugs Act, 1875, where any article of food or drug is exposed for sale in an unopened tin or packet duly labelled, no person shall be required to sell it except in the unopened tin or packet in which it is contained. Articles sold in tins or packets. 38 & 39 Vict. c. 63.

19.—(1.) When any article of food or drug has been purchased from any person for test purposes, any prosecution under the Sale of Food and Drugs Acts in respect of the sale thereof, notwithstanding anything contained in section twenty of the Sale of Food and Drugs Act, 1875, shall not be instituted after the expiration of twenty-eight days from the time of the purchase. Time for proceeding and regulation as to summons.

(2.) In any prosecution under the Sale of Food and Drugs Acts the summons shall state particulars of the offence or offences alleged, and also the name of the prosecutor, and shall not be made returnable in less time than fourteen days from the day on which it is served, and there must be served therewith a copy of any analyst's certificate obtained on behalf of the prosecutor.

20.—(1.) A warranty or invoice shall not be available as a defence to any proceeding under the Sale of Food and Drugs Acts unless the defendant has, within seven days after service of the summons, sent to the purchaser a copy of such warranty or invoice with a written notice stating that he intends to rely on the warranty or invoice, and specifying the name and address of the person from whom he received it, and has also sent a like notice of his intention to such person. Provisions as to use of warranty or invoice as defence, and proceedings against the warrantor.

(2.) The person by whom such warranty or invoice is alleged to have been given shall be entitled to appear at the hearing and to give evidence, and the court may, if it thinks fit, adjourn the hearing to enable him to do so.

(3.) A warranty or invoice given by a person resident outside the United Kingdom shall not be available as a defence to any proceeding under the Sale of Food and Drugs Acts, unless the defendant proves that he had taken reasonable steps to ascertain and did in fact believe in the accuracy of the statement contained in the warranty or invoice.

(4.) Where the defendant is a servant of the person who purchased the article under a warranty or invoice he shall, subject to the provisions of this section, be entitled to rely on section twenty-five of the Sale of Food and Drugs Act, 1875, and section seven of the Margarine Act, 1887, in the same way as his employer or master would have been entitled to do if he had been the defendant, provided that the servant further proves that he had no reason to believe that the article was otherwise than that demanded by the prosecutor. 38 & 39 Vict. c. 67.
50 & 51 Vict. c. 29.

(5.) Where the defendant in a prosecution under the Sale of Food and Drugs Acts has been discharged under the provisions of section twenty-five of the Sale of Food and Drugs Act, 1875, as amended by this Act, any proceedings under the Sale of Food and Drugs Acts for giving the warranty relied on by the defendant in such prosecution, may be taken as well before a court having jurisdiction in the place where the article of food or drug to which the warranty relates was purchased for analysis, as before a court having jurisdiction in the place where the warranty was given.

(6.) Every person who, in respect of an article of food or drug sold by him as principal or agent, gives to the purchaser a false warranty in writing, shall be liable on summary conviction, for the first offence, to a fine not exceeding twenty pounds, for the second offence to a fine not exceeding fifty pounds, and for any subsequent offence to a fine not exceeding one hundred pounds, unless he proves to the satisfaction of the court that when he gave the warranty he had reason to believe that the statements or descriptions contained therein were true.

21. The justices or court referred to in section twenty-two of the Sale of Food and Drugs Act, 1875, shall on the request of either party under that section cause an article of food or drug to be sent to the Commissioners of Inland Revenue for analysis, and may, if they think fit, do so without any such request. Duty of court to send article for analysis. 38 & 39 Vict. c. 63.

22.—(1.) At the hearing of the information in any proceeding under the Sale of Food and Drugs Acts, the production by the defendant of a certificate of analysis by a public analyst in the form prescribed in section eighteen of the Sale of Food and Drugs Act, 1875, shall be sufficient evidence of the facts therein stated, unless the prosecutor requires that the analyst be called as a witness. Provisions as to certificates of analysis.

(2.) A copy of every such certificate shall be sent to the prosecutor at least three clear days before the return day, and if it be not so sent the court may, if it thinks fit, adjourn the hearing on such terms as may seem proper.

23. This Act shall apply to Scotland with the substitution for "the Local Government Board" of "the Local Government Board for Scotland," and all powers and duties Transfer of powers from

Secretary for
Scotland to
Local Govern-
ment Board.

Application
to Ireland.

Interpreta-
tion of terms.

Definition of
"food."

Repeal of
enactments in
schedule.

Short title
and com-
mencement.
38 & 39 Vict.
c. 63.
42 & 43 Vict.
c. 30.
50 & 51 Vict.
c. 29.

vested in or imposed on the Secretary for Scotland in relation to the Sale of Food and Drugs Acts shall be transferred to, vested in, or imposed on the Local Government Board for Scotland.

24. This Act shall apply to Ireland with the substitution for "the Board of Agriculture" of "the Department of Agriculture and Technical Instruction for Ireland," and for "the Local Government Board" of "the Local Government Board for Ireland," and for "the London and Edinburgh Gazettes" of "the Dublin Gazette."

25. In this Act, unless the context otherwise requires—

The expression "margarine-cheese" means any substance, whether compound or otherwise, which is prepared in imitation of cheese, and which contains fat not derived from milk :

The expression "cheese" means the substance usually known as cheese, containing no fat derived otherwise than from milk :

The expression "local authority" means any local authority authorised to appoint an analyst for the purposes of the Sale of Food and Drugs Acts, and the expression "public analyst" means an analyst so appointed :

Other expressions have the same meaning as in the Sale of Food and Drugs Acts, and an offence under this Act shall be treated as an offence under those Acts.

26. For the purposes of the Sale of Food and Drugs Acts, the expression "food" shall include every article used for food or drink by man, other than drugs or water, and any article which ordinarily enters into or is used in the composition or preparation of human food ; and shall also include flavouring matters and condiments.

27. The enactments in the schedule to this Act are hereby repealed to the extent mentioned in the third column of that schedule.

28.—(1.) This Act may be cited as the Sale of Food and Drugs Act, 1899, and the Sale of Food and Drugs Act, 1875, and the Sale of Food and Drugs Act Amendment Act, 1879, and the Margarine Act, 1887, and this Act may be cited collectively as the Sale of Food and Drugs Acts, 1875 to 1899, and are in this Act referred to as the Sale of Food and Drugs Acts.

(2.) This Act shall come into operation on the first day of January one thousand nine hundred.

SCHEDULE.

ENACTMENTS REPEALED.

Session and Chapter.	Short title.	Extent of Repeal.
38 & 39 Vict. c. 63 . . .	The Sale of Food and Drugs Act, 1875.	In section two, the definition of the term "food." In section fourteen, the words "offer to," and the words "proceed accordingly and shall." Section fifteen. In section twenty-seven, the words from "Every person who shall give a false warranty in writing" to "a penalty not exceeding twenty pounds."
42 & 43 Vict. c. 30 . . .	The Sale of Food and Drugs Act Amendment Act, 1879.	Section ten.
50 & 51 Vict. c. 29 . . .	The Margarine Act, 1887.	In section six, the words "or with," and the words "not less than a quarter of an inch square."
54 & 55 Vict. c. 46 . . .	The Post Office Act, 1891.	Section eleven.

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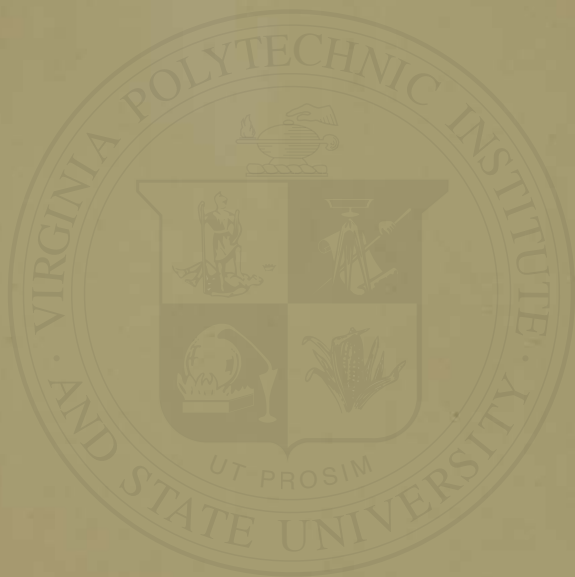
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