THE ANALYST

PROCEEDINGS OF THE SOCIETY OF PUBLIC ANALYSTS AND OTHER ANALYTICAL CHEMISTS

AN Ordinary Meeting of the Society was held at the Chemical Society's Rooms, Burlington House, on Wednesday, February 6th, Mr. John Evans, President, being in the chair.

Certificates were read in favour of:—Albert Edward Andrews, A.I.C., William Lewis Davies, Ph.D., M.Sc., F.I.C., George William Ferguson, B.Sc., Ph.D. A.I.C., Oswald Hitchen, B.(Tech.)Sc., A.I.C., John Knaggs, B.Sc., Ph.D., A.R.C.S., D.I.C., F.I.C., Norman Ratcliffe, F.I.C.

The following were elected members of the Society:—Henry Dryerre, Ph.D., M.R.C.S., L.R.C.P., Ronald William Hoff, A.R.C.S., A.I.C., Laurance John Sidney Lane, B.Sc., Alfred Edward Wright.

The following papers were read and discussed:—"The Chemical Examination of Furs in Relation to Dermatitis, Part V, The Action of Acid on Bandrowski's Base," by H. E. Cox, D.Sc., Ph.D., F.I.C., and J. U. Lewin, B.Sc., F.I.C.; "The Use of Infra-Red Rays in the Examination of Inks and Pigments," by C. Ainsworth Mitchell, M.A., D.Sc., F.I.C.; and "Vitamin Potency and Associated Characteristics of Cod-liver Oil," by R. S. Morgan and H. Pritchard.

NORTH OF ENGLAND SECTION

THE Tenth Annual General Meeting of the Section was held in Manchester on February 9th, 1935. The attendance was thirty-six; the Chairman (Prof. W. H. Roberts) presided.

The Secretary read the report and financial statement for 1934, which were adopted.

The following appointments for the coming year were made:—*Chairman*, Prof. W. H. Roberts; *Vice-Chairman*, A. R. Tankard; *Committee:* Prof. T. P. Hilditch, A. O. Jones, C. H. Manley, H. M. Mason, A. Scholes, C. J. H. Stock, R. W. Sutton; *Honorary Auditors:* U. A. Coates, J. W. H. Johnson; *Honorary Secretary and Treasurer*, J. R. Stubbs.

The following papers were read and discussed:—"Difficulties in Determining the *p*H values of Various Liquids," by D. Burton, M.B.E., D.Sc., F.I.C.; "The Determination of Water in Foodstuffs," by H. M. Mason, M.Sc., F.I.C.; "The Determination of Moisture in Cereal Products by Distillation with Tetrachloroethane," by J. M. Tucker, B.Sc., F.I.C., and T. E. Burke, A.I.C.

Death

WITH great regret we record the death of Frank Edward Day, who joined the Society in 1913.

Obituary

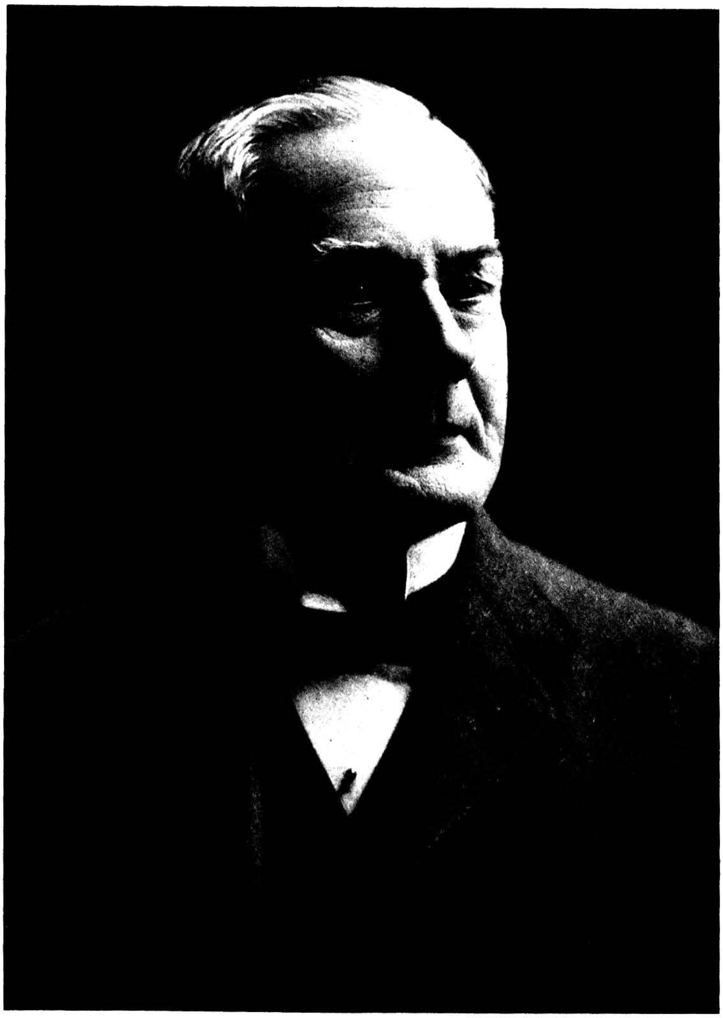
ROBERT RATTRAY TATLOCK

ROBERT RATTRAY TATLOCK was born in Glasgow on May 18th, 1837, and died at Helensburgh on December 22nd, 1934. He was the elder son of Robert Tatlock, a manufacturer in Glasgow, and on the maternal side he could trace his ancestors, who were engaged in commerce, back to the early years of the seventeenth century.

Tatlock received his early education in Greyfriars' School and later in the Trades House School, but from his youth his aim was to be an analytical chemist, and his motto was that he must excel in his chosen science, or leave it alone. With this object in view, he studied chemistry under Dr. Penny in Anderson's College (the site of which is now occupied by the Glasgow Royal Technical College), and later he was appointed chief assistant to Dr. Penny, a post which he held for over eight years. During this period, perhaps his most important undertaking was the analysis of over 100 "productions" in the trial for the murder by Dr. Pritchard of his wife and mother-in-law. This investigation was carried out entirely by Tatlock, the result being that antimony was found in quantity, having been administered in the form of tartar emetic.

About 1865 Tatlock severed his connection with Anderson's College, and accepted the post of manager in one of the departments of the Kames Gunpowder Company, Kyles of Bute. He remained in this post for only a short period, and in 1867 began his career as an analytical and consulting chemist in a laboratory in George Street, a short distance from Anderson's College. There success came slowly but surely; it was gained only by determination and very hard work, as those well knew who were intimate with him. In addition, he had classes for students, one of whom, in 1869, was the late Sir William Ramsay. In 1909 Ramsay wrote a short autobiography, in which he said: "In 1869 I entered the laboratory of Robert Tatlock, who had been assistant to Professor Penny. Mr. Tatlock was (and is) an eminent analytical chemist, and during a year with him I had a course of qualitative analysis, and got through a good part of quantitative analysis."

In 1868 Tatlock read his first paper on analytical chemistry before the Chemical Section of the Glasgow Philosophical Society. This was "On the Estimation of Potassium," and was the result of a laborious series of trials in collaboration with his friend James Chalmers, who was also an excellent chemist. They came to the conclusion that the results reported by some chemists were too high in potassium, and they traced this to the fact that considerably different atomic weights for platinum were in use. Working on chemically pure potassium chloride, they found that the atomic weights required to obtain an accurate result were those



published by Stas in 1865. They were: platinum, 197·193; potassium, 39·137; chlorine, 35·457. These results, which have been corroborated by other analysts, mean that if the International Atomic Weights for 1934 are adopted, the result would be 100·36 per cent. for pure potassium chloride. The new atomic weights are: platinum, 195·23; potassium, 39·096; chlorine, 35·457; the last is identical with Stas's figure of 70 years ago, while that of platinum has been oscillating. Tatlock had his results re-tested and confirmed, and, to the last, adhered to the old atomic weights.

Another pet subject of Tatlock's was that of graduated vessels used in chemical analysis, and indeed he was almost finical about them. He had an apparatus by which a tube (a burette for example), although of unequal calibre throughout, could be graduated so that each division delivered exactly the same volume of the same fluid. In 1870 he read a paper on "Some Sources of Error in Volumetric Analysis," dealing with this subject.

In 1870 Tatlock joined in partnership with Dr. William Wallace and Dr. John Clark, both of them eminent analysts, under the designation of Wallace, Tatlock & Clark. The firm was dubbed the "Chemical Trinity" by *The Bailie*, a Glasgow comic weekly. They proved to be a very strong combination, and practically monopolised all the analytical work required in the City of Glasgow.

A good deal of work was done on the analysis of Scottish blackband and clayband ironstone, but this source of iron gradually dwindled, and Spanish iron ore began to be imported. Tatlock managed to obtain the sampling and analysis of these ores, and in time it formed a large part of the analytical business, which has continued, with ups and downs, especially the latter, in recent years. Happily for the iron trade in Scotland, improvement has begun. Tatlock was the first to undertake this work in Scotland, and, like everything he took in hand, he carried it on to success.

In 1877 he was, along with his partners, appointed Public Analyst for Glasgow, and he held that post till his death. In 1888 he dissolved the partnership with Wallace and Clark, and joined with Dr. J. B. Readman, an Edinburgh analyst well known at that time, the new firm being R. R. Tatlock & Readman, and on Dr. Readman's retiral, the firm of R. R. Tatlock & Thomson was formed. In 1888 Tatlock was appointed Gas Examiner to the City of Glasgow.

During these co-partneries, and later, the usual routine work of a laboratory was carried on, but there were four outstanding matters to attend to. In 1896 Tatlock was appointed Sewage Analyst to the Corporation of Glasgow, the object being to determine the method of treatment that would be most suitable, when all the circumstances were taken into account. Shortly before the Boer War he was requested to go to South Africa, and at Pretoria he gave expert evidence in favour of the Macarthur–Forrest process for the extraction of gold from ores by potassium cyanide. He was also employed to give evidence for Nobel's Explosives Co. in their case, "Ballistite *v.* Cordite," against H.M. Government.

The fourth important business was to give evidence before the Royal Commission which was appointed, in 1907, to answer the question "What is Whisky?" This arose out of cases in which prosecutions were instituted against samples that were not up to the standard in secondary products for pot-still whisky. It was

assumed that the pot-still variety only was genuine whisky, although the patent-still product had been in use for 73 years, and was always called whisky. Another point was that a rumour went about that patent-still whisky was poisonous, and a chemist, who shall be nameless, said that he gave pot-still whisky to one monkey and patent-still whisky to another, the result being that the former became benevolently drunk, and the latter beastly drunk. Tatlock experimented upon himself, but neither of these results occurred. The Royal Commission decided in favour of the distillers, and left matters as they were.

Tatlock also contributed a number of papers to THE ANALYST and other publications, such as those on Tea, Coffee, Water, etc., and one on "The Determination of small proportions of Bromine and Chlorine in Iodine," this being a subject in which he was much interested.

From 1873 to 1884, besides attending to his duties in the laboratory, Tatlock lectured on chemistry in the College of Science and Arts, now incorporated in the Royal Technical College. He did not profess to be an orator, but the clearness and precision of his exposition of the principles of chemistry could not be surpassed. He was particularly careful in the preparation of his experiments shown on the lecture table, and they never failed to come off. At the beginning of a course, he always gave several lectures on physics, dealing with sound as well as with heat and electricity. As an example of his minute care about experiments, the well-known singing flames may be taken. He had three or four which gave different sounds, and adjusted the surrounding tubes exactly on the points where they gave a sound at a certain pitch. He then struck the required note on a violin and immediately the flame responded and sang, as we say in popular language.

In 1867 he was elected Fellow of the Chemical Society, in 1870 Fellow of the Royal Society of Edinburgh, in 1877 Fellow of the Institute of Chemistry, and in 1903, when the Association of Public Analysts of Scotland was formed, he was elected first President. From 1888 to 1890 he acted as Examiner in the University of Glasgow. He became a member of the Society of Public Analysts in 1876, served on the Council in 1887–8 and 1904–5, became Vice-President in 1891–2, and was elected President in 1908–9.

Tatlock loved his work, and was thorough, perhaps excessively so, in preparing his evidence for important cases, but he did not lose by such care. He would not only prepare arguments for his own side, but would also discuss any the other side might bring forward. As an example, in one case at a meeting with Counsel, he gave his facts and arguments for his clients, and then was asked what evidence he would give against them. He did so, and Counsel told him that he should be on the other side, and that, their case being bad, it would be advisable to admit liability and settle the matter out of Court.

Outside his profession he was excellent company, and had always some good stories to tell, but when in a somewhat flippant mood he would talk nonsense for fun. On one such occasion, one of his best friends, a brother analyst, told him that he was the most frivolous man he ever knew. He certainly loved to talk nonsense sometimes, but that is really a saving grace in a serious-minded man like him.

Tatlock was no specialist in the narrow sense of the word, for, to the last, he took an interest in politics, music and literature. In politics he held liberal principles, but was not a rigid party man, and recently he described his views as those of a sensible socialist, but he never took any active part in politics.

In his earlier days he was an excellent violinist, being very fond of music, and in his later days, when he could not go to concerts, he listened to them on the wireless. He specially loved Scots songs, and even when going round the laboratory among his assistants he would be singing some melody in a low crooning voice.

Tatlock had the courtly manners towards others that we called the conduct of a gentleman, the true meaning of which is lost in our more unconventional age. He was no hero worshipper, but towards Faraday he came very near that attitude. He admired his great work as a chemist, but he could never forget a Sunday in a little Glassite Chapel in Glasgow, when he heard Faraday preach a simple and beautiful sermon on "Let brotherly love continue."

Tatlock is survived by a son and by a daughter—the wife of the Very Rev. Charles Warr, Dean of the Thistle.

May I close on a personal note? For about 50 years I was in daily business relations with Tatlock, and during all that time we worked harmoniously together, without a note of disagreement—a record, he used to tell his friends. Then in his retirement we kept up the old friendly relations, and to the last he took an intense interest in our business and in all that went on in the chemical world.

R. T. THOMSON

The Question of Tannin in Maté

BY W. A. WOODARD AND A. N. COWLAND

(WORK DONE UNDER THE SOCIETY'S ANALYTICAL INVESTIGATION SCHEME)
(*Read at the Meeting, December 5*, 1934)

INTRODUCTION.—In South America and associated countries a beverage is made from maté, the dried leaves of various species of *Ilex* (Fam. *Aquifoliaceae*)—shrubs indigenous to Brazil and Argentina. It is usual to take the drink as an aqueous infusion, prepared in a silver cup or gourd, from which the name "maté" is derived.

Owing to the discovery of the shrub in Paraguay by early Jesuit missionaries in the seventeenth century, all varieties of maté, irrespective of species, have come to be known as *Ilex paraguayensis*. The genus *Ilex*, however, contains two-hundred-and-eighty species, and Wehmer[1] gives the names of nine different varieties of *Ilex* leaves sold under the name of maté. To-day, the finest variety obtainable is derived from *Ilex paraguariensis* (St. Hilaire), and in Parana it is forbidden by law to export any other.

The process for curing maté is distinct from that used for common "tea"; moreover, it is unlikely to produce any appreciable change in the phenolic

constituents of the plant. Tea is rolled and fired, maté is not. In several publications it is stated that maté is dried by means of smokeless heat.*

According to the descriptions given in abstracts from this literature and elsewhere the modern curing process may be summarised roughly as follows:— The twigs and leaves are collected between May and September, this period being fixed by law to avoid damage to the shrubs. First, there is a preliminary toasting and drying by means of hot air; direct heat is carefully avoided, because smoke would give the finished product an undesirable taste and odour. Secondly, after a definite period of storage, the leaves and twigs are separated by means of a special threshing process. Lastly, the leaves are subjected to a final drying by means of hot air conveyed through pipes or tunnels. The final product should be green, hard and resistant to fermentation and deterioration.

The presence in maté of so-called tannin, either identical with, or analogous to, that in tea, has been definitely reported by several chemists, but it should be pointed out that little or no work has been done on the phenolic constituents of the drug since 1922. This was before the advent of such specific tests as that requiring the use of goldbeaters' skin, and we have therefore made a critical examination of the results of earlier work. The more important references are as follows:— Rochleder and Hlasiwetz[2] concluded that the main phenolic constituent of maté was identical with the caffetannin of coffee. Arata[3] found the plant phenol to be analogous to, but not identical with, that of coffee. Peckolt[4] published a confirmation of Arata's work. Kunz-Krauze[5] described the main constituent as a glucoside of either caffeic or chlorogenic acid containing an unknown optically inactive hexose. Peacock and Peacock[6] concluded, on very slender evidence, that maté contained an astringent principle which they isolated and identified as a phlobaphene.

At the suggestion of the Society's Analytical Investigation Scheme Committee we have made a further investigation, dealing primarily with the question of the presence of genuine tannin; secondary considerations were the establishment of a difference between the plant phenols of maté and common "tea" and an attempt to separate colouring matter from the main plant phenol in maté, with the object of obtaining data relating to their constitution and structure.

PRELIMINARY EXAMINATION.—Thirty individual samples of cured maté were examined, including imports dating as far back as 1920 and 1927, as well as current imports from wholesale houses and purchases from London stores. A cursory botanical examination showed that all the samples were typical of the genus *Ilex*, although there was undoubtedly some variation in species.

A small and limited supply of each kind of maté for comparison purposes was obtained by courtesy of the authorities at Kew. Coffee beans (*Coffea arabica*) and Asiatic tea leaves (*Thea chinensis*) were also compared with maté, with a view to bringing out essential similarities and differences. Except in certain instances, aqueous extracts (1 in 20) were used for the tests. The extracts were prepared by decocting the comminuted material with water for fifteen minutes; when necessary,

* "Maté: An Important Brazilian Product," by C. R. Cameron (*Bulletin of the Pan-American Union*, Oct., 1929, pp. 988–1005); "Il Mate o Tè del Paraguay," by Dr. C. Micastro (*L'Agricoltura Coloniale*, Nov., 1928, to Aug., 1929); "A Exploracao do Mate" (*Boletin do Ministerio da Agricultura, Industria e Commercio*, Brazil, for April and May, 1929).

chlorophyll, caffeine, fats, waxes, resins, etc., were removed by treatment with organic solvents.

It will be realised by those familiar with the analysis of tannins that the available tests capable of giving specific results are very few. In this work, particular emphasis has been laid on the results obtained with goldbeaters' skin,[7] gelatin solution, and certain tests devised by Ware, all of which, with certain exceptions, are described in *Allen's Commercial Organic Analysis*, Volume V, under the section dealing with tannins.[8] It was found necessary to employ a limited number of colour reactions, but only those were used with which it was considered possible to carry out control tests.

Aqueous extracts of maté had the following general characteristics:—Yellowish-brown in colour, giving an acid reaction to litmus (pH 5 to 6); they gave precipitates with solutions of cinchonine and quinine sulphates and a copious yellow precipitate with basic lead acetate; ferric iron (with increasing pH) gave a grass-green colour, whereas ferrous iron (with carefully controlled decreasing pH) gave green, violet, purple and deep brown colours; aqueous ammonia (10 per cent.) gave a beautiful emerald-green which deepened rapidly on shaking or allowing the mixture to stand in contact with air.

The reactions with iron salts were particularly interesting, especially when considered in conjunction with the results of tests for genuine tannin detailed in the next section of this paper. The results given with ferrous iron (Mitchell's ferrous tartrate reagent)[9] have special significance when interpreted according to Ware.[10] Ware[11] classified plant phenols by the colours and precipitates obtained with the ferrous tartrate reagent in the presence of an appropriate quantity of $N/50$ sodium bicarbonate solution or a weak solution of ammonia; if no colour is obtained, the phenol is provisionally assigned to class A; a violet colour indicates probably a phenol of class B, and a deep brown colour probably a phenol of class C.

Maté extracts gave results typical of classes B and C; the more important phenols belonging to class B are pyrocatechol, protocatechuic acid, the catechinols (catechins), chlorogenic acid, caffetannin, ipecacuanhic acid, adrenalin and catechol tannins; class C (containing most of the pyrone and quinonoid phenols, and some benzophenone phenols) is represented chiefly by the anthoxanins, such as the flavone, luteolin, and the flavonols morin, quercetin, quercetrin, rutin, myricetin, etc.

It is not practicable to set out here in full all the tests used to show the absence of various phenols from maté; it is sufficient to state that the following substances were thoroughly tested for, particular attention being paid to the possible presence, in the free state, of the first two in the list:—Gallic acid, catechol, protocatechuic acid, resorcinol and phloroglucinol. There was no evidence indicating the presence, in the free state, of any of these substances.

Another qualitative reaction of extracts of maté is the beautiful emerald-green colour obtained with ammonia. We believe this reaction to be characteristic of maté, and probably of caffetannin; the colour is very intense, persisting for several hours and slowly changing, on long contact with air, to a deep brown. This is undoubtedly an oxidation reaction, because the colour can also be obtained with acid permanganate, and, conversely, can be completely and rapidly destroyed

by reduction with zinc and hydrochloric acid. Aqueous extracts of coffee beans gave a similar reaction, but the colour was very much less intense.

It is well known that, under suitable conditions, many phenols (notably gallic acid, gallotannin, catechol, protocatechuic acid and resorcinol) will give a green colour with ammonia or other alkali, but the colour is usually very transient and much less intense than that given by maté extractives; moreover, it must be recalled that four of the above-mentioned substances have been shown by many tests to be absent from maté.

There has been a general tendency in the past to associate chlorogenic acid with this peculiar green colour given by various extracts with ammonia solution; furthermore, it has been customary to identify caffetannin with chlorogenic acid, in spite of the fact that the evidence for their identity is very slender. Nierenstein,[12] dealing with hydrolysable tannins, gives good reasons for the conclusion that caffetannin and chlorogenic acid are not identical. Gorter,[13] describing the reactions of chlorogenic acid isolated from coffee, makes it clear that it gives with ammonia not a green colour, but a definite yellow which, on standing, acquires a reddish tint.

The preliminary examination of maté extracts indicated the presence of two plant phenols, *viz.* an iron-greening anthoxanin (probably a flavone or flavonol derivative) which was responsible for the deep brown colour given with Mitchell's reagent in the presence of sufficient alkali; and secondly, what is here described as a pseudo-tannin (probably a caffetannin), which gave the violet colour with Mitchell's reagent, also in the presence of sufficient alkali; appreciable quantities of these two phenols undoubtedly exist in maté. As will be seen, the results recorded at a later stage of this work tend to substantiate this suggestion.

EXAMINATION OF FRESH AND CURED MATÉ FOR TANNIN.—The more important results are given below in tabular form, the tests used being all fairly well known, with, perhaps, the exception of the antipyrine test devised by Ware.[14] This routine test for tannins, used in conjunction with a suitable phosphate buffer, gave excellent results with controls containing very small amounts of gallotannin.

TABLE I

Test	Observation	Inference
Goldbeaters' skin		
(*a*) Treatment for tanning	No opacity	Tannin absent
(*b*) Staining with ferrous sulphate	Greyish-green stain	Indication of iron-greening anthoxanin
(*c*) Decolorisation with acid	No final stain	Phlobaphen absent
Gelatin solution	Complete absence of turbidity or precipitate	Tannin absent
Ware's antipyrine test	No turbidity or precipitate	Tannin absent
Ware's iron and ammonium citrate test	No precipitate	Tannin absent
Ware's modification of Stiasny's reaction	No characteristic precipitate Iron-greening and iron-browning filtrate No blue colour produced	Phlobatannin absent Gallotannins absent
Ware's modification of Mitchell's ferrous tartrate test	No blue or violet colour produced	Gallotannin absent

In addition to the tests described above, a hide-powder test, kindly made by Dr. E. W. Merry of The British Leather Manufacturers' Research Association, gave noteworthy results. The comminuted material (25 g.) was extracted with 1 l. of water in a Procter extractor at a temperature not exceeding 60° C. The sample contained 33·2 per cent. of water-soluble substances, of which 12 per cent. was absorbed by the hide powder, as used in the official method of tanning analysis; the remaining 21·2 per cent. was not absorbed. Lack of sufficient fresh material made it impossible to make a control test; but since hide powder will absorb substances other than tannins, including natural colouring matters and non-volatile organic acids, we suggest that the absorption in the case of maté was due to the presence of one or both of these constituents in the leaves of the plant.

The results of sensitive spot-tests on filter-paper, devised by Ware,[15] confirmed the foregoing conclusions; the same author's tests with iodine and ammonia were also confirmatory.

It is noteworthy that the goldbeaters' skin test, the most specific test known for detecting genuine tannin, gave a negative result with each of the thirty samples of maté examined. We conclude that there is not, and never has been, any genuine tannin in maté, for the following reasons:

(*a*) First and foremost there is no phlobaphen present.
(*b*) There is no gallic acid present.
(*c*) The modern process by which the leaf is cured is unlikely to produce tannin or to destroy it.

A Comparison of Fresh Maté with Fresh Tea.—These tests bring out certain pronounced differences between maté and tea. Little can be said about the plant phenols in tea, because the chemistry of tea-tannin is a mass of contradictions and requires further elucidation. Comparisons were also made with fermented tea, but the results were so similar to those given in Table II that there was no point in recording them. The only noticeable difference between fresh and fermented tea was that the cured material gave strong positive results for phlobaphen, whereas the fresh tea did not.

It is clear from these results that, so far as qualitative tests go, there is little similarity between maté and tea. Tea undoubtedly contains genuine tannin, but maté does not, and this constitutes the chief and most far-reaching difference between the two.

A Comparison of Fresh Maté with Fresh Coffee.—These tests bring out differences and similarities between maté and coffee. A comparison of the cured materials also gave very similar results, except that with coffee the reactions were much less intense; this was probably due to destruction of plant phenols during roasting.

Coffee beans are known to contain caffetannin and chlorogenic, caffeic, and gallic acids. From the comparison given on p. 140 it seems probable that a little genuine tannin is also present; this would be quite feasible, since it is rare to find gallic acid unassociated with tannin in a plant. It is noteworthy that the remarkable reaction with ammonia (p. 137) is also shared by extracts of coffee.

TABLE II

Test	Observation	Inference
Goldbeaters' skin	Maté: Already described. Tea: Definite opacity with bluish-green stain	— Tannin present; gallotannin, gallic acid or catechol tannin present
Gelatin solution	Maté: Already described Tea: Characteristic precipitate	— Confirmation of presence of tannin
Ware's modification of Stiasny's reaction	Maté: Already described Tea: Characteristic precipitate, iron-blueing and iron-greening filtrate	— Phlobatannin present: gallotannin, gallic acid or catechol tannin present
Osmium tetroxide solution (1 per cent.) + appropriate buffer	Maté: Reddish-brown, no violet or blue Tea: Purplish-violet	Confirmed absence of gallic acid Probable presence of a pyrogallol tannin
Bromine water	Maté: No precipitate on standing Tea: Precipitate on standing	Absence of catechol tannin Probable presence of catechol tannin
Nierenstein's test (0·5 per cent. diazobenzene chloride)	Maté: No precipitate on standing Tea: Precipitate on standing	Absence of catechins or catechol tannin Presence of one, or both, of these substances

SEPARATION OF THE PLANT PHENOLS IN MATÉ.—The preliminary examination of maté extracts indicated the presence of two plant phenols: one classified as a pseudotannin, and the other as a member of the large family of natural yellow colouring matters. Perkin[16] has called attention to the fact that many natural colouring matters and tannins associated in plants often show a marked resemblance to each other in constitution and chemical reactions. This seemed probable with

TABLE III

Test	Observation	Inference
Goldbeaters' skin	Maté: Already described Coffee: Slight opacity, bluish-green stain	— Indication of a little tannin; gallotannin, gallic acid or a catechol tannin present
Solution of gelatin	Maté: Already described Coffee: Slight turbidity	— Confirmation of above result
Ware's modification of Stiasny's reaction	Maté: Already described Coffee: Very slight precipitate, iron-greening and iron-blueing filtrate	— Confirmation of above results
Ammonia solution, 10 per cent.	Maté: Beautiful emerald-green intensified on shaking Coffee: Very similar to maté only less intense	Caffetannin present Caffetannin present
Osmium tetroxide solution (1 per cent.) + appropriate buffer	Maté: Already described Coffee: Same as maté, but tinged with violet	— Probably a little gallic acid or tannin present
Nierenstein's test (0·5 per cent. diazobenzene chloride)	Maté: Already described Coffee: No precipitate on standing	— Absence of catechins and catechol tannin

maté, since the reactions of one phenol nearly always tended to be masked by those of the other.

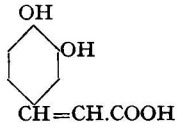
For this reason attempts were made to separate the two constituents. Fractional precipitation with lead gave poor results; various attempts to remove the colouring matter by adsorption or oxidation were equally unsuccessful. Extraction with different organic solvents yielded, towards the end of the work, some measure of success. It was noticed that neutral acetone extracted some colouring matter from maté, but very little or no pseudotannin; alkaline acetone extracted a fair amount of colouring matter, but again, little or no pseudotannin; acidified acetone extracted both plant phenols in sufficient amount to afford hope of a good separation when the extract was treated with dry ammonia gas for a definite period. The process actually used was briefly as follows:

One kg. of dried and powdered maté was allowed to stand in contact with dry chloroform for six hours with occasional shaking. The chloroform was then decanted, and the leaves were pressed; washing with several portions of ether and a final pressing of the leaves completed the removal of caffeine, chlorophyll, fats, resins, waxes, etc. It goes without saying that the plant phenols in maté had previously been found to be almost insoluble in chloroform and ether. Next, the pressed material was spread out on a tray and dried at a temperature not exceeding 60° C. Treatment with acidified acetone followed (pH 3 to 4, adjusted by means of concentrated sulphuric acid), sufficient solvent being added to cover the leaves completely, and twenty-four hours being allowed for the extraction. The solution was filtered through a Buchner funnel, and a fairly rapid current of dry ammonia gas was passed through the filtrate for about two minutes. The pinkish-red precipitate obtained was quickly washed with neutral acetone (several times), and then transferred to a desiccator to dry. The filtrate, containing colouring matter, was again treated with ammonia gas, any precipitate obtained being neglected; after re-filtering, if necessary, the new filtrate was evaporated to dryness under reduced pressure, and the residue was transferred to a desiccator.

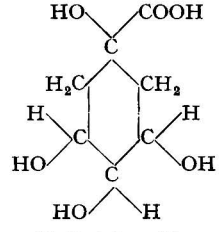
This represents a rough attempt to separate the colouring matter from the plant phenol or pseudotannin, but the method will probably need some re-adjustment before being applicable on a large scale.

Examination of the Precipitate.—The precipitate obtained by the method described had the following general characteristics:—It was readily soluble in water, forming a clear reddish-brown solution; the reaction to litmus was markedly acid, and the colour given with ammonia was the deep emerald-green already described; ferric iron (with increasing pH) gave a grass-green colour, whereas ferrous iron (with decreasing pH) gave green, violet-purple, and finally a deep wine-red colour, but no brown colour was noticed. According to Ware,[11] this reaction is typical of pseudotannins in general. Precipitates were obtained with cinchonine and quinine sulphates, heating by itself yielded catechol, and fusion with alkali gave catechol together with a volatile acid not yet identified. It is worth noting that the preliminary reactions for colouring matter, and also others described later in detail, gave negative results when applied to an aqueous solution of the above precipitate.

Caffetannins give, on hydrolysis, (i) caffeic and (ii) quinic acid, and a residue not yet fully investigated.

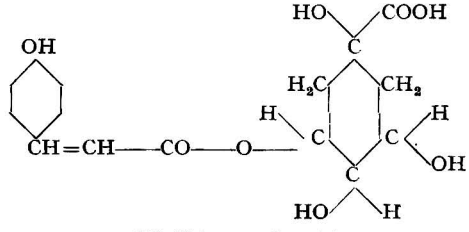


(i) Caffeic acid



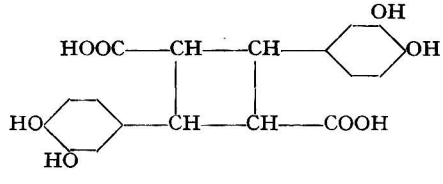
(ii) Quinic acid

Nierenstein (*loc. cit.*) is in favour of regarding caffetannin as a condensation product of (iii) chlorogenic acid, the established formula of which is given below.

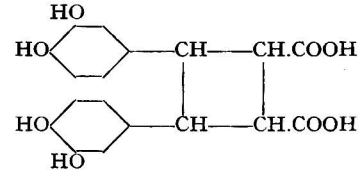


(iii) Chlorogenic acid

If this view be accepted, the residue obtained on hydrolysis would be either (iv) tetrahydroxy-truxillic acid or (v) tetrahydroxy-truxinic acid.



(iv) Tetrahydroxy-truxillic acid



(v) Tetrahydroxy-truxinic acid

At present little is known of either of these acids; moreover, Nierenstein[12] points out that in accepting the suggested formula for caffetannin it must be realised that the production of caffeic acid would be entirely due to the presence of some unchanged chlorogenic acid in the caffetannin. Finally, he agrees with Gorter,[13] who is of the opinion that caffetannin is a mixture of several substances, including chlorogenic acid.

An attempt was made to produce caffeic acid from the precipitate obtained from acidified acetone. The precipitate was treated with a slight excess of 10 per cent. aqueous potassium hydroxide solution and hydrolysed under reduced pressure for thirty minutes; after cooling, the brown solution was acidified with dilute sulphuric acid and then shaken out with several portions of ether. The ethereal layers were mixed, the solvent evaporated, and the residue was taken up in a little water. This aqueous solution gave positive results in the following tests devised by Gorter[13] for caffeic acid obtained by him from coffee beans: a grass-green colour with weak ferric iron solution; a lemon-yellow colour with

lead acetate solution and baryta water; ready reduction of silver nitrate, and a yellow colour with ammonia solution, changing to a reddish tint on standing. This last reaction is also shown by chlorogenic acid.

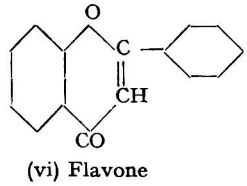
The same tests were applied to a residue obtained by a method proposed by Griebel[17] for determining chlorogenic acid in coffee. The method depends on the conversion of chlorogenic acid into caffeic acid by alkaline hydrolysis; the results were positive for caffeic acid.

An attempt to produce quinic acid from the precipitate under discussion was based on the knowledge that this substance, when distilled with sulphuric acid and manganese dioxide, yields quinone. The precipitate was hydrolysed with excess of 30 per cent. aqueous potassium hydroxide solution for one hour; subsequent distillation with concentrated sulphuric acid and manganese dioxide, gave, on standing, a yellow deposit which was collected and identified as quinone.

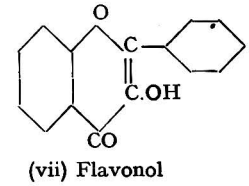
We fully realise that the results just described need further confirmation; nevertheless, we maintain that there is, at this stage of the work, evidence for concluding that caffetannin, or some closely allied substance, is present in maté. As previously mentioned, the search for a practical method of separating the two phenols in maté has only recently been successful, and work is still in progress with the object of obtaining the pseudotannin and colouring matter in the crystalline state. Attempts were made to crystallise the pinkish-red precipitate described above, but the final product was invariably a hard brittle mass. This may have been due to impurity, or, on the other hand, possibly the precipitate is a mixture of substances; if so, this would be another point in favour of caffetannin being present.

Another possibility is that chlorogenic acid exists in the free state in maté. The tests described by Charaux[18] and by Gorter,[19] depending on colour reactions with iron salts after acid hydrolysis of fresh extracts, gave results indicating the presence of chlorogenic acid. Hoepfner[20] describes a colorimetric method for determining chlorogenic acid in coffee; this was tested with maté, coffee being used as a control, and the result was positive. In spite of this, we think that it is difficult to state definitely whether the reactions just described would be given by chlorogenic acid existing in the free state in maté, or by the same acid existing as a component of a mixture such as caffetannin may be. This must be left for further investigation.

EXAMINATION OF THE FILTRATE.—The presence of an iron-greening anthoxanin, probably a (vi) flavone or (vii) flavonol derivative, was indicated by the results obtained in certain tests in the preliminary examination.



(vi) Flavone



(vii) Flavonol

Attention has been called to the fact that the reactions of one phenol interfered with those of the other; owing to this confusion, it was thought at one time

that the colouring matter in maté was almost certainly a flavonol derivative, but the separation process has made it possible to correct this error.

The filtrate from the acid acetone precipitate should contain the colouring matter; a residue obtained from such a filtrate in the manner already described, gave the following general reactions: It dissolved in water to give a pale yellow solution, being slightly acid to litmus and giving a copious yellow precipitate with lead acetate; with ferric iron (increasing pH) a green colour was obtained; with ferrous iron (decreasing pH) there was a green colour, which, on adding sufficient $N/50$ sodium bicarbonate solution, became deep brown; no violet or purple colour was seen, thus indicating, according to Ware,[11] that phenols belonging to class B had been eliminated; in other words, plant colouring matter had been separated from pseudotannin. When heated alone, the dry residue gave catechol; when fused with alkali, it gave protocatechuic acid and phloroglucinol.

The following results, presented in tabular form, sum up the more important reactions of the colouring matter in maté.

TABLE IV

Test	Observation	Inference
Calico or wool mordanted with aluminium	Bright yellow—no reddish tint —fairly fast to light; fluoresced in ultra-violet light	Flavone or flavonol present; probably a flavone
Ware's iron and ammonium citrate test	Production of a heavy brown precipitate	Presence of a catechol, phloroglucinol, flavone or flavonol
Willstätter's reduction reaction	Complete absence of any colour	Flavonols absent; flavone derivative probably present
Perkin's test (air-oxidation of an alkaline solution, followed by acid precipitation)	Definite precipitation after acidification	Flavone derivative present

All natural hydroxyflavones dye fabrics mordanted with aluminium a yellow shade, the intensity of which is dependent upon the position of their hydroxyl groups. The yellow given by flavonols to mordanted calico is usually slightly tinged with red, which serves as a useful distinction from flavones. Willstätter's reduction test with zinc and hydrochloric acid is another useful reaction for distinguishing flavones from flavonols. A third reaction, based on Perkin's observation that hydroxyflavones are not, as a rule, readily oxidised in alkaline solution, serves as a useful confirmatory test. The tests for colouring matter in maté were controlled with oak-bark containing quercetin and onion-skin containing apigenin.

We thus consider that there is good evidence pointing to the existence in maté of a flavone derivative. In further work it is hoped to obtain confirmation of this by preparing the colouring matter in the crystalline state and investigating its chemical constitution.

CONCLUSIONS.—It has been shown that maté is completely free from genuine tannin. This is a matter of considerable importance, in view of the effects of ordinary tea upon the digestion.

It has also been shown that maté contains an appreciable amount of a natural

yellow plant colouring matter; it is reasonably certain that this colouring matter is a derivative of flavone.

Certain evidence has been obtained pointing to the presence in maté of caffetannin or some closely allied compound.

Comparisons have been made with coffee and tea, and attention has been drawn to important differences and similarities.

We wish to express our thanks to Dr. C. A. Mitchell and Dr. H. E. Cox for their help and for many useful suggestions received during the course of the work. Thanks are also due to Dr. M. Nierenstein and Mr. Alan Ware for confirming certain results, and to Prof. O. L. V. de Wesselow, Director of the Medical Unit Laboratory, St. Thomas's Hospital, for kindly providing facilities for the work to be done.

References

1. C. Wehmer, "Die Pflanzenstoffe," 1911, p. 457. Jena.
2. F. Rochleder and H. Hlasivetz, *Ann. Chem. Pharm.*, 1848, **66**, 39; 1850, **76**, 339; 1867, **142**, 219.
3. P. N. Arata, *Gazz. Chim. Ital.*, 1877, **7**, 520.
4. T. Peckolt, *Pharm. J.*, 1884, **14**, 121.
5. H. Kunz-Krauze, *Arch. Pharm.*, 1893, **231**, 613.
6. J. C. and B. L. de G. Peacock, *J. Amer. Pharm. Assoc.*, 1922, **11**, 609.
7. P. Price, ANALYST, 1924, **49**, 25.
8. *Allen's Commercial Organic Analysis*, 6th Ed., Vol. V, pp. 1–204.
9. C. A. Mitchell, ANALYST, 1923, **48**, 2.
10. A. Ware, *Quart. J. Pharm.*, 1928, **3**, 365.
11. *Ibid.*, 1926, **1**, 377; ANALYST, 1929, **54**, 58.
12. M. Nierenstein, "The Natural Organic Tannins," 1934, Chapter VI, p. 207.
13. K. Gorter, *Annalen*, 1908, **358**, 327; 1908, **359**, 217.
14. A. Ware, *Quart. J. Pharm.*, 1933, **3**, 460; *Pharm. J.*, 1933, **131**, 148; ANALYST, 1933, **58**, 703.
15. A. Ware, *Quart. J. Pharm.*, 1930, **3**, 460.
16. A. G. Perkin and A. E. Everest, "Organic Colouring Matters," 1918, p. 17.
17. C. Griebel, *Chem.-Ztg.*, 1933, **57**, 353; ANALYST, 1933, **58**, 621.
18. C. Charaux, *J. Pharm. Chim.*, 1910, **102**, 292.
19. K. Gorter, *Rec. Trav. Chim. Pays-Bas*, 1912, **31**, 281.
20. W. Hoepfner, *Chem.-Ztg.*, 1932, **56**, 991; ANALYST, 1933, **58**, 100.

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Discussion

Dr. H. E. Cox congratulated the authors on adding so valuable a chapter to the study of maté. He drew attention to Capt. T. A. Joyce's lecture on maté (*Nature*, 1934, **134**, 724), from which it appeared, among much interesting matter, that the first chapter had been written in 1617 by Cornejo, and now Mr. Woodard and his colleague had added one more. It was particularly important to note that the authors had established the absence of true tannin, as in commercial circles it was still asserted that about 7 per cent. of tannin was present. He thought it was a matter for congratulation that the authors had established the nature of the chlorogenic acid and the absence of tannins by definite chemical study, and had not depended solely on colour reactions which were known to be somewhat erratic. It was significant that the hydrolysis yielded caffeic acid, which was known to be a product of caffetannin, and he hoped that they would succeed in establishing the identity of the hydrolysis products of the chlorogenic acid, and also that they would continue their good work, so that we might get to know something of the real constitution of the substances present.

Mr. WOODARD replied that they had been able to produce quinic acid on distillation with sulphuric acid. They had also been able to show that caffeic acid was present.

Dr. C. A. MITCHELL said that the authors had made a valuable addition to the long series of papers that had been brought before the Society by way of the Analytical Investigation Scheme. When considering the conflicting statements upon the presence or absence of tannin in products such as maté, it was necessary to have a clear conception of what was meant by "tannin." Definitions for tannin were notoriously vague, but it seemed reasonable to regard a tannin as a substance that would "tan," and for this reason great importance attached to the results of the goldbeaters' skin test, which was essentially a "tanning" reaction. If this test gave negative results, as it did with maté, there could be no true tannin present, but it would be interesting to learn whether the authors had ascertained the nature of the substances that were adsorbed from the extract of maté by hide-powder. In view of the results obtained with goldbeaters' skin, they were evidently not tannins.

Mr. R. L. COLLETT asked whether the green colour obtained with ammonia was also obtained with other alkalis.

Mr. WOODARD, replying, said that he very much appreciated the remarks made by Dr. Cox. In the first place he would like to emphasise the fact that the work dealt primarily with the question of genuine tannin in maté, and that most of the time at their disposal had been devoted to this purpose. Also, he was of opinion that before any useful knowledge bearing on the chemistry of the plant phenols in maté could be gained, it would be necessary first to separate colouring matter from essential plant phenols and then to purify the products so obtained. This was not an easy matter, although he was pleased to say that reasonable success had been attained in making a separation of the two phenols, and some evidence was given in support of their chemical identity. The green colour given by maté extracts with ammonia was also obtained with solutions of sodium and potassium hydroxide, although the colour was much less intense. He had observed that the green colour was destroyed by acids alone, as well as by reduction with zinc and hydrochloric acid. With regard to the points raised by Dr. Mitchell, he had not considered it necessary to make a full investigation of the substances adsorbed by hide-powder. Since all tests for genuine tannin gave negative results with maté, and also since hide-powder was known to adsorb both colouring matter and non-volatile organic acids, he considered the adsorption in this case to be due to the presence of one or both of these constituents in the leaves. The green colour given by alkalis with aqueous maté extracts was similar in many respects to the bluish-green colour given by coffee extracts under identical conditions. This reaction was commonly attributed to viridic acid, but he thought that this conclusion was a little too hasty. Viridic acid was a substance of unknown constitution, since, according to Vlaaderen and Nhulder (*J. prakt. Chem.*, 1858, **67**, 261), there were at least six different kinds of viridic acid in coffee beans. His own observations led him to regard this interesting colour reaction as an indication of the presence of caffetannin or some closely allied compound.

The Hortvet Freezing-point Process for the Examination of Milk: Correction Factors and the Influence of Stirring: I

By J. R. STUBBS, M.Sc., F.I.C.

(*Read at the Meeting of the North of England Section, December* 8, 1934)

Introduction

Hortvet, with his collaborators under the scheme adopted by the American Association of Agricultural Chemists, began work in the year 1917 on the problem of evolving an apparatus and a technique for the determination of the freezing-point of milk. He aimed at producing a cryoscope which would be convenient to use and by which results could be rapidly obtained with sufficient accuracy for the purpose of examining milk. Other workers, notably Raoult and Monier-Williams, had described forms of apparatus by which accurate results could be obtained, but their cryoscopes were elaborate and difficult to manipulate, and a considerable expenditure of time was necessary to carry out an observation, thus rendering them quite unsuitable for routine work. The position cannot be better described than in the words of Monier-Williams[1]:—"The use of such comparatively complicated apparatus as that shown on page 8 is quite out of the question except for purposes of research"; and again, "The apparatus that I used was too elaborate for routine work."[2]

Hortvet, on the other hand, remarks, "There does not appear to be any fairly defined uniformity in respect to design and construction of apparatus. . . . In order to satisfy practical requirements an attempt has been made to unify the conditions which have been outlined by giving attention chiefly, first, to the construction of a suitable cryoscope and thermometer; and second, to the method of manipulation. The cryoscope . . . has been designed to serve the purposes under discussion; . . . the application of correction factors may, for all practical purposes, be avoided by means of a carefully standardised method of procedure."[3]

This standardised form of instrument and technique was completed in the year 1921, accepted by the A.O.A.C., and became an official method.[4] It has since been fairly extensively used in this country, and in the year 1933 the Council of the Society of Public Analysts recommended that it should be adopted for the examination of milk.[5] It is interesting to observe, in passing, that both Raoult and Monier-Williams felt the need for a less elaborate form of apparatus which would serve for ordinary purposes, and each produced a simplified design of cryoscope.

Experience seems to show that by the use of the Hortvet apparatus reliable and concordant results can be obtained with a minimum expenditure of time and trouble, and, provided that the technique laid down by the regulations is followed, the results of one observer are comparable with those obtained by others. After a somewhat extensive use of the cryoscope for several years I have no hesitation in saying that the process is as rapid as any other suggested process; indeed, it is probably the speediest of all; it is economical, requiring very little ether; it is much more convenient and agreeable to work than processes in which mixtures of

ice and salt are employed as the cooling agent; it is under strict control, and it is possible and usual to obtain concordant results in duplicate experiments both by the same observer and also by different observers; and, finally, the degree of accuracy is more than sufficient for the practical purposes for which the examination of milk is undertaken.

But the process has been criticised in some quarters, particularly by Monier-Williams, who has raised objections both to the type of stirrer and to the method of stirring. Thus there are raised the questions whether the results obtained by the use of the Hortvet process will be affected materially by varying rates of manipulation of the Hortvet stirrer, and whether the use of a more efficient stirrer will cause differences in the results; if so, what will be the extent of the variations? The work described in the first part of this paper was undertaken with the object of answering these questions.

It has also been stated, both in the literature and during discussions I have had with other workers, that the Hortvet apparatus and technique do not give a figure from which there can be readily ascertained the "true" freezing-point of milk, *i.e.* the temperature at which milk and ice are in equilibrium with each other, so that the quantity of ice remains constant in amount; that it is difficult, if not impossible, with the standard Hortvet apparatus to arrive at the corrections which should be applied to the observed or apparent freezing-point, on account of supercooling and of heat transference while the temperature is rising after crystallisation begins. These points are dealt with in the second part of this paper.

The apparatus used in all this work was the modified Hortvet cryoscope, referred to in a previous paper,[6] and explained in the second part of the present paper. For the experiments, recorded in the first part of the paper, to ascertain the influence of stirring, the alcohol was retained, as is the case in the usual method of working with the standard Hortvet cryoscope. Unless otherwise stated, stirring was in all cases effected by means of an electric motor causing the stirrer to work at the rate of 40 complete strokes (each stroke consisting of an up-and-down movement) in a minute.

The thermometer used throughout the first series of experiments recorded in the first and second parts of the paper was an American one, which still continues to be very satisfactory after being in use for about three years. It is the one whose good performance is referred to in the paper mentioned above and, as there stated, its indications of the freezing-point of water appear to be nearly independent of any influence, excepting variations in the barometric pressure. The readings were taken with a small hand-lens by which the graduations are in focus when the distance between the lens and the thermometer is about 44 mm. from the thermometer.

The bulk of the work was repeated after a lapse of about three months, when the readings of the thermometer were observed by means of the telescope of a cathetometer. The instrument was clamped to a heavy table, which in turn was screwed to the floor. Two spirit levels were attached to the frame and one to the telescope for the purpose of adjustment. The distance between the end of the telescope and the thermometer was twenty-two inches. The thermometer was adjusted to the vertical position by two plumb lines attached to a movable arm

by means of which they could be removed when not required. After a reading had been taken the position of the thermometer was checked, to ascertain if tapping had displaced it from the vertical position.

The time required to make a reading with this levelled telescope is very little, if any, longer than that needed when a hand-lens is used. While the cooling of the milk is taking place the instrument and telescope are levelled and the cross-wires of the telescope brought to bear on the thermometer at about the expected position of the mercury at the freezing-point.

Three readings were usually taken with both the telescope and the hand-lens, and it was found that there was no appreciable difference between readings of the same position of the mercury by the two methods. Experience showed that, after adequate practice, the hand lens gave readings sufficiently accurate for all practical purposes.

Another thermometer, also of American manufacture, which has proved very satisfactory for some months, was used for the purpose of repeating the work.

The zero-point of the thermometer was checked frequently, at least once each working day.

The samples of milk used in all the following work were of good quality as judged by the chemical analysis, and their smell and acidity indicated that they were quite fresh. The temperature of the cooling-bath was maintained at $-3{\cdot}0°$ C. throughout all the experiments on milk, except in one instance mentioned later; in the case of water, a temperature of $-2{\cdot}5°$ C. was used.

PART I

Investigation of the Effect of Different Methods of Stirring

(1) *Comparison of the use of the Hortvet stirrer with that of a flat and more efficient one.*

The Hortvet stirrer is described in the *Methods of Analysis of the A.O.A.C.* (3rd Edition, p. 220) as composed of non-corrodible low-conductivity metal . . . the lower end provided with a horizontal loop. It is essentially a length of wire having a loop at right angles at one end, whilst the other end is provided with a handle of insulating material.

This stirrer is to be used with a steady up-and-down motion at the rate of approximately one stroke in one or two seconds. When the desired amount of super-cooling has been obtained, freezing is induced and the thermometer indicates a rise of temperature. The instructions are that, when the freezing-point of water is being ascertained, the stirrer should be manipulated slowly and carefully three or four times as the mercury approaches its highest point. In the case of milk, the stirrer is to be similarly used two or three times at the corresponding point. In effect, then, when the instructions are followed the stirrer is at rest from the moment when freezing begins until the column of mercury "approaches" its highest point.

It will be obvious that, within reasonable limits, it can be of little consequence what rate of stirring is adopted while cooling is taking place, provided that it is effective in mixing the liquid in the freezing-tube so that the temperature indicated

by the thermometer is the same as that of the different parts of the liquid; it might be expected that an increase in the rate of stirring would have the effect of reducing the time needed for cooling, in spite of the slight amount of extra heat generated by the more vigorous agitation, and that a reduction in that rate would produce the opposite effect.

It would appear that, in the Hortvet process, the question of stirring, while the temperature is rising after freezing has begun, might assume greater significance, for the final temperature attained is the resultant of at least two opposing effects, *viz.* the abstraction of heat from the freezing liquid by the cooling-bath and the addition of heat due to the latent heat of formation of ice. Vigorous stirring might be expected, by exposing fresh layers of the liquid under examination to the walls of the freezing-vessel, themselves cooled by the cooling-bath, to promote extraction of heat while, at the same time, increasing the rate at which the liquid is heated up by bringing the particles of ice more quickly into contact with the different parts of the liquid.

It must be admitted that the procedure as regards the use of the stirrer in the Hortvet technique during the period now under consideration stands alone and in distinct contrast with that adopted by most prominent observers. Raoult, when examining dilute solutions, used a large platinum stirrer, fitted with two helical vanes, which was fixed over the bulb of the thermometer, and both stirrer and thermometer were rotated together at the rate of 300 revolutions per minute. Monier-Williams, in the experiments recorded in his work published in the year 1914, employed a glass spiral stirrer rotating at a speed of 1300 revolutions per minute, while in his later experiments in 1933 he used an up-and-down flat or disc stirrer manipulated at the rate of one up-and-down stroke every two or three seconds.

From a consideration of the above it will be quite clear that no claim can be put forward that, in the Hortvet process, the mixing together of the ice and the liquid will bear comparison, as regards thoroughness and speed, with that adopted in the technique of Raoult and of Monier-Williams. This point, indeed, would appear so obvious that it can hardly have escaped the notice of Hortvet and his collaborators; it seems reasonable, on the other hand, to presume that vigorous stirring was intentionally avoided.

Monier-Williams definitely suggests that the stirring in the Hortvet method of procedure is "weak," and in his later design of apparatus he himself uses, as mentioned above, a flat stirrer which may be described as a disc with a central hole for the thermometer and two smaller holes, in place of the ring of wire used by Hortvet. Moreover, Monier-Williams directs that this more efficient stirrer should be operated at the rate of one stroke (1 up-and-down motion) in two or three seconds all the time the temperature is rising until the tapping of the thermometer is carried out. So that Monier-Williams not only uses a more efficient stirrer, but also uses it continuously.

For the following experiments there was employed a stirrer, made of thin sheet brass, fastened to one end of the stem of a Hortvet stirrer in place of the ring of wire. The metal disc was 0·7 mm. in thickness and 29·5 mm. in diameter, with a central hole 12·5 mm. in diameter and two small holes, each 4 mm. in diameter,

opposite to each other. When this was substituted for the ordinary Hortvet stirrer it was quite obvious, in manipulating it, that a much more vigorous and thorough stirring was obtained; this was proved by the effort needed to work it, and also by the appearance of the milk, which was agitated to such an extent that it was inclined to froth.

In the experiments, the results of which are given in Table I, a comparison was made between the effect of the use of the Hortvet stirrer and the disc stirrer described above. The same portion of milk was used for an experiment with both types of stirrer; in the first experiment the Hortvet stirrer was used first, followed by the flat one; in the second, this order of use was reversed, and so on alternately. Supercooling was carried out to approximately the same extent in every case, and never differed by more than 0·02° for the experiments on the same milk.

In those cases where the Hortvet technique was used no stirring at all was done while the mercury was rising, until it was approaching the highest point, when three up-and-down strokes were slowly made just previous to tapping. Where the flat or disc stirrer was employed, stirring was carried out all the time the temperature was rising from the lowest to the highest point, and at the rate of one complete (1 up- and 1 down-movement) in 2 seconds. This manipulation was done by hand and carefully timed.

The cooling of the milk in the freezing-vessel was much more rapid when the disc stirrer was used than when the Hortvet stirrer was employed. It was found also that when the flat stirrer was used, spontaneous freezing, *i.e.* freezing without the introduction of a particle of ice, took place rather frequently, rendering, sometimes, several attempts necessary before the desired amount of supercooling could be obtained. This never happened in those instances where the Hortvet stirrer was used. The freezing-point of water was taken with each of the two methods of stirring, and identical readings were obtained.

The results of the experiments are given in Table I.

TABLE I

Effect of different methods of stirring

No. of sample	Hortvet stirrer Δ*	Flat stirrer Δ
Reading by hand-lens		
20863 L.B.D.	0·552	0·552
20867 L.B.D.	0·553	0·553
83583 Ws.D.	0·541	0·541
83589 Ws.D.	0·548	0·546
93438 Wgn.D.	0·533	0·533
53477 R.D.	0·541	0·541
Reading by levelled telescope		
53562 R.D.	0·546	0·548
83665 Ws.D.	0·550	0·551
83668 Ws.D.	0·550	0·548
76548 S.D.	0·538	0·538
76552 S.D.	0·548	0·548
76554 S.D.	0·539	0·539

* Δ is the depression of the freezing-point of milk below that of water, the reading of the thermometer being corrected for zero-point and irregularities of bore. It is of the same numerical value as the freezing-point, but opposite in sign.

It will be seen from the above table that in eight cases identical results were obtained by the use of the two different stirrers, and that in the remaining cases small variations of not more than 0·002° were obtained.

This result was unexpected. The explanation may be that the flat, efficient stirrer used as described, distributes quickly through the liquid the heat liberated on the formation of ice, but, at the same time, the thorough stirring of the liquid increases the rate at which heat is abstracted from the contents of the freezing-tube. On the other hand, when the Hortvet stirrer and technique are used, the particles of ice and liquid are not so quickly and thoroughly brought into contact with each other, causing the heating-up to take place more slowly, but the comparatively quiescent state of the outer layers of liquid nearest the cooling-bath hinders the process of abstraction of heat, causing the resultant to be about the same in both cases. Another factor, the accession of heat from the atmosphere, is probably not very different, whichever method of stirring is employed, and contributes its quota to both in much the same amount.

There is one distinct difference which was invariably observed in the experiments described above. In the cases where the Hortvet stirrer was employed, the time occupied by the mercury of the thermometer in rising from the lowest to nearly the highest point was definitely greater than the corresponding period when the efficient stirrer was used. This time, measured by means of a stop-watch from the onset of freezing until tapping was carried out, was, in the case of the Hortvet technique, about 90 seconds, and, with the flat stirrer, about 30 seconds; that is, about three times as long in the one case as in the other. The obvious explanation of this would appear to be that vigorous stirring, by exposing quickly fresh surfaces of ice and liquid to each other, increases the rate of heating, while the comparative slowness with which mixture must take place where there is practically no stirring has the opposite effect.

Monier-Williams[6] suggests that "in the Hortvet apparatus steadiness of temperature at the freezing-point is secured by a fortunate oversight in the matter of stirring." But may it not be that Hortvet has made an attempt, more or less successful, if the above experiments are taken as an indication, to diminish the action of the cooling-bath during the time freezing is in operation without the removal of the alcohol from the jacket surrounding the freezing-tube, and so to arrange matters that the mercury column remains constant at the observed freezing-point sufficiently long to enable several tappings and readings to be made? It is suggested that he may have intentionally avoided energetic stirring, in order to secure a longer period of rest of the mercury than would otherwise have been the case. There can be no doubt that he aspired to produce an apparatus which would be capable of giving results with a minimum of manipulation, and one which would, therefore, be suitable for routine use.

If, in the process, the alcohol need not be removed and a satisfactory result can still be obtained for the freezing-point, it is a very definite recommendation for the adoption of non-removal in a routine process. The reduction in the amount of manipulation and the saving of time, consequent on the non-removal of alcohol, will be acknowledged, it is confidently thought, by all who have had experience of the two methods when undertaking the examination of a number of milks.

There was nothing in these experiments to warrant any suggestion that the use of a flat efficient stirrer would be preferable to that of the standard Hortvet pattern used according to the Hortvet technique with, possibly, the slight variation suggested below.

(2) *The Hortvet stirrer—comparison between its use as prescribed in the A.O.A.C. regulations and its continuous use.*

In this series of experiments I employed the Hortvet stirrer throughout, comparing the effects of using it as prescribed in the A.O.A.C. regulations and also continuously, while warming-up by means of the latent heat of formation of ice was in progress. It has been shown that vigorous stirring and practically no stirring gave the same, or nearly the same, readings for the freezing-point but, since two different types of stirrers were used, there still remained the question if, when using the Hortvet stirrer, different rates of manipulation would affect the results.

There appeared, too, to be another reason which would justify the carrying out of this comparison. It was noticed during the work described in the previous section that the rate at which the mercury "approached" the highest point, as judged by the use of a hand-lens, after freezing had been induced, was at first rapid and then gradually became slower and slower, especially when the Hortvet technique was being followed, and that the last 0·035° in that case took about 45 seconds, compared with the whole time of rising of about 90 seconds—that is, half the period was taken by the last 0·035°. The watching for the highest point reached is, therefore, apt to become tedious, and some considerable practice is desirable because the column rises so slowly. It might be an advantage if this time could be shortened, which, it was thought, might possibly happen if stirring were resorted to during the time the temperature was rising. It was therefore decided to carry out the work described below, comparing the effects of continuous stirring with those of the procedure laid down in the Hortvet technique.

Both varieties of stirring were practised on the same quantity of milk, the sequence of the two methods being alternated in consecutive experiments. The supercooling was practically the same in every case; the greatest difference in the experiments on any one milk did not exceed 0·02°. Table II gives the results. The freezing-point of water was taken with the two methods of stirring, and identical readings were obtained.

It will be seen from this table (p. 154) that the results are in close agreement, and in no case differ by more than 0·002°, an amount so small that it cannot be regarded as significant.

There was observed, however, as in the case where two different types of stirrers were used, a difference in the time required by the mercury column to rise, after freezing had begun, to nearly the highest point, when tapping was needed. With the Hortvet method this period is about 90 seconds, and, with continuous stirring, roughly 60 seconds, and, for the last 0·035° or so, about 45 and 21 seconds respectively. Although the difference between the two latter figures is only 24 seconds, it is very noticeable in working, and the shorter time is a distinct advantage in judging the attainment of the nearly stationary position. It might,

TABLE II

Comparison of the effect of different methods of using the Hortvet stirrer

No. of sample	Hortvet's method Δ	Continuous stirring Δ
Reading by hand-lens		
53479 R.D.	0·535	0·536
25321 H.B.D.	0·535	0·537
960 Westhoughton	0·542	0·542
970 ,,	0·547	0·547
973 ,,	0·536	0·537
30669 C.D.	0·550	0·551
Reading by levelled telescope		
76547 S.D.	0·540	0·542
76549 S.D.	0·539	0·539
76550 S.D.	0·539	0·537
25428 H.B.D.	0·543	0·542
25430 H.B.D.	0·532	0·532
25433 H.B.D.	0·541	0·539

therefore, be worthy of consideration whether the Hortvet technique should be altered so as to permit of a certain amount of stirring during the ascent of the mercury column.

REFERENCES

1. G. W. Monier-Williams, *Food Reports*, 1914, No. 22, p. 22.
2. G. W. Monier-Williams, ANALYST, 1933, **58**, 254.
3. J. Hortvet, *J. Ind. Eng. Chem.*, 1921, **13**, 200.
4. *Journal of the Association of Official Agricultural Chemists*, 1922–3, **6**, 264.
5. ANALYST, 1933, **58**, 318.
6. ANALYST, 1934, **59**, 592.
7. G. W. Monier-Williams, ANALYST, 1933, **58**, 260.

PARTS II AND III TO FOLLOW.

Tests for Elements in Organic Compounds

BY H. MIDDLETON, M.Sc., A.I.C.

SINCE the use of an alkali metal (Laissaigne's method), or of Castellana's mixture of magnesium powder and anhydrous potassium carbonate, is attended with certain disadvantages and some danger, the following substitutes are proposed:

I. *An intimate mixture of pure anhydrous sodium carbonate and one-tenth of its weight of pure sucrose.*

An ignition tube, about 2¼″ long, made from ordinary glass tubing, ¼″ outside diameter, with a bulb slightly wider than the stem, is suitable for the test.

If the organic substance is a solid, an amount roughly twice the bulk of a rice grain is mixed with about 5 times its bulk of the reagent; the mixture is then introduced into the bulb of the tube and the reagent is added until, after tapping down, there is a column about 1″ long above the bulb.

For a liquid, two or three drops are introduced into the bulb, and the reagent is added until, after tapping down, the column extends to within about $\frac{1}{2}''$ of the mouth of the tube.

The tube is now held horizontally and heated in a flame about $2\frac{1}{2}''$ high, at first just above the mixture, in order to prevent movement of the latter along the tube. The column of mixture is then gradually heated to redness, the tube being turned over periodically to prevent undue bending. When a portion of the column equal in width to the flame has been heated, the tube is held at an angle, so that while this portion still remains in the flame the heating is gradually extended until the bulb as well as the stem is in the flame.

The idea is to heat the organic substance very gradually by bringing it nearer and nearer to the flame, any vapours evolved passing through the red-hot column of mixture.

Finally, the whole tube is heated to redness in a large flame for a minute or more and then plunged into about 10 ml. of distilled water in a porcelain dish. The contents of the dish are then heated to boiling, and filtered.

As when sodium is employed, halogens are converted into sodium halides, sulphur into sodium sulphide, and nitrogen (in a limited number of cases) into sodium cyanide.

The proportion of sugar employed does not cause excessive fumes when the mixture is heated, yet provides sufficient carbon for the reduction of sulphates, etc., to sulphide. The final heating to redness is essential, in order to cause any such reduction, to decompose the sugar thoroughly, and to remove any deposit from the mouth of the tube, so that a perfectly colourless filtrate is obtained.

Tests for sulphur, nitrogen, and halogens are carried out with the alkaline filtrate as in Laissaigne's method, except that in the nitrogen test a few drops of aqueous sodium hydroxide solution are added before the ferrous sulphate.

RESULTS WITH THE ALKALI-SUGAR TEST.—(*a*) *The test for sulphur.*—A brown or black precipitate on the addition of one drop of lead acetate solution to about 1 ml. of the alkaline filtrate has been obtained with 50 compounds, representing the following classes:

Sulphates of bases, alkyl sulphates, bisulphite compounds, thioureas, thiocyanates and isothiocyanates, sulphones, sulphonic acids and their salts, chlorides, esters, amides and substituted amides, derivatives of phenthiazine and thioindigo.

(*b*) *The test for nitrogen.*—A deep blue solution or precipitate was obtained with only a limited number of compounds, which included the following:

p-Nitrotoluene, *m*-dinitrobenzene, 2 : 4-dinitrotoluene, trinitrotoluene, *o*-nitrochlorbenzene, 2 : 4-dinitrochlorbenzene, uric acid, caffeine, theobromine, theophyllin, barbitone, phenylbarbitone, urea and its nitrate, hydrochloride, and oxalate, ethyl urethane, oxamide, succinamide, chloralformamide, thiourea, glycine, hippuric acid, hexamine, ethyl cyanacetate, acetone semicarbazone, antipyrine and amidopyrine.

With a number of other compounds a green solution was obtained, which, since a blank test with the mixture gave a solution of pure yellow colour (*i.e.* the colour of ferric chloride acidified with hydrochloric acid), is a sufficient indication of the presence of nitrogen in the organic compound.

If a negative or doubtful result is obtained, the alkali-zinc test (II) must be tried.

With the exception of thiourea, poor or negative results were obtained with substances which contain sulphur in addition to nitrogen.

Negative results were usually obtained with aromatic bases, and their salts and derivatives.

If a negative result is obtained (although the subsequent alkali-zinc test proves the presence of nitrogen), there is the compensating advantage that the alkaline filtrate to be tested for halogens contains no cyanide.

(*c*) *The tests for halogens.*—The usual tests with the alkaline filtrate have been carried out, with highly successful results, with 40 chloro-compounds, 15 bromo-compounds, and 6 iodo-compounds, exclusive of salts of bases with the halogen acids.

II. *An intimate mixture of zinc dust and half its weight of anhydrous sodium carbonate* (*ordinary commercial products*).

This mixture is employed only if a negative result for nitrogen has been obtained in the alkali-sugar test.

The test is carried out in exactly the same way as when the alkali-sugar mixture is used. The gradual heating is particularly necessary in this case, as otherwise negative results may be obtained with salts and derivatives of aromatic bases.

Nitrogen only is tested for in the alkaline filtrate.

RESULTS WITH THE ALKALI-ZINC TEST FOR NITROGEN.—The results obtained with 220 compounds varied from dense blue precipitates to blue-green solutions, yielding a blue residue after boiling (to coagulate the precipitate), cooling and filtering.

The substances tested represent the following classes:—Ammonium salts, amides, urea and its salts, aryl substituted ureas, urethanes, ureides, purine group, nitriles, thiocyanates and isothiocyanates, compounds of aldehydes with ammonia, oximes, phenylhydrazones and semicarbazones, esters of nitric and nitrous acids, amino-compounds and their salts, acetyl, benzoyl, benzylidene and *p*-toluene sulphonyl derivatives, aromatic and heterocyclic secondary and tertiary bases, nitro-, azo-, and diazo-amino compounds, picrates of organic compounds, simple and substituted sulphonamides, alkaloids, derivatives of pyrazolone, indigo and indanthrene.

The alkali-zinc mixture, with constituents of A.R. quality, may also be used for sulphur and halogens, provided that the tests for chlorine and sulphur are made in comparison with a blank test.

The test for sulphur is carried out as follows:—After the hot tube has been plunged into water, the mixture is heated to boiling and allowed to settle, and the liquid is then decanted through a filter. To the residue in the dish (containing insoluble zinc sulphide if sulphur is present in the organic compound) are added about 10 ml. of dilute hydrochloric acid, and a filter paper (upon the centre of which a drop of sodium plumbite solution has been poured) is immediately placed over the dish.

If sulphur is present in the organic compound, a dark brown stain, visible on the upper surface of the paper, will be formed.

Blank tests gave only a slight brown stain, visible only on the under surface of the paper.

The alkali-sugar test appears preferable, however, since the constituents of the mixture can be obtained quite free from chlorine and sulphur; moreover, the test for sulphur is simpler, and involves less time.

ADVANTAGES OF THE ALKALI-SUGAR AND ALKALI-ZINC MIXTURES.—(i) The mixtures are readily prepared (it is not necessary to dry the sodium carbonate) and convenient to handle, and may be kept without any special precautions.

(ii) The tests are applicable without modification to very volatile substances, *e.g.* ethyl bromide (b.p. 38° C.).

(iii) Except in the case of picric acid and picrates of organic compounds the reactions are perfectly quiet, even with nitrates and polynitro compounds.

With picric acid and picrates, the mixture, or a portion of it, is harmlessly ejected from the tube, this behaviour affording an indication of this class of compound.

In the event of the tube bursting there is no startling flash, as is the case with the magnesium mixture.

(iv) The plunging of the hot tube into water is attended with less danger, or with less startling results, than when an alkali-metal or the magnesium mixture is used.

(v) No cyanide is formed when either mixture is heated alone. When the magnesium mixture in the recommended proportions of 1 part of magnesium to 2 parts of potassium carbonate is strongly heated alone, under the conditions essential for carrying out the test with a very volatile substance, *i.e.* in an ignition tube, sufficient cyanide is formed to give a dense blue precipitate when the nitrogen test is applied. Apparently, atmospheric nitrogen plays a part in the reactions.

When the proportions of 3 parts of magnesium to 1 part of potassium carbonate are employed, however, only a slightly green solution is obtained.

(vi) The alkali-zinc test has the great advantage over all other methods, that the test for nitrogen is not interfered with by the presence of sulphur in the organic compound.

On applying the test to 30 substances containing sulphur in addition to nitrogen, no sulphide or thiocyanate was detected in the alkaline filtrate containing the cyanide.

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The Specific Gravity of Fatty Oils Shipped in Bulk

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In cases where the specific gravity or density of an oil is required to be determined with great accuracy at a given temperature, the measurement must obviously be made at that temperature. In practice, however, it is customary to make the determination at some neighbouring temperature and to calculate the required specific gravity or density from the figure so obtained by the use of a correction. The usual correction is derived from the experimentally-determined fact that an alteration of 1° C. in temperature causes a change in specific gravity of 0·00069 in the case of most, if not all, liquid oils.

This method has certain practical advantages, and works very well so long as it is applied only to liquid oils. Unfortunately it has also been the custom to proceed in a similar manner with semi-solid fats and oils which contain stearine at normal temperatures; the determinations are made at a raised temperature with the oil completely liquid, and "specific gravity" at 15·5° C., or other lower temperature, is calculated by means of the above correction. This leads to gross inaccuracies, as the correction is valid only for liquid oils and becomes modified as soon as stearine crystallises and solidification begins. The true correction varies and depends on the proportion and nature of the stearine present.

We have obtained the following values for a few of the corrections to illustrate this:

Sardine oil	0·00080 to 0·00105
Pilchard oil	0·00090 (approx.)
Palm oil	0·00140
Crude sperm oil	0·00187

It will be seen that these figures vary up to nearly three times the conventional figure. In a recent case which came to our notice the specific gravity of an oil had been returned as 0·880 instead of the correct figure of 0·894, solely because the wrong correction had been employed.

A further difficulty arises because stearine is much denser in the solid than in the liquid form and crystallises slowly from oils, with the result that supercooling without crystallisation may occur. It is quite possible to have an oil at a given temperature in a liquid state or in varying degrees of solidification. The specific gravity of each is quite different, as is exemplified in the following table, all the figures in which were obtained on a normal palm oil at 32·5° C.

Specific gravity of a palm oil at 32·5° C./15·5° C.

Condition of oil	Observed specific gravity
Liquid	0·9006
Some stearine present	0·9025
Semi-solid	0·9050
Solid	0·9084

The shipment of very large tonnages of oil in bulk is becoming more and more popular, and many problems have arisen in connection therewith. Among these, perhaps, the most important concern the determination of the weight of oil in a ship's tank or in the shore-tank into which it is discharged.

This weight is obtained by calculation from the volume occupied by the oil in the tank at a known temperature, and an accurate specific gravity or density of the oil determined at the same temperature. It is necessary also to know the condition of the oil (*e.g.* partly solid and partly liquid) when its volume is measured.

As has been previously pointed out (E. R. Bolton, "Oils, Fats and Fatty Foods," 1928, p. 33, and A. Torisawa, "Bulk Oil," 1929, p. 47), the factor required for converting the volumes to weights depends on the units in which they are expressed. In Great Britain it is based on the statutory relations between gallons and pounds, and for practical purposes is the ordinary *specific gravity*, in which the weight of a given volume obtained at the required temperature in air is compared with the weight of an equal volume of water obtained at 15·5° C. in air. On the Continent the weight is required in kilograms, although it may be subsequently converted into tons by a conventional factor (1 ton = 1016 kg.). The volume is measured in litres, and the factor required is a *density—viz.* the weight (in air) in kilograms of one litre of oil measured at the required temperature. The reference standard is the weight *in vacuo* of a standard volume of water measured at 4° C.

The difference between the specific gravity and the density figures for a given oil amounts to about 0·2 per cent.

Five thousand tons of palm oil is a quantity commonly dealt with in a bulk shipment, and with such a quantity the difference between the extreme figures shown in the above table would involve an error of about 43 tons in the apparent out-turn; and with palm oil at £17 per ton this would involve a sum of £731.

In most cases the specific gravity or density required is that of the oil when it contains the maximum amount of stearine (*i.e.* during cold weather). In order to obtain this figure it is most satisfactory to fill the specific gravity bottle with oil in a liquid condition, cool it to a temperature some 5 to 8 degrees below that at which the determination is required, and keep it at that temperature until as much stearine as possible has separated (usually this requires 24 hours). Precautions should be taken to see that no air is sucked into the mass of oil during crystallisation. After allowing it to stand in this manner, the stopper is inserted and the bottle and its contents are warmed to the required temperature in a thermostat until expansion ceases; the stopper is then wiped and the outside of the bottle cleaned as usual; the weight of the bottle and contents is obtained after it has attained the room temperature.

This method has now been adopted by many leading authorities, and is not claimed to have any novel features; but we feel that it is desirable that it should receive wide attention, as disputes continually arise about specific gravities and densities, and we find, almost always, that they are due to a lack of understanding of the facts we have mentioned above.

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Preliminary Notes on the Sterol Iodine Values of Oils and Fats by the Bolton and Williams Method

By A. C. BOSE, Ph.D., A.I.C.

In the present investigation an attempt has been made to determine the sterol iodine values (S.I.V.) of various oils and fats of authentic origin by the method of Bolton and Williams. In addition to samples of the oils and fats of known purity included in each of the four groups by Bolton and Williams in Table I[1], a few samples of other oils in common use in India have also been analysed.

The objects of the investigation were to corroborate for Indian oils the Bolton and Williams values, and to ascertain whether the method can be used for detecting and estimating the degree of adulteration of oils and fats in one group with those of the other groups, with special reference to olive oil.

Although the method of Bolton and Williams affords the means of classifying oils and fats of both animal and vegetable origin, yet I have found that with some Indian samples the sterol iodine values do not fall exactly within any of the four specified groups, but occupy intermediate positions between them. (*Cf.* Table I.)

In the process of extraction of oils from different varieties of mustard, rape and similar oil-seeds, niger-seed (surgônja) is generally added as an adulterant, since, being harder, it helps to increase the yield of the extracted oil. It is thus obvious that, since the sterol iodine value of niger-seed oil is fairly close to those of the mustard oils (*cf.* Tables I and II), it would be difficult to employ the Bolton and Williams method to detect or estimate the percentage of adulteration in them. It is also evident from Table II that there is no correlation between the sterol iodine value and the butyro-refractometer reading or saponification value of oils and fats.

The usual adulterants of ghee are hydrogenated vegetable oils, coconut oil, rapeseed oil, and almond and similar edible oils. During the winter in tropical countries ghee can be readily adulterated with coconut oil, which solidifies with the ghee on cooling, whereas other edible oils which do not solidify can be used as adulterants only during the summer.

The estimation, or even detection, of such adulterants as occur in Group I by this method is obviously impossible, because the sterol iodine values of such mixtures will be of the same order after adulteration of the ghee, in any proportion, with any of the oils of the first group.

An authentic sample of "vanashpati," which consists of a mixture of hydrogenated vegetable oils, was analysed. It had the appearance of ghee, and gave a sterol iodine value of 93·5, which is characteristic of oils in Group II. A mixture in equal proportions of "vanashpati"* and ghee was also analysed. The analytical results show that easy detection and approximate estimation of adulterants that occur in groups other than the first may be effected, provided that the sterol iodine value of the actual adulterant is known. (*Cf.* Table III and the example.)

A sample of cow-ghee prepared in the laboratory was separated, by ordinary

*An Indian name for vegetable ghee.—Editor.

filtration, into a solid and a liquid fraction at a temperature of about 30° C., and the physical properties of the two fractions were examined. The liquid fraction gave higher values for all the constants determined.

The sterol iodine value of cow-ghee, on storage, appears to diminish, whilst the saponification value remains nearly the same. (*Cf.* Table IV.) Judging by the results given by two samples, there appears to be a relationship between the sterol iodine value and the Reichert–Wollny value; this will be investigated later.

It will be seen from the results in Table IV that the sterol iodine values for old ghee range from 52 to 58. If this range for cow-ghee, or for any other oils or fats, can be confirmed on a large number of samples, it is suggested that such fats should be placed in Group "Zero" with reference to olive oil, so as to take their place in Bolton and Williams Table I.

Detailed work on the sterol iodine values of cow-ghee and buffalo-ghee is in progress.

EXPERIMENTAL.—Two quantities (2·5 g. each) of the oil or fat were hydrolysed for an hour in roomy conical flasks with 25 to 30 ml. of $N/2$ alcoholic potash. The alcohol used for this solution was purified by treatment with silver nitrate and potassium hydroxide and re-distillation; the alcoholic potash made with it remained brilliant and did not develop a yellow tint. After neutralisation (determination of the saponification value), the contents of the flasks were transferred to two separators, and each solution was washed with 50 ml. of water, and then extracted with three successive portions of 50 ml. of ether. The ethereal extracts were washed, gently at first and then vigorously, three times with 20 ml. of water, then three times with $N/2$ potassium hydroxide solution, and again with water in 20-ml. portions until free from alkali. The washed extracts were transferred to small, weighed conical flasks, and the ether was distilled off completely. The residues were treated with 2 to 3 ml. of acetone, which was then evaporated at as low a temperature as practicable, the last traces being drawn off by means of a current of dry air, and the flasks with the residues were dried in an air-oven at 80° C. until constant in weight (about 30 minutes). The unsaponifiable matter was dissolved in 5 ml. of chloroform, the solution was treated with 20 ml. of $N/10$ pyridine sulphate bromide solution and left for exactly 5 minutes, after which it was treated with 5 ml. of 10 per cent. potassium iodide solution and 40 ml. of water, and titrated with $N/20$ sodium thiosulphate solution.

A blank determination was made simultaneously, and the strength of the sodium thiosulphate solution was determined against $N/10$ potassium dichromate solution.

All the figures given in the following tables below have been verified by duplicate, and in some instances, four determinations.

TABLE I

Sample	Butyro-refracto-meter reading	Iodine value (Wijs)	Saponification value	Unsaponifiable matter Per Cent.	Sterol iodine value	Group
Poppy-seed oil	67·0	—	196·0	0·644	82·15	I–II
Niger-seed oil (surgônja)	63·0	132·1	190·4	0·964	109·30	II–III
Pila Sarso (yellow mustard) oil	58·6	101·8	172·5	0·960	130·97	III–IV

TABLE II

Sample	Butyro-refracto-meter reading	Iodine value	Saponification value	Unsaponifiable matter Per Cent.	Sterol iodine value	Group
Cocogem	34·6	—	255·0	0·276	70·2	I
Coconut oil	34·6	—	258·7	0·296	67·4	I
Vanashpati	51·0	—	188·7	0·460	93·8	II
Linseed oil	72·8	177·0	189·8	0·920	122·8	III
Tori-seed oil (mustard)	59·0	101·1	173·6	0·890	117·5	III
Mixed mustard oil ..	59·0	101·4	173·0	0·970	121·5	III
Arachis oil	55·5	93·8	187·0	0·520	117·3	III
Castor oil	68·6	—	180·3	0·490	125·0	III
Cod-liver oil*	68·5	—	184·8	0·940	100·4	II–III

* Obtained from a medical store.

TABLE III

Sample	Butyro-refracto-meter reading	Reichert–Wollny value	Saponification value	Unsaponifiable matter Per Cent.	Sterol iodine value	Group
Ghee (A)	40·5	38·2	235·8	0·42	63·2	I
Vanashpati (B)	51·0	0·2	188·7	0·46	93·5	II
(A+B) in 50 per cent. mixture	45·1	—	213·6	0·44	77·8	I–II
Theoretical value [mean of (A) and (B)]	45·8	19·2	212·3	0·44	78·35	I–II

Example: In a mixture of fats and oils, say of "B" with A, a is the per cent. of adulterant. If the S.I.V. of the adulterant, B, is known, a can be calculated by the equation,

$$\frac{B \times a}{100} + \frac{S(100-a)}{100} = F \quad .. \quad .. \quad .. \quad .. \quad (1),$$

where B represents the S.I.V. of the adulterant (which must be known); S, the standard S.I.V. for the sample obtained from the Table; and F, the S.I.V. of the mixture (found by actual determination). Then, using the actual data from Table III, we have B = 93·5 (Vanashpati); S = 63·2 (Ghee); F = 77·8 (Mixture A + B) in 50 per cent. Substituting the values in (I), we get

$$\frac{93{\cdot}5 \times a}{100} + \frac{63{\cdot}2(100-a)}{100} = 77{\cdot}8, \text{ or } a = 48{\cdot}2. \quad \text{Percentage error} = -1{\cdot}8.$$

Again, when the data are taken reversely, we get

$$\frac{63{\cdot}2 \times a}{100} + \frac{93{\cdot}5(100-a)}{100} = 77{\cdot}8, \text{ or } a = 51{\cdot}8. \quad \text{Percentage error} = +1{\cdot}8.$$

TABLE IV

Sample	Reichert-Wollny value	Butyro-refracto-meter reading	Saponification value	Unsaponifiable matter Per Cent.	Sterol iodine value	Group
Ghee A	38·2	40·5	235·8	0·43	63·2	<I
Ghee B (old)	35·1	40·5	234·1	0·49	52·05	<I
Cow-ghee (C) (old) ..	—	41·0	222·5	0·46	58·48	<I
New cow-ghee (C) (solid fraction)	—	42·4	220·6	0·39	60·8	<I
New cow-ghee (C) (liquid fraction)	—	42·7	223·4	0·43	65·4	I

SUMMARY.—The Bolton and Williams values for fats and oils have been corroborated, and those for some new samples of Indian origin have been added.

Adulteration of ghee with such fats and oils as occur in Group I cannot be detected or estimated, whilst adulteration with fats of groups other than the first can be easily detected and approximately estimated, provided that the S.I.V. of the adulterant is known.

I wish to thank the Director of Public Health, Bihar and Orissa, for his kind permission to carry out this investigation in his laboratory.

REFERENCES

1. E. R. Bolton and K. A. Williams, ANALYST, 1930, **55**, 5.
2. "Report of the Sub-Committee on the Determination of Unsaponifiable Matter in Oils and Fats and of Unsaponified Fat in Soaps, to the Standing Committee on Uniformity of Analytical Methods," ANALYST, 1933, **58**, 205.

The Determination of Small Amounts of Sulphur in Certain Organic Compounds

BY N. STRAFFORD, M.Sc., F.I.C., AND H. CROSSLEY, A.M.C.T.

THE determination of small amounts of sulphur in certain dyestuffs intermediates, *e.g.* nitrobenzene, proved to be a matter of some difficulty. As the accurate determination of amounts of sulphur of the order of 0·01 per cent. was called for, it was evidently necessary to provide a method for the decomposition of a considerable quantity of the material under examination, with quantitative recovery of the sulphur.

The possibility of employing "wet" methods, such as oxidation with bromine and nitric acid or with nitric and perchloric acids, or reduction with sodium and amyl alcohol, was explored, but without success.

Attention was next directed to combustion methods. Existing methods for the determination of sulphur in organic liquids by combustion may be classified broadly under (i) the "lamp" method in which the liquid is burned directly from a wick*, and (ii) combustion of the vaporised material in a heated tube. In

* *Cf.* "Standard Methods for Testing Tar and its Products," 1929: Published by Standardisation of Tar Products Test Committee, 166, Piccadilly, London, W.1. Determination of sulphur in light benzol, serial LB 11, p. 90.

methods of the latter type the liquid may be vaporised (*a*) from a boat contained in the combustion tube,[1] or (*b*) by passing a current of air through the liquid contained in an auxiliary tube which may be suitably heated.[2]

The lamp method is not directly applicable to liquids such as nitrobenzene, for, even after dilution of the liquid with five times its volume of alcohol, combustion is not complete, the flame being very smoky.

The existing methods of combustion in a heated tube were found to be inadequate for the determination of traces of sulphur in liquids such as nitrobenzene. Volatilisation of the liquid from a boat in the combustion tube suffers from the defect that only relatively small amounts of the substance can conveniently be handled, and it is, moreover, very difficult to maintain conditions giving regular and complete combustion. Volatilisation from an auxiliary tube, which has been applied with success to a volatile liquid such as benzene[2] fails completely with liquids of higher boiling-point. A modification of the method in which vaporisation was effected by passing air through the nitrobenzene, maintained at or near its boiling-point, gave unsatisfactory results.

Attention was therefore directed to devising an apparatus for the production, at a steady rate, of a correctly proportioned mixture of substance and air. The apparatus ultimately evolved consists essentially of a spraying apparatus of a well-known type in which the spray is produced by a jet of air blown across the orifice of a narrow tube dipping into the liquid. The spray is carried by the air-current into the combustion tube.

In the preliminary experiments the spray was operated by applying a pressure above that of the atmosphere, but it was found later that quite satisfactory spraying with more uniform control was obtained by aspirating by means of a water vacuum-pump.

By this means, rapid, even and complete combustion was realised, the time required for the combustion of 20 g. of nitrobenzene being about 30 minutes. By contrast, in the Davidson lamp method, the (imperfect) combustion of 2 g. of nitrobenzene, diluted with 8 g. of alcohol, takes about 6 hours.

RESULTS OBTAINED.—Experiments in which the method (described in detail later) was applied to different samples of technical nitrobenzene gave the results shown in Table I.

TABLE I

Determination of sulphur in samples of technical nitrobenzene

Sample	Weight of sample taken g.	Weight of barium sulphate g.	Sulphur found Per Cent.
(*a*)	14·15	0·0857	0·083
	15·00	0·0767	0·070
	18·91	0·1031	0·075
	23·91	0·1329	0·075
(*b*)	14·01	0·0231	0·023
	23·33	0·0339	0·020
(*c*)	12·99	0·0103	0·011
	16·48	0·0131	0·011
	15·04	0·0171	0·016

In a further series of experiments in which to a sample of nitrobenzene were added known quantities of (*a*) carbon disulphide and (*b*) thiophen, the results shown in Tables II and III were obtained:

TABLE II

Determination of sulphur in nitrobenzene with known amounts of carbon disulphide added

(*a*) The nitrobenzene contained 0·013 per cent. of sulphur.

	Weight of sample g.	Weight of barium sulphate g.	Total sulphur found Per Cent.	Sulphur (corrected for S present in nitrobenzene) Per Cent.	Sulphur added Per Cent.	Error
(i)	13·15	0·1853	0·194	0·181	0·207	−0·026
	14·45	0·2198	0·208	0·195	0·207	−0·012
(ii)	14·04	0·1136	0·111	0·098	0·102	−0·004
	12·31	0·1019	0·098	0·100	0·102	−0·002
(iii)	16·30	0·0951	0·080	0·067	0·057	+0·010
	13·71	0·0744	0·074	0·061	0·057	+0·004
(*b*)	The nitrobenzene contained 0·011 per cent. of sulphur.					
(iv)	15·99	0·1161	0·100	0·089	0·097	−0·008
	14·50	0·1010	0·095	0·084	0·097	−0·013
	14·67	0·0985	0·092	0·081	0·097	−0·016

TABLE III

Determination of sulphur in nitrobenzene with a known amount of thiophen added

The nitrobenzene contained 0·013 per cent. of sulphur.

Weight of sample g.	Weight of barium sulphate g.	Total sulphur found Per Cent.	Sulphur (corrected for sulphur in nitrobenzene) Per Cent.	Sulphur added Per Cent.	Error in sulphur
14·15	0·0660	0·064	0·051	0·057	−0·006
16·49	0·0825	0·068	0·055	0·057	−0·002

The results in the preceding tables show that, when applied to nitrobenzene, the method is capable of giving results accurate to within ± 10 per cent. for sulphur-contents from 0·05 to 0·2 per cent.

Subsequent investigation showed that the method was applicable to a number of other organic substances. With certain hydrocarbons, *e.g.* paraffins, naphthalene, etc., and bodies other than nitro compounds, with a low oxygen-content, it was found necessary to dissolve the sample in about an equal weight of nitrobenzene of known sulphur-content before combustion. Combustion of solid nitro bodies, *e.g. p*-nitrotoluene and nitronaphthalene, may be effected either by dissolving the substance in nitrobenzene, or by heating it to a temperature slightly above its melting point and pre-heating the air passed into the spray apparatus.

Satisfactory results have been obtained with the substances shown in Table IV.

TABLE IV

Substance	Sulphur found Per Cent.	Substance	Sulphur found Per Cent.
Benzene	0·016 0·017	α-Nitronaphthalene	0·083 0·082 0·084 0·086
Naphthalene (sample *a*)	0·176 0·177		
Naphthalene (sample *b*)	0·087 0·087	α-Naphthylamine	0·021 0·016
Kerosene	0·036 0·040	Aniline	0·005 0·002
Toluene	0·05 0·04	*o*-Dichlorobenzene	0·23 0·24
Pyridine	0·015 0·014	*p*-Nitrotoluene ..	0·0081 0·0082
α-Picoline	0·058 0·056	*p*-Chloronitrobenzene	0·003 0·003
		Methyl alcohol* ..	0·0008

* The weight of methyl alcohol burned was 168 g., the time required being 2 hours 10 minutes.

The method has been employed successfully in an independent laboratory for the determination of small amounts of sulphur in ethyl alcohol.* Some of the results are given in Table V, together with those obtained by a modified lamp-method.

TABLE V

Sample	Lamp method (modified) Sulphur Per Cent.	Combustion (spray) method Sulphur Per Cent.
Ethyl alcohol, sample (i)	0·0050	0·0061 0·0069
,, ,, ,, (ii)	0·0015	0·0022 0·0020 0·0017
,, ,, ,, (iii)	0·0015	0·0020 0·0015 0·0015
Alcohol, to which was added 0·005 per cent. of thiophen and 0·005 per cent. of H_2SO_4. Theory, 0·0048 per cent. of sulphur.	0·0041	0·0045 0·0048

METHOD.—The following is a description of the method:

Apparatus.—This (see Fig. 1) consists of a glass spraying vessel (G) connected by a ground-in joint with a silica tube (H). This in turn is connected with the absorption bottles (K, L and M), in which the combustion products are retained. Details of the construction of the spraying vessel are shown in Fig. 2. The silica tube, which is of translucent silica, is approximately 38 cm. long with a neck 12·5 mm. internal diameter, and walls 1 to 2 mm. thick. A disc of platinum gauze of about 36 S.W.G. is placed in the silica tube about 6·5 mm. above the ends of the spraying tubes. A second disc of platinum gauze is then placed about

* We are indebted to Messrs. W. G. Bailey and T. Thompson, of Imperial Chemical Industries, Ltd. (Explosives Group), for these determinations.

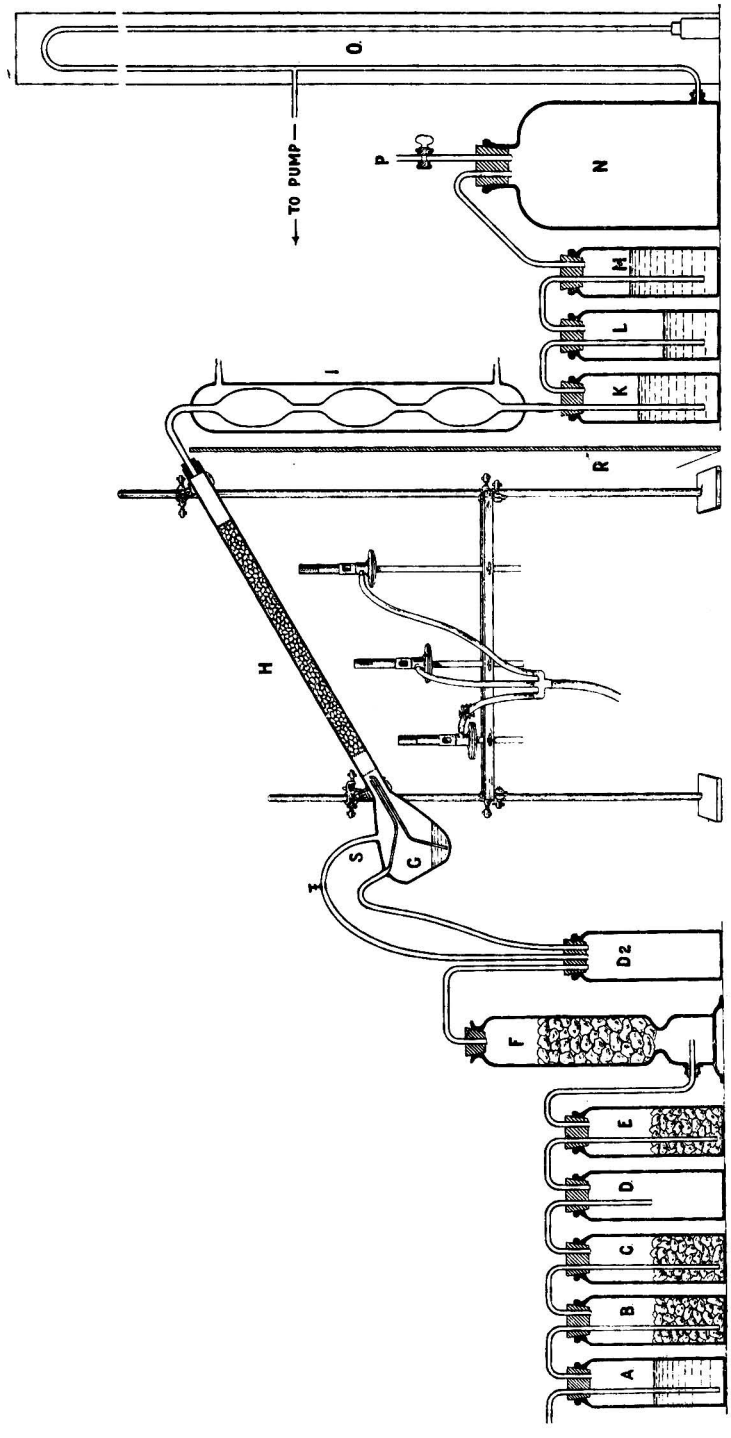

Fig. 1

3·8 cm. above the first, and the tube is filled to about three-quarters of its total length with quartz particles, about 2 mm. in size, platinised by immersing them in 5 per cent. platinum chloride solution, draining off the excess of solution, and igniting them.

A third platinum gauze is placed above the quartz packing, and a short piece of glass tubing, of such length as to come within half an inch of the end of the tube, rests on the last gauze. The last gauze and the glass tube are introduced to prevent the packing from moving in the silica tube.

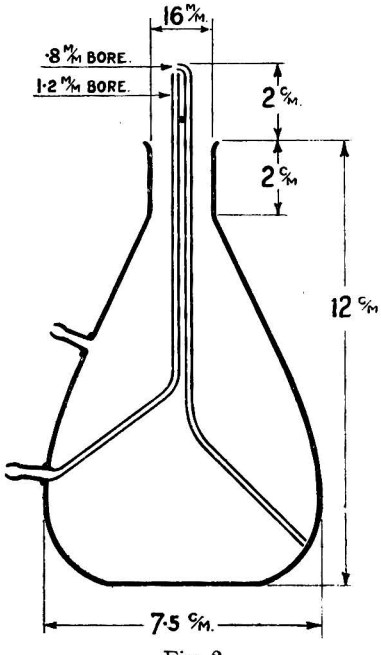

Fig. 2

The quartz tube is inclined at an angle of about 30° to the horizontal, and its upper end is connected, by means of a cork, with a small water-cooled condenser (I), the lower end of which enters the first absorption bottle.

The absorption bottles (K), (L), (M) are each of approximately 200 ml. capacity, and contain freshly prepared 3 per cent. sodium peroxide solution; in (K) 50 ml. and in each of (L) and (M) 25 ml. of the peroxide solution and 25 ml. of water; (M) is connected with an aspirator (N), of about 2 litres capacity, provided with an adjustable air-leak (P); this is employed in regulating the pressure in the apparatus. The aspirator is connected [with a manometer (O) in circuit] with the vacuum pump. A shield of asbestos (R) protects the condenser from the heat of the burners.

The quartz tube is heated by three burners, one placed under the space between the first and second gauzes, the second at a point about 3·8 cm. above the second gauze, and the third at a point about two-thirds of the way along the quartz tube.

The air entering the apparatus is drawn through a purifying train constituted as follows:—(A) containing alcoholic potash (2 *N*) (to absorb carbon disulphide); (B) and (E) containing pumice soaked in 5 per cent. lead acetate solution; (C) and (F) containing pumice soaked in 5 per cent. aqueous potassium hydroxide solution; an empty bottle (D) is placed between (C) and (E) to prevent the solution in (C) being blown over into (E). A trap-bottle (D2) carries the tubes connected with the spraying vessel, the one attached to the auxiliary air inlet (S) being fitted with a screw clip.*

PROCEDURE.—(*a*) *Nitrobenzene and similar bodies.*—A portion of the sample, weighing approximately 15 to 20 g., is placed in the spraying vessel, and the vessel and its contents are weighed. The parts of the apparatus are connected as described, and a gentle stream of air is aspirated through the vessel. The burners are lighted, and the tube is heated to bright redness, except at the point

* The apparatus is obtainable from Messrs. Griffin and Tatlock, Ltd., Kemble Street, Kingsway, London, W.C.2, and at Manchester, Edinburgh, Glasgow and Liverpool.

between the first two platinum gauzes at the lower end of the tube; this is maintained at a temperature just sufficient to prevent the collection of liquid above the first gauze. (For benzene and other easily-volatilised liquids the burner under this gauze may be omitted.)

When the tube is red hot the rate of aspiration is gradually increased until the spraying commences. The rate of air entering through the main and auxiliary inlets is adjusted by means of the tap (P) and the screw clip on (S), so as to maintain a state of bright incandescence at the point above the second burner. The reading of the manometer (O) will vary somewhat with different sets of apparatus, but once the optimum conditions have been established for a particular apparatus, they will serve as a guide in subsequent determinations.

When practically all the sample has been burned the burners are removed and the apparatus is allowed to cool, a gentle current of air being meanwhile drawn through it.

After cooling, the condenser is washed into a 500-ml. beaker, and the contents of the absorption bottles are transferred to the same vessel. The contents of the beaker are boiled to decompose the excess of sodium peroxide, cooled slightly, cautiously acidified with 10 ml. of concentrated hydrochloric acid, and filtered. The filtrate and washings are diluted with water to about 450 ml. and heated to boiling, and the sulphate is separated and weighed as barium sulphate in the usual manner.

The spraying vessel is weighed after the combustion, the weight of the sample used in the determination being found by difference. A well-constructed vessel should retain only about 0·5 g. of the sample.

(*b*) For liquids which do not contain nitro groups, and for solids, the sample is dissolved in about an equal weight of nitrobenzene of known sulphur-content; this solution is then treated exactly as described under (*a*).

Note I.—A small amount of nitrogen acids is formed during the combustion, but it has been found that this is insufficient to interfere with the barium sulphate precipitation.

Note II.—As the glow from the combustion is extremely bright, it is advisable for the operator to wear dark goggles.

The method shows promise of wide application, and work is at present in progress to extend it to the determination of sulphur as a major constituent in some organic compounds.

We wish to record our thanks to Imperial Chemical Industries Limited (Dyestuffs Group), in whose Research Analytical Laboratory this work was carried out, for permission to publish the results of this investigation.

References

1. P. Klason, *Ber.*, 1886, **19**, 1910; C. S. Leonard, *J. Amer. Chem. Soc.*, 1923, **45**, 255; E. Wertheim, *ibid.*, 1930, **52**, 1086.
2. British Engineering Standards Association Specification for Motor Benzol, 1921 [135], Appendix III.

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Notes

The Editor wishes to point out that the pages of the Journal are open for the inclusion of short notes dealing with analytical practice and kindred matters. Such notes are submitted to the Publication Committee in the usual manner.

WATER IN BONE MEAL AND IN MEAT AND BONE MEAL

THE proximate analysis of samples of genuine bone flour and of genuine meat and bone meal usually adds up to 98·5 to 99 per cent., but samples of both substances are occasionally met with in which the sum of the moisture, oil, protein and mineral matter amounts only to 90 to 95 per cent. My attention was first drawn to this fact by Mr. John Evans, and the matter has since been investigated here on a number of samples.

Apparently all the meals exhibiting this phenomenon contain a proportion of steamed bone, and the undetermined constituent is undoubtedly chemically combined water that has been introduced during the steaming process. This water is not expelled during the determination of moisture as prescribed by the Fertilisers and Feeding Stuffs Regulations.

The following account of tests made on one sample illustrates the presence of combined water, and the results are typical of those obtained with a number of samples examined:

Steamed bone flour gave the following analytical results:—Moisture, 14·2; oil, 0·2; albuminoids (protein), 3·2; mineral matter, 77·8; total 95·4 per cent.

Five g. of the meal were weighed and subjected to analysis in the following order:

(i) Drying to constant weight at 100° C.:—Moisture, 14·2 per cent.

(ii) Heating for 2½ hours in a closed vessel contained in an oil-bath at 180° C. A current of dry air was drawn over the meal and led into a weighed calcium chloride tube. By this means an additional 3·3 per cent. of water was found.

(iii) Determination of oil on the chemically dry residue. An increase of 0·05 per cent. was found.

(iv) Determination of protein in the oil-free residue. A decrease of 0·2 per cent. was found.

(The determinations of oil and protein in the chemically dried material were made, of course, to prove that the additional water had not been produced by oxidation of the organic matter of the meal.)

The above results clearly indicate the presence, in the sample, of water not detected by the prescribed method for the determination of moisture, and the matter becomes of some importance if a meal contains added carbohydrate in addition to steamed bone. In such a case it is obviously quite inaccurate to estimate the "carbohydrate" by a difference figure between 100 and the sum of the moisture, oil, albuminoids and mineral matter.

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THE DETERMINATION OF ESTERS IN ALCOHOLIC LIQUIDS

VARIOUS organisations, governmental and otherwise, in different parts of the world, which are concerned with the determination of esters in alcoholic liquids, have laid down "official" methods for the purpose. All agree in fundamental principles, and all those whose methods concern me in the course of my duties agree also on one detail, *viz.* that the refluxing of the spirit with excess of hot alkali should continue for an hour. In *Allen's Commercial Organic Analysis* (5th Ed., Vol. I,

p. 257) the suggested time is reduced to thirty minutes. My experience is that even this shorter time greatly exceeds what is analytically necessary.

The following tabulation of some determinations made recently brings out the point excellently:

Sample No.	Volume of $N/10$ sodium hydroxide solution required for hydrolysis of esters: Boiled for 10 minutes ml.	Boiled for 30 minutes ml.	Boiled for 60 minutes ml.
1	—	0·25	0·25
2	—	0·30	0·30
3	—	0·60	0·60
4	—	0·80	0·80
5	1·10	1·1	—
6	10·30	—	10·30
7	4·40	—	4·40
8	0·25	—	0·25
9	0·70	—	0·70
10	0·40	—	0·40
11	0·80	—	0·80
12	5·30	—	5·30

The nature of the samples varied from rectified spirit of good quality to illicit spirit. Strengths varied from 26·84 to 61·8 per cent. of alcohol by volume at 60° F./60° F.

In this Laboratory it is often necessary for legal reasons to certify that an analysis has been made by an "official method" of some organisation not under our control. Where a spirit analysis is concerned, this entails a waste of fifty minutes, accompanied by a corresponding "tie-up" of apparatus and bench-space, for every individual sample examined. This is a serious matter for a busy analyst, and it seems very desirable that the organisations concerned in establishing "official methods" should revise some of their preconceived ideas.

This note is published with the approval of Dr. Alfred Tingle, the Chief of this Laboratory.

JAMES HOSSACK

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COLOUR REACTIONS OF CARBAZIDES AND CARBAMIDES WITH DIACETYL AND DIACETYLDIOXIME

ON warming semicarbazide hydrochloride and diacetyldioxime with hydrochloric acid it was found that an intense permanganate-red colour developed, reaching a maximum intensity after a few minutes. The colour was permanent and apparently stable for several days. A similar colour was produced by using diacetyl in aqueous solution instead of diacetyldioxime, but the time of development was not appreciably shortened. On the subsequent addition of ammonia bluish-violet colours appeared.

The intensity of the red colour varies with the relative proportions of diacetyl or the dioxime and semicarbazide, and with the acidity of the solution; increasing the acidity favours the formation of a more intense colour.

The following results were obtained in an attempt to discover to what extent these effects were characteristic of semicarbazide:—Hydrazine, guanidine, nitroguanidine, aminoguanidine, dicyandiamide, oxamethane, semi-oxamazide, oxalic

dihydrazide, sym. diphenylthiocarbamide, phenylthiosemicarbazide and asym. dibenzylhydrazine gave very pale yellow or practically colourless solutions when gently heated with either diacetyldioxime or diacetyl in the presence of hydrochloric acid.

Carbamides, semicarbazides and certain other compounds gave definite colours, as shown in the following table:

Substance	Diacetyldioxime and hydrochloric acid	Diacetyl and hydrochloric acid
Semicarbazide	Red	Red
4-Methyl- ,,	Red	Red
4-Phenyl- ,,	Red	Red
4-*m*-Tolyl- ,,	Red	Very pale pink
4-*p*-Tolyl- ,,	Reddish-brown	Red
4-*p*-Bromphenyl- ,,	Reddish-brown	Red
4-Benzyl- ,,	Red	Reddish-yellow
4-β-Naphthyl- ,,	Brown	Brown
4-as.*m*-Xylyl- ,,	Red	Red
1-Phenyl- ,,	Reddish-brown	Red
1-β-Naphthyl- ,,	Brown	Brown
Carbamide	Brown	Bright yellow
Methylcarbamide	Reddish-brown	Pale pink turning to bright yellow
o-Tolylcarbamide	Brown	Red
m-Tolylcarbamide	Brown	Red
Benzylcarbamide	Brown	Reddish-brown
Diphenylcarbazide	Red	Red
Phenylhydrazine	Brown	Yellow
Phenol	Pink	Yellow
Aniline	Pale violet	Very pale pink
m-Toluidine	Pale violet	Very pale pink

1-Phenylsemicarbazide gives a red colour with hydrochloric acid alone, but this does not survive a few minutes' heating in a boiling-water bath; if diacetyl is present, a permanent deep red colour is produced.

The addition of a slight excess of ammonia after the development of a colour in acid solution yielded a pale yellow or colourless solution in all cases except with semicarbazide and 4-methylsemicarbazide (blue or violet), 1-phenylsemicarbazide (reddish-brown), and carbamide (bright yellow).

The results shown above should be useful for rapid qualitative tests; thus, semicarbazide is readily differentiated from hydrazine, semi-oxamazide, aminoguanidine and oxalic dihydrazide by the formation of a red colour, and also from all the other compounds mentioned, with the exception of 4-methylsemicarbazide, if the solution is finally made ammoniacal. The sensitivity with semicarbazide is such that about 0·1 mg. may be detected in a few ml. of a solution made strongly acid with hydrochloric acid.

I wish to thank the Air Ministry for granting facilities for carrying out this work.

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BUTYRISED FATS: BUTTER-AROMA

WHEN a full-flavoured butter is distilled with steam, free butyric acid can be identified in the distillate, which also contains a neutral compound yielding butyric acid on saponification. The use of butyric cultures, ethyl butyrate or butyric acid in margarine has been tried experimentally, but with unsatisfactory results. Butyric acid itself is too fugitive, and search was therefore made for a parent substance which would continuously yield traces of the free acid. In experiments

with margarine which had been prepared with skim-milk properly soured with lactic streptococci, including aroma bacteria, it was found that a small addition of tributyrin caused the mass to be gradually pervaded by a pleasant butter-aroma. Unfortunately, the flavour was spoilt by the bitterness of the tributyrin; this was obtained from various sources, and was synthesised, fractionated and refined, but in no instance was the bitterness avoided. Glycol dibutyrin produced a similar aroma in margarine, but had also an inherent nasty flavour. Mixed butyric triglycerides were then prepared, and the flavour of these indicated that the butyric acid in butter-fat must be present as a mono-butyric triglyceride, with butyric acid in the β-position. The bitterness of tributyrin is due to the butyric acid in the α-position.

Coconut oil was butyrised by heating it with butyric acid at 160° C. After 2 hours' treatment the neutralised fat gave Gilmour butyric numbers equivalent to 3 per cent. of butter, and to 116 per cent. after 36 hours' treatment. Fat can also be butyrised by heating it with tributyrin in presence of a suitable catalyst, such as 1 per cent. of stannous hydroxide, at 200° to 250° C., the mixture being agitated by causing an inert gas to bubble through it. Coconut fat containing from 5 to 15 per cent. of tributyrin was free from bitterness after $2\frac{1}{2}$ to 7 hours, whilst arachis oil with 25 per cent. of tributyrin required 11 hours' treatment.

The results of several investigations of aroma-producing substances of the acetoin (acetyl-methylcarbinol), 2·3-butylene glycol, and diacetyl-type have recently been published. The quantity of diacetyl (the substance to which the aroma of butter is mainly due) in butter ranges from 0 to 5 parts per million, this depending on the presence (and proportion) of aroma bacteria in the souring cream. My experiments (in which I was assisted by Mr. Kenneth Wallis) have definitely indicated, however, that the composition of the fat is an important factor in the production of aroma.

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Legal Notes

Under this heading will be published notes on cases in which points of special legal or chemical interest arise. The Editor would be glad to receive particulars of such cases.

SOLUTION OF IODINE : USE OF METHYLATED SPIRIT

AT Salford City Police Court on the 4th January summonses were heard in connection with the sale of a bottle of solution of iodine. The sample was certified by the Public Analyst to contain 0·7 per cent. of iodine, 0·7 per cent. of potassium iodide, 72·2 per cent. of methylated spirit (90 v/v), 2·5 per cent. of boric acid, and 23·9 per cent. of water, and, upon comparison with the British Pharmacopoeia formula for Liq. Iodi. Mitis, to be 71 per cent. deficient in iodine, and 53 per cent. deficient in potassium iodide, to contain the foreign ingredient, boric acid, and to have the alcohol of the Pharmacopoeia replaced by a smaller amount of methylated spirit. The retailer was summoned under Section 2 of the Food and Drugs (Adulteration) Act, and the makers under Section 5 of the Summary Jurisdiction Act, 1848, for aiding, abetting, counselling and procuring the sale. The makers were also summoned under Section 30 of the Food and Drugs (Adulteration) Act for wilfully applying a false label. Both retailer and maker pleaded guilty.

A feature of the case was the statement by the makers that their formula had been approved by the Commissioners of Customs and Excise. One of the objects of the prosecution was to make it clear that when the formula of a preparation containing industrial methylated spirits is approved by the Commissioners of Customs and Excise, such approval merely signifies that they are satisfied that the spirits will be used in accordance with the provisions of the Spirits Act, 1880, and amending Acts, and does not imply that the resulting article will comply with the Food and Drugs Act or with any similar Act.

Mr. P. Butlin, appearing for the prosecution, said that this case was intended to serve as a warning. The Stipendiary Magistrate, Mr. Percy Macbeth, therefore imposed nominal penalties of five shillings in each charge, but ordered the makers to pay seven guineas costs.

The same penalties were inflicted in two similar cases concerning other brands of iodine solution.

"REAL CREAM" TARTS

At the West London Police Court, on August 9th, 1934, a confectioner was summoned by the Hammersmith Borough Council for selling "Real Cream Tarts" and "Real Cream Cookies" which were not of the nature, substance and quality demanded. Evidence was given that samples were purchased at the defendant's premises from a quantity of cakes and pastries displayed in the window and bearing tickets with the words "Real Cream, 4 for 3½d." The certificates of the Public Analyst (Mr. F. W. Edwards, F.I.C.) showed that the samples contained a filling resembling cream, made from a fat having the characteristics of margarine fat, and therefore consisted of a substance other than real cream.

The defendant, who pleaded guilty, was fined £2 10s.

At the same Court a firm of bakers was summoned for a similar offence in respect of the sale of a "Real Cream Sandwich." The defendants, who pleaded guilty, stated that a wrong ticket had been used by an assistant.

The summons was dismissed on payment of £2 2s. costs.

A summons was also brought against another Hammersmith confectioner in respect of the sale of "Real Cream Éclairs." In this case the pastries were displayed in the shop window bearing a ticket "Real Cream Éclairs—2d. each." The Public Analyst certified that the filling contained 51·7 per cent. of fat other than milk-fat, and therefore consisted of a substance other than real cream.

The defendant, who pleaded not guilty, was fined £5.

Department of Scientific and Industrial Research

REPORT FOR THE YEAR 1933–4*

IN the introduction to the Report of the Committee of the Privy Council for Scientific and Industrial Research (pp. 1–5) it is mentioned that the most important of the proceedings during the year concerned the provision of further grants to Research Associations to be offered on conditions that would secure a larger measure of support from the industries benefited. The Building Research Board has been reconstituted to conform with the general principles adopted for Research Boards, and a Standing Conference on Timber Utilisation has been established.

During the year fifteen British patent applications have been filed, and ten British patents have been allowed to lapse; the subject-matter of these patents is given.

The Report of the Advisory Council to the Committee of the Privy Council (pp. 7–22) discusses the question of grants to the various Research Associations, and lays emphasis on the immediate practical advantages that industry can derive from adequate support and use of research associations. Certain researches have been transferred from Government control to the supervision of industry, and co-operation in research with the Dominions overseas has been continued, notably in connection with the storage and transport of food.

The researches for Government include the investigations of the conditions of safety for the transport of gas cylinders, and for the production of cheap and efficient respirators to be used against the inhalation of dust. Another important development has been the establishment of a large-scale fire-testing station in London by the Fire Offices Committee of the insurance offices in consultation with the Building Research Station. The third section of the Report (pp. 25–95) gives a summary of the research work done during the year.

NATIONAL PHYSICAL LABORATORY.—Work has been continued on the fundamental standards of length, mass, electrical resistance and current, candle-power, temperature, and *X*-ray intensity.

It has been found that up to about 900° C. iron has a negative temperature coefficient of thermal conductivity, but that gamma iron appears to possess a positive temperature coefficient.

A further advance in the standardisation of *X*-ray measurement has been made by the completion of the investigation of calibration by measurement of the colour-change of barium platinocyanide pastilles when exposed to *X*-rays. In the Photometry Division photo-electric methods of photometry have been developed, *e.g.* for the detection of non-uniformity in the candle-power of a lamp.

Metrology Department.—The researches having for their ultimate object the establishment of a wave-length standard of length have reached a further stage in the determination of the lengths of the yard and the metre in terms of the red radiation of cadmium, both in air and *in vacuo*.

Metallurgy Department.—Progress in the study of the physical structure of metals and alloys has been made. The production of iron of high purity has received further attention; in recent samples the total impurities, excluding oxygen, amount to only 0·012 per cent., the nickel present having been reduced to 0·0006 per cent. The study of the properties of molten metals and alloys has also been continued.

FUEL RESEARCH.—The Report of the Fuel Research Board for the year ended March 31st, 1934, has recently been published (see ANALYST, 1934, **59**, 540).

* H.M. Stationery Office, pp. 183. Cmd. 4787. 1935. Price 3s. net.

FOOD INVESTIGATION.—For the Report of the Food Investigation Board for the year 1933 see ANALYST, 1934, **59**, 696.

BUILDING RESEARCH.—Details of the work carried out at the Building Research Station during the year are given in the Annual Report of the Building Research Board (*cf.* ANALYST, 1934, **59**, 755). The general research is now steadily advancing to the stage at which there is a definite scientific basis for the utilisation of building materials and methods of construction.

STEEL STRUCTURES RESEARCH.—The Committee has issued a second report, in which the investigations are dealt with under four headings: the measurement of strains in existing buildings; the examination of bolted and riveted connections; stress analysis, and the study of new methods of design.

ROAD RESEARCH.—The comprehensive scheme of road research visualised by the Road Research Board has been described in the first Annual Report of the Board. The subjects dealt with include road construction, processes, road usage, and the development of special testing plant.

FOREST PRODUCTS RESEARCH.—The sixth report of the Forest Products Research Board deals *inter alia* with the factors determining the technical qualities of timber, the investigation of the insects and fungi that destroy wood, and the application of analytical methods to anatomical measurements of various timbers and to the study of the variations that occur in the wood-cells.

WATER IN WOOD.—It has been found that practically all the water in wood up to the fibre saturation point is held by adsorption, and that the method employed in the investigation has immediate applications of a practical nature in affording a means of measuring directly the fibre saturation point of various woods. Experiments have demonstrated that the common belief that timber becomes less affected by changes of moisture-content as it ages has no foundation in fact, and that there is no material difference between old and new timbers in the amount of shrinkage or expansion that takes place with a given variation in moisture-content.

METALLURGICAL RESEARCH.—The work is surveyed under the following headings: Behaviour of Materials at High Temperatures, Research on Light Alloys, Steel Castings, Alloy Steels, Gases in Metals, Cracking of Boiler Plates, Oxides in Steel, Oxidising Power of Basic Slags, Rust Prevention (see Engineering Research Special Report No. 12, H.M. Stationery Office, 1934; price 6d. net).

WATER POLLUTION RESEARCH.—The progress in the Department's work in relation to water supplies and the prevention of pollution is summarised in the Annual Report of the Research Board (ANALYST, 1935, 37).

CHEMICAL RESEARCH.—Among the activities of the Laboratory to which special reference is made are corrosion research, chemical reactions under high pressure, tar research, chemotherapy, water pollution, microbiology, dental investigation, and chemical engineering.

COLOURING MATTER IN HEAVY OILS.—A long investigation into the nature of the colouring matter of heavy oils of low-temperature tar has shown that these coloured substances are hydrocarbons of the naphthacene class.

The mixture of higher tar acids used as a wetting agent under the name of "Shirlacrol" has been obtained in considerable quantities from low-temperature tars and vertical-retort tars.

CHEMOTHERAPY.—The outstanding feature of research in chemotherapy has been the preparation of a potent arsenical drug, succinanilidomethylamide-*p*-arsonic acid; this is being tested clinically.

Synthetic Resins.—In connection with the study of synthetic resins it has been found that polyhydric phenols condense readily with formaldehyde to form

resins with remarkable adsorptive power for metallic cations—a property of significance in water purification.

Sensitive Test for Ferrous Iron.—In comparative experiments on the dehydrogenation of pyridine in the presence of different anhydrous metallic chlorides two products, 2:2′-dipyridyl and 2:2′:2″-tripyridyl, were found to afford a delicate test for ferrous iron. Samples of these have been distributed to various enquirers.

ILLUMINATION RESEARCH.—Work on the glaring effect of coloured light sources and the "time effect" of glare is continuing, and a glare-meter has been designed. Problems of mine lighting have been discussed, and the investigations in progress for the British Standards Institution on the fundamental properties of glassware have now been completed. The colorimetric examination of artificial daylight units on behalf of the B.S.I. has also been completed, and a report has been communicated to that Institution.

LUBRICATION RESEARCH.—The problems investigated include journal friction, oscillating bearing experiments, and experiments on the oxidation of oils.

ATMOSPHERIC POLLUTION.—The Report of the Atmospheric Pollution Research Committee (ANALYST, 1934, **59**, 280) includes a new method for obtaining an indication of sulphur pollution in the air.

FABRICS RESEARCH.—A report, including the large-scale tests of methods for fire-proofing fabrics, is to be published.

INDUSTRIAL RESPIRATORS.—A new design of respirator, which will not interfere with the vision of the wearer, is being developed.

RESEARCH ASSOCIATIONS.—As in previous years a brief account is given of the work of Research Associations during the year (pp. 97–134). Among these the Reports of the British Association of Research in the Chocolate, Sugar, Confectionery and Jam Trades and of the British Food Manufacturers' Association contain numerous points of special analytical interest.

Gelatin.—The effects of variations in the physical properties of commercia gelatins have been studied, with special reference to the manufacture of marsh mallows.

Pectin.—Pectin may vary in composition with the kind of fruit from which it is extracted and the method of extraction used. The formation of bubbles in jelly and premature setting of jelly, which are a frequent source of trouble to the jam manufacturer, are caused by the extracts of certain fruits. Purified pectins are therefore being prepared from these fruits, and their chemical and physical properties are being studied with a view to finding an explanation and a remedy.

Starch.—Starch, like pectin, is a complex substance, and its properties vary according to its source and method of preparation. An investigation of the properties other than its jellifying power is in progress.

Determination of Salt in Cured Meats.—An apparatus has been adapted to the purpose of determining the proportion of salt in bacon, etc. In the modified form it will be possible to read the percentages of salt direct from an indicator, the test taking only a few minutes. By means of this instrument it has already been found possible to determine the thickness of the fatty layers in bacon or pork, and thus to grade carcases in this respect.

Salt Tolerance of Bacteria.—Work on this subject has been continued, and information on variations in the percentages of salt required to inhibit the growth of various bacteria has been accumulated. Fortunately, some of the most objectionable forms appear to be the most easily suppressed.

Ministry of Agriculture and Fisheries

RETAIL TRADE NAMES FOR FISH

THE following letter has been sent by the Ministry to the Clerks of Local Authorities administering the Food and Drugs (Adulteration) Act:

RETAIL TRADE NAMES FOR FISH. *Re* FISHERIES NOTICE No. 23

SIR,

I am directed to state that the Ministry of Agriculture and Fisheries has had under consideration the possibility of effecting some measure of standardisation of the names under which different kinds of sea fish are sold to the public.

Your Authority are probably aware that there have recently been a number of prosecutions of fishmongers by certain Local Authorities under the terms of the Merchandise Marks Acts and the Food and Drugs (Adulteration) Act, 1928, for selling fish under false trade descriptions. As a result the National Federation of Fishmongers approached the Ministry and suggested that a standard list of retail names of all the principal varieties of fish should be drawn up for the information of fishmongers who were in many cases in doubt at the present time as to what names might properly be used. It was explained to the Federation that neither the Ministry nor any other body had power to prescribe or legalise the use of particular names; the legality of any name in the event of dispute could only be determined by the Courts. Nevertheless, the Ministry thought that certain advantages would accrue if an agreement could be reached among the principal Associations of fish merchants and traders on the use of a standard list of names which would be recommended by the Associations to their members.

With this object in view a list has been drawn up after consultation with the principal Associations concerned. So far as possible the ordinary recognised names for the various kinds of fish have been adopted. In a few instances, however, where these names were liable to be confused with those of other kinds of fish or were so unpleasing in themselves as to be likely to prejudice the sale of the fish to the public (*e.g.* cat-fish or dog-fish) a different name has been suggested, care being taken in no case to propose a name which might be confused by the public with that of some other known variety of fish.

In a few cases where different names are known to be in common use in particular localities for the same kind of fish, alternatives have been shown. In order to simplify the list and to secure as much uniformity as possible, alternative names have been introduced very sparingly, but it is of course possible that other established local names may be in use which are not misleading to the public and to which therefore no objection could reasonably be raised on that ground.

As previously indicated, this list is being recommended by the various Associations to their members for general adoption. The Ministry thinks, therefore, that the list will be of use to those Local Authorities by whom action under the Merchandise Marks Acts or the Food and Drugs (Adulteration) Act is likely to be taken in the event of fish being sold under false trade descriptions. Further copies of the list may be obtained from the Ministry on request.

I am, Sir, your obedient servant,

(*Signed*) H. G. MAURICE

To the Clerk of the
Council or Town Clerk

FISHERIES NOTICE No. 23

The list contains in six pages the suggested trade names, the scientific names, and the general English equivalents of 53 species of fish, with synonyms in brackets. The suggested new trade names include the following:

Argentine (*Argentina sphyraena* and *A. silus*) for Lesser silver smelt, Great silver smelt.

Atherine (*Atherina presbyter* and *A. boyeri*) for Silver sides, Smelt, Sand smelt, Atherine, Sea smelt.

Forkbeard (*Urophycis blenoides, Phycis blenoides, Raniceps raninus*) for Greater forkbeard, Forked hake, Lesser forkbeard.

Lance (*Ammodytes lanceolatus, A. tobianus, A. cicerulus*) for Greater sand eel, Small sand eel, Smooth sand eel, Lance.

Redfish (*Sebastes marinus, S. norvegicus, S. viviparus, S. heltzeni, Scorpaena dactylopera*) for Norway haddock, Soldier.

Roughback (*Hippoglossus platessoides, H. linianoides, Drepanopsetta platessoides*) for Long rough dab.

(See also Hattersley, ANALYST, 1935, 69.)

Cyprus

ANNUAL REPORT OF THE GOVERNMENT ANALYST FOR THE YEAR 1933

THE Government Analyst (Dr. S. G. Willimott) states that, of the 2342 samples examined, 2321 were submitted officially, 1073 being samples of food and drugs, and 229 exhibits in connection with criminal cases. For the purpose of the administration of the Food and Drugs Law the Island is divided into seven districts. Samples are taken by the District Sanitary Inspectors, and the control over the food supply is increased by authorising certain sanitary inspectors, who hold the Local Sanitary Certificate, to take official samples in their sub-districts.

MILK.—Fifty-four of the 172 samples examined were adulterated, and there is ample evidence that the adulteration of fresh milk is widespread throughout the Island. A more stringent requirement for fat-content than the present 3 per cent. might prove effective (*cf.* Annual Report for 1932; ANALYST, 1934, **59**, 41).

OLIVE OIL.—Forty of the 68 samples of olive oil examined were adulterated, and it was difficult to obtain a genuine article in the town of Nicosia. Owing to the drought the harvest of olives was poor, and the fact that the supply of olive oil was restricted was regarded as an opportunity and justification for wholesale adulteration. In some cases material sold as olive oil was found to consist entirely of soya-bean oil.

CANNED FISH.—The District Sanitary Inspectors, acting on instructions, again devoted particular attention to the stocks of canned foods, and it was found necessary to condemn some thousands of cans of unfit material. Merchants are now showing more caution by ordering smaller stocks, and thus avoiding the exposure of large consignments to long-continued heat. It has been found, for example, that canned fish in tomato sauce is very liable to be spoiled by exposure to the Cyprus summer, and shopkeepers have been advised accordingly. As a result of the last four years' work there is now a definite improvement throughout the Colony in the condition of canned food.

COFFEE.—Starch continues to be the favourite adulterant, 37 of the 187 samples examined containing from 5 to 60 per cent.

NITROBENZENE POISONING.—An unusual case of non-fatal poisoning at Amiandos was investigated. A tin of water-proofing composition, containing nitrobenzene as solvent, had been left for some time in a closed office during the summer, and vapour had escaped and produced symptoms of faintness and nausea in a young clerk. It is known that nitrobenzene vapour is toxic (*Taylor's Medical Jurisprudence*, 1928, Vol. II, p. 699), and this appears to be the explanation of the effects in this case.

CAUSTIC SODA POISONING.—It is somewhat remarkable that in one year there should have been 8 cases of poisoning by caustic soda in Cyprus, an island with a population of only 348,000. Four of these cases were the results of accidents to young children, and the remainder were cases of suicide. In every instance the alkali was derived from supplies used for domestic washing purposes, to soften the generally prevailing hard waters of Cyprus, but, as the result of recent experience, it was advised that caustic soda and caustic potash should be placed on the Poisons Schedule. This recommendation was adopted by Government in February, 1934. Formerly the alkali used as a water softener consisted exclusively of wood ashes, but within recent years this has been replaced by washing soda, and during the last two years by caustic soda.

The first case of poisoning occurred in December, 1932, and an account of it was published in *The Lancet*, August 19th, 1933. The caustic soda, as sold, consisted of fused lumps, and analysis showed an average of 77 per cent. of sodium hydroxide with 10 per cent. of sodium carbonate. Four cases of this series, all affecting adults, have been studied in some detail. The fatal dose of caustic soda is unknown, but Starkenstein (*Toxikologie*, Berlin, 1929, p. 91) considers it to be from 10 to 20 grams of the solid material. From our experience it must be much less than this, because even five grams has produced the most serious complications of stricture, culminating fatally in 74 days. There was perforation of the oesophagus in one fatal case, and possibly perforation of the stomach in another; in another three cases stricture had developed. The pathology of certain of these cases has been studied by the Bacteriologist, and a joint paper embodying these results was published in the *British Medical Journal* (June 9, 1934, p. 1022).

CYPRUS WATER SUPPLY.—Owing to the continuance of the drought, the water problem remained urgent throughout the year, and many old sources went dry during the summer. There was intensive search for water, and a number of schemes were launched, some of which were successful. Altogether 67 samples, mostly new supplies, were analysed, of which 14 were returned as unsatisfactory for drinking purposes. It is encouraging to observe a more extensive use of iron pipes, in contrast with the old system of ground aqueducts, for the conveyance from the source to the point of distribution of public and private water supplies. Water conveyed by aqueduct is nearly always open to contamination, especially in Cyprus, where chlorination is not practised, while the loss by leakage and theft must be considerable. A marked improvement in the quality of the water supply, both in town and village, can be expected when the system of piping supplies has become general.

The character and quality of water supplies in Cyprus vary widely from the pure soft water of the mountain spring to the hard, saline and often polluted waters of the plains. The constituents of the water also frequently vary widely with the different seasons of the year.

British Guiana

REPORT OF THE GOVERNMENT ANALYST, 1933

THE Government Analyst (Mr. K. Wallis) reports that the total number of samples analysed during the six months ended June 30th, 1933, was 2485, of which 106 were adulterated. As usual, the bulk of the samples consisted of milk, 98 of the 2204 samples being adulterated. The average percentage of fat in the genuine samples was 4·0, showing that the minimum of 3·25 per cent., fixed by the Sale of Food and Drugs Ordinance, is ample for average milk from the cows in the Colony.

Surprise raids at frequent intervals, on Sundays, Public Holidays and at times outside official hours, are necessary. Forty-three of the 568 samples of milk thus taken on 25 occasions were adulterated. All milk is strained by the vendors, so that none of the 52 samples tested contained dirt.

Of the other 281 samples of food examined, 8 did not comply with the requirements of the Ordinance, three samples of butter consisting wholly of margarine. Two samples of aerated drinks contained saccharin, the use of which in such products is prohibited.

ABSTRACTS OF PAPERS PUBLISHED IN OTHER JOURNALS

Food and Drugs

Detection of Trichlorotribenzylidenesorbitol by means of Acetone. G. Reif. (*Z. Unters. Lebensm.*, 1934, **68**, 468–473.)—The method of detecting benzylidenesorbitol by means of acetone and sulphuric acid (ANALYST, 1934, **59**, 44) for ascertaining the presence of fruit-wine in wine may be adapted to apply to Litterscheid's method of detecting sorbitol (ANALYST, 1932, **57**, 178). When treated with sulphuric acid, the trichlorotribenzylidenesorbitol formed in this method gives *o*-chlorobenzaldehyde, and this, with acetone, gives *o*:*o*-dichlorodibenzylideneacetone, which dissolves in the sulphuric acid to form an orange-red solution. Trichlorotribenzylidenesorbitol is, however, more difficult to resolve into its components than the corresponding chlorine-free compound, so that stronger sulphuric acid is required. Prior to its decomposition with acid the chlorobenzylidene compound must be purified by thorough washing with methyl alcohol. Details of the procedure are given. T. H. P.

Determination of Rancidity in Flours, Semolinas and Italian Pastes. J. Berlie. (*Ann. Falsif.*, 1934, **27**, 552–553.)—Further experience of the method for determining rancidity (ANALYST, 1934, **59**, 629) has led to certain modifications being adopted. With substances poor in fat, such as flour, 5 g. of material should be taken, and where a high percentage is present, as in wheat germ, powdered egg, etc., 0·5 to 1 g. The extraction method should follow the French official method for flours, but 40 ml. of a mixture of 40 parts chloroform, 30 parts ether, and 30 of 95 per cent. alcohol, should be used, and to the collected extract 5 ml.

of an alcoholic solution of potassium iodide are added (5 g. in 200 ml. of alcohol and 50 ml. of water). After standing for 24 hours in the dark, the mixture is titrated with 0·002 *N* sodium thiosulphate solution. The degree of rancidity for 1 g. of fatty substance is the number of ml. of thiosulphate required for 100 g. of the product, divided by the percentage of fatty substance present, and the index of rancidity is the above-mentioned figure multiplied by 0·000254. From a number of experiments rancidity appears to manifest itself when the index for 1 g. of fatty substance is 0·01 or more. D. G. H.

Preliminary Tests for the Detection of Small Amounts of Hydrogenated Oils, Tallow and Fats of the Palm-fat Group in Lard. A. Peter. (*Z. Unters. Lebensm.*, 1934, **68**, 521–530.)—For the detection of hydrogenated oil or tallow, use is made of the fact that the glycerides crystallising from lard under certain conditions have a lower specific gravity than the crystals of α-palmito-distearin of tallow or the tristearin of hardened oils. The lard is filtered through a dry filter-paper and 5 g. are weighed into a thick-walled bacteriological test-tube, 170 mm. long and of 15 mm. bore, which is then left for 20 to 24 hours at 10 to 18° C. In order to ensure uniformity of conditions, not more than 12 samples should be tested in one batch. A stand holding the samples is left in an oven at 105° C. for 30 to 40 minutes, after which the stand is removed from the oven and the tubes immersed singly in a water-bath at 45° to 46° C. Exactly 5 minutes after the removal of the stand from the oven the first tube is taken from the bath and quickly wiped. Exactly 5 ml. of ether at 22° to 23° C. are run in rapidly from a burette, the tube being then closed with a rubber stopper, shaken five or six times, and placed in a bath at 20·5° C. The other tubes are similarly treated in order. Twenty minutes after removal from the bath at 45° to 46° C. the first tube is taken from the 20·5° C. bath and wiped, 6 ml. of alcohol (sp.gr. 0·80783 at 15° C.) at 22° to 23° C., being run in from a burette, and the tube closed and shaken as before, and placed vertically in a stand. All the tubes are left for 2½ hours in an incubator at 23·5° to 24° C., and afterwards shaken by four or five brisk movements and returned to the stand. Any tube in which the upper phase undergoes rapid de-mixing with sudden clearing, while the lower phase forms a turbid emulsion, is held by the stopper and inverted two or three times. After the lapse of 10 to 15 minutes, the tubes are examined. Samples forming no deposit contain at most small proportions of tallow or hardened oil, but if any appreciable thick deposit forms, the sample should be tested for tallow or hardened oil by Bömer's method (ANALYST, 1914, **39**, 84). When hardened oil is present in only small amount, the lower phase often demixes rapidly, whilst the upper phase forms an emulsion including the solid glycerides. In this case, hardened oil is indicated by the powdery, crystalline deposit forming in the lower phase and keeping the liquid above the deposit turbid for some time. To ensure that the alcohol used is of correct strength, 1·5 litre of 96 per cent. alcohol is distilled with 300 g. of lime from a 2-litre fractionating flask until the whole of the alcohol has passed over. To the distillate 96 per cent. alcohol is added until 6 ml. of the product, 5 ml. of ether and 5 g. of fat melted from bacon and cooled to about room temperature shows the mixing temperature 31·5° to 32° C. (see below).

As a preliminary test for coconut oil or palm-kernel oil in lard, use is made of the mixing temperature. Two grams of the anhydrous lard are placed in a bacteriological test-tube (170 × 15 mm.), 6 ml. of glacial acetic acid of definite water-content are run in from a burette, and then exactly 1 ml. of xylene from a micro-burette. The tube is at once closed with a rubber stopper carrying a small-bulbed thermometer reading to 0·1° C., the bulb being at the middle of the column of liquid. If the fat sets after being weighed out, it is re-melted and cooled to about room temperature before the acetic acid and xylene are added. The tube is then heated and carefully shaken over a flame until a homogeneous liquid phase forms; it is then fixed, by means of a cork, inside a wider tube. The liquid is next shaken until it becomes permanently turbid, the mixing temperature being read off to within 0·1° C. Results are given for a number of lards of various origins, after addition of 5 or 20 per cent. of coconut oil or palm-kernel oil. For Yorkshire lard, with the refractometer reading (Zeiss butyro-refractometer) 49·0 at 40° C. and the mixing temperature 46·6° C., this temperature becomes 43·6° (43·0°) or 35·3° (34·8°) on addition of 5 or 20 per cent. of palm-kernel oil (coconut oil). To ascertain the suitability of the acetic acid, use is made of fat rendered from bacon and having the acid value 0·6 to 1·2, and the butyro-refractometer value 50 to 50·4 at 40° C. The acetic acid should contain so much water that the mixing temperature of 2 g. of this fat, 1 ml. of xylene and 6 ml. of the acetic acid is 43·5° C. If the temperature found differs from this by less than 1°, the acetic acid may be used and the results obtained corrected accordingly. The theoretical aspects of the above tests are discussed at length. T. H. P.

Deterioration Value [Verdorbenheitszahl]. Iodimetric Determination of the Oxidised Products in Fats and Oils. J. Gangl and W. Rumpel. (*Z. Unters. Lebensm.*, 1934, **68**, 533–539.)—The deterioration of edible fats and oils which finally renders them unfit for food is due essentially to oxidation processes, and in its early stages consists of combination of oxygen at particularly reactive double linkings. If dried in the air with a large surface exposed, beef-fat and lard may take up quantities of oxygen representing an increase in weight of 3 per cent. The extent to which this change has proceeded may be determined iodimetrically (*cf.* Taffel and Revis, *J. Soc. Chem. Ind.*, 1931, **50**, 87T; abst. ANALYST, 1931, **56**, 323), and is termed the "deterioration value," this being defined as the number of milligrams of potassium iodide decomposable by 10 g. of the fat.

For the determination the following solutions are required: (i) 0·01 *N* potassium iodate solution. (ii) A solution of 4·4 g. of potassium iodide in propyl alcohol (free from impurities reacting with iodine). The titre is ascertained by diluting 5 ml. with 20 ml. of hydrochloric acid (1 : 1), adding 10 ml. of 10 per cent. potassium cyanide solution and 3 ml. of starch solution, and titrating and shaking with the iodate solution (i) to decolorisation. (iii) Ten per cent. potassium cyanide solution. (iv) Two g. of soluble starch and 0·01 g. of mercuric iodide (as preservative) are ground with a little water and poured into 1 l. of boiling water. The clear solution undergoes no change even on long storage in a colourless glass vessel, but, if for any reason, it gives a violet instead of a pure blue colour with dilute iodine solution it should be discarded.

Ten grams (5 if the fat has undergone much change) of the fat are treated, in a 200-ml. flask with a ground stopper, with 5 ml. of solution (ii) and 2 drops of glacial acetic acid. The flask is kept for about 10 minutes (5 minutes suffice with a much-altered fat) in a bath at about 50° C., with occasional vigorous shaking. After being well shaken with 20 ml. of hydrochloric acid (1 : 1) and left for a short time, the liquid is poured through a moistened filter and the flask and filter are washed with repeated small amounts of the hydrochloric acid (1 : 1), care being taken to wash the fat thoroughly. The filtrate, which may be slightly turbid, is treated with 10 ml. of solution (iii) and about 3 ml. of (iv), and is titrated with solution (i), the end-point being sharp: 1 ml. of 0·01 *N* iodate ≡ 1·66 mg. of potassium iodide.

With practice the titration may be carried out, although not so easily, in presence of the fat. The end-point is then reached when a drop of the iodate solution just decolorises the aqueous layer, subsequent appearance of a blue colour being neglected. Fresh edible fats give deterioration values of about 3 to 5, whilst, for fats which have become rancid and nauseous, values up to 20 may be obtained. In judging the condition of fats, these values are considered in conjunction with the acidities and the Kreis values. T. H. P.

New Colour Reaction of Quinine, Quinidine and Cupreine and its Application to the Determination of Quinine. **J. A. Sanchez.** (*J. Pharm. Chim.*, **1935**, **21**, 24–32.)—This method overcomes the difficulty of the lack of a selective solvent for quinine, and also avoids the complicated technique of the B.P. (1932) method. It is based on the fact that since quinine is the methyl ether of a phenolic alkaloid (cupreine), the latter may be produced by demethylation and coupled with a diazo-compound to form a coloured substance. The material (0·1 g.) is macerated with 0·2 g. of powdered lime and 2 ml. of water, the mixture is dried on the water-bath, and the residue is ground and heated for 10 minutes, with shaking, in a tube containing 5 ml. of boiling chloroform. The mixture is filtered, the residues being washed with chloroform, and the filtrate is evaporated, the new residue being warmed with 30 drops of 30 per cent. sulphuric acid and 2 ml. of water, and the mixture filtered when cold. The residue is washed with water until the total filtrate amounts to 10 ml. A tube containing 1 ml. of this filtrate is immersed in glycerin at 120° C. until no water remains, when the temperature is raised to 180° C. and maintained there for 5 minutes. The tube is cooled, and 2 ml. of water and 1 ml. of a fresh mixture of 10 ml. of a saturated solution of *p*-nitraniline in 1 per cent. sulphuric acid and 1 drop of 10 per cent. sodium nitrite solution are added. The mixture is shaken and made alkaline with 10 drops of 30 per cent. sodium hydroxide solution, after which it is acidified with 10 drops of 30 per cent. sulphuric acid and shaken with 2 ml. of 95 per cent. alcohol. The resulting orange colour is matched against that obtained from a solution of pure quinine sulphate in water (0·145 per cent. $C_{20}H_{24}N_2O_2,3H_2O$; 1 ml. ≡ 0·001 g. of quinine). The standards are prepared by adding to 10 tubes containing 0·1, 0·2, etc., to 1·0 ml. of quinine solution, respectively, 3 drops of the sulphuric acid and sufficient water to make 1 ml. These mixtures are then evaporated on a glycerin-bath and the colour is produced as described above. If a tincture is to be tested,

1 ml. is mixed with 0·2 g. of lime and 10 drops of water, the mixture is evaporated, and the above procedure is followed; fluid extracts should first be diluted with alcohol. The method is also applicable to quinidine (after demethylation), and, if it is necessary to separate this compound from quinine, it may be precipitated by means of a solution of an alkali iodide from the residue obtained after evaporation of the chloroform. Cupreine is determined directly, *i.e.* without demethylation. J. G.

New Adulterant of Cocaine. E. Collard. (*J. Pharm. Chim.*, 1935, **127**, 57–60.)—A sample of cocaine, m.p. 94° C., was found to contain about 33 per cent. of an insoluble substance which, on heating, sublimed with partial decomposition and emitted the odour of benzoic acid. It proved to be ethyl *p*-amino benzoate (anaesthesine), which, according to the German Pharmacopoeia, melts at 90°–91° C., although a sample used for comparison had m.p. only 88° C. A solution of anaesthesine in dilute hydrochloric acid differs from one of cocaine in that it does not give a precipitate with iodine in potassium iodide solution, but resembles it in being precipitated by picric acid. If sufficiently dilute solutions are used and precipitation is brought about slowly, crystals shaped like palm leaves are produced in both cases, but the two picrates differ in appearance (illustrations are given); in stronger solutions the tufts of crystals are very similar. D. G. H.

Determination of Certain Local Anaesthetics derived from Amino-Alcohols. F. and J. Girault. (*J. Pharm. Chim.*, 1934, **126**, 584–586.)—The anaesthetic is determined by precipitating the amino alcohol base by ammonia, extracting with ether, and washing and shaking with excess of acid. The excess of acid is isolated and titrated in the presence of methyl red, and the quantity of anaesthetic is determined from the proportion of combined base found by difference. About 0·001 of the molecular wt. of the anaesthetic is weighed, and washed into a separating funnel with the smallest possible amount of water. Fifty ml. of ether are added, and then, after shaking, 0·5 ml. of ammonia (French off.), followed by 10 minutes' shaking. The water fraction is drawn off into another funnel, 30 ml. of ether added, and, after shaking as before, the water is withdrawn. The ammonia is removed from the united ethereal extracts by washing with water at least four times, using 10, 5, 5, and 5 ml. respectively, and 20 ml. of 0·1 *N* hydrochloric acid are added. After shaking for 10 minutes the aqueous acid layer is withdrawn, and the ethereal solution washed three times with 10, 5, and 5 ml. of water, and the washings are added to the acid. Ether is driven off by heating the acid solution, and the excess of acid is titrated. Detailed examples are given for stovaine, novocaine, and larocaine. D. G. H.

Carbohydrates of Tobacco and their Significance. C. Pyriki. (*Z. Unters. Lebensm.*, 1934, **68**, 554–566.)—Most of the tobaccos examined—of Oriental, overseas and German origin—contain below 0·5 per cent. of invertible sugar, the maximum found being 0·9 per cent. The tobacco extracts obtained contained large proportions of salts, and the sugars could not be isolated free from these.

By comparison of the optical activity with the reducing power, by examination of the phenylosazones, by the reaction with bromine, and by colour reactions, the sugars were, however, identified as dextrose and laevulose, the latter being in excess. Most of the Oriental tobaccos contained relatively large proportions of sugars, the better qualities showing from 5·5 to 13, and the poorer ones from 0 to 4 per cent. German and cigar tobaccos proved almost free from sugar. Oriental tobaccos rich in sugar contained comparatively little nicotine, and those with higher nicotine-contents smoked mild; the varieties with no or very little sugar were usually strong and bitter. The sugar-content had a marked influence on the acidity of the smoke from Oriental tobaccos, but is not regarded as the sole cause of the acid reaction. Near the stalk of the leaf the amount of sugar is greater than near the tip, and the distribution of nicotine in the leaf is related inversely to that of the sugar. The starch-content of the tobaccos examined varied from 1·1 to 8·3 per cent., and with Oriental tobaccos, those free from sugar contained but little starch. In general, the percentage of pentosans present ranged only from 3·8 to 4·9, but German tobaccos showed from 5·5 to 6·4 per cent. No relationship was observable between starch- and pentosan-contents and the quality of the tobacco. T. H. P.

Colorimetric Test for Quillaia Saponin. J. Rae. (*Pharm. J.*, 1935, **134**, 59.)—If 10 ml. of a 1 per cent. solution of quillaia saponin are treated in a Nessler glass with 1 ml. of 10 per cent. sodium nitrite solution and 2 drops of sulphuric acid, followed, after 30 seconds, by 20 ml. of *N* sodium carbonate solution (a few drops of ether being used to prevent frothing), a yellow colour, reaching its maximum intensity in 5 minutes, is produced, and the depth of colour is proportional to the amount of saponin present. The reaction was positive with 5 different samples of quillaia bark saponin, but negative with a sample of unknown origin and with digitonin, which were also negative to Mitchell's test (ANALYST, 1926, **51**, 181). So far, the test is not directly applicable to quillaia bark, owing to the interference of substances extracted with the saponin, and preparations of senega give a wine-red colour, but saponin used as a denaturant in toilet and perfumery preparations made with industrial spirit may be quantitatively determined. One per cent. of saponin was added to petroleum emulsion, and the amount determined colorimetrically was 0·92 per cent. Phenols and other substances reacting with the reagents used must be absent. D. G. H.

Saponin from Soya-bean. R. C. Burrell and E. D. Walter. (*J. Biol. Chem.*, 1935, **108**, 55–60.)—The saponin was extracted by refluxing 4 kg. of finely ground soya-bean meal with 5 l. of 80 per cent. alcohol, at 50–70° C., in a specially-made copper boiler. From the extract the solvent was removed by distillation, and the heavy liquid remaining was dialysed in an electro-dialyser with 200 ml. of water at 100 volts for 24 hours. The supernatant liquid was siphoned off, and the precipitate was collected on a filter and dried on a steam-bath. The dried residue was extracted with ether in a Pickel extractor for 24 hours. The residue from the extraction was dissolved in 70 per cent. alcohol, and the solution was clarified with "Darco," and evaporated to about half its volume. From this

solution, crystals of the saponin separated. When first removed from the mother liquor they had the form of micro-rosettes, but, after being dried over sulphuric acid they became very thin plates. These crystals melted at 220–225° C. with decomposition, were soluble in methyl and ethyl alcohols, in acetone and in caustic alkali, giving solutions which foamed on shaking. An average analysis was C = 59·24 per cent. and H = 8·52 per cent. with a molecular equivalent of 769. The saponin on long-continued hydrolysis yielded a sapogenin, which probably contained a terpene grouping, galactose and possibly rhamnose. It is suggested that it is probable that several of the recently reported saponins from soya-bean are identical, and also that the hydroxy acid reported by Muramatsa (*Bull. Imp. Coll. Agric. and Forestry*, 1924, **7**, 31) was a difficultly hydrolysable, insoluble, acid saponin. S. G. S.

Sodium Dinitrophenate. J. C. Bird, Z. Panciera and E. G. E. Shafer. (*Amer. J. Pharm.*, 1934, **106**, 462–466.)—To determine the water of crystallisation in sodium dinitrophenate, 1 to 2 g. of the sample are heated in an air-oven at 160–170° C., for at least 2 hours, or until constant in weight. Percentage loss was found to be 8·05 per cent., corresponding with 1 H_2O. Sodium may be determined by applying heat in such a way that the entire surface of the crucible is heated gently and uniformly, otherwise simple ignition invariably results in explosion. Methods of reduction and precipitation involve complicated procedures. The sample taken for the water determination may also be used for the determination of sodium (provided that a platinum crucible or dish is used), and the dehydrated material is moistened with about 5 ml. of concentrated sulphuric acid, and placed in a "chimney heater." The temperature is gradually raised until the material is charred and there is no further tendency to "creep," after which heating is continued over a small open flame, and, finally, the material is strongly ignited until a pinkish-white residue is obtained. This is cooled and moistened with sulphuric acid, and, after further heating and cooling, the anhydrous sodium sulphate is weighed. An average of 10·29 per cent. of sodium was found (the calculated figure being 10·27), allowing for one mol. of water. The "chimney heater" used was made from a 5-lb. ether can, with the top removed, a nichrome triangle to support the crucible being inserted about midway up by running the ends through holes in the sides of the can. When heated, yellow sodium dinitrophenate becomes increasingly red, fusing to a deep purple or black mass; if heating is rapid, explosion follows immediately upon fusion. If heated slowly, the material appears to "boil," and explosion eventually follows with even greater violence. D. G. H.

Two New Crystalline Principles from Indian Species of Artemisia (from the Laboratories of Messrs. T. and H. Smith, Ltd.). (*Pharm. J.*, 1935, **134**, 3–5.)—Two new crystalline principles, distinct from santonin or artemisin, have been found in species of *Artemisia* collected on the N.W. frontier of India. These were isolated in the ordinary course of an assay for santonin and the names pseudo-santonin and *k*-santonin were suggested. Clemo (*J. Chem. Soc.*, 1934, 1343) has found that *k*-santonin is a laevo-isomer of santonin, but pseudo-santonin

has not yet had its constitution determined. The following table shows the relationship existing between these compounds:

	True santonin	*k*-santonin	Pseudo-santonin	Artemisin
M.P.	172° C.	216–218° C.	184–186° C.	200° C.
Specific rotation	−172·5° C.	−140° C.	−172·5° C.	−84·3° C.
Sensitivity to light ..	Very sensitive	Less sensitive	Insensitive	Less than santonin
Alcoholic potash ..	Carmine-red	Pale carmine-red	Brownish-yellow	Pale carmine-red
Sulphuric acid conc. ..	No colour	No colour	Dark brown	No colour
Solubility in:				
Alcohol (90 per cent.) ..	1–50	1–116	1–15	—
Ether (0·720)	1–140	1–625	1–312	—
Chloroform	1–2·5	1–4·2	1–4·2	Forms a compound
Boiling water	1–416	1–590	1–45	1–60
Cold water	Almost insoluble	Almost insoluble	1–300	—

S. G. S.

Oxidation of Rotenone by Copper in an Alkaline Medium. R. M. Whittaker and I. Glickmann. (*Rec. Trav. Chim. Pays-Bas*, **1934**, **53**, 1145–1150.)—This reaction of rotenone serves for its determination in alcoholic solution, but is not found adaptable to the assay of plant material for rotenone-content. With derris-root extracts, for example, other reducing substances present cause high results. It may, however, be possible to establish an arbitrary relationship between the reducing values in terms of dextrose and the toxic powers of such extracts. The reagents required are: (i) Aldehyde-free alcohol. (ii) Alkaline copper solution: Dissolve 2·5 g. of pure copper sulphate ($+5H_2O$) in about 100 ml. of warm water, cool, add a solution of 5 g. of pure sodium potassium tartrate and 7·5 g. of sodium hydroxide in 100 ml. of water, and make up to 500 ml. This should not be used after it is three days old. (iii) Folin's reagent: Dissolve 150 g. of sodium molybdate in 300 ml. of water, filter the solution through a small quantitative paper and wash the residue with about 75 ml. of water, add 0·1 to 0·2 ml. of bromine and shake till this is dissolved. After an hour, add, with shaking, 225 ml. of 85 per cent. phosphoric acid, and then 150 ml. of dilute sulphuric acid (1+3). Remove the free bromine by means of a current of air, add 75 ml. of 99 per cent. acetic acid, and make up to 1 litre. (iv) Dissolve 1 g. of pure, anhydrous dextrose in water, add 40 ml. of aldehyde-free alcohol and dilute to 200 ml. with water; 10 ml. of this stock solution and 210 ml. of aldehyde-free alcohol are made up to 250 ml. with water.

Standard rotenone solutions are prepared from the commercial product, first recrystallised several times from alcohol, dried, and stored over calcium chloride. Ten ml. of the standard rotenone solution are treated with 6 ml. of reagent (ii) in a test-tube, which is kept for 45 minutes in a bath at 78° C., and then for 3 minutes in a bath at 20° C. Ten ml. of reagent (iii) are added and, after 3 minutes, the solution is diluted to 90 ml. with water, filtered through a thick asbestos pad in a Gooch crucible, and diluted to 100 ml. This solution is at once compared colorimetrically with the solution obtained from 10 ml. of the diluted dextrose standard in a similar manner, except that this need not be filtered.

The relation between the amounts of rotenone and dextrose is found to be: $Y = 0{\cdot}6195 - 0{\cdot}006858X + 0{\cdot}008371X^2$, Y and X being the numbers of mg. of dextrose and rotenone, respectively. Results accurate to about 0·2 per cent. are thus obtainable. T. H. P.

Organic

Reactions and Reagents for the Detection of Organic Compounds. III. E. Eegriwe. (*Z. anal. Chem.*, 1935, **100**, 31–36; *cf.* ANALYST, 1932, **57**, 584.)—*Glycerol.*—Two drops of an aqueous solution to be tested for glycerol are placed in a test-tube, which is then filled with bromine vapour, covered with a small funnel with sealed-up end, and heated for 10 minutes in a water-bath at 85° to 90° C., and, after removal of the funnel, for 10 to 15 minutes longer. Any residual trace of bromine is rendered ineffective by addition of a small crystal of sodium sulphite. A little concentrated sulphuric acid is poured down the wall of the inclined tube, which is meanwhile, and during the subsequent mixing, kept cool under the tap. Further sulphuric acid is added to give a total volume of 2 to 3 ml., a little solid *m*-hydroxybenzoic acid is introduced, and the tube is swirled and then left for 10 to 15 minutes in a water-bath at 65° to 70° C. The presence of glycerol is shown by the appearance of a green fluorescence, which appears even with one drop of solution containing 0·005 mg. of glycerol. The fluorescence obtained with 0·024 mg. of glycerol is apparent in presence of twice as much sucrose or six times as much glycol or dextrose; laevulose has a somewhat greater disturbing action.

Allyl Alcohol.—This alcohol (0·003 mg.), which also gives dihydroxyacetone on oxidation, may likewise be detected as described above.

Glyoxylic Acid.—One drop of the aqueous solution is mixed in a test-tube with a little solid pyrogallol-4-carboxylic acid, and one or two drops of concentrated sulphuric acid are poured down the wall of the inclined tube, which is kept cool. The volume is made up to 0·5 to 0·75 ml. with the sulphuric acid, and the tube is heated for 30 minutes in a bath at 40° C. A blue colour is formed with as little as 0·001 mg. of glyoxylic acid in one drop of the aqueous solution.

Oxalic Acid.—As this is reducible to glyoxylic acid, the previous test may be applied. To one drop of oxalic acid solution (or one drop of a solution of calcium oxalate in dilute sulphuric acid) in dilute sulphuric acid is added a small quantity of powdered magnesium, and when this has dissolved the pyrogallolcarboxylic acid reaction is used. The blue colour appears with 0·02 mg. of oxalic acid in one drop of solution. This reaction is not given by the near homologues of oxalic acid or by pyruvic, laevulinic, or acetonedicarboxylic acid, but is shown by mesoxalic (not precipitable by calcium sulphate solution) and dihydroxytartaric acids. T. H. P.

Action of Nessler's Reagent on some Ketonic Alcohols and Ketonic Acids. G. Schuster. (*J. Pharm. Chim.*, 1935, **21**, 32–43.)—The following results were obtained:—*α-Ketonic Alcohols.*—Dimethylacetol: oxidation is complete after 15 minutes, the oxygen consumed being equivalent to 2 atoms of iodine. Dihydroxyacetone: 90 minutes, 8 atoms. Propionin: 4 hours for total oxidation (*i.e.* of the secondary alcohol and of the diacetone), 4 atoms. Butyroin, 90 minutes.

Iso-butyroin, rapid oxidation on the water-bath, 4 atoms. Capronoin, 90 minutes, 4 atoms. Hydroxyacetophenone (in methyl alcohol containing hydrochloric acid), 15 minutes, 8 atoms. Benzoin (in methyl alcohol), 15 minutes below 10° C.; dibenzyl is produced. *β- and γ-Ketonic Alcohols.*—Chloralacetophenone, chloralpropanone and benzoylacetone are not oxidised, but a green precipitate of mercurous salts is formed and is accompanied by what is probably a product of the transformation from the ketonic to the enolic form, with the occurrence of a double-linkage. Acetonylacetone produced a yellow precipitate similar to that obtained with all the ketones, but there was no evidence of reduction of the reagent. *α-Diacetones and α-Ketonic Acids.*—Diacetyl and dibenzyl are oxidised slowly at 20° C., but dibutyryl is oxidised only on the water-bath, and then very slowly. Pyruvic acid, ethyl-phenylglyoxylate and ethyl-aceto-acetate are not oxidised, although the yellow precipitate is produced. Nessler's reagent may therefore be used to determine the α-ketonic alcohols (if these are allowed to react at the temperature of the water-bath), the final procedure being to acidify the mixture with hydrochloric acid, and to add 20 ml. of a 0·1 *N* solution of iodine, the excess of which is titrated with 0·1 *N* sodium thiosulphate solution (*cf.* Bouveault, *id.*, 1906, **35**, 629; Bougault and Gros, ANALYST, 1922, **47**, 405). Methods of preparation and reaction-rate data are given in each case. J. G.

Detection of Diazonium Salts and Primary Amines by means of Resorufin. H. Eichler. (*Z. anal. Chem.*, 1934, **99**, 348–350.)—To test for diazonium salts, the filtered, slightly acid liquid is added to 5 to 10 ml. of water rendered distinctly fluorescent by adding a solution of 0·2 g. of resorufin and 0·2 g. of sodium carbonate in 100 ml. of water. The acid liquid is then made alkaline with sodium carbonate. If a diazonium salt is present, the liquid becomes brown and non-fluorescent; otherwise, the red fluorescence of the resorufin persists unchanged. In some cases formation of the colouring matter with the resorufin proceeds slowly, addition of ammonium chloride or sulphate or replacement of the sodium carbonate by the bicarbonate or ammonium carbonate then being advisable. Diazotised toluidines are best detected in ammoniacal solution. In all cases the reaction is hastened by heating the liquid. Any metal salts present must be removed by sodium acetate or a small quantity of sodium carbonate before the above test is applied. Diazotates are first treated with dilute hydrochloric acid to convert them into the corresponding diazonium salts. Under the above conditions, fluorescein forms sparingly soluble, non-fluorescing dyes only with certain diazonium salts, *e.g.* those of *p*-toluidine. This behaviour may be utilised to detect and identify such amines or diazonium salts in presence of substances which yield no dyes with fluorescein.

Nitrous acid or nitrites may be detected by the author's Magdala red method (ANALYST, 1934, **59**, 303). Diazonium salts should first be destroyed by gently heating the faintly acid liquid, where possible in presence of added copper salt or alcohol. Any mineral acid present should be annulled by addition of sodium acetate.

To detect primary amino groups in aromatic compounds, the hydrochloric acid solution is diazotised by sodium nitrite at 0° to 10° C., and the resorufin test

then applied to the diazonium chloride formed. Prior to diazotisation, difficultly soluble aminocarboxylic acids may be obtained in finely divided form by dissolving them in alkali and precipitating with hydrochloric acid. Primary amino groups in readily diazotised organic dyestuffs may be detected similarly. Dyestuffs of low solubility may be dissolved in concentrated sulphuric or glacial acetic acid and precipitated in finely divided form with ice-water. Dyestuffs on fibres are extracted by means of water, glacial acetic acid, dilute acids or sodium carbonate solution. T. H. P.

Brazilian "Cedro" Wood Oil. F. W. Freise. (*Perf. and Ess. Oil Record*, 1935, **26**, 11–12.)—Brazilian "Cedro" wood yielding essential oil may be derived from the trees of the N.O. *Meliaceae*, genus *Cedrela*; *C. odorata*; *C. angustifolia*, D.C., *C. montana* Karst., *C. macrocarpa* Ducke, *C. fissilis* Vell., *C. australis* Juss., St. Hil., and *C. glaziovii* D.C., the frequency of their occurrence being in the order cited. The roots of old and very strong individual trees contain from 2·0 to 3·5 per cent., and yield 1·6 to 3·2 per cent. of oil, whereas chips contain 0·8 to 1·8 per cent., and yield 0·5 to 1·4 per cent. of oil. Sawdust yields only 0·08 to 0·2 per cent., and leaves when fresh may give 0·03 to 0·05 per cent. of oil. Exceptionally, however, the yields may be considerably higher. The fruit capsules of *C. macrocarpa* and *C. glaziovii*, in the fresh half-green state, contained 0·04 to 0·085 per cent. of oil, but practically none when ripe; the seeds contain only a very small proportion of a fixed oil. Crude commercial "Cedro" wood oil, which is extracted with steam, is of a bluish colour, but so far no such colour has been observed in any oils obtained in the laboratory. The crude oil has a sp.gr. at 20° C. of 0·900 to 0·965°, whilst the optical rotation, which is positive or negative, ranges from +6° 35′ to −4° 25′. Solubility is complete in 6 vols. of 90 per cent. alcohol. Figures obtained for various samples extracted in the laboratory were as follows:

Original plant and part employed	Yield on dried material Per Cent.	Sp.gr. 20°/4° C.	$\alpha^{20°}$	$n_D^{20°}$	Acid value	Ester value	Main constituents
C. fissilis, young roots	3·37	0·933	−11° 15′	1·4985	1·55	26·5	Cadinene, cineol, α-pinene
,, woodchips	2·86	0·922	− 5° 35′	1·5055	2·51	38·3	ditto; pinene in traces
,, leaves	0·13	0·896	− 2° 50′	1·4826	1·82	41·5	Camphene
C. odorata, fresh sawdust	0·22	0·911	−13° 30′	1·5210	Not determined	N.D.	
,, raspings	0·31	0·921	− 9° 30′	1·4955	1·93	38·8	Cadinene, borneol
C. montana, roots	2·72	0·944	−14° 10′	1·4474	2·11	44·4	
,, shavings	1·88	0·931	Not determined		3·13	38·3	Cadinene, α-pinene
,, old chips	0·92	0·915	Not determined		2·83	44·5	Pinene, cineol in traces
C. macrocarpa, capsules	0·09	0·927	− 4° 35′	1·5228	1·06	26·8	Camphene, α-pinene

All these oils were soluble in 4 vols. of 90 per cent. alcohol, and were of a clear yellow colour, except that the oil from *C. odorata* gave a slightly opalescent alcoholic solution, and the oil from leaves and capsules had a greenish tinge. D. G. H.

Biochemical

"Digestibility" of Common Foodstuffs as Determined by Radiography. W. C. D. Maile and K. J. L. Scott. (*Lancet*, 1935, 21–23.)—X-ray screening methods were applied, 1 ounce of "barium," mixed either with the food or with a little water which was taken with the food, being used. It was found that an ordinary meal leaves the stomach in about 4 hours, and a large meal in 5 hours; if the meal contains much fat (especially as butter or cream) it may be retained much longer, even if the quantity is not unduly large. Concentrated carbohydrates (*e.g.* sugar) leave the stomach more rapidly than natural carbohydrates (*e.g.* banana or potato). With some foods (*e.g.* milk) cooking shortens the time of stomach digestion, but with others (*e.g.* eggs) the time is lengthened. The period of stomach digestion of different foods cannot be taken as a measure of their "digestibility," and, since the sensation of hunger does not necessarily result from an empty stomach, there is no support for the view that it is due to vigorous peristaltic contractions of the stomach. In general the figures indicate a rather longer emptying-rate than "digestibility" tables based on tube experiments (*cf.* Rehfuss and others, *Amer. J. Physiol.*, 1919, **49**, 174, 204, 222, 254; 1920, **51**, 332; 1920, **52**, 1, 28, 248). This may be explained by the fact that barium delays the emptying of the stomach or is left as a coating to the lower part of the greater curvature after the food has passed through the pylorus; a more likely explanation is that the presence of a Rehfuss tube in the stomach acts as an irritant, and causes exaggerated peristalsis.

J. G.

Influence of Freezing Temperatures on Haddock's Muscle. G. A. Reay. (*J. Soc. Chem. Ind.*, 1934, **53**, 413–416T.)—The freezing and thawing of muscle are accompanied by a partial irreversible physico-chemical breakdown of muscle-plasm, with increased insolubility of the proteins. The process is also associated with an increase in the amount of fluid that can be easily expressed from the muscle (termed "drip"). Minced haddock muscle (149 g.) was packed into circular tinned cans (9·6 × 2 cm.), and the ends were hermetically sealed on. After freezing and thawing, the ends were removed and pads of filter paper were applied to the flat surfaces, close contact being assured by means of a lead plate weighing 0·5 lb. placed on top of the paper. After standing 17 hours at 5° C. the cake of muscle was removed, and the loss in weight determined. The maximum formation of "drip" occurred between −1° and −3° C., and the quicker the freezing and thawing the less the drip. The amount of the various kinds of change brought about by freezing is closely dependent on the time spent within the zone of maximum change, −1° to −5° C., during any complete temperature time-cycle. The increasing insolubility of the proteins, brought about as a result of freezing and thawing, is not held to be the cause of the "drip" which develops from one hour's exposure to temperatures in the critical range, −1° to −5° C. (accounting for one-third of the total "drip" obtainable by the longest exposure at the temperature range, whereas measurable denaturation does not occur until after 7 hours' exposure). The secondary slow increase in "drip" with a period of exposure up to 10 days or so may, however, be attributable

to the corresponding denaturation development. The initial rapid formation of "drip" is regarded as due to the structural changes brought about by the formation of ice. D. G. H.

Use of Tartrazine in the Determination of Chlorides in Biological Material. W. R. Fearon and W. A. Gillespie. (*Biochem. J.*, 1934, **28**, 1629–1630.)—Chlorides may be determined in protein-free solutions, such as sea-water and normal urine, by mixing 10 ml. of 0·1 *N* silver nitrate solution with five drops of pure nitric acid and 2 to 5 drops of 0·5 per cent. aqueous tartrazine solution. The solution under analysis is used to titrate this mixture, 0·1 to 0·5 ml. being added at a time, with shaking. The precipitate is at first buff in a colourless solution. The end-point occurs when the precipitate becomes white and the solution greenish-yellow. If only a small quantity of liquid is available, one ml. may be added to the acid solution of silver nitrate and indicator, and the mixture titrated with 0·1 *N* hydrochloric acid. Proteins affect the solution colour and, if they are present, the second method should be used. Proteins also affect the silver nitrate solution by altering its chloride value, and, if possible, should be removed by coagulation or precipitation. S. G. S.

Influence of Antiseptics on Yeast Autolysis. H. Haehn and H. Leopold. (*J. Inst. Brewing*, 1935, **41**, 44.)—If yeast is incubated at 50° to 52° C., no development of foreign organisms takes place, even in the absence of antiseptics. At this temperature autolysis occurs, the whole liquefying in 24 hours. This was allowed to proceed for 21 days, both alone and with 5 per cent. of toluene, chloroform, and ethyl acetate, respectively. The *p*H value, titratable acidity, inorganic phosphates, total soluble nitrogen, and formol and ammoniacal nitrogen were determined at intervals, the results indicating that proteolysis and the formation of acid products and inorganic phosphates are retarded more by chloroform and ethyl acetate than by toluene, but that the last has a retarding effect on the increase in titratable acidity and the fall of the *p*H value. Toluene has but a slight effect on proteolysis. Ethyl acetate is hydrolysed to some extent, but the acetic acid formed also retards autolysis. S. G. S.

Detection of Volutin in the Living Yeast Cell by means of Neutral Red. L. Heucke and W. Henneberg. (*Woch. Brau.*, 1934, **90**, 425; *J. Inst. Brewing*, 1935, **41**, 43.)—The volutin in yeasts, moulds and bacteria was rendered visible by staining with neutral red. One drop of a 0·001 per cent. aqueous solution of neutral red was mixed with a drop of yeast suspension on the microscope slide. Dead cells were stained uniformly throughout, but in the living cells the colour was absorbed by the volutin globules, which were situated both in the vacuole and in the protoplasm. The vacuoles (but not the protoplasm) gradually became pale red and then dark red. Those cells having red vacuoles or volutin died after a short time. When specimens of living organisms prepared by this method were compared with Löffler-blue formaldehyde preparations, cells from a fresh fermentation being used for both, it was found that in the living preparations the volutin was in the form of one large globule (resting form), whereas in the other preparation the volutin consisted of many small globules (working form). Yeast from old agar cultures gave the resting form by both methods. When living

yeast spores were stained with neutral red, one volutin globule in each spore was rendered visible. Wort-agar containing 0·001 to 0·005 per cent. of neutral red was a satisfactory medium for the growth of yeast, even though many cells contained stained volutin; but the presence of the dye in a concentration of 0·01 per cent. inhibited the growth of beer yeast. S. G. S.

Determination of Sodium in Human Red Blood Cells. F. W. Oberst. (*J. Biol. Chem.*, 1935, **108**, 153–160.)—The sodium-content of red blood cells was determined by drawing venous blood into a 50-ml. centrifuge tube, treating it with heparin (0·1 ml. of a solution of 75 mg. of heparin in 5 ml. of water per 5 ml. of blood and evaporating off the water), and centrifuging for 30 minutes, at 3000 R.P.M. The plasma with 0·5 ml. of cells was removed by suction and the remaining cells thoroughly mixed. Some cells were analysed unwashed, and others were washed with isotonic potassium chloride solution, or with sodium-free dialysed serum made isotonic with potassium salts and equilibrated with approximately 40 mm. of carbon dioxide. Protein was removed either by treatment with trichloroacetic acid or by ashing, and phosphates were removed by means of powdered calcium hydroxide. The sodium-content was determined colorimetrically by the sodium-zinc-uranyl acetate method. Tables are given showing the effect of the various treatments on the sodium-content, the average value of which was 16·6 mg., with a range of from 14·1 mg. to 20·3 mg., per 100 g. of cells. S. G. S.

Method for the Determination of Sucrase Activity. J. B. Sumner and S. F. Howell. (*J. Biol. Chem.*, 1935, **108**, 51.)—The definition, by Willstätter, of a unit of sucrase activity based on the time necessary for the rotation of a sucrose solution to be reduced to 0, is criticised, and a new definition and method of determination are suggested. The suggested unit is based on the number of mg. of invert sugar formed in 5 minutes at 20° C., at *p*H 4·5. This may be determined by placing 5 ml. of 6·5 per cent. sucrose in an acetate buffer solution in a test-tube and heating the mixture to 20° C. in a thermostat. One ml. of sucrase solution or yeast suspension at 20° C. is added from a Folin–Ostwald pipette which delivers rapidly, and the solutions are mixed and kept in the bath for 5 minutes, after which 5 ml. of 0·1 *N* sodium hydroxide are added. The invert sugar in the mixed liquids is determined in 1 ml. by the dinitrosalicylic acid method (*J. Biol. Chem.*, 1925, **65**, 393), a 1 mg. glucose standard being used. The number of mg. of invert sugar, multiplied by 11, gives the number of sucrase units per ml. of enzyme solution. If more than 1 mg. or less than 0·5 mg. of invert sugar is found per ml., the analysis must be repeated with an adjusted amount of sucrase solution or a longer or shorter time for digestion. Yeast preparations containing sugar must be washed before being tested. The sucrose and acetate buffer solution is prepared by mixing 43 ml. of *N* sodium acetate solution with 57 ml. of *N* acetic acid and diluting to 1 l. with water redistilled from glass. If 6·5 g. of sucrose are dissolved in 96 ml. of this solution and a few drops of toluene added, the result will be a product which has *p*H 4·5, and will remain serviceable for 3 days. Glucose is used for the sugar standard because it gives the same reducing value as invert sugar by the dinitrosalicylic acid method. S. G. S.

Research on Vitamin *A* in Animal and Plant Cells. P. Joyet-Lavergne. (*Compt. rend.*, 1935, **200**, 346.)—The well-known antimony trichloride reaction for vitamin *A* has been applied to living cells from animal and vegetable sources. In sporozoite forms the blue colour was localised in the elements of the chondriome. In all cells examined the chondriome gives the blue colour, but the intensity of this may vary from cell to cell. Hepatic cells give a strong colour, and the salivary gland cells of *Chironomus* gave a poor colour. Between these extremes a wide range of intensity was obtained. Studies on species of *Coccidia* and *Gregarina* revealed the fact that the chondriome conserves its vitamin *A* even throughout the varying morphology of the animal's life cycle. Vitamin *A* was also found in the cytoplasm (outside the chondriome) in the liver cells of certain salt-water fish. No vitamin *A* was found in the nucleus unless a nucleole was present. It is suggested that in plant cells the reaction is given by carotene, $C_{40}H_{56}$, but that in animal cells it is the true vitamin, $C_{20}H_{30}O$, that reacts. S. G. S.

Influence of the Solvent on the Biological Assay of Vitamin *A*. K. C. Lathbury and G. N. Greenwood. (*Biochem. J.*, 1934, **28**, 1665–1673.)—In the biological assay of carotene and vitamin *A* concentrates the values found may vary with the oil used, and the results may be very contradictory. Results of experiments on various batches of coconut and arachis oils, described in detail, show that the suitability of a particular oil can be determined only biologically, for the variation is independent of the stability of the dissolved substance in the oil. The carotene or vitamin *A* had not lost its activity as shown by colour tests, and yet the biological results were very variable. This is thought to be due to an unknown factor in certain oils. The addition of quinol will not render an inferior oil suitable for biological assay. S. G. S.

Properties of Carotenes from certain Roots and Leaves at Various Stages of Development. G. MacKinney. (*J. Biol. Chem.*, 1935, **108**, 45–49.)—Carotene was isolated in crystalline form, from carrot roots, the leaves of English ivy (*Hedera helix*), and coastal redwood (*Sequoia sempervirens*) at various stages of development. It was found that both α- and β-components may be present together, and that the ratio $\alpha : \beta$ showed no significant change with the degree of maturity of the organ containing them. The mixed crystals gave optical rotations $\alpha_{667\cdot8}^{20°}$ from $+59°$ to $+97°$ in benzene, and melted at 162° to 174·6° C. according to the proportion of each component present. S. G. S.

Concentration of Vitamin *G* (B_2) by Adsorption and Elution from Fuller's Earth. S. Lepkovsky, W. Popper and H. M. Evans. (*J. Biol. Chem.*, 1935, **108**, 257–265.)—Vitamin B_2 derived from fresh liver extracts is capable of being adsorbed on fuller's earth in neutral or strongly acid solutions, such adsorption being complete in 10 minutes (above *p*H 8·0 the adsorption is poor). The adsorbates may be eluted with diluted diethylamine (10 to 50 per cent. in aqueous solution) or diluted caustic soda solution (0·2 per cent. in water), with little loss of activity. S. G. S.

Erratum.—Dialysis of Milk. L. H. Lampitt and J. H. Bushill, abst. ANALYST, 1934, **59**, p. 828, line 12, *for* "250 ml." *read* "25 ml."

Inorganic

Solubility of Lead Sulphate in Water and Aqueous Sulphuric Acid. H. D. Crockford and D. J. Brawley. (*J. Amer. Chem. Soc.*, 1934, **56**, 2600–2601.)—The solubility of lead sulphate has been determined in pure water, and in sulphuric acid solutions containing up to 80 per cent. of the acid, at 0°, 25°, 35° and 50° C., with the following results:

Sulphuric acid Per cent. by vol.	Lead sulphate, p.p.m.			
	0° C.	25° C.	35° C.	50° C.
0·00	33·0	44·5	49·7	57·7
0·005	8·0	10·0	11·0	24·0
0·01	7·0	8·0	10·0	21·0
0·02	6·4	7·0	8·0	18·0
0·05	5·2	6·0	6·6	15·0
0·10	4·6	5·2	5·6	13·0
0·20	3·4	3·8	4·5	12·0
0·50	2·0	2·5	4·3	11·5
1·00	1·8	2·2	4·2	11·3
5·00	1·6	2·0	4·0	10·3
10·00	1·2	1·6	3·8	9·6
20·00	0·5	1·2	2·8	8·0
30·00	0·4	1·2	2·0	4·6
40·00	0·4	1·2	1·8	2·8
50·00	0·4	1·2	1·8	2·8
60·00	0·4	1·2	2·0	2·8
70·00	1·2	1·8	2·4	3·0
75·00	2·8	3·0	3·8	6·6
80·00	6·5	11·5	24·0	42·0

For the determination of lead sulphate the following methods were used, (*a*) in *pure water*; the lead was precipitated as chromate and then titrated iodimetrically according to the method of Kolthoff and Rosenblum (*id.*, 1933, **55**, 2656); (*b*) in *acid solution* (lead concentration less than in pure water): the lead was precipitated as chromate in 100-ml. Nessler tubes, and the turbidity produced was compared with standards prepared as nearly like the unknowns as possible; this method was found suitable for determining 0·01 to 0·15 mg. of lead sulphate. The solid phase in equilibrium with the solutions was in all cases normal lead sulphate. S. G. C.

Volumetric Determination of Copper. E. Voyatzakis. (*Bull. Soc. Chim.*, 1934, **1**, 1356–1357.)—The process is a modification of Weil's method, in which cupric salt is reduced in hydrochloric acid solution by stannous chloride. The author replaces stannous chloride by potassium chlorostannite, a known quantity of which is added to the copper solution, the excess being titrated with iodine. The reducing agent is prepared from 15·5 g. of potassium chloride and 14·9 g. of stannous chloride, both in saturated solution, and a little hydrochloric acid. If necessary, the liquid is concentrated and left to cool during agitation. For the determination of copper, the sulphate solution freed from nitric acid is diluted with an equal bulk of strong hydrochloric acid in a flask, and the air is expelled

by addition of sodium bicarbonate. An accurately weighed amount of powdered potassium chlorostannite is added, the cupric salt being immediately reduced to the cuprous state. The excess of stannous salt is at once titrated in the usual manner with iodine solution. If the concentration of copper in the solution is higher than 0·022 per cent., cuprous iodide is precipitated; hence the titration is carried out in dilute solution containing an equal volume of strong hydrochloric acid. Potassium chlorostannite is very stable and is easy to prepare in pure conditions (*cf.* Richardson, *Amer. Chem. J.*, 1892, **14**, 91). W. R. S.

Colorimetric Determination of Copper as Copper Sulphide. L. de Brouckère and S. Solowiejczyk. (*Bull. Soc. Chim. Belg.*, 1934, **43**, 597–625.)—In the absence of other metals which give coloured sulphides under the conditions employed, copper in solution may be determined by the addition of hydrogen sulphide water in the presence of 1 per cent. of gelatin as protective colloid. The method is applicable to solutions containing 0·003 to 0·03 g. of copper per l. and being 0·1 *N* to 0·5 *N* in sulphuric, hydrochloric, or nitric acid. The addition of the hydrogen sulphide water (2·5 ml. of saturated solution to 25 ml. of test solution) should be made simultaneously (using both hands) to the sample and to the standard solution of copper contained in colorimeter glasses, the colorimetric comparison being made after about 15 minutes with the aid of a Duboscq colorimeter. Results accurate within 1 per cent. were obtained in the presence of a large amount of the following salts added to the standard solution: alkali chlorides, nitrates and sulphates, the chlorides of magnesium, calcium and barium, and the sulphates of manganese, zinc and aluminium. Iron interferes. The results cited of test experiments occupy 19 tables. S. G. C.

Application of the Benzoate Method for the Separation of Iron, Aluminium, and Chromium in Qualitative Analysis. L. Lehrman and J. Kramer. (*J. Amer. Chem. Soc.*, 1934, **56**, 2648–2649.)—The recent method of Kolthoff, Stenger and Moskowitz (*id.*, 1934, **56**, 812; ANALYST, 1934, **59**, 435, 572) has been tested with regard to its use in qualitative analysis. Solutions containing 500 mg. of aluminium, tervalent chromium or tervalent iron, require respectively 90 ml., 60 ml. or 40 ml. of 10 per cent. ammonium benzoate reagent for complete precipitation after a 1-minute period of boiling. The bulk of the precipitate obtained with 500 mg. of the tervalent metals is rather large, but the precipitate can be contained in an 11-cm. filter paper, if filtered with the aid of suction. Two or three washings with 10 ml. of 5 per cent. ammonium nitrate solution remove practically all bivalent metal ions. One mg. of bivalent metal can be detected in the filtrate after precipitation of 500 mg. of any of the tervalent metals, alone or in combination. Before the filtrate is tested for the bivalent metals, the benzoic acid is removed by evaporating the acidified liquid until crystallisation begins, cooling, and filtering with the aid of suction. S. G. C.

Volumetric Determination of Niobium. J. A. Tschernichow and M. P. Karssajewskaja. (*Z. anal. Chem.*, 1934, **99**, 398–402.)—The authors have repeated the work of V. Schwarz (*Z. angew. Chem.*, 1933, **46**, 552), who reduced strongly acidified tartrate solutions by means of liquid zinc amalgam in complete

absence of air; the reduced niobium compound was re-oxidised with ferric chloride, and the excess of ferric salt titrated with titanous chloride. Schwarz claims to have obtained concordant stoichiometric results, which the authors were quite unable to reproduce; the results of their 26 published tests differ too much even for the calculation of an empirical factor. They conclude that Schwarz has not solved the problem of the volumetric determination of niobium, and they concur with Schoeller and Waterhouse (ANALYST, 1924, **49**, 215) in considering oxidimetric methods for niobium unreliable. W. R. S.

Determination of Molybdenum as Silver Molybdate. L. W. McCay. (*J. Amer. Chem. Soc.*, 1934, **56**, 2548–2549.)—The solubility of silver molybdate in water is 0·044 g. per l. (25° C.), but when a moderate amount of silver nitrate is present in the water, the solubility is practically nil. The following method was employed for the determination of molybdenum in alkali molybdates, the element being precipitated and weighed as silver molybdate:—The solution (150 ml.) is acidified with sulphuric acid, methyl orange being used as indicator; 1 g. of sodium acetate is added, the solution is heated to boiling, and silver nitrate is added; the liquid is allowed to cool, and is stirred from time to time while cooling. The precipitate of silver molybdate is yellowish-white and curdy. It is filtered off on an asbestos filter with the aid of suction, and washed seven or eight times with 4 to 5-ml. quantities of silver nitrate solution (0·5 per cent.), the silver nitrate being finally removed by washing three times with 5-ml. quantities of alcohol (96 per cent.). It is dried at 110° C., then heated at 250° C., to remove the last traces of water, and weighed as Ag_2MoO_4. During the drying the precipitate changes in colour through pink to purple. Various samples of ammonium molybdate gave results in close agreement with the ignition method. The method is stated to be easier and more rapid than the mercurous molybdate and lead molybdate precipitation methods. S. G. C.

Volumetric Determination of Potassium by the Cobaltinitrite Method. C. S. Piper. (*J. Soc. Chem. Ind.*, 1934, **53**, 392–396T.)—As a result of a study of the method, the following technique has been evolved, yielding results in close agreement with the theoretical with 0·1 to 50 mg. of potassium oxide. The solution is evaporated to dryness. After cooling, 1·5 ml. of glacial acetic acid and 10 ml. of saturated sodium chloride solution are added, followed, after 5 to 10 minutes, by 5 ml. of sodium nitrite solution (35 per cent.). The liquid is stirred until all soluble matter is dissolved. After 5 to 10 minutes, but not longer, 5 ml. of cobalt nitrate solution (20 per cent.) are added rapidly (in less than 2 seconds), with stirring, from a burette with a large jet. The liquid is stirred for 40 to 60 seconds and kept overnight. The precipitate is filtered off in a Gooch crucible with an asbestos filter, or a crucible of sintered glass (G4) or unglazed porcelain, and washed either (*a*) 5 times with 10-ml. portions of freshly-prepared approx. saturated solution of potassium cobaltinitrite (this solution does not keep for more than about 30 minutes), or (*b*) 3 times with 10-ml. portions of 35 per cent. alcohol, then 3 times with 3-ml. portions of cold water. The crucible and precipitate are placed in 150 ml. of water to which have been added 5 ml. of concentrated sulphuric acid and a volume of standard permanganate solution (0·05 *N* or 0·02 *N*)

about 5 ml. in excess of what the precipitate is likely to consume; the liquid is heated just to boiling and removed from the source of heat for 5 minutes; standard oxalic acid solution in slight excess is added, and the excess is back-titrated with standard permanganate solution. Eleven equivalents of oxygen are required for the oxidation of the cobaltinitrite molecule. The result is calculated with the aid of the empirical equation established by the author's work: K_2O (mg.) = permanganate value $\times$ 0·354 + (permanganate value)^2 $\times$ 0·00034; the "permanganate value" is the number of ml. of 0·05 *N* permanganate solution required in the titration. The composition of the precipitate was found by analysis to correspond with the formula $M_2Co(NO_2)_6$, where *M* represents *e.g.* sodium and potassium. The ratio of potassium to sodium varies according to the amount of potassium being determined, the amount and nature of the precipitating reagent, the amount of sodium salts present, and the temperature of precipitation. These variables are taken into account in the method of calculation adopted. Calcium, magnesium, iron, aluminium, sulphate, chloride, and phosphate do not interfere with the precipitation. S. G. C.

Microchemical

Volumetric Determination of Calcium in Serum. F. Rappaport and D. Rappaport. (*Mikrochem.*, 1934, **15**, 107–110.)—The calcium is precipitated from the serum (0·2 ml.) with ammonium oxalate, the precipitate is washed and dissolved in dilute sulphuric acid, and the liberated oxalic acid is oxidised with a measured quantity of 0·001 *N* ceric sulphate solution. The excess of cerium sulphate is then determined by titration with 0·001 *N* thiosulphate solution. *Reagents.*—(i) Cold saturated ammonium oxalate solution; (ii) 4 *N* sulphuric acid; (iii) a solution of 2 ml. of concentrated ammonia diluted to 100 ml.; (iv) cerium sulphate solution (0·001 *N*), made by dissolving 1 g. of the finely-powdered salt in 100 ml. of water containing 30 ml. of concentrated sulphuric acid, and diluting the solution to about 750 ml. (This solution is standardised against 0·001 *N* thiosulphate solution, diluted to approximately the correct volume, and again standardised; the solution will keep for a long time.); (v) 1 per cent. potassium iodide solution; (vi) 0·25 per cent. starch solution. *Method.*—The sample of serum (0·2 ml.) is accurately measured into a small centrifuge tube, treated with 0·5 ml. of the ammonium oxalate solution, allowed to stand overnight, and centrifuged (15 minutes at 3000 rev. per minute) after the addition of 1 ml. of distilled water. The precipitate is washed three times with 3 ml. of the ammonia solution, then dissolved in 0·5 ml. of the sulphuric acid on the hot water-bath, cooled, and treated with 2 ml. of the ceric solution. After 3 minutes a few drops of the potassium iodide solution and the indicator are added, and the liquid is titrated with thiosulphate solution. The results thus obtained agreed very closely with those given by determinations on the macro-scale. J. W. M.

Volumetric Micro-determination of Sugar in Blood (Plasma, etc.). F. Rappaport and R. Pistiner. (*Mikrochem.*, 1934, **15**, 111–113.)—The Hagedorn-Jensen method is used for 0·02 ml. of blood, with slightly smaller apparatus and amounts of reagents, and with a phosphate-buffered potassium

ferricyanide solution, as used by Akiji and Iwatake (*Biochem. Z.*, 1932, **242**, 43); this consists of (*a*) 0·9 g. of potassium ferricyanide dissolved in 1 l. of water, and (*b*) 21·0 g. of anhydrous potassium phosphate and 63·75 g. of tertiary potassium phosphate dissolved and diluted to 1 l. Equal parts of (*a*) and (*b*) are mixed just before use, and 2 ml. of the mixture are taken. For the titration 0·001 *N* thiosulphate solution is used in a micro-burette. J. W. M.

Colorimetric Micro-Determination of Caffeine. G. Denigès. (*Compt. rend.*, 1934, **199**, 1622–1623.)—The chloroform solution of the caffeine is evaporated to dryness at a low temperature in a porcelain casserole about 5 cm. in diameter, and the residue is treated with six drops of saturated bromine water and with a little *N* hydrochloric acid. By continuous circular movement of the vessel over the tip of a Bunsen flame, the liquid is evaporated to dryness, the residual film being similarly heated until it becomes completely orange-red (no trace of yellow). Ten ml. of water and 1 drop of 5 per cent. mercuric acetate solution (5 g. of the acetate dissolved, with heating, in 100 ml. of water containing 2 ml. of glacial acetic acid) are introduced and stirred until the residue is dissolved. The garnet-red liquid is transferred to a test-tube, 12 to 15 mm. wide, and its colour compared with those of standards similarly prepared and containing from 0·1 to 1·2 mg. of caffeine; such standards retain their colour well. If the mercuric acetate used is replaced by a zinc acetate solution of the same strength, the resultant colour is yellow and also serves for colorimetric comparison. T. H. P.

Mercurimetric Iodine Determination with Diphenylcarbazide as Indicator. J. V. Dubsky and J. Trtílek. (*Mikrochem.*, 1934, **15**, 95–99.)—Diphenylcarbazide, in 2 per cent. alcoholic solution, is used as a sensitive indicator for either micro- or macro-mercurimetric determinations of iodine. The intense violet colour formed with $Hg^{\bullet\bullet}$ is very easy to see, even in the presence of the insoluble mercury iodide formed in the titration. J. W. M.

Macro- and Micro- Tests for Nitrous Acid. J. V. Dubsky, J. Trtílek and A. Okáč. (*Mikrochem.*, 1934, **15**, 99–106.)—Chrysean ($C_4H_5N_3S_2$) can be used as a spot test reagent to identify 0·25γ of nitrite in 1 part in 2 millions dilution. A drop of the test solution is treated on the spot plate with the cold alcoholic solution of the reagent and a drop of dilute hydrochloric acid. To simplify the procedure the reagent may be used in acidified solution. Fairly concentrated nitrite solutions give a red-brown precipitate at once, whilst very dilute solutions give only a red solution, from which a precipitate slowly separates; the precipitation is accelerated by warming. The reaction is specific only in the absence of cations of the hydrogen sulphide and ammonium sulphide groups. Large amounts of nitrate give a yellow-red to red colour, but no precipitate, even on warming; in solutions of nitrates of concentrations below 2 *N* no reaction takes place. As nitrous acid is quantitatively precipitated by chrysean, and the precipitate is readily filterable, the reaction is recommended for the separation of nitrous acid from nitric acid. For this purpose an excess of a saturated solution of chrysean is used, and the precipitate filtered after the mixture has been warmed. The nitrate is identified in the filtrate by the diphenylamine reaction.

Similar diazotisation tests were also carried out with a number of other dyestuffs, but, except with safranine, an ammoniacal solution of α-naphthol was used for the coupling, instead of the coupling being effected with a further molecule of the same compound. The sensitivity of the tests is shown in the following table:

Dyestuff	Limit of identification γ	Concentration limit	Colour change
Primuline	0·3	1:100,000	Bright yellow→red
Pure yellow S	0·7	1:43,000	Yellow→blue
Safranine	1·5	1:20,000	Violet→blue
Victoria violet	15·0	1:2000	Red violet→blue

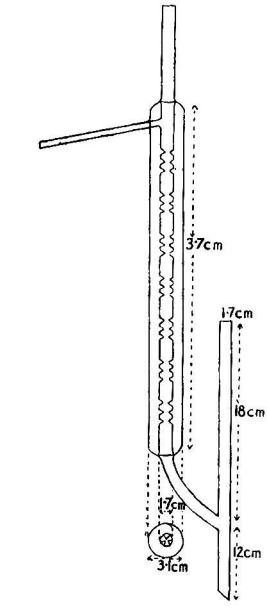
It should be noted that, whilst the limit of identification remains similar, the concentration limit varies with the depth of solution observed. J. W. M.

Collected References. Fluorescence Methods in Micro-analysis. M. Haitinger. (*Mikrochem.*, 1934–35, **16**, 321–356.)—The account consists of a description of general methods and sources of light (13 references), and then more detailed description of the methods applied to inorganic analysis (40 references), organic analysis (75 references), and the examination of biological material (10 references). Among the inorganic elements and compounds detected by fluorescence methods are:—Sodium, aluminium, beryllium, zinc, zinc oxide, arsenic and antimony, bismuth, manganese and antimony, cadmium, boric acid, sulphurous acid, the rare earths, and uranyl compounds. The general types of fluorescence of various aliphatic and aromatic organic compounds are briefly described, and there is an outline of the determination of hydrogen-ion concentration in organic compounds. Fluorescence tests for the following compounds are described in detail:—β-naphthol and resorcinol, malic acid, tartaric acid, citric acid, 1.2-dicarboxylic acids, aliphatic amines, pyrrole derivatives, phthalic acid, diethyl esters, and pyridine bases. J. W. M.

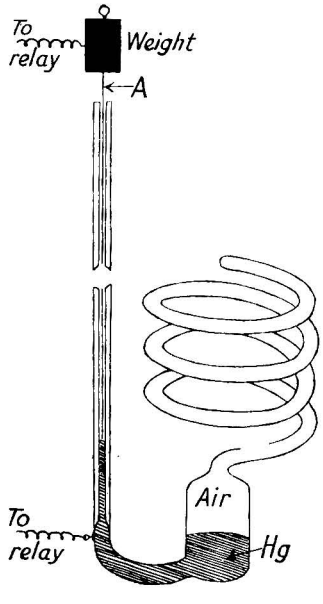
Physical Methods, Apparatus, etc.

Light Filters for the Mercury Lamp. E. J. Bowen. (*J. Chem. Soc.*, 1935, 76.)—In order to obtain a concentrated beam of light, a filter *A*, *B* or *C* is placed in a 500-ml. flask, 10 cm. in diameter (glass for the visible region and 366mμ, and silica for the ultra-violet), near the lamp, followed by *D* or *E* and/or the other filters mentioned, the test solution being behind a diaphragm about 24 cm. from the lamp. *A*.—145 g. of nickel sulphate (6 to 7 H_2O) and 41·5 g. of cobalt sulphate (7 H_2O) in 1 l. of water (10 cm.). *B*.—4·4 g. of copper sulphate (5 H_2O)and 150 ml. of ammonia (sp.gr. 0·88) in 1 l. of water (10 cm.). *C*.—13 g. of copper sulphate, 0·44 g. of potassium dichromate and 50 ml. of concentrated sulphuric acid in 1 l. of water (10 cm.). *D*.—Chlorine gas at 1 atm. (3 cm.). *E*.—A saturated solution of uric acid (1 cm.). For 248 and 254mμ, *A*, *D* and 0·108 g. of iodine and 0·155 g. of potassium iodide in 1 l. of water (1 cm.). For 265 and 270mμ, *A*, *D* and carbon tetrachloride (2 mm.) or 4·5 per cent. mercuric chloride solution (1 cm.). For 275 and 280·5mμ, *A* only. For 289·5, 292·5, 297, and 303mμ, *A* and 2 per cent. oxalic acid (1 cm.) or 1·5 per cent. copper sulphate (1 cm.). For 313·5mμ, *A*, 0·5 per cent. potassium hydrogen phthalate (1 cm.), and oxalic

acid as above. For 334$m\mu$, *A*, *E*, phthalate and oxalic acid as above. For 366$m\mu$, *B* and Chance's black "ultra-violet" filter (2 to 3 mm.). For 405$m\mu$, *B*, a 0·75 per cent. solution of iodine in carbon tetrachloride (1 cm.), and a 1 per cent. solution of quinine hydrochloride in water (2 cm.). For 436$m\mu$, *B* and 75 per cent. sodium nitrite (1 cm.). For 546$m\mu$, *C* and Corning glass No. 512 (5 mm.). For 577 and 579$m\mu$, *C* and Corning glass No. 344 (3·4 mm.). The solutions containing iodine, carbon tetrachloride, phthalate, quinine or oxalic acid should be renewed frequently. Filter *A* (cf. *id.*, 1932, 2236 and Bäckström, *Naturwiss.*, 1933, **13**, 251) must be free from iron salts; analytical reagents are suitable. J. G.



Fractional Distillation under Reduced Pressure. A. E. Bradfield. (*J. Soc. Chem. Ind.*, 1935, **54**, 6T.)—The Pyrex glass column shown was used successfully for the separation of two constituents of a viscid oil (b.p. 160° and 170° C. at 10 mm. pressure), which choked the ordinary types of column and caused the liquid to siphon over. It is based on the Widmer design, an essential feature being the alternate plain and "pushed-in" sections. In the present instance the outer jacket was evacuated to about 1 mm. pressure and sealed, the light jacketing of the exposed portion of the column and of the distillation-flask being sufficient to control the rate of reflux. J. G.



Thermo-Regulator for Heating and Cooling Baths. A. E. Bradfield. (*J. Soc. Chem. Ind.*, 1935, **54**, 6T.)—In the apparatus shown the weighted steel rod, *A*, is hung from a cord winding on (or unwinding from) a drum which is attached to the main spindle (lengthened if necessary) of a clock mechanism. The other end of *A* passes down the capillary stem of the regulator to the mercury surface and makes and breaks the relay operating the heating circuit, the former being connected with the mercury in the regulator. For most purposes air is a suitable medium for filling the regulator, and for a bath of 1 litre capacity a spiral of 6-mm. glass tubing, 8 to 10 ml. in capacity is required. The bath may thus be held at any desired temperature, or the rate of

heating or cooling may be adjusted by means of a movable weight on the pendulum of the clock; the apparatus is therefore particularly suitable for studying inflexions and arrests in cooling- and heating-curves of mixtures. J. G.

Reviews

DIE RÖNTGENSPEKTROGRAPHIE ALS UNTERSUCHUNGSMETHODE. By J. R. KATZ. Abderhalden, Handbuch der biologischen Arbeitsmethoden. Lfg. 436. Pp. 315. Berlin: Urban & Schwarzenberg. 1934. Price RM. 20.

The output of grotesquely misnamed "Handbooks" continues steadily, and the authoritative *Handbuch der biologischen Arbeitsmethoden*—indispensable to advanced students in the biological sciences—has increased in volume by a fascicule, paged, be it noted, from 3401 to 3716, dealing exclusively with X-ray spectrography.

In few branches of applied physics have there been of recent years advances more spectacular than in that which deals with the elucidation of the structure of organic fibres and the like bodies. In the present volume the methods of attack and the results obtained are discussed clearly and fully.

The author is not afraid to begin at the beginning, and his first principal section, "Die Versuchsmethodik der Röntgenspektrographie," is an excellent example of elementary but thorough exposition.

Succeeding sections deal with "Die Ergebnisse der Röntgenspektrographie in den einzelnen Gruppen der hochmolekularen Substanzen," "Polymorphie und Quellung der hochmolekularen Substanzen," "Die Röntgenspektrographie als Untersuchungs Methode (i) bei niedrigmolekularen Kolloiden, (ii) der feinstruktur von Muskeln, Nerven und anderen Organen," and "Ratschläge bei der Ausführung von kleineren röntgenspektrographischen Untersuchungen auf dem Gebiet der hochmolekularen Substanzen und der Kolloide."

The work is fully documented and well illustrated; it may be recommended unreservedly. A. FERGUSON

THE PRACTICE AND SCIENCE OF BREADMAKING. By D. W. KENT-JONES, Ph.D., F.I.C. Pp. 184. Liverpool: The Northern Publishing Co., Ltd. 1934. Price 7s. 6d.

This is essentially an elementary book written for bakery students and those engaged in the bakery trade. A book of this nature must encourage the application of scientific principles in the baking industry. The price places it within the reach of all.

"The baker must understand the character of the raw materials he is using and the nature of panary fermentation." This is an aim that might defeat the most astute student of cereal chemistry, but Dr. Kent Jones brings our incomplete knowledge into a simple form which should be easily understood by all. Starch, gluten and the proteins, water, cellulose, fat, sugar, dextrin, the enzymes and mineral matter are all dealt with in the compass of 15 pages. Strict accuracy is

sacrificed for simplicity in the statement that "In panary fermentation the yeast ferments the cane sugar present"; invertase, however, is also mentioned.

Bakers' raw materials are referred to, including wheats of world-wide origin; perhaps a little more space might have been given to flour-milling; the care taken in the cleaning of wheat and the general purity of flour are not emphasised as they should be. Bakery practice is dealt with fairly fully, and the chemical changes in dough fermentation are explained. The reason for the important "knock-back" is adequately stressed, as is also the need for control of yeast, temperature and humidity, but how many fermentation rooms can be maintained at 80–90 per cent. relative humidity? The comments on bread faults should be helpful to the student, although the baking conditions which accentuate the faults of high maltose are not detailed. No emphasis is laid on the matter of dough-mixing; correct mixing is a first essential, and numerous cases of badly made bread can be traced to insufficient mixing at the outset.

The questions of flavour and staling are discussed and admirably summed up as follows: "Generally speaking, correct fermentation and the use of cool doughs with sufficient yeast will produce bread which has good flavour and keeping qualities."

The chapter on nutritive value should help the baker to understand the importance of bread in the country's diet. In this simple presentation criticism may be unjustified, but the importance of absorbability of foodstuffs is not dealt with, and the statement: "If vitamin *A* is absent from a diet, a deficiency disease, as it is called, occurs," does not seem quite complete by itself.

An outline of bakery machinery includes several helpful hints, and the chapter on "Bakery Management and Laws Relating to the Baking Industry" will be useful to many. The final analytical chapter is elementary, but suffers from the lack of specific detail so necessary for beginners. Full details of procedure are given for determining the effective acidity of neutralising agents, but not for determining moisture and gluten. In the maltose test the previously published order of adding the sodium tungstate and the sulphuric acid is reversed.

Various repetitions detract somewhat from pleasurable reading, although these may be included for emphasis, but the type is clear and well set, and typographical errors are singularly few. From a perusal of the book the chemist should appreciate many of the interesting and unsolved problems of flour and breadmaking. The practical baker will get a new view of his art, and will realise to what extent the chemist can help him in his daily effort to produce bread of the highest quality.

C. W. HERD

CLINICAL AND PATHOLOGICAL APPLICATIONS OF SPECTRUM ANALYSIS, with Notes on Spectrography in Chemistry and Mineralogy, and Tables for Qualitative Analysis; being the Authorised Translation of Part II of "Die Chemische Emissionsspektralanalyse," by Dr. WALTHER GERLACH and Dr. WERNER GERLACH. Trans. by JOYCE HILGER TWYMAN. Illus. London: Adam Hilger, Ltd. Price 14s. 6d.

Mr. D. M. Smith, in his valuable monograph on "Metallurgical Analysis by the Spectrograph," states that "physical methods are now so largely used in

chemical laboratories that the technique of spectrographic analysis should not present any formidable obstacles." The translation of Gerlach and Schweitzer's *Die Emissionsspektralanalyse*, however, records that the appearance of certain papers "resulted in an avalanche of enquiries respecting details, and—which was still more significant—in prolonged visits from colleagues attached to industrial establishments, who wished to become practically acquainted with the methods." These two quotations illustrate the undoubted truth that, although spectrum analysis is essentially simple and straightforward, it requires experience and skill. The present work is of great value, since it should appreciably shorten the time required to master the technique of spectrographic analysis.

As a general guide to methods of exciting arcs, sparks, etc., to the recognition of "Raies Ultimes" and the interpretation of spectra, the book is concise and well-ordered. It is, moreover, noteworthy for a new and extremely interesting advance in technique, namely, the systematic use of the high-frequency spark with a Tesla coil. This method of excitation permits of spectrum analysis of biological material *in the form received*—bone splinters, calculi, tissue sections, etc.—without the use of chemical reagents. In addition, the distribution of metals over the different parts of a specimen can be determined, *e.g.* the centre and periphery of a calculus.

The collaboration of Gerlach the spectroscopist and Gerlach the pathologist has resulted in a significant broadening of the field of usefulness of spectrum analysis, and it is very desirable that toxicologists and pathologists should learn of these advances.

The translation is good, and the illustrations add greatly to the value of the work.

R. A. MORTON

QUALITATIVE ANALYSE MIT HILFE VON TÜPFELREAKTIONEN. By FRITZ FEIGL. Second Edition. Pp. xi+513, 24 figures and 35 tables. Leipzig: Akademische Verlag G.M.B.H. 1935. Price: R.M. 28, bound; R.M. 26.40, stitched.

The first edition of this work has already been reviewed in THE ANALYST (1931, **56**, 492). In view of the very great development of spot tests during the past three years, the increase in their number, and the widening of their application, a new edition of this, the only standard work on the subject, was much needed. The new edition has been very considerably enlarged, and now contains about 130 more pages than the old; this corresponds with even more matter, as the spacing of the letters is closer in the new edition than in the old, the type size remaining the same. This change, however, does not, in the reviewer's opinion, render the type any less legible, and enables the book to be produced in a convenient size.

The increase in matter is not so much in the theoretical section (142 pages, as compared with 122), which was already very carefully and amply written in the first edition, but in the number of tests described and the amplification of descriptions of tests in the presence of interfering ions. The neglect of precautions when interfering substances may be present is one of the greatest dangers attending the use of spot tests, and the author very rightly deals with this point with

considerable thoroughness under the description of each test, it being usually assumed that the general group separations have been carried out.

The most important addition to the book is the new section on spot tests in organic qualitative analysis (135 pages). This describes very recent work, most of which has been published only during the past few months by the author and his co-workers, and also includes a considerable amount of previously unpublished work. Many of these organic spot tests are adaptations of well-known tests; some are the same as tests for inorganic ions, with organic reagents, used in the reverse direction, and some are entirely new colour tests.

All the tests are described with admirable clarity and great attention to detail. This is due to the fact that the author either has originated most of the tests himself, or has first-hand experience of them. The tabular summary of tests at the end of the book is very useful, although had a complete list of all the accessory reagents for the tests also been included, the tables would have been more useful to those equipping for the first time a laboratory for spot tests. The addition would, however, have entailed considerable expansion of the tables. The book is to be strongly recommended to all analysts. J. W. MATTHEWS

Publications Received

A HISTORY OF FOOD ADULTERATION AND ANALYSIS. By F. A. FILBY. With a Foreword by Dr. BERNARD DYER. Pp. 269. London: George Allen & Unwin, Ltd. Price 10s. net.

THREE PHILOSOPHERS (LAVOISIER, PRIESTLEY AND CAVENDISH). By W. R. AYKROYD. Pp. xii+227. London: William Heinemann, Ltd. Price 10s. 6d. net.

A GERMAN-ENGLISH DICTIONARY FOR CHEMISTS. By A. M. PATTERSON. Second Edition. Pp. 411. London: Chapman & Hall. Price 15s. net.

ANNUAL REPORTS OF THE SOCIETY OF CHEMICAL INDUSTRY ON THE PROGRESS OF APPLIED CHEMISTRY. 1934. Vol. XIX. Pp. 836. Price 12s. 6d. (Members 7s. 6d.)

THE JOURNAL OF THE IRON AND STEEL INSTITUTE. Vol. 129, No. 1. 1934. Edited by K. HEADLAM-MORLEY.

BRITISH ASSOCIATION FOR THE ADVANCEMENT OF SCIENCE. REPORT OF THE ANNUAL MEETING, 1934. London: Burlington House, W.1. Price 15s.

BRITISH CHEMICALS AND THEIR MANUFACTURERS. London: Association of British Chemical Manufacturers. 1935.

INTERNATIONAL TIN RESEARCH AND DEVELOPMENT COUNCIL. FIRST GENERAL REPORT, 1934.

OFFICIAL YEAR BOOK OF THE SCIENTIFIC SOCIETIES OF GREAT BRITAIN AND IRELAND, 1934. Pp. vii+164. London: Charles Griffin & Co., Ltd. Price 8s. 6d. net.