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THE ANALYST

PROCEEDINGS OF THE SOCIETY OF PUBLIC ANALYSTS AND OTHER ANALYTICAL CHEMISTS

A JOINT Meeting of the Society with the Food Group of the Society of Chemical Industry was held at the Chemical Society's Rooms, Burlington House, on Wednesday, February 5th, 1936. The President of the Society, Mr. John Evans, was in the chair, supported by Dr. L. H. Lampitt, Chairman of the Food Group.

Certificates were read in favour of:—Archie Hector Cameron, B.Sc., A.I.C., A.R.T.C., Frederick T. W. Carman, Basil William Clarke, B.Sc., A.I.C., A.R.C.S., D.I.C., Evelyn Beryl Daw, B.Sc., A.I.C., William Edward James Hansford, Cyril Charles Harris, B.Sc., A.R.C.S., Arthur George Jones, B.Sc., A.I.C., Reginald William Money, M.Sc., A.I.C., Horace Edward Newton, Kenneth Sams, B.Sc., Ph.D., A.R.C.S., A.I.C., D.I.C., Winifred Edris Welton, B.Sc., A.I.C., Donald Major Wilson, M.C., B.Sc., A.I.C.

The following were elected Members of the Society:—George Edward Boizot, B.Sc., F.I.C., Frank Ward Bury, M.Sc., F.I.C., George Henry Croft, M.Sc., A.I.C., George John Cunningham, M.R.C.S., L.R.C.P., M.B., B.S., Reginald Stanley Garlick, B.Sc., A.I.C., Robert Thomas Moline Haines, M.A., Philip John Courtney Haywood, B.Sc., Douglas Thurlow Lucke, B.Sc., F.I.C., Hugh Clouston Moir, B.Sc., A.I.C., Jack Leake Pinder, B.Sc., A.I.C., Oswald Victor Richards, Ph.D., Henry Geoffrey Smith, B.Sc., Ph.D., A.I.C., William Warren, B.Sc., F.I.C., Kenneth Wallis, B.Sc., A.I.C.

The following papers were read and discussed:—"The Constitution of Tannins, including those of Tea and Coffee," by Peter Maitland, B.Sc., Ph.D.; "A Survey of the Methods of Analysis for Tannins," by C. Ainsworth Mitchell, M.A., D.Sc., F.I.C.; "Experimental Work on Tea Tannin," by M. Nierenstein, D.Sc., Ph.D.; "The Pharmacology of Caffeine, and of Tea and Coffee," by G. Roche Lynch, O.B.E., M.B., B.S., F.I.C.; "The Tannins in Tea," by P. J. Norman, B.Sc., A.I.C., and E. B. Hughes, D.Sc., F.I.C.; "Coffee Extracts," by E. Hinks, B.Sc., F.I.C.; and "A Note on 'Tanninless' Teas," by H. H. Bagnall, B.Sc., F.I.C.

Obituary

THOMAS HENRY POPE

By the death of Thomas Henry Pope, on January 12th, the Society has sustained a very heavy loss and the Publication Committee of the Analyst, in the words of Dr. J. A. Voelcker, has lost "an invaluable friend and worker." From the date when he joined the Society of Public Analysts in 1921 he served as an abstractor to the Analyst, of which he was appointed assistant editor in 1933. In this capacity his wide knowledge of foreign languages and of scientific literature was of very special value. He impressed his colleagues on the committee by his evident grasp of the multitudinous questions which came up for consideration, and by his quiet and methodical way of dealing with them. Although thoroughly familiar with his subject, his extreme modesty led him to refrain from imposing his personal views on others, but his work throughout was characterised by thoroughness and efficiency.

Pope was born on February 1st, 1875, so that he had nearly completed his 61st year. After receiving his early education at the Central Foundation Schools, Cowper Street, Finsbury, and at the Finsbury Technical College, he passed to the Central Technical College, South Kensington, as a chemical student under Professor Armstrong in 1893. He took the diploma of A.C.G.I. in 1896, and then became research assistant to Mr. (now Sir) Robert Mond until 1898, when he was invited by Mr. Julian L. Baker, who had succeeded Professor A. R. Ling as chief chemist of the Beetroot Sugar Association, to act as his assistant. In 1900 he became, himself, chief chemist to the Association, and shortly afterwards, in 1901, succeeded Mr. J. H. Millar as Lecturer in the British School of Malting and Brewing, Birmingham University, under the late Professor Adrian Brown, F.R.S. He stayed at Birmingham until September, 1917, lecturing not only in brewing, but in general inorganic and organic chemistry to the degree students. He also had charge of the Analytical Department, in which, in those days, much analytical work was done, principally for brewing purposes. Professor Hopkins writes to me that "he was exceedingly popular with the students and his colleagues on the staff, and I have already heard many expressions of great sorrow from his old students."

On October 1st, 1917, he joined Messrs. Calder's, Ltd., working at the distilleries at Bo'ness and Gartloch on alcohol and yeast production. This firm became an associated company of the Distillers' Company, Ltd., in 1921. Early in 1922 he was transferred to the Vauxhall Distillery at Liverpool, and in 1925 to Bankhall Distillery as assistant to the Works Manager. In 1927 he became a

member of the Research Department of the Distillers' Company, then newly formed at Great Burgh, Epsom, by Dr. J. Vargas Eyre, taking charge of the section devoted to industrial and potable alcohol.

Pope was a B.Sc. of Birmingham University, a Fellow of the Institute of Chemistry since 1903, and of the Chemical Society (1899) and a member of the Society of Chemical Industry (1899). From the time he joined them until his death he did valuable service for the last two societies as an abstractor, dealing particularly with papers in the (to British chemists) less-known languages, Italian and Russian. Although he published little original work—one paper in 1900, with J. L. Baker, on Two New Polysaccharides (*J. Chem. Soc.*, 1900, 696), and another with A. R. Ling, in 1901, on the Refractometric Analysis of Beer—he did much literary work in the form of translations of standard textbooks. The English edition of Euler's *Chemistry of the Enzymes* was published in 1912; two editions of Molinari's comprehensive *General and Industrial Chemistry*, the last, Inorganic in 1920 and Organic (two volumes) in 1921 and 1923; and Villavecchia's *Applied Analytical Chemistry* (2 vols.) in 1918. He also revised the section on Starch and its Isomerides in the Fifth Edition of *Allen's Commercial Organic Analysis* (1924), and compiled a valuable Bibliography on Heavy Metals in Food and Biological Material for THE ANALYST (1932–4).

His knowledge was not, however, mere book knowledge; he had acquired a very wide range of experience on the practical sides of brewing and distillery work, such as few others possessed in this particular field.

His charming personality—an old-world dignity and courtliness, combined with unusual gentleness and modesty—endeared him to all with whom he came in contact. The writer, who first came to know him well in his student days and worked with him as colleague for many years, and many others, mourn his passing as a great personal loss.

Dr. Hughes and Dr. Mitchell represented the Society at the funeral.

W. A. DAVIS

The Determination of Cocaine Alkaloids in Mixtures with other Alkaloids and Local Anaesthetics

BY JOHN RALPH NICHOLLS, B.Sc., F.I.C.

FOR legal purposes it is often necessary to determine if a local anaesthetic or other preparation comes within the provisions of the Dangerous Drugs Acts, 1920–1932. As far as the cocaine alkaloids are concerned, these Acts do not apply to preparations containing less than 0·1 per cent. of cocaine or of ecgonine; and for the purposes of the Acts "the expression 'ecgonine' means laevo-ecgonine and includes any derivative of ecgonine from which it may be recovered industrially."

In this paper the term "cocaine alkaloids" refers to those ecgonine derivatives which are present in coca leaves and are of similar constitution to cocaine. These are cocaine (methylbenzoylecgonine), cinnamyl cocaine (methylcinnamylecgonine) and the truxillines (methyl-α-truxillylecgonine and methyl-β-truxillylecgonine). Of these, only cocaine is prepared pure for use in medicine; mixtures of the cocaine alkaloids with other coca alkaloids occur in medicaments containing extracts of coca leaves. No other ecgonine derivative is a commercial article.

There are thus two types of preparations which may be encountered:—(i) Those in which pure cocaine, either as such or as a salt, is an ingredient, and (ii) those in which an extract of coca leaves is employed, thus containing the mixed cocaine alkaloids.

I. Preparations containing Pure Cocaine.—As its nitrogen group is attached to two closed rings, cocaine is a relatively weak base. Although sodium bicarbonate does not precipitate cocaine from solutions of its salts, the free alkaloid can be extracted with an immiscible solvent after the bicarbonate is added. Other alkaloids and local anaesthetics which are stronger bases are only very slowly, if at all, extracted under such conditions; and if the immiscible solvent is light petroleum, few are extracted. Since cocaine is readily soluble in light petroleum, the use of this solvent in conjunction with sodium bicarbonate enables a separation to be made. For example, cocaine can be completely separated from procaine (novocaine) by such treatment, the extracted cocaine being sufficiently pure to crystallise and to give the correct melting-point.

In those few cases where traces of alkaloids other than cocaine are extracted, the extract may be treated with potassium permanganate to decompose interfering substances. Cocaine in slightly acid solution is not appreciably affected by permanganate, whilst almost all other alkaloids and local anaesthetics are attacked. Such a treatment can often be applied directly to a sample, but may be tedious if much material has to be oxidised. For carrying out the oxidation, the extracted residue (or original sample) should be dissolved in $N/10$ sulphuric acid, and 3 per cent. potassium permanganate in $N/2$ sulphuric acid solution added until an excess is indicated by the colour. After the solution has been decolorised with oxalic acid and sodium bicarbonate added in excess, any cocaine may be extracted with light petroleum. The manganese comes out of solution only slowly, and does not interfere with the extraction if this is carried out immediately after the addition of the bicarbonate. Oxidation with permanganate can be hastened by keeping the solution in a water-bath at about 60° C., and, under these conditions, 0·2 g. of procaine gave no extractable matter, whilst 0·052 g. of cocaine hydrochloride yielded, after half-an-hour's treatment, 0·0455 g. of cocaine base, equivalent to 0·051 g. of cocaine hydrochloride.

Any other alkaloids or local anaesthetics remaining in a solution from which cocaine has been extracted, as above described, may be liberated by adding excess of ammonia, and can then be extracted by a suitable solvent. Such substances will not, of course, remain after treatment with permanganate.

II. Preparations containing Mixed Coca Alkaloids.—In addition to the cocaine alkaloids, coca leaves contain appreciable proportions of other bases, *e.g.* the hygrines. Only the cocaine alkaloids come within the provisions of the

Dangerous Drugs Acts, and it is only these that are physiologically active. The usual methods of assay of coca leaves and coca preparations give the total ether-soluble alkaloids liberated by ammonia, including the hygrines, as well as the cocaine alkaloids. The hygrines all have their nitrogen groups attached to single closed rings and are, consequently, stronger bases than the cocaine alkaloids which resemble cocaine. If, in the assay, sodium bicarbonate is used instead of ammonia, and either light petroleum or a mixture of equal parts of ether and light petroleum replaces the ether, the hygrines are not extracted, whilst the cocaine alkaloids can be completely removed, and in this way the content of cocaine alkaloids may be obtained. In certain cases it may be desirable to check the purity of these alkaloids; treatment with permanganate, suggested above for cocaine, decomposes the other cocaine alkaloids and is inapplicable, but a method involving the determination of the acids (benzoic, cinnamic and truxillic) obtained after hydrolysis may be used, as indicated later.

The British Pharmaceutical Codex provides a standard and a method of assay for Extractum Cocae Liquidum. This standard is expressed as a percentage of ether-soluble alkaloids calculated as cocaine, and the assay determines, by titration with acid, the total alkaloids liberated by ammonia and extracted with ether. It would appear desirable that such a preparation should be standardised and assayed on the physiologically active constituents only, *i.e.* on the cocaine alkaloids. With certain types of coca leaves the alkaloids, extracted as in the B.P.C. method and calculated as cocaine, may be double the actual content of cocaine alkaloids as defined above. The B.P.C. assay could be readily modified to give only the cocaine alkaloids by replacing the fourth, fifth and sixth sentences by the following:

"To the combined acid liquids add 1 g. of sodium bicarbonate and 20 ml. of a mixture of equal parts of ether and light petroleum. Shake vigorously, separate the aqueous layer, and extract it with two successive quantities of 15 ml. of a similar mixture of ether and light petroleum. Filter the combined extracts, evaporate the solvent, and dissolve the residue in 10 ml. of $N/10$ sulphuric acid."

With such an assay it would be necessary to provide a fresh standard for cocaine alkaloids calculated as cocaine.

EXAMINATION OF THE COCAINE ALKALOIDS.—The following considerations and results will indicate that the suggested method of assay does, in fact, determine the cocaine alkaloids only. Each molecule of a cocaine alkaloid yields, on hydrolysis, one molecule of benzoic, cinnamic or truxillic acid. The titration of such alkaloids with standard acid should, therefore, equal the titration of the hydrolysed and extracted acids with standard alkali. Also, the proportion of ecgonine, calculated from either of these titrations, should equal that calculated from the optical rotation of the completely hydrolysed alkaloids. The hygrines give no acids on hydrolysis and are not optically active.

Two samples of coca leaves (one from Java and one from Peru) were examined as follows:—An acid solution of the total alkaloids from 20 g. of leaves was extracted in one case with ether after being made alkaline by the addition of ammonia, and in another with a mixture of ether and light petroleum after the addition of sodium bicarbonate. The extracted alkaloids were dried in a vacuum desiccator and

weighed. They were dissolved in 10 ml. of neutral acetone (alcohol leads, subsequently, to losses through ester formation), 10 ml. of water were added, and the solution was titrated with $N/10$ acid, methyl red being used as indicator. The solution was then mixed with 10 ml. of approximately N sodium hydroxide solution and boiled under a reflux condenser for 10 minutes. The acetone was evaporated on a water-bath, the solution was cooled and acidified, and the acids were extracted with a mixture of equal parts of ether and light petroleum. The solvent was evaporated under slightly reduced pressure in a flask placed in a water-bath at about 30° C. The flask was closed with a two-holed stopper, carrying the connection to the pump, and a capillary tube through which a current of air impinged upon the surface of the solvent. Under these conditions the solvent was rapidly evaporated without deposition of moisture or loss of acids, and the acids separated in a dry crystalline form. The residue was dried in a vacuum desiccator, dissolved in neutral alcohol and titrated with $N/10$ alkali, with phenolphthalein as indicator. Other portions of the alkaloids similarly extracted from the leaves were hydrolysed by boiling with hydrochloric acid for 5 hours, and the ecgonine was calculated from the optical rotation of the solution; for ecgonine $[\alpha]_D = -57°$. (This determination cannot be made after an alkaline hydrolysis, as *l*-ecgonine is then partly changed to *d*-ecgonine.) Control experiments were carried out with pure cocaine hydrochloride to ensure that each extraction, hydrolysis, etc., was satisfactory.

The following results were obtained:

Origin of leaves	Java	Peru
I. *Alkaloids liberated by ammonia.*		
Weight of alkaloids, g.	0·3230	0·2500
Titration of alkaloids, ml. $N/10$ acid	11·50	10·40
equivalent ecgonine, g.	0·213	0·192
Titration of acids, ml. $N/10$ alkali	7·90	5·45
equivalent ecgonine, g.	0·146	0·101
Ecgonine from rotation, g.	0·176	0·100
II. *Alkaloids liberated by sodium bicarbonate.*		
Weight of alkaloids, g.	0·2545	0·1750
Titration of alkaloids, ml. $N/10$ acid	7·85	5·50
equivalent ecgonine, g.	0·145	0·102
Weight of acids, g.	0·1150	0·0740
Titration of acids, ml. $N/10$ alkali	7·85	5·45
equivalent ecgonine, g.	0·145	0·101
Ecgonine from rotation, g.	0·143	0·102

The sodium bicarbonate solutions, after extraction, were made distinctly ammoniacal and re-extracted with ether. In each case liquid alkaloids were obtained which fumed strongly on heating to 100° C., but which gave no extractable acids on hydrolysis and were, therefore, not cocaine alkaloids. The extract from the Java leaves was separated into two fractions, one corresponding with a mixture of α- and β-hygrines, and the other consisting of a small residue not giving alkaloidal reactions but having laevo-rotation. A similar extract from the Peruvian leaves had no rotation and consisted largely of cuscohygrine.

From the weights and titrations of either the alkaloids or the acids obtained

after hydrolysis it is theoretically possible to calculate the proportions of cocaine and of cinnamyl cocaine or truxilline, or both. But the differences between the equivalent weights are so small that the results are not very accurate. (Molecular equivalent weights:—cocaine 303, cinnamyl cocaine and truxilline 329; benzoic acid 122, cinnamic and truxillic acids 148.) From the weights of the acids obtained in II the following may be calculated:

JAVA LEAVES

g.			g.			g.	
0·0055	benzoic acid	=	0·008	ecgonine	=	0·014	cocaine
0·1095	cinnamic and/or truxillic acid	=	0·137	,,	=	0·243	cinnamyl cocaine and/or truxilline
0·1150			0·145			0·257	
			0·143 = ecgonine (from rotation)			0·2545 = alkaloids found	

PERU LEAVES

g.			g.			g.	
0·0313	benzoic acid	=	0·0475	ecgonine	=	0·078	cocaine
0·0427	cinnamic and/or truxillic acid	=	0·0535	,,	=	0·095	cinnamyl cocaine and/or truxilline
0·0740			0·1010			0·173	
			0·102 = ecgonine (from rotation)			0·175 = alkaloids found	

It is clear from all these results that the true coca alkaloids can be separated from the other alkaloids in coca leaves by suitable extraction from a solution made alkaline with sodium bicarbonate.

SUMMARY.—1. Cocaine can be separated from most other alkaloids and local anaesthetics by extracting with light petroleum from solutions made alkaline with sodium bicarbonate. A method of treatment with potassium permanganate described may be used when complete separation is not effected.

2. A method is described for determining the true cocaine alkaloids in preparations derived from coca leaves.

I have to thank Sir Robert Robertson, Government Chemist, for permission to publish this paper.

GOVERNMENT LABORATORY
W.C.2

The Amino Acids of the Mixed Proteins of Ox-Muscle: The Basic Amino Acids

By HENRY GEORGE REES, PH.D., D.I.C., A.I.C.

CONSIDERING the economic importance of the proteins of ox-muscle, there has been surprisingly little systematic investigation of these substances since the analysis carried out by Osborn and Jones.[1] In general, most of the published work has been carried out on lean meat, which has been extracted with water, alcohol and ether; this product, which is essentially the beef powder or desiccated beef of commerce, was selected for the present investigation in preference to one of the individual muscle proteins on account of its ease of preparation and of its importance in nutrition. An excellent summary of the present position of the chemistry of the muscle proteins is given in a recent paper by Smith.[2]

In the present paper we are concerned only with the basic amino acids, and the values obtained by Osborn and Jones will be quoted here:—Arginine, 7·5; histidine, 1·8; lysine, 7·6 per cent. These results were obtained by the Kossel-Patten method, and were based on the nitrogen-content of purified arginine and histidine fractions and on the lysine isolated as picrate. During recent years Vickery and his co-workers[3–6] have effected considerable improvement in the Kossel procedure for the quantitative determination of the individual members of this group, the precipitation of the silver salts of arginine and histidine being carried out under more precise conditions than formerly, and both bases finally estimated as crystalline flavianates. The determination of lysine is based, as has always been customary, on the isolation of the crystalline picrate. As a rule, the results obtained by the Vickery method do not show a great difference from those by the Kossel method, since in the latter the mutual interference of arginine and histidine was practically equal.

Other published work includes the following, which have been carried out by the Van Slyke method; Thrum and Trowbridge[7] give a series of results for various fractions obtained by precipitation methods; Moulton[8] has analysed the heat coagulable proteins, and Rosedale[9] the peptic digest of ox-muscle.

In view of this apparent lack of reliable data and the improvement in analytical technique during recent years, it was decided to re-investigate the amino acids of ox-muscle, particularly those of the basic fraction.

EXPERIMENTAL

PREPARATION OF THE MIXED PROTEIN.—Fresh steak, freed from fat as completely as possible by trimming, was exhaustively extracted by boiling with alcohol, dilute acid, and finally with water. The dried residue was ground and extracted in small batches with ether in a Soxhlet extractor.

The analytical figures on the bulk dried sample were as follows:—Total nitrogen, 15·87; moisture, 0·06; ash, 0·01; fat, nil.

DISTRIBUTION OF NITROGEN.—The distribution of nitrogen into five groups (the Hausmann number) was determined by the usual methods.[10] Normally, the mono-amino nitrogen is determined by difference. In this case, however, an

actual determination of this fraction was made on the filtrate after precipitation of the bases, the procedure of Van Slyke[10] being used. The results, expressed as percentage of the total nitrogen, were as follows:—amide nitrogen, 7·2; humin nitrogen, 1·6; basic nitrogen, 24·3; mono-amino nitrogen, 66·6; non-amino nitrogen, 0·8 per cent.

DETERMINATION OF THE BASIC AMINO ACIDS.—Five analyses in all were made. In Nos. 1 to 3 arginine and histidine were determined; the lysine values would have been low on account of incomplete removal of sulphuric acid in the later stages. Consequently, in analyses 4 and 5 lysine only was determined, with a slight modification to be described later.

PROCEDURE USED IN ANALYSES 1 TO 3.—Approximately 70 g. of protein were hydrolysed for 30 hours in an oil-bath with 450 ml. of conc. hydrochloric acid and 425 ml. of water at about 110° C. Hydrochloric acid was removed as completely as possible by concentrating the liquid to a syrup three or four times, the bulk was made up to 1 litre, and aliquot parts were taken for the determination of total nitrogen. The remainder of the hydrochloric acid was then removed by the addition of sulphuric acid and silver oxide in excess, the precipitated silver chloride being thoroughly washed by digestion with dilute hydrochloric acid and water, and this operation was repeated to remove the last traces of acid after the final digestion.

The first Silver Precipitation was then carried out in a strongly alkaline solution, containing excess of silver. The excess of silver ion was introduced by using silver oxide and sulphuric acid in dilute solution, as recommended by Vickery and Shore.[6] At *p*H 12–13 arginine and histidine are obtained in the silver precipitate, which is filtered off and decomposed with hydrogen sulphide at *p*H 4. The volume of the filtrate (A), which is preserved for the lysine fraction, must be recorded for calculation of the soluble arginine silver compound.

SEPARATION OF HISTIDINE FROM ARGININE.—This was obtained by precipitation twice at *p*H 7·2 after the introduction of excess of silver. The precipitate, after decomposition at *p*H 4·0 with hydrogen sulphide, was treated as described below for histidine, whilst the two filtrates composed the crude arginine fraction.

PRECIPITATION OF ARGININE.—The two filtrates mentioned above were combined and concentrated at *p*H 4·0, and the silver salt of arginine was again precipitated at *p*H 12. The filtrate, the volume of which was again noted for the solubility correction, was added to filtrate (A), the crude lysine fraction. The arginine silver was decomposed at *p*H 5 to 6 with hydrogen sulphide, and the filtrate was concentrated and made up to 500 ml. at *p*H 6. Aliquot parts were removed for the determination of total nitrogen, and the arginine was precipitated in further aliquot portions with the calculated quantity of flavianic acid (1 g. of arginine nitrogen ≡ 5·61 g. of flavianic acid). The arginine flavianate was collected, dried at 105° C., and weighed. In each determination arginine was calculated from the highest yield of flavianate obtained from the aliquot parts taken. The conversion factor to arginine is 0·3566, and the solubility factor for the arginine silver compound 0·036 g. of arginine per l. of solution.

PRECIPITATION OF HISTIDINE.—The crude fraction, dissolved in 5 per cent. sulphuric acid, was treated with Hopkins' reagent (10 per cent. mercuric sulphate

in 5 per cent. sulphuric acid), and, after standing for several days, the precipitate was filtered off and decomposed with hydrogen sulphide. Cystine was then removed by precipitation with freshly prepared copper hydroxide at *p*H 5, and the filtrate (faintly acid and free from copper and barium) was made up to 250 ml. Total nitrogen was determined on aliquot parts, and, after concentration to about 30 ml., flavianic acid was added in 15 per cent. excess to the main bulk (1 g. of histidine nitrogen ≡ 14·91 g. of flavianic acid). After standing for 48 hours at 0° C. the histidine diflavianate was filtered off, the mother liquor was evaporated to 5 ml., and a second crop was obtained on further standing. The conversion factor is 0·1979.

THE LYSINE FRACTION.—Silver was removed, the fraction was concentrated to 1 litre, and the ammonia removed by distillation *in vacuo* after addition of baryta and alcohol. Excess of barium was removed, and the solution was treated with phosphotungstic acid (Kahlbaum) and 5 per cent. sulphuric acid. After decomposition of the precipitate in 5 per cent. acetone with baryta, barium and sulphate ions were completely removed, and the bulk was concentrated to 500 ml. Aliquot parts were taken for total nitrogen, and the main solution, after concentration to 15 ml., was first treated with absolute alcohol until a slight turbidity was produced, and then with alcoholic picric acid. The lysine picrate was filtered off, and the mother liquor was evaporated to obtain a second crop. The volume of the final filtrate was recorded for the solubility correction of 0·54 g. of lysine picrate per 100 ml. The conversion factor for picrate to the base is 0·3895.

According to Vickery,[4] Crop 1 "explodes" at about 265° C., whilst, to be acceptable, Crop 2 should "explode" above 250° C. The first crops obtained in these determinations all "exploded" between 261° and 264° C., and the second crops between 254° and 258° C.

PROCEDURE IN ANALYSES 4 AND 5.—As mentioned previously, the lysine values in analyses 1 to 3 were very low compared with the value obtained from the nitrogen distribution in the protein. This was traced to incomplete removal of sulphuric acid prior to precipitation as picrate, and the values obtained for lysine in these analyses were ignored. Consequently, two further analyses were made in which lysine only was determined.

Forty g. of protein were hydrolysed and the silver precipitate obtained at *p*H 12, as described above. The precipitate was decomposed and re-precipitated at *p*H 12. The filtrates were then worked up in the normal manner for lysine, the arginine and histidine fractions being disregarded. The following tables show the results obtained:

TABLE I

ARGININE FRACTION

Analysis	Protein taken for analysis g.	Arginine in protein (from total *N* in fraction) Per Cent.	Vol. of arginine silver solutions ml.	Solubility correction for arginine g.	Wt. of flavianate for total fraction g.	Arginine recovered g.	Arginine Per Cent.
1	69·88	7·70	8350	0·300	11·500	4·401	6·29
2	70·75	7·70	8950	0·322	10·615	4·170	6·02
3	70·31	7·44	7400	0·266	11·540	4·381	6·23

Average 6·18 per cent.

TABLE II

HISTIDINE FRACTION

Analysis	Protein taken for analysis g.	Histidine equivalent of total *N* in fraction Per Cent.	Diflavianate Crop 1 g.	Diflavianate Crop 2 g.	Diflavianate Total g.	Histidine recovered g.	Histidine Per Cent.
1	70·75	0·81	1·589	0·202	1·791	0·385	0·57
2	70·31	1·29	1·902	0·237	2·139	0·440	0·64
3	70·75	0·46	0·892	0·248	1·140	0·245	0·35

Average 0·52 per cent.

TABLE III

LYSINE FRACTION

Analysis	Protein taken for analysis g.	Wt. of picrate Crop 1 g.	Wt. of picrate Crop 2 g.	Solubility correction g.	*N* content of fraction g.	Lysine equivalent in protein from *N* Per Cent.	Yield of lysine Per Cent.
4	37·80	5·970	0·570	0·110	0·729	10·5	7·14
5	37·80	6·276	0·408	0·043	0·995	14·2	7·22

Average 7·18 per cent.

In every instance these results are slightly lower than those obtained by Osborn and Jones (*cf.* p. 160), which are the only available comparable figures.

A comparison of these results with those obtained from the distribution of nitrogen shows that the value for the total basic nitrogen is 23·1 per cent., as compared with 24·3 per cent. In accordance with the general findings, the value for the total bases, determined from the distribution of nitrogen, is slightly higher than that obtained by direct determination; in other words, there is no appreciable amount of basic material present, other than that determined in the protein. It must be remembered, however, that the value for the basic nitrogen obtained from the distribution of nitrogen includes the cystine value, which is not accounted for in the present determination.

The discrepancy between these results and those of Osborn and Jones, after allowance has been made for any possible difference in the experimental material, is not great. As Osborn's results, with the exception of lysine, were obtained from nitrogen determinations on the final fractions, it was to be expected that a method based on actual isolation of crystalline derivatives would give lower results. It is of interest to compare the arginine and histidine values obtained from the nitrogen determinations on the respective fractions with those of Osborn and Jones which are given in parentheses:

Arginine	7·61 per cent.	(7·5 per cent.)	From nitrogen
Histidine	0·52 ,, ,,	(1·8 ,, ,,)	,, ,,
Lysine	7·78 ,, ,,	(7·6 ,, ,,)	Isolation of picrate

The results now approach more closely to those of Osborn and Jones, and it may be concluded that the observed difference is due primarily to the improved methods employed rather than to any variation in the material.

SUMMARY.—Arginine, histidine and lysine have been determined in the mixed proteins of ox-muscle by means of Vickery's modification of Kossel's procedure. The results obtained were:—arginine, 6·18; histidine, 0·52; lysine, 7·18 per cent.

I wish to express my thanks to Professor A. C. Chibnall, of the Imperial College, and to Dr. A. H. Salway, for their continual interest in this work, and to Messrs. Oxo Limited, for permission to publish the results.

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Diphenylcarbazide. An Internal Indicator for Use in the Titration of Iron with Dichromate

BY H. E. CROSSLEY, M.Sc., A.I.C.

THE use of diphenylcarbazide, $(C_6H_5.NH.NH.)_2CO$, in qualitative analysis was described by Cazeneuve,[1] and his work was continued by Brandt[2] in the application of diphenylcarbazide to iron titrations, a subject still further investigated by Barneby and Wilson.[3] Adoption of the method has been retarded because of difficulties encountered in the titration of less than 0·2 g. of iron—the technique of the method had been incompletely worked out, and the authors disagreed on the question of applying a correction for the amount of indicator oxidised. The method has therefore been investigated in order to devise a technique of general application and to correct the errors that had previously arisen.

PRINCIPLE OF METHOD.—A measured amount of diphenylcarbazide, dissolved in dilute acetic acid, is added to the ferrous solution, with a limited amount of hydrochloric acid present, and "manganous sulphate mixture" (containing sulphuric and phosphoric acids) is added to retard the oxidation of the indicator until the true end-point is reached. With small amounts of iron, a solution of ferric chloride is added, to ensure that the indicator oxidation is not delayed after the true end-point, but no definition of critical amounts has hitherto been stated. According to Barneby and Wilson (*loc. cit.*) a correction is necessary for the amount of dichromate used by the indicator; Brandt originally stated that no dichromate was consumed by the indicator, but he afterwards modified this, and admitted a negligible correction. No colour is given by the indicator until one or two drops of dichromate have been added; a violet colour then appears, which fades to

lavender near the end-point of the titration. The end-point is shown by decolorisation of the indicator to leave only the colour of dissolved iron and chromium salts.

EXPERIMENTAL.—The research followed systematic lines, each variable factor being investigated in turn. Several hundred experimental titrations were made, but space permits of the inclusion of only a few selected results to illustrate the principal findings.

The reagents were prepared according to the directions of Barneby and Wilson (*loc. cit.*), with the addition of a standard ferric chloride solution and approximately $N/10$ ferrous ammonium sulphate solution, the two having the same iron-content.

Solutions containing various amounts of $N/10$ iron solution were titrated with dichromate—the acidity, amount of indicator, and the amount of manganous sulphate mixture being varied. In some experiments standard ferric solution was added. The indicator was added before the titrations, and in certain experiments further quantities were added towards the end of a titration (to allow for colour fading).

Titrations of Indicator in the Absence of Ferrous Solutions.—Unless otherwise stated, each titration was carried out on a total solution bulk of 500 ml., containing 40 ml. of conc. hydrochloric acid, 25 ml. of ferric solution and 20 ml. of manganous sulphate solution. The results were as follows:

TABLE I

Experiment number	Indicator ml.	Dichromate solution (0·1 *N*) Total ml.	Dichromate solution (0·1 *N*) Per ml. of indicator ml.	Remarks
1	2·0	0·31	0·155	Titration proportional to amount of indicator
2	4·0	0·60	0·150	
3	6·0	0·92	0·153	
4	4·0	0·70	0·175	Only 10 ml. of HCl present
5	4·0	0·61	0·153	Fresh indicator solution
6	4·0	0·50	0·125	Indicator stored 14 days (nitrogen)
7	4·0	0·61	0·153	Added before other reagents: $SnCl_2$+excess $HgCl_2.ZnCl_2$
8	4·0	0·60	0·150	

The claim of Barneby and Wilson, that a correction is necessary for the amount of indicator oxidised, was substantiated, and also the order of the amount. This correction is dependent on the purity of the diphenylcarbazide, but no variations were observed in successive preparations from the same batch of solid (Expts. 2, 5). Also the unit correction was found to be independent of the amount of indicator titrated (Expts. 1, 2, 3). The storage of indicator under nitrogen was found to be more satisfactory than the use of Barneby and Wilson's carbon dioxide method. In whatever gas indicator is stored, however, it is necessary to standardise it before use (Expt. 6). Hence it appears more desirable to prepare indicator daily when required, as the procedure is simple, and only occasional preparations from one batch of solid will require standardising. The titration of diphenylcarbazide was found to be unaffected by the presence of tin and mercury or of zinc (Expts. 7, 8).

The method of determining the indicator value by addition of more indicator at the end of an iron titration, and then continuing the titration, gave inaccurate results (0·14 to 0·19 ml. of dichromate per ml. of indicator). Any error in the iron titration is directly transmitted to the indicator titration.

In the titration of ferrous solutions a stronger colour was obtained by adding only 10 ml. of hydrochloric acid before the titration, with a further addition of 30 ml. when the indicator began to fade. Titrations completed in the presence of only 10 ml. of hydrochloric acid showed high results, due to the slow rate of oxidation of the indicator.

Titrations of Various Amounts of Ferrous Solution.—In Table II, unless otherwise stated, each titration was carried out on a total solution bulk of 500 ml., containing 10 ml. of conc. hydrochloric acid and 20 ml. of manganous sulphate solution. A further 30 ml. of hydrochloric acid was added towards the end-point in each case. The ferrous solution was stored under nitrogen and titrated with $N/10$ potassium permanganate solution; 25·00 ml. of ferrous solution ≡ 24·50 ml. permanganate. The amount of indicator added before the titration and also (when required) during titrations is shown. The amounts of $N/10$ dichromate solution shown are corrected for the indicator used.

TABLE II

Exp. number	Indicator addition: Before titr. ml.	Indicator addition: Towards end ml.	Ferrous solution ml.	Dichromate solution (0·1 N) ml.	Remarks
9	1·0		10·0	9·80	
10	1·0	1·0	10·0	9·81	
11	1·0	1·0	10·0	9·78	15 ml. of ferric solution added
12	1·0		10·0	9·85	Mn solution increased to 40 ml.
13	1·0	1·0	25·0	24·50	
14	1·0	1·0	25·0	24·48	
15	1·0	1·0+1·0	50·0	48·50	
16	2·0	1·0	50·0	48·99	40 ml. of Mn solution
17	2·0	1·0	50·0	48·95	Further 20 ml. of Mn solution added during titration
18	2·0	1·0	50·0	49·00	40 ml. of Mn solution, total bulk diluted to 750 ml.
19	1·0		1·0	—*	(*cf.* p. 167.)
20	1·0		1·0	—*	Total bulk only 250 ml.
21	1·0		1·0	1·50	No manganese present
22	1·0		1·0	1·15	10 ml. of ferric solution added
23	1·0		1·0	0·98	As in 22, but slower titration
24	1·0		1·0	0·98	25 ml. of ferric solution added

Ten ml. of Ferrous Solution.—It was possible to titrate satisfactorily with only 1 ml. of indicator present (Expt. 9), although the colour was not strong during the final ml. of titration. An improvement was effected (Expt. 10) by adding a further ml. of indicator after adding 9 ml. dichromate, and allowing a minute between each subsequent drop addition of dichromate. Slow titration was avoided (Expt. 11) by increasing the iron-content. An increased manganese-content (Expt. 12) gave rise to very slow fading towards the end-point; the result was slightly high.

Twenty-five ml. of Ferrous Solution.—Experiments 13 and 14 were check determinations of what was found to be the most satisfactory order of iron-content.

Fifty ml. of Ferrous Solution.—In Experiment 15 the persistence and strength of colour was poor; a further ml. of indicator was necessary after the addition of 35 ml. of dichromate and again near the end-point. Great improvement was effected by doubling the manganese-content and the amount of indicator originally added (Expt. 16); the colour-change was easy to follow, although not very strong, and, in contrast with Experiment 15 (which shows evidence of too rapid oxidation), a good result was obtained. Experiment 17 demonstrated that the accuracy of the result was unaffected by adding the additional 20 ml. manganous solution half way through the titration. A more intense colour was obtained by diluting the solution before titration (Expt. 18).

One ml. of Ferrous Solution.—No definite result was obtained (Expt. 19*) even after concentrating the bulk of solution (Expt. 20*). To favour oxidising conditions manganese was omitted (Expt. 21). Fading was still slow; the result was high, despite a delayed rate of titration. Even in the presence of 10 ml. of ferric solution moderately rapid titration (over 2 to 3 minutes) gave a high result (Expt. 22). However, good results were obtained in the presence of 10 ml. of ferric solution by allowing 30 seconds between subsequent drop-additions of dichromate (Expt. 23) and by a faster rate of titration in the presence of 25 ml. of ferric solution (Expt. 24).

The amount of iron present (ferrous or ferric) is of great importance in the control of the oxidation of diphenylcarbazide by dichromate. In experiments with iron solutions having an iron-content equivalent to $N/10$ ferrous solution, the presence of less than 10 ml. caused a slow fading of colour and a consequent tendency for high results in the titration. With larger amounts the fading of colour was more rapid until over 25 ml. the oxidation of diphenylcarbazide passed the coloured stage too quickly; larger amounts of indicator were required, and titration results tended to be low. The last-named faults could be corrected by increasing the amount of manganous sulphate mixture and diluting the solution. Except with very low iron concentrations the addition of an excess of manganese had no appreciable effect; the absence of the reagent had an effect similar to that with higher iron concentrations.

End-points.—Three kinds of end-point were observed: *viz.* (i) recurring colour from colourless, obtained with stored indicator (Expt. 6), or, in certain experiments involving high iron concentration (Expt. 15), (ii) fading without continued addition of dichromate, where less than 5 ml. iron solution was present (Expt. 21), and (iii) an extremely sharp decolorisation when the last drop of dichromate could be sub-divided into "split drops" of about 0·01 ml., a sharp, permanent decolorisation being observed between the addition of two such "split drops." The last type of end-point was obtained when the promoting and retarding factors had been correctly balanced (Expts. 11, 13, 14, 18, 24).

Proposed Method

The trustworthiness of the following method, intended for general application, has been carefully checked. Apart from the known advantages of potassium

dichromate as a standard, a sharp change is provided at the end-point, the end-point does not involve the arbitrary selection of one tint in a range of colours, and the titration is not affected by the previous use of stannous chloride and mercuric chloride, or zinc, for reduction. In contrast with the methods of Brandt and of Barneby and Wilson, foreknowledge of the approximate result is not required, but when an approximate result is known, as in duplicate analysis, the control may sometimes be simplified, as is indicated.

PREPARATION OF REAGENTS.

Indicator.—Diphenylcarbazide (0·1 g.) is dissolved in 30 ml. of glacial acetic acid without warming, and the solution is diluted to 100 ml. with distilled water.

Manganous Sulphate Mixture.—This consists of an aqueous solution of 200 g. of anhydrous manganous sulphate, 250 g. of sulphuric acid (sp.gr. 1·84), 170 g. of phosphoric acid (sp.gr. 1·75), diluted to 2 l.

Ferric Solution.—An aqueous solution of ferric alum or ferric sulphate, with a little sulphuric acid added to promote stability, containing 5·584 g. of iron per l. (equivalent in iron concentration to $N/10$ ferrous solution).

PREPARATION FOR TITRATION.—Reduce the iron, if necessary, either by the stannous chloride and mercuric chloride method, or by the solution of zinc in acid, in the usual manner. The only free acid present should be hydrochloric acid, and there should be an excess of approximately 10 ml. of that conc. acid after reduction. Dilute the solution to approximately 500 ml., and add 20 ml. of manganous sulphate mixture, 10 ml. of ferric solution, and 1·0 ml. of indicator. At this stage there should be no indicator colour.

THE TITRATION.—The first four drops of $N/10$ dichromate solution should be added at the rate of one drop per 30 seconds. The rate of development of the violet colour will, with experience, provide an approximate estimate of the order of the titration; if the colour is faint, add 1 ml. more of indicator before proceeding. Continue adding dichromate, one drop every 30 seconds, for the first ml., one drop every 2 seconds afterwards, unless there is sufficient indication of a medium or high titration (of 10 ml. or over). For all titrations up to 10 ml. no addition of indicator must be made near the end-point; beyond that stage the final colour-change is strengthened by the addition of 1·0 ml. of indicator a few drops before the end-point. If the titration is not complete after the addition of 15 ml. of dichromate solution, dilute the titrated solution to approximately 750 ml., and add a further 20 ml. of manganous sulphate mixture. With a prolonged titration add more indicator, if necessary. The imminence of the end-point is shown by the rapid fading of the indicator colour; when this is observed, add 30 ml. of conc. hydrochloric acid. The final few drops of dichromate are added at the rate of one each 30 seconds until it appears that one more drop will discharge the colour. At this stage add incomplete drops of dichromate, each about 0·01 ml., on the side of the titration vessel, and wash in, until the indicator colour just disappears and does not re-appear during a further 30 seconds. This is the end-point. The normal practice of shaking the titrated solution between additions of dichromate is observed. Deduct from the titration the amount of dichromate used in oxidising the indicator (to be determined as described later).

SUGGESTED MODIFIED TECHNIQUE FOR DUPLICATE EXPERIMENTS.—If it is known that the amount of iron corresponds with less than 20 ml. of $N/10$ solution, add the equivalent ferric solution to make approximately 20 ml. before the titration, thereby allowing the advantage of indicator addition towards the end-point.

If the first titration exceeded 20 ml., ferric solution is unnecessary in the second titration; if the first titration exceeded 25 ml., dilute the solution to 750 ml., increase the manganous sulphate solution to 40 ml., and add 2·0 or 3·0 ml. of indicator before the second titration.

The rate of titration should be adjusted to take a minimum period of 8 minutes. With these modifications, further additions during titration, other than indicator and hydrochloric acid near the end-point, are avoided.

THE DICHROMATE VALUE OF THE INDICATOR.—To a mixture of 40 ml. of conc. hydrochloric acid, 25 ml. of ferric solution and 20 ml. of manganous sulphate mixture, add water to give approximately 500 ml., and then add 6·0 ml. of indicator. Titrate with $N/10$ dichromate solution, adding the first 10 drops at 10-second intervals, and concluding the titration with 30-second intervals. Shake the solution after each addition of dichromate. Approach the end-point with "divided drops" of dichromate solution, with no further addition of acid.

Examples of the standard procedure for unknown iron concentrations (*a* experiments) and also where the approximate iron-content is known (duplicate or *b* experiments) are given in Table III. In these experiments 25·0 ml. of the ferrous solution required 24·02 ml. of $N/10$ dichromate solution in titrations with the use of the old external indicator (potassium ferricyanide), and 24·10 ml. of $N/10$ permanganate solution. In each experiment the solution contained 10 ml. of hydrochloric acid, a further 30 ml. being added when the end-point was approached. The dichromate figures are corrected for indicator oxidation.

TABLE III

Exp. number	Dilution ml.	Manganese solution ml.	Ferrous solution ml.	Ferric solution ml.	Indicator addition		Dichromate solution (0·1 *N*) ml.
					Before titr. ml.	Towards end ml.	
25*a*	500	20	1·0	10·0	1·0	—	0·96
25*b*	500	20	1·0	20·0	1·0	1·0	0·93
26*a*	500	20	10·0	10·0	1·0	—	9·61
26*b*	500	20	10·0	10·0	1·0	1·0	9·63
27*a*	500+250	20+20	25·0	10·0	1·0	1·0	24·08
27*b*	500	20	25·0	—	1·0	1·0	24·08
28*a*	500+250	20+20	50·0	10·0	1·0	1·0+1·0	48·15
28*b*	750	40	50·0	—	2·0	1·0	48·16
29*a*	500+250	20+20	100·0	10·0	1·0	1·0+1·0+1·0	96·29
29*b*	750	40	100·0	—	3·0	1·0	96·32

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Colorimetric Analysis by the Photo-electric Cell

BY NORMAN STRAFFORD, M.Sc., F.I.C.

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INTRODUCTION.—The instrument here described was first demonstrated at a meeting of the Hull Chemical and Engineering Society on February 19th, 1935. Since that date Bolton and Williams[1] have described a photo-electric instrument which they employ for the measurement of the colour of oils and other liquids.

DESCRIPTION.—The general plan of the instrument is shown schematically in Fig. 1. The source of light is a 6-volt 3 ampere motor headlamp bulb, set at

SCHEMATIC DIAGRAM OF PHOTO-ELECTRIC COLORIMETER

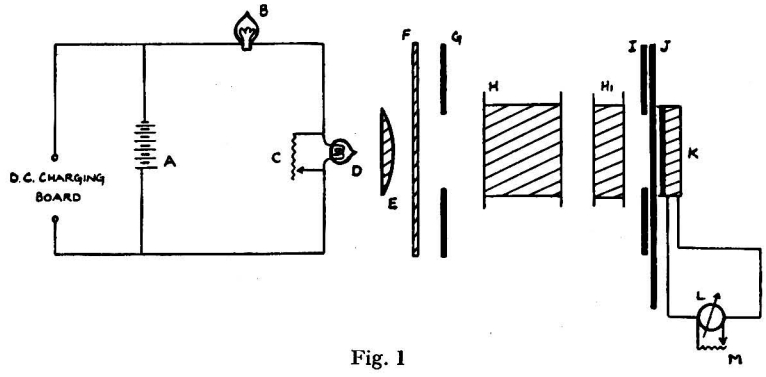


Fig. 1

FIG. 1. A, accumulator. B, Barretter lamp. C, variable shunt (approx. 50 ohms max.). D, lamp. E, lens. F, colour filter. G, stop. H, cell for copper sulphate solution (if required). H1, cell for liquid under test. I, stop. J, movable shutter. K, photo-electric cell. L, galvanometer. M, variable shunt.

the focal point of a lens of short focus—about 1½ in. By the use of a lens of this type, maximum light intensity is ensured. An important feature of the lamp circuit is the incorporation of a Barretter or current-regulating lamp (Philips' model No. 1120†). This eliminates with remarkable efficiency changes in the light intensity due to minor fluctuations of the source of supply. The lamp and Barretter device are fed from an accumulator (12 to 18 volt), preferably maintained throughout the tests at a charge at 3 to 4 amperes. By this means constancy of light intensity is attained. Alternatively, the mains alternating current, transformed down to 12 to 20 volts, may be used as a source of supply, provided that the mains voltage is reasonably steady.

The current output of the Weston photo-electric cell No. 594‡ is measured on a galvanometer such as the Tinsley portable galvanometer§ with combined lamp and scale (Model No. 530; sensitivity approximately 10 mm. per microampere).

* A demonstration of the instrument was given.
† Philips Lamps, Ltd., 145, Charing Cross Road, W.C.2.
‡ Weston Electrical Instrument Co., Kingston By-Pass, Surbiton, Surrey.
§ H. Tinsley & Co., Werndee Hall, South Norwood, S.E.25.

To permit of measurement of light of relatively high intensity, which would cause more than full-scale deflection, the galvanometer is provided with a variable shunt (1000 ohms maximum).

THEORY OF COLOUR MEASUREMENT.—From a consideration of the Beer–Lambert law it is evident that the most convenient measure of relative colour intensity is afforded by the use of *absorption density* rather than *percentage transmission* or *percentage extinction*, since the resulting calibration curve connecting light absorption with concentration of coloured substance is rectilinear under suitable conditions.

For a system which obeys the Beer–Lambert law* we may write

$$I_0 = I \times 10^{ecd}$$

where I_0 and I represent the intensity of the incident and emergent light, respectively, c the concentration of coloured substance, d the thickness of solution, and e the molecular extinction coefficient *for a given wave-length of light*, or,

$$\log \frac{I_0}{I} = \text{absorption density} = \Delta_c = ecd$$

From this it follows that, for light of a given wave-length, the absorption density for a constant thickness of a solution is directly proportional to the concentration of the coloured substance.

$$\text{For } \Delta_c = ecd \quad \text{and} \quad \Delta_{c1} = ec_1d$$

$$\therefore \quad \frac{c}{c_1} = \frac{\Delta_c}{\Delta_{c1}}$$

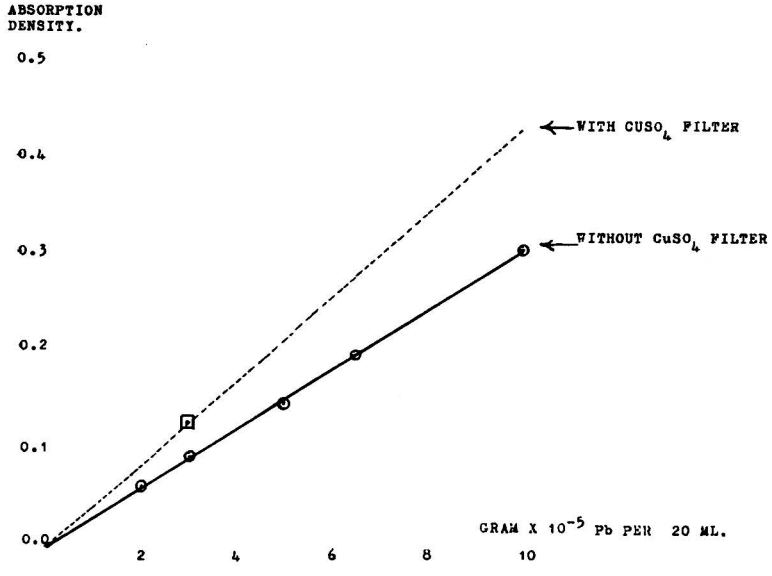


Fig. 2

Colorimetric Determination of Lead by means of Diphenylthiocarbazone
0·5 cm. Cell: Wratten Green-Blue (h) Filter

* That is to say, one in which no alteration in colour is caused by chemical changes occurring on diluting the solution.

For monochromatic light, therefore, the absorption density at constant thickness of liquid is a linear function of the concentration of the coloured substance (*cf.* Fig. 2).

A typical curve showing variation in absorption density of a coloured solution with wave-length of the light is shown in Fig. 3. It is evident that the curve connecting absorption density and concentration of coloured substance (see Fig. 2) has the greatest slope relative to the concentration axis when the light employed has the wave-length at which the solution under examination exhibits maximum absorption.

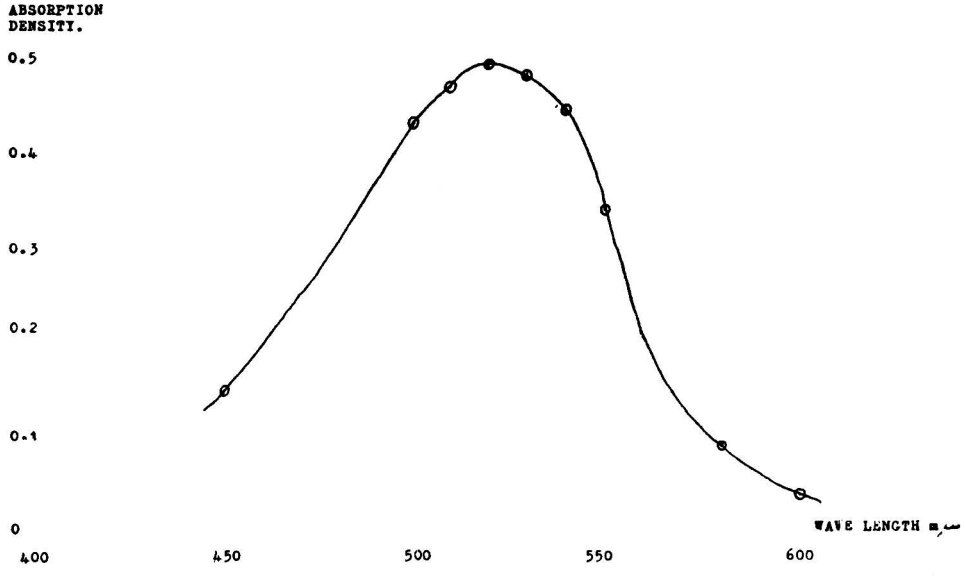


Fig. 3

Absorption of Light by Lead Diphenylthiocarbazone (measured on Hilger Spectrophotometer)
3 G.M. × 10^{-5} in 20 M^{s}
1·0 CM.Cell

It may be noted here that no over-all measure of colour is afforded by determining merely the values at one or two arbitrary points on the absorption density/wave-length curve. It is only by taking into account in some suitable manner the whole of this curve that a quantitative measure of the total colour of the substances is obtained. In the special case of colorimetric analysis, however, which is based on determining the ratio of the colour depths of solutions of the *same substance* in known and unknown concentrations, measurement of absorption densities at any one wave-length where absorption occurs is entirely adequate.

MEASUREMENT OF ABSORPTION DENSITY.—So far it has been assumed that the absorption density of a solution can be measured at any desired wave-length.

This in practice can be done with great accuracy by the Hilger absorption spectrophotometer, the actual width of the wave-length band being only about 5$m\mu$. With the present instrument, however, it is possible to measure only the average absorption density at a limited number of wave-length *ranges*, since the

light transmitted by the best commercial filters is far from monochromatic. Evidently, therefore, the absorption density/wave-length curve is necessarily only an approximation to the truth, even after the disturbing effect of the infra-red rays has been eliminated by the use of a copper sulphate filter,* as employed by Bolton and Williams.

For the *relative* measurements required in chemical analysis by colorimetry, however, this factor is of no practical significance, and within reasonable limits the use of filters which transmit a comparatively broad band of the spectrum does not reduce the accuracy of the results. Moreover, there is often no appreciable gain in sensitivity as a result of using the additional "minus infra-red" filter; see, for example, the calibration curves shown in Fig. 2.

ADVANTAGES OF USING THE PHOTO-ELECTRIC CELL IN COLORIMETRIC ANALYSIS. —These may be summarised briefly as follows:—(1) Under suitable conditions, the calibration curve is virtually a straight line, and can therefore be constructed with a minimum of effort.

(2) The calibration curve, once constructed, is always available, so that in subsequent colorimetric analysis no standard comparison solutions need be prepared.

(3) Measurements are independent of the human eye, and are of appreciably greater accuracy than those afforded by the simpler visual colorimeters.

CHOICE OF BEST WORKING CONDITIONS IN COLORIMETRIC DETERMINATIONS.— (i) *Choice of Filter.*—For a given coloured solution, the filter transmitting light approximating to that of maximum absorption by the solution should generally be chosen. But, since filters vary in the percentage of the total light they transmit, it does not necessarily follow that the one showing the highest absorption density gives the most accurate results. Often, in fact, it is best to compromise by choosing a filter which gives a full scale I_0 reading, although it may give a lower absorption density than another filter or filter combination, with a lower total transmission. In the latter case the galvanometer readings may be crowded into a smaller range.

For example, in a determination of lead by colorimetric measurement on a chloroform solution of lead dithizone the following figures were obtained:

Filter	Wave-band $m\mu$	I_0	I	I_0–I	log. $\frac{I_0}{I}$
Wratten H	430–550	7·30	5·80	1·50	0·10
Wratten B+H	480–550	2·00	1·25	0·75	0·20
Wratten G	510–700	10·05	8·80	1·25	0·06

Of course, if a more powerful light were available, the I_0 reading with the B + H filters could be increased to a point where the sensitivity would be greater than with the H filter, owing to the higher absorption density given with the B + H combination.

(ii) *Best Working Range.*—The most suitable filter having been found, the next point is to discover the best working range, *i.e.* the depth of colour giving the biggest change of galvanometer reading per unit percentage change in concentration of coloured substance.

* Alternatively, infra-red light may be removed by using a "minus infra-red" (glass) filter obtainable from Ilford, Ltd., London.

From a consideration of the fact that the absorption density, a logarithmic function of the *reciprocal* of the transmission, varies in a linear manner with the concentration of the coloured substance, whilst the galvanometer readings are directly proportional to the transmission, it can be shown that the maximum sensitivity of the instrument is obtained when the transmission lies between the limits 70 and 10 per cent. (corresponding with absorption densities of 0·15 and 1·0, respectively). The depth of liquid examined should, therefore, preferably be such that the observed absorption density lies between these limiting values. This means, in practice, that if the initial galvanometer reading be 10·0, conditions should be chosen such that the readings with the test solutions lie between 1·0 and 7·0 (see Fig. 4). In practice, this is attained by the use of a solution of quite a pale colour in a thickness of 0·5 cm. or 1 cm.

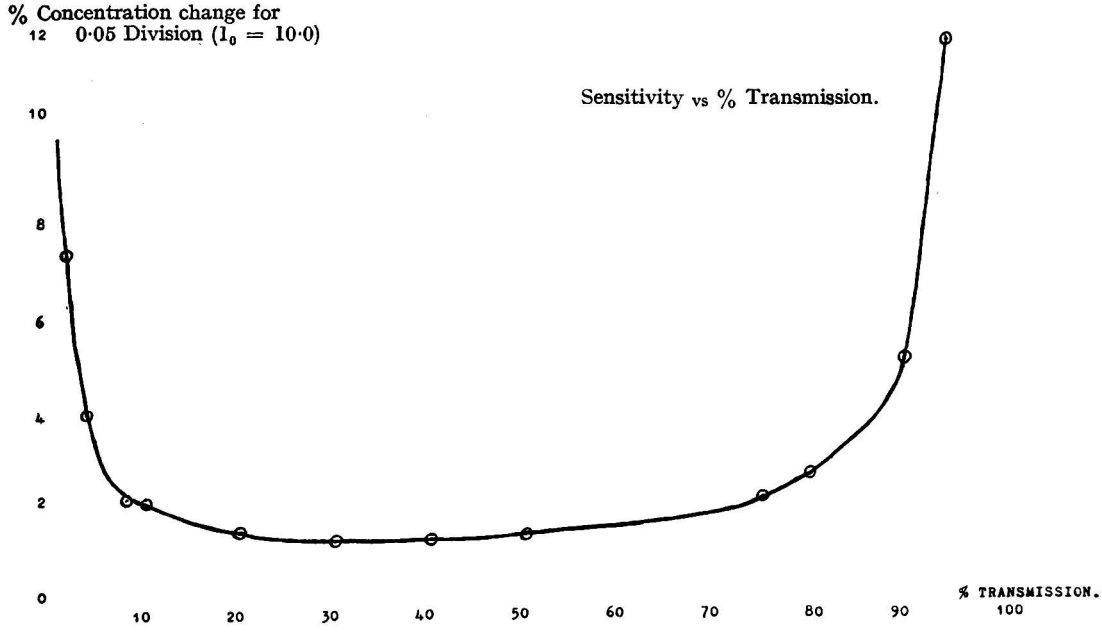


Fig. 4

In carrying out accurate colorimetric determinations the following precautions should be observed:

(1) Readings should not be taken until the photo-electric cell has assumed a condition, after the initial "fatigue" effect, in which a light of constant intensity gives a steady galvanometer reading.

(2) The Barretter lamp requires 5 to 10 minutes to reach a steady condition, and the apparatus should therefore be allowed to "warm up" for this length of time before any measurements are made. During this period the lamp providing the light must be protected by the shunt from temporary excessive current.

(3) In determining I_0, the initial intensity, the measurement should be carried out with an absorption cell similar to the one used in the test, *filled with pure solvent*, in the path of the light.

NOTES ON THE APPLICATIONS OF THE INSTRUMENT.—Accurate colorimetric determinations can be made only on true coloured *solutions*. It is interesting to note that the classical method for determination of lead as the colloidal sulphide does *not* come within this category. Photo-electric measurements applied to a solution containing the colloidal sulphide give entirely erratic and non-reproducible results. It is all the more surprising, therefore, that in the usual visual Nessler cylinder method the error is not greater than it actually is, namely, ±8 per cent. absolute.[2] Whatever the explanation may be, however, it is certainly clear that the colloidal sulphide method should be replaced by a truly colorimetric one.

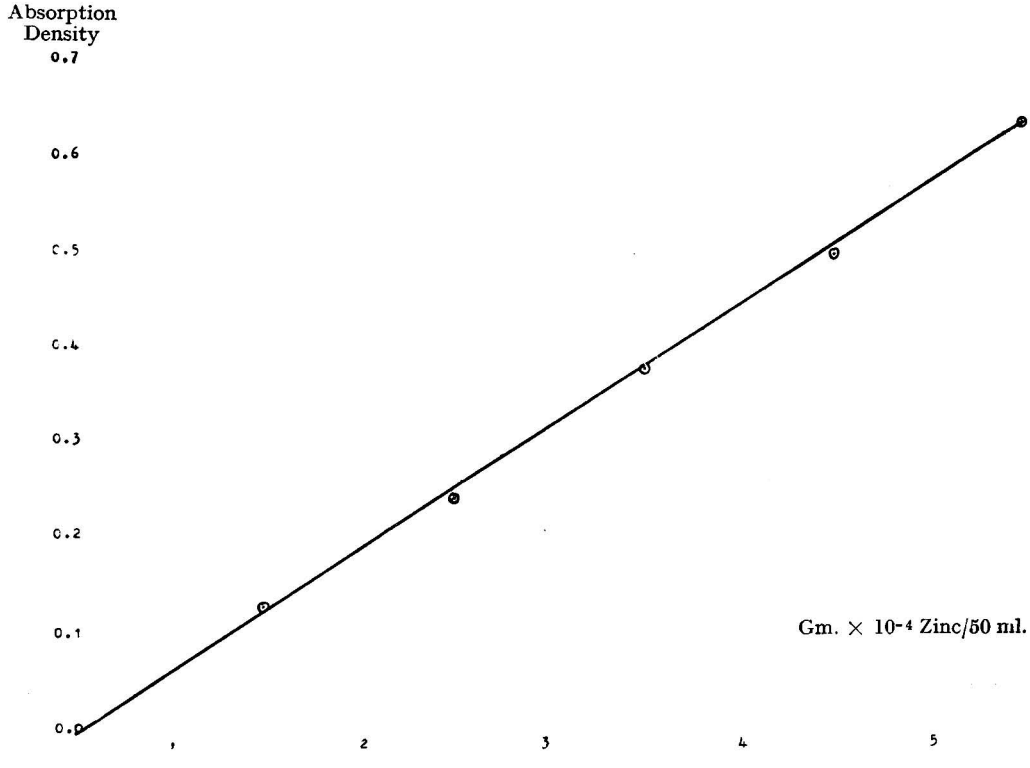


Fig. 5
Nephelometric Determination of Zinc, as Ferrocyanide
2-inch Cell. White Light

Such a method is afforded by the use of the diphenylthiocarbazone reagent.[3] The lead is measured in the form of the red dithizone complex in chloroform or carbon tetrachloride solution. The calibration curve is readily reproducible, and the over-all error of the measurement with the photo-electric cell is of the order of ±2 per cent. absolute.

Although the photo-electric cell method fails with the pseudo-colorimetric colloidal sulphide method for lead, it can be applied with success to true nephelometric measurements. For example, in my experience the error of the nephelometric determination of zinc as ferrocyanide by the visual method is approximately ±25 per cent. absolute. By using the photo-electric equipment the error is reduced

to about ±4 per cent. or less. A calibration curve for this determination is shown in Fig. 5.

An interesting application of the instrument is the measurement of the relative turbidities of commercial liquids. Where it is desired to measure turbidity only, any effect due to variations in depth of colour may be eliminated almost entirely by using a filter of similar colour. For example, varnishes would be measured in a yellow light, red wines with a red filter, and so on.

My thanks are due to Imperial Chemical Industries, Limited, for permission to publish this communication.

References

1. E. R. Bolton and A. K. Williams, ANALYST, 1935, **60**, 447.
2. Second Report of the Sub-Committee on the Determination of Arsenic, Lead, etc., *id.*, 541.
3. Fischer and Leopoldi, *Z. ang. Chem.*, 1934, 90. Winter, Robinson, Lamb, and Miller, *Ind. Eng. Chem. Anal. Ed.*, 1935, **7**, 265.

RESEARCH DEPARTMENT, ANALYTICAL SECTION
I.C.I. LTD. (DYESTUFFS GROUP)
MANCHESTER

Discussion

Mr. E. R. BOLTON said that he was naturally very interested to hear this paper, as Mr. Williams and he had been working on similar lines to the author. On reading the literature they had found that many endeavours had been made to use photo-electric cells for colour measurement, and that various devices had been used to obtain an absolutely steady light. The first difficulty they had encountered was that a very large proportion of infra-red rays was recorded by the photo-electric cell, and they therefore introduced a copper sulphate filter to remove all infra-red rays. As Mr. Strafford had evidently tried this suggestion, he would like to know with what success. The wave-bands used by Mr. Strafford were wider than they had employed, and consequently Mr. Strafford was putting more light through the instrument; had he tried narrower wave-lengths to see whether there was the same effect of variation of light?

Mr. R. MILTON said that a valuable point about this paper was the attempt to make the source of light constant. In his experiments this had been difficult. If the source of light varied, the galvanometer reading varied. One small point worth mentioning was that if the Barretter lamp took 4 minutes to settle down after the voltage fluctuation, how was one to ensure that such a change was not in progress while the reading was being made?

Mr. K. A. WILLIAMS said that Mr. Strafford was using the photo-electric cell for a different purpose from the one that Mr. Bolton and he had in view. His aim was colorimetric analysis; theirs was to fix or measure the colour of liquids. For that reason, Mr. Strafford was able to use wider wave-bands than they were. The effective wave-band of the half-a-dozen filters Mr. Bolton and he used was something of the order of 20 to 30$m\mu$, and was as narrow a wave-band as was obtainable by means of filters. The photo-electric cells they used did not show any fatigue at all in their instrument, owing to the very small energy output. The instrument they had brought there that evening to show the meeting was one that could be

plugged into the electric mains almost anywhere, and could be run for five hours on end without noticeable change in the zero reading.

Mr. D. M. WILSON said that one point upon which no speaker had touched was the difficulty that no two Weston cells gave exactly the same output. The "flickering" of the electric light source raised a difficulty in an industrial area; he had had to put in an independent generating set to supply the current.

Mr. STRAFFORD, replying, said he entirely agreed as to the value of the Bolton and Williams method for the removal of infra-red light by copper sulphate. However, for the *relative* measurements required by colorimetric analysis, as distinct from absolute colour measurement, there was often no appreciable advantage in removing the infra-red light, as was shown by Fig. 2. He had found that the Barretter lamp, fed from an accumulator maintained on charge, extremely effective in maintaining constancy of the light-source over long periods, as indicated by the steadiness of the galvanometer needle within ±0·02 scale division. He was not prepared to say much about the "fatigue" of the photo-electric cell. Probably a new cell showed more fatigue than one that had been in use for some time. In his experience it was best not to take a reading until the cell had passed the fatigue stage, and gave a steady galvanometer deflection for a light of constant intensity. This condition was usually realised within 5 minutes of the initial exposure of the photo-electric cell to light.

Notes

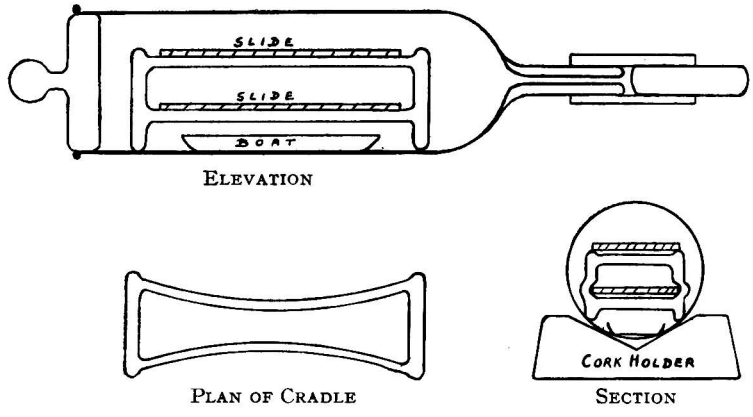
The Editor wishes to point out that the pages of the Journal are open for the inclusion of short notes dealing with analytical practice and kindred matters. Such notes are submitted to the Publication Committee in the usual manner.

NOTES ON THE BROMINE VAPOUR METHOD FOR THE DETERMINATION OF THE HALOGEN ABSORPTION OF OILS

SINCE this method was first published (ANALYST, 1928, **53**, 69) there have been some criticisms of the crudeness of the original apparatus. Hence an improved form has been devised with the object of eliminating the risk of contamination of the film with brominated cork and wax, and also the unpleasant effects of unused bromine vapour escaping into the atmosphere of the laboratory. The new apparatus has also the advantages that two determinations can be made simultaneously, and that, being entirely of glass, it is easy to keep clean.

The apparatus (see Fig. 1) consists essentially of a cylinder closed at one end with a ground-glass stopper, and at the other (being a capillary) with a glass rod held in place by a piece of pressure-tubing. Inside is a glass cradle, made to carry two microscope slides, and a porcelain boat to hold the bromine. A determination is made exactly as previously described (*loc. cit.*), except that at the end of the absorption the capillary is attached to a water-pump, the stopper removed, and the excess of bromine drawn away. The slides may then be removed and "dried"

at a suitable temperature, or the whole apparatus (still attached to the pump) may be placed on a warm plate and thus heated in a current of warm air until all free bromine has gone.*



ELEVATION

PLAN OF CRADLE

SECTION

Fig. 1

It should, perhaps, be mentioned here that some commercial samples of bromine have not given satisfactory results, whereas "Anala R" bromine has always been satisfactory.

H. TOMS

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THE EXTRACTION OF LEAD BY MEANS OF DIPHENYLTHIOCARBAZONE

A NOTE by Garratt (ANALYST, 1935, **60**, 817) on "The Extraction of Lead by means of Diphenylthiocarbazone" deals with a difficulty that has arisen in the colorimetric matching of the final solutions obtained when foodstuffs and biological materials are examined for traces of lead by wet oxidation with nitric and sulphuric acids, followed by the diphenylthiocarbazone extraction method of Allport and Skrimshire (*id.*, 1932, **57**, 440). The same problem has also been met with in the examination of biological tissues by Roche Lynch, Slater and Osler (*id.*, 1934, **59**, 787).

In an investigation on the amount of lead present in the blood of "normal" patients and in the blood of patients suffering from malignant tumours, who are undergoing treatment with certain lead preparations, the "Titrimetric-Extraction" method of Wilkins, Willoughby, Kraemer and Smith (*Ind. Eng. Chem., Anal. Ed.*, 1935, **7**, 33) is being used. During the preliminary experiments an attempt was made to combine this procedure with a colorimetric method, whereby the lead was finally estimated as lead sulphide. This modification, however, was not further proceeded with, because although the estimation could be carried out in less time, the results obtained were not quite as accurate. Nevertheless, since no difficulty was experienced at any time in making the colorimetric comparison, the method used may be of interest.

The first lead-dithizone complex (Wilkins *et al.*, *loc. cit.*) is extracted, as described, with two 10-ml. portions of 1 per cent. nitric acid, the nitric acid extract being transferred to a Pyrex boiling-tube (6 in. × ¾ in.) and the small amount of

* The apparatus is obtainable from C. L. Müller, 6, Parton Street, London, W.C.1.

chloroform in the liquor evaporated. The residual solution is transferred to a 25-ml. graduated flask, the washings (with re-distilled water) of the boiling-tube are added, and the volume is made up to 25 ml. with re-distilled water. The whole volume or an aliquot part can then be prepared for colorimetric comparison as usual.

With the small amounts of lead present in blood, the lead-dithizone complex need be extracted only with two 5-ml. lots of 1 per cent. nitric acid, and the combined volumes, after evaporation of the chloroform, added directly to the Nessler tube and prepared for comparison as before.

Of course, as Garratt (*loc. cit.*) suggests, the interfering colour might possibly be avoided by using other methods for the oxidation of the original material, and Roche Lynch *et al.* (*loc. cit.*) only experienced the difficulty when the method was applied to tissues, and not to blood. However, extraction of the lead-dithizone complex from two specimens of liver with nitric acid also gave colourless solutions.

Should this method be applicable in general to any dithizone extract, it is undoubtedly much simpler to carry out than the complete oxidation of the dithizone.

This work was carried out on behalf of the Liverpool Medical Research Organisation.

L. ELLIS

DEPARTMENT OF ORGANIC CHEMISTRY
THE UNIVERSITY OF LIVERPOOL

Report of the Essential Oil Sub-Committee to the Analytical Methods Committee

REPORT No. 12

THE DETERMINATION OF ASCARIDOLE

THE Essential Oil Sub-Committee makes the following recommendations with regard to the determination of ascaridole in chenopodium oil.

The method recommended depends on the titration of the iodine liberated under specified conditions by the ascaridole from a strongly acidified solution of potassium iodide. In order that accurate and concordant results may be obtained, it is essential to adhere to the conditions laid down, as the reaction which takes place is complex and cannot be expressed by a simple equation.[1] The conditions governing the reaction have been studied, and those adopted result in the maximum liberation of iodine. This is considerably in excess of the theoretical amount liberated by a normal peroxide; the factor used for calculating the results is an empirical one and is based on the titration of a sample of pure ascaridole, which itself had been standardised by titration with titanous chloride.

The following reagents are required:

Glacial acetic acid.
90 per cent. acetic acid.
5 *N* potassium iodide solution in water (83 per cent. w/v).
Hydrochloric acid, B.P. (32 per cent.).
N/10 sodium thiosulphate solution.

METHOD OF DETERMINATION

About 2·5 g. of the oil are weighed out into a 50-ml. graduated flask, and the flask filled to the mark with 90 per cent. acetic acid. Some of this solution is

placed in a narrow burette graduated in 20ths of a ml., with the divisions sufficiently far apart to enable the readings to be estimated to 1/100th ml. The burette should be fitted with a stop-cock and jet of sufficiently wide bore to enable 5 ml. to be run out in not more than 5 seconds.

The reaction is carried out in a stoppered tube[2] as used for the determination of aldehydes and ketones—approximately 150 mm. long by 25 mm. in diameter. In this tube are placed 3 ml. of the potassium iodide solution, 10 ml. of glacial acetic acid, and 5 ml. of hydrochloric acid; the tube is stoppered and placed in a freezing-mixture until the temperature is reduced to −3° C. Approximately 5 ml. of the acetic acid solution of the oil are then run in from the burette *as rapidly as possible*; the tube is immediately stoppered and the contents mixed and allowed to stand in a cool place (not in a water-bath) for 5 minutes, during which time their temperature will rise slowly, but must not exceed 10° C.

The exact volume run out of the burette should be noted after about 2 minutes, when the contents have drained down completely.

At the expiration of 5 minutes, the contents of the tube, now dark brown in colour, are titrated directly with $N/10$ sodium thiosulphate solution. The end-point is sharp, the final liquid being *white* and turbid, owing to the finely-dispersed minute oil-globules. Starch must not be used as an indicator.

At the same time, a blank experiment is carried out under the same conditions of temperature, 3 ml. of the potassium iodide solution, 10 ml. of glacial acetic acid, and 5 ml. of hydrochloric acid being used, as a small amount of iodine is always liberated by the reagents. Before titrating this blank, however, it is necessary to dilute it with 20 ml. of water. The amount indicated by the blank should be deducted from the titration. Each ml. of $N/10$ sodium thiosulphate is equivalent to 0·00665 g. of ascaridole.

From the results of our experiments, we are of opinion that the maximum variation in the percentage of ascaridole, as determined by this method, should not exceed ±1 per cent.

Several series of determinations have been carried out by members of the Sub-Committee, and these have shown that certain precautions are necessary in order to obtain correct results.

The following should be noted:

(1) The mixing of the acetic acid solution of the oil with the cooled reagent must be as rapid as possible, and the reading of the burette must not be recorded until it has been allowed to drain down completely.

(2) The reaction mixture must not be allowed to stand for longer than 5 minutes, and the final temperature must not exceed 10° C.

(3) The reaction mixture must not be diluted before titration.

(4) The end-point should be sharp and the final liquid white. If the end-point is not sharp and the liquid has a yellow colour, which an excess of thiosulphate does not remove, the determination has not been carried out correctly, and a low result will be obtained, or, if an excess of thiosulphate has been added in an attempt to decolorise the solution, the result will be high.

(5) The blank determination on the reagents must be diluted with 20 ml. of water before titration.

Results

The following results have been obtained by six members of this Sub-Committee, the determinations being carried out at the same time with the same solutions and burettes.

The acetic acid solution of the oil contained 4·748 per cent. w/v of chenopodium oil, and the blank required 0·2 ml. of $N/10$ sodium thiosulphate solution.

Member	Amount of solution of the oil taken for the test ml.	Corrected volume of $N/10$ $Na_2S_2O_3$ required ml.	Percentage of Ascaridole found
1	4·91	28·2	80·4
2	4·95	28·1	79·5
3	4·96	28·42	80·25
4	4·95	28·37	80·25
5	4·96	28·42	80·25
6	4·89	28·87	81·15

Range	79·5 to 81·15
Max. difference	1·65
Mean	80·30
Standard deviation	0·49

Further results were obtained on another sample of oil, when the determinations were carried out independently in the various members' laboratories, each using his own reagents and apparatus.

Member	Percentage of Ascaridole found			
1	81·0	81·9	82·6	
2	80·8	80·9	81·2	81·3
assistant (1)	81·5	81·7	81·9	82·7
assistant (2)	81·3	81·8	82·3	82·5
3	80·7	80·8	80·9	81·7
4	82·1	82·3	82·6	
5	81·3	81·4	81·9	82·4

Range	80·7 to 82·7
Max. difference	2·0
Mean	81·7
Standard deviation	0·62

References

1. T. Tusting Cocking and F. C. Hymas, ANALYST, 1930, **55**, 183.
2. Report No. 10, ANALYST, 1932, **57**, 774.

(Signed),

W. H. Simmons (Chairman), C. T. Bennett, S. W. Bradley, L. E. Campbell, Thos. H. Durrans, J. W. Harrison, Ernest J. Parry, C. Edward Sage, John H. Seager, T. Tusting Cocking (Hon. Secretary).

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.

CARBOLIC OINTMENT

ON January 17th three firms of druggists were summoned by the Fulham Borough Council at the West London Police Court for having sold carbolic ointment deficient in phenol to the extent of 38, 21 and 25 per cent., respectively. The analyses were not disputed, and it was agreed that all the cases should be taken together.

Mr. Davey, for the Fulham Borough Council, said that the contention of the prosecution was that carbolic ointment should be based on British Pharmacopoeia formula, and contain 3·0 per cent. of phenol. In one of the samples the percentage was only 1·54.

Mr. T. Tickle, B.Sc., F.I.C., called by the defence, said that he did not agree that the proportion of phenol in the samples did not conform to the British Pharmacopoeia, since the proportion of phenol that should be present in carbolic ointment was not specified, and no directions as to the method of storage or the length of time the ointment should be kept were given. Only the proportion prescribed for the purpose of manufacture was given, and in successive editions of the B.P. the percentage of phenol had been reduced and was now 3 per cent. In the U.S. Pharmacopoeia it was 2 per cent. Phenol was rapidly volatilised, and there was steady evaporation both during and after preparation. The loss of 0·5 per cent. allowed by the Fulham Public Analyst was a wholly arbitrary standard; the loss might be greater or less than that during manufacture, and there was a progressive loss during storage. In his (the witness's) opinion an ointment containing 1·54 per cent. of phenol would still be carbolic ointment.

Mr. C. E. Corfield, B.Sc., F.I.C., said that he had made analyses of successive layers of a jar of carbolic ointment which had been stored for three years under ordinary conditions in a jar covered with parchment and a grease-proof cover and wrapped in sealed transparent paper. The top layer ($\frac{1}{2}$ inch) of the ointment showed an average percentage of 1·5 per cent. of phenol, whilst at a depth of 1 inch the average percentage was 2. In his opinion a standard of 2·5 per cent. was unreasonable, because it took into account only the loss of phenol during the preparation of the ointment.

Dr. P. J. Hamill said that he was not aware of any point at which the proportion of phenol would cease to be effective. Carbolic ointment was largely used to allay irritation, but if too strong, phenol would burn some skins. Where it was necessary for the protection of the public that a preparation should retain definite quantities of a constituent, the proportions were defined by the B.P., and directions were given for retaining them. He did not consider 1·54 per cent. of phenol too small a proportion for the ointment to be effective.

The magistrate (Sir Gervais Rentoul) said that, in view of the expert evidence that had been given, he could not hold that the prosecution had proved its case, and the summonses would be dismissed.

Two guineas costs were awarded in each case.

Ministry of Health

CIRCULAR 1518*

Draft Milk (Special Designations) Order, 1936

THE following circular has been sent to the Clerks of County and County Borough Councils, the Common Council of the City of London, and of Metropolitan Borough Councils:

1. I am directed by the Minister of Health to transmit for the information of the Council a draft of the Milk (Special Designations) Order, 1936. Notice of intention to make the Order in the form of this draft has been inserted in the *London Gazette* of the 24th January, 1936, and it is intended that the Order shall come into operation on the 1st April, 1936.

2. It will be seen that the draft Order prescribes four special designations for milk, namely, "Tuberculin Tested" and "Accredited" for raw milk, and "Certified (Pasteurised)" and "Pasteurised" for milk which has been pasteurised. The new designation "Tuberculin Tested" will replace the existing designations "Certified" and "Grade A (Tuberculin tested)"; "Accredited" will replace "Grade A"; and "Certified (Pasteurised)" milk will be Tuberculin Tested milk which has been treated by the pasteurising process.

3. The draft Order further provides that licences authorising the use of the designation "Tuberculin Tested" by producers shall be granted by County Councils (outside London) and the Councils of County and Metropolitan Boroughs and of the City of London. It is hoped, therefore, that the Council will make the necessary arrangements to enable them to deal without delay with any applications that may be received for such licences to come into operation on or after the 1st April next. The Minister proposes to refer to the appropriate Council any application for a licence for the sale of milk as "Certified" or "Grade A (Tuberculin tested)" under the existing Order which is received by him too late to be dealt with before the 1st April.

4. Licences granted by the Minister and in operation on the 1st April authorising the sale of milk as "Certified" or "Grade A (Tuberculin tested)" will continue in operation (subject to the provisions of the new Order) until the 31st December, 1936, as if they were licences authorising the use of the new designation "Tuberculin Tested." During that period the Minister will continue to exercise the necessary supervision and control over the licences, but their renewal at the end of the year will be a matter for the Council.

5. The Minister proposes to address a further circular to all Local Authorities when the Order is finally made.

6. A copy of this circular is being sent to the Medical Officer of Health, and additional copies may be obtained from His Majesty's Stationery Office at any of the addresses shown below or through any bookseller.

* H.M. Stationery Office, Kingsway, London, W.C.2; 120, George Street, Edinburgh, 2; York Street, Manchester, 1; 1, St. Andrew's Crescent, Cardiff; 80, Chichester Street, Belfast. 1936. Price 1d. net. The Draft Order, itself, is also obtainable from H.M. Stationery Office.

Department of Scientific and Industrial Research

REPORT FOR THE YEAR 1934*

THE Advisory Council of the Department, the Chairman of which is Lord Rutherford, is able to point to a very encouraging response to the offer of increased Government support which the Department made last year to Research Associations, provided that the Associations, on their part, obtained correspondingly increased contributions from the industries they serve. Negotiations with 13 out of 18 Research Associations in receipt of financial assistance from the Department have been completed and, in every instance, offers of increased grants on a new basis have been made and accepted. The report shows that work of interest to practically every industry in the country is being carried out either in the Department's own establishments or in the laboratories of the research associations it has fostered.

NATIONAL PHYSICAL LABORATORY.—A comprehensive account of the work is given in the Report of the Laboratory for the year 1934 (*cf.* ANALYST, 1935, **60**, 469).

FOOD INVESTIGATION.—The Board's own Report (*cf.* ANALYST, 1935, **60**, 687) gives a brief review of the work as a whole, and the progress of individual investigations is described by the Director of Food Investigation in the present Report. Among subjects of interest mentioned are the following:—

Oxidation of Fat.—Work has been continued on the effect of the *p*H of the aqueous medium in contact with fat on its rate of oxidation, and on the pro- and anti-oxidant properties of different substances dissolved in the aqueous phase. In connection with the possibilities of reducing microbial growth on meat by the introduction of traces of ozone into the store, experiments are being carried out on the rate of oxidation of various fats exposed to definite concentrations of the gas. Low concentrations of ozone, *e.g.* five parts per million for five hours a day, greatly accelerate the oxidation of films of pure fat (lard, beef-fat and egg-oil) at 0° C.

Rate of Growth of Fungal Hyphae.—A microscopical technique has been devised for estimating the rate of growth of fungal hyphae in sections of living tissues from the apple fruit. Measurements by this method are highly correlated with the rate of radial advance determined by the inoculation method. By this new method comparisons of resistances may be made within a short time after sectioning the fruit. The microscopical method, unlike the inoculation method, is capable of wide applications to plant material other than the apple.

BUILDING RESEARCH.—A full account of the recent work of the Building Research Station is given in the Annual Report of the Building Research Board (ANALYST, 1935, **60**, 321).

FOREST PRODUCTS RESEARCH.—Marked progress has been made in the investigation of the relation between the structure and the technical properties of wood. In connection with woodworking and seasoning tests, an abnormal condition of the fibre walls has been found to affect the machining properties and seasoning qualities of certain timbers. The phenomenon will be further investigated. Chemical analysis of matched samples of timber has been carried out with the object of determining whether any direct relation exists between variations in the chemical composition and the technical properties of the timber.

Fungal Decay.—The temperature relations of further species of fungi which attack wood have been determined. A paper containing the results which had previously been obtained in work on a number of important species was published during the year. A chemical investigation of the effect on oak heartwood of the fungus responsible for the production of "brown" oak has been undertaken. The

* H.M. Stationery Office, Adastral House, Kingsway, price 3s. net.

results so far achieved suggest some action by the fungus on the tannin in the wood, but there is also evidence that the major wood components may be affected. Tests of the strength properties of "brown" oak tend to indicate that the action of the fungus is slow in affecting the strength properties in the early stages of infection.

A further study of the effect of heat treatment for the destruction of the furniture beetle (*Anobium punctatum*) has been in progress, with satisfactory results.

METALLURGICAL RESEARCH.—In the investigation of the behaviour of materials at high temperatures, data have been obtained concerning the creep and growth of five types of cast iron at temperatures up to 538° C. Both growth and creep of ordinary cast iron have been found to be reduced considerably by a preliminary heat treatment at 650° C. or even at 600° C. Work is now in hand on tests on magnesium and aluminium alloys. The experimental difficulties involved in the determination of the solubility of gas in liquid aluminium have been surmounted, and measurements have indicated that the solubility is very low; this work links up closely with the study of the nature of oxide films and their permeability to gases. Another branch of the problem under investigation is the correlation of the density with actual gas-content of aluminium alloys.

WATER POLLUTION RESEARCH.—A summary of the results in the different investigations is given in the Annual Report of the Board (ANALYST, 1935, **60**, 37).

CHEMICAL RESEARCH.—A detailed review of the work of the Chemical Research Laboratory since its inception has been published (*cf.* ANALYST, 1935, **60**, 613).

Road Tar Research.—Changes which occur in tars during exposure under various conditions are being investigated in order to develop an artificial ageing test similar to outdoor exposure. The formation of benzene-insoluble material in the surface layers of tar by exposure to sunlight has been confirmed by a visual proof and the slow absorption of oxygen by tar in the dark has been measured by a volumetric method. Methods of increasing the rate of set of tars are being examined.

Ruthenium Red.—In the course of work on the chemistry of ruthenium, the constitution of "ruthenium red" has now been shown to be $(RuOH.H_2O.4NH_3)Cl_2$, thus bringing this inorganic colouring matter into line with modern views of co-ordination compounds. In addition, a variety of ruthenium compounds containing 2:2′-dipyridyl and 2:2′:2″-tripyridyl have been isolated.

ATMOSPHERIC POLLUTION.—A special investigation on a larger scale than any at present in progress is contemplated by the Atmospheric Pollution Research Committee at a specially selected industrial centre. It is hoped that this investigation will also throw light on a further problem of general interest, namely, the extent to which one area is affected by pollution coming from another area; as a preliminary step an investigator has been appointed to take charge of the investigation on the selected site in due course, as well as to assist in the solution of the general problems which arise.

A record of the work for the year ended 31st March, 1934, was published as a special report (*cf.* ANALYST, 1935, **60**, 409).

INDUSTRIAL RESPIRATORS.—A respirator has been developed which, while being an efficient protection against the inhalation of harmful dusts, is designed to allow the greatest possible range of vision for the worker. At the close of the period covered by this report trials were being carried out in selected factories, mines and quarries, in collaboration with the Home Office and the Mines Department.

Laboratory work on the detection, in the atmosphere, of small quantities of toxic gases commonly occurring in industrial processes is almost complete. Methods

have been developed for the detection and estimation of hydrogen sulphide, arsine, organic halogen compounds, carbon monoxide, sulphur dioxide, nitrous fumes, aniline vapour, hydrogen cyanide, chlorine, carbon disulphide vapour, and benzene vapour, and are now being examined for their suitability for use under industrial conditions. Work has been begun with the object of standardising performance tests for respirators for use in industry as a protection against such gases.

These investigations have been undertaken at the request of the Home Office, and carried out on behalf of the Department by the Chemical Defence Research Department.

RESEARCH ASSOCIATIONS.—As in previous Reports, brief summaries are given of the investigations of the Research Associations during the year.

In the condensed summary issued with this Report special attention is directed to the following points *inter alia*:

Food Research.—A method has been developed for determining the kind and quantity of acids in a jam, whereby it is also possible to estimate, *e.g.* the quantity of raspberries in the jam.

The true explanation of why sausages lose their fresh appearance and become pale under certain conditions of storage has also been found.

Flour Milling.—A process of hot aeration, developed by the Association, has given results exceeding all expectations. The process improves very markedly the baking quality of wheat flour and wheat meal, sterilises the flour against mites, and partly sterilises it against bacteria and mould fungi.

Cotton.—The fundamental work on the new principle of opening and cleaning cotton is making rapid progress, and the first machine is now on the market. This machine, known as the Shirley Lint Recoverer, is for extracting from all kinds of mill wastes the good spinnable fibre that they contain.

Wool.—During the year the Wool Research Association has made considerable progress in perfecting, for use on a commercial scale, the process developed for the production of shrinkage-resisting wool. The process has advantages over existing well-known wet methods of treatment, which produce in knitted or woven material a degree of unshrinkability. It will be used almost entirely for experimental weaving, and preparation machinery, looms and cloth rooms will be near at hand.

Rubber.—The Rubber Association, in collaboration with the gas industry, is investigating suitable rubber joints for gas-mains, which will withstand the vibration due to modern traffic conditions. Another investigation deals with the development of a "smell-proof" rubber tubing, for attaching to gas appliances.

Leather.—The Leather Research Association has been studying methods of preserving leather from attack by micro-organisms at all stages of the tannery process. It has also devised a quick and easy optical method for judging the value of sole leather.

Boots and Shoes.—In the study of worn shoes by the Boot and Shoe Research Association it was found that a person's health is frequently reflected in the condition of his footwear. Sometimes the upper leather has been badly affected by uric acid, apparently from the foot perspiration, whence it is not unreasonable to conclude that the wearer had a constitutional tendency towards rheumatism or gout. In at least one instance a person was led to discover that he was diabetic by the fact that the Association found his boots to be impregnated with sugars.

Cyprus

ANNUAL REPORT OF THE GOVERNMENT ANALYST FOR THE YEAR 1934

THE Government Analyst (Dr. S. G. Willimott) reports that the total number of samples analysed was 2255, as compared with 2342 in 1933. For the administration of the Food and Drugs Law the island is divided into seven districts. During the last decade there has been no decrease in the adulteration of foods. The explanation is that dishonest shopkeepers and contractors will continue their usual falsification so long as they know that the legal penalty, if detected, will be only a matter of 5s. to 20s. including costs. Obviously it is often very profitable to continue adulteration on such terms. Furthermore, the sharp practice of labelling any foodstuffs as "adulterated," whether or not the commodity is genuine, appears to render the seller immune from legal action. It would seem, therefore, that little improvement can be expected until a new Food and Drugs Law is enacted to replace the present out-of-date measure.

OLIVE OIL.—The olive harvest was again not good, so that genuine supplies of the oil were restricted, and these conditions offered opportunity for wholesale adulteration. Of the 47 samples examined, 18 were adulterated, a variety of imported vegetable oils, notably soya bean and cotton seed oils, and refined solid fats used in cooking, being employed for the purpose.

ACORN FLOUR IN COFFEE.—Of the 219 samples of coffee examined, 44 were adulterated, some form of starch, in quantities ranging from 4 to 65 per cent., being the favourite adulterant. In the last quarter of the year a number of samples from Paphos district were found to contain up to 10 per cent. of starchy matter, originating from roasted acorn flour. Paphos is the only district where the oak tree flourishes, and, in fact, boasts a pig industry in which the acorns are used as a feeding stuff.

MILK.—The adulteration of fresh milk of all kinds continues to be widely practised throughout the Colony. Nine of the 45 samples examined were adulterated by watering, skimming or both.

CRIMINAL CASES.—Two hundred and twenty-one exhibits in 38 criminal cases were examined on behalf of the Police. The malicious poisoning of the wells of Eptitagonia village was found to be due to Paris green.

It was possible to substantiate a charge of stealing poultry by the isolation of chicken fat from the broken potsherds, although the cooking pot had been cleaned and smashed on the approach of the Police.

Caustic Soda Poisoning.—A case of attempted suicide was traced to poisoning with caustic soda. This case was not included in the series of poisoning published in the *British Medical Journal* (June 9th, 1934, p. 1022). It is remarkable that the suicides who use caustic soda are nearly always women. Caustic alkalis have been scheduled as poisons.

Sugar-coated Tablets containing Poison.—There have been fewer cases of accidents to children through eating sugar-coated tablets in mistake for sweets. An example and a safeguard has been set by the Government in discontinuing the importation of all sugar-coated tablets containing poison.

New Zealand

ANNUAL REPORT OF THE DOMINION ANALYST FOR 1934

In this, the Sixty-eighth Annual Report of the Dominion Laboratory, Mr. W. Donovan gives the following totals for the numbers of samples analysed in main and branch laboratories:—Wellington, 5656; Auckland, 3124; Christchurch, 2416; and Dunedin, 1190. The samples examined at the branch laboratory were mainly of milk submitted by the Department of Health. Increasing use is being made by the Police of scientific assistance in connection with criminal and other investigations.

Sale of Milk "as it comes from the Cow."—In the Auckland District 30 samples of milk below the legal standard for solids-not-fat, but not containing added water, were examined, and in one case in which the solids-not-fat were 8·0 per cent. (legal minimum 8·5 per cent.) the supplier was successfully prosecuted. This was an important decision, as it is often contended that it should be lawful to sell milk which is below the legal standard, provided it is sold as it comes from the cow. Whilst at first sight this appears reasonable, it must be borne in mind that such milk is cheaper to produce than milk of higher quality, and the seller of the former has therefore an advantage over the seller of the latter. If the sale of such sub-standard milk were allowed, the tendency would be towards lowering the average quality of milk. The legal standard is a very reasonable one, and is readily obtained, as is shown by the average composition of the milk sold (fat, 4·1; solids-not-fat, 9·1 per cent.). From the consumers' point of view it matters little whether milk of poor quality is naturally so or has been made so by the addition of water.

Saltpetre in Milk.—Saltpetre was present in six samples of milk purchased from two vendors. This is a very unusual form of adulteration, and the chemical was probably added as a preservative, though it would not be effective for the purpose.

Antimony in Rubber Beer-hose.—Red rubber hose used for conveying beer into casks was found to contain antimony compounds. Although the risk of poisoning would be very slight, it was recommended that its use be discontinued.

Anti-Obesity Preparations.—Eight preparations recommended for the treatment of obesity were examined for thyroid or other iodine-containing material. Most of them consisted of well-known laxatives, and in no instance was a significant amount of iodine found.

Identification of Paint in a Burglary Case.—In a case of breaking and entering, a tire-lever, on which was a small fragment of red paint, was found in the possession of the suspect. It was shown that this fragment was similar in composition to paint on a door which had been forced. It was also convincingly demonstrated by means of low-power photography that the marks on the door had been made with the tire-lever in question.

Application of Copper Sulphate to Lake Water.—A problem of special interest was the treatment of Lake Pupuke (Auckland) with copper sulphate to kill a growth of *Ceratium*, which was causing odour and flavour troubles in the Devonport water supply. The Lake holds 21,000,000 tons of water, and the copper sulphate was applied in the ratio of 0·3 part per million, a strong solution being sprayed from a launch running on parallel courses over the lake. The rate of application was varied according to the depth. The treatment took ten days to complete, and resulted in the entire destruction of the *Ceratium*. The worst samples examined prior to treatment contained 1000 *Ceratium* per ml. The average count was 400. Since the completion of the treatment samples have been examined regularly each month, and no trace of *Ceratium* has been found.

MISCIBILITY OF LOCOMOTIVE-BEARING AND CYLINDER OILS.—A sample of gelatinous sediment from an oil-feeder, sent in by the New Zealand Railways, contained 32 per cent. of saponifiable matter, about half the fatty acids being oxidised acids, while the locomotive-bearing oil had 18 per cent. of compounding materials (blown rape oil). According to Archbutt and Deeley (*Lubrication and Lubricants*, 1927, pp. 158–164) a certain minimum of blown oil, depending on the character of both the mineral oil and the blown oil, is necessary to give a clear solution, and if the amount of mineral oil is increased, separation occurs.

A method of testing the miscibility of the two types of oil was devised. Samples were mixed in various proportions in weighed 100-ml. thick-walled centrifuge tubes, which were then heated for an hour in the water-oven and centrifuged until cold. The oil could then be poured from the stiff gelatinous sediment. The tubes were allowed to drain inverted, wiped free from remaining oil, and weighed. Mixtures of bearing oil with equal amounts of cylinder oil gave 7 per cent. of sediment for two different samples of cylinder oil, but another cylinder oil gave no sediment. Of four subsequent samples of locomotive-bearing oils, each containing about 20 per cent. of blown rape oil, two gave about 5 per cent. of sediment, and two gave none at all, on mixing with various samples of cylinder oil.

CARDBOARD PACKING FOR PEARS.—Corrugated wrapping which had caused brown stains on pears, with which it had been in contact, was examined for the Scientific and Industrial Research Department. Rose and Lutz (*J. Agric. Res.*, August, 1933) found that such stains could be produced by alkali from the sodium silicate adhesive used. The wrapping examined, when soaked in water, yielded solutions with alkalinity equivalent to 1·1 per cent. of sodium hydroxide calculated on the weight of the cardboard. Another sample gave 0·70 per cent. of sodium hydroxide. It appeared that the injury was due to the use of sodium silicate adhesive.

CITRUS TAINT IN SHIP'S HOLD.—The ships that carry produce to England have much insulated space not used on the return voyage, and it has been found that some of this can be profitably used to carry oranges from Jamaica or Palestine. To prevent the volatile citrus oils from tainting the hold, a small proportion of ozone is produced, in the air circulating in the hold, by means of an electric ozoniser, which is kept at the lowest power during the voyage, and after the removal of the fruit is kept running for several days on full strength. In test examinations, generally no citrus odour could be detected after the hold had been well cleaned by a current of air.

In order to test for taint, dishes containing charcoal were distributed in the locker and left as long as the ship's loading arrangements permitted, from a few days upwards. On the return of the charcoal to the laboratory it was heated in a flask in an oil-bath to 130° C. and distilled with steam, the distillate being collected in wide-mouthed stoppered bottles, in quantities of a few drops, a few ml., 10 ml., and 50 ml. The bottles were warmed to 37° C. and the smell noted. The charcoal used in some cases was coconut-shell charcoal, but charcoal "special for gas absorption" was also tried. As received from the suppliers, however, this material gave on steam-distillation a strong smell of chlorinated hydrocarbons, and before use had to be twice distilled with steam. In one or two of the first tests the distillates gave a slight smell of essential oils, but not definitely of orange oils. On later occasions the smell did not differ markedly from that of the distillate from charcoal kept as a control in the ship's butter store. As there was no experience on the utility of the test, it was considered that, although fruit and meat could be safely carried in the holds, there might possibly be some risk of taint for butter. Since these tests were made, a locker that chanced to have no cargo was used to carry a box of butter to England. On arrival it was found to be unaffected, so that probably butter would also be safe in such a hold.

ABSTRACTS OF PAPERS PUBLISHED IN OTHER JOURNALS

Food and Drugs

Composition of Meat Extracts and Meat Cubes. H. E. Cox. (*Chem. and Ind.*, 1936, **55**, 69–71.)—The influence of these products as gastric stimulants outweighs their food value, and it is generally agreed that, of their constituents, the meat bases, particularly creatine and creatinine, are of primary importance, the proportion of albumoses and peptones being also significant, whilst meat fibre, coagulable proteins and gelatin are of subordinate importance. Meat bases are taken as the nitrogenous constituents other than gelatin, albumoses and peptones, and the coagulable and insoluble proteins, the factor usually adopted being 3·12. When comparing different varieties of extract the moisture-content must, of course, be taken into account. Analyses, such as those of Stutzer and of Hehner (1895–1897), made before methods for the determination of creatine and creatinine were perfected, show that the total nitrogen-content of the products of that time was similar to that of the modern products, but that there was more meat fibre. Shortly after 1905, when Folin's method for determining creatine and creatinine was published, it was established that the proportion of creatine and creatinine amounted to upwards of 10 per cent. of the dry matter, and figures of 9 and 10 per cent. on an ordinary sample containing 18 per cent. of moisture were commonly obtained. The exhaustive analyses of Bigelow and Cook (1906), when examined in the light of Emery and Henley's investigation of meat extracts prepared from different parts of the animal, are seen to refer to extracts made not from muscle fibre only, but also from other organs not now used in the preparation of the better grades. The author gives the following analyses of typical products procured during the last two years. Nos. 1 and 2, representing the best grades sold to-day, are dry extracts; 3, 4 and 5 are well-known semi-fluid brands, No. 5 contains substantial amounts of vegetable proteins; 7 and 8 are of Australian origin. In the best grades, *viz.* 1, 2, 3, 7, and 8, there is no appreciable amount of gelatin, the albumoses and peptones are insignificant, meat fibre is low and the creatine and creatinine satisfactorily high. A standard of 6 per cent. of creatine and creatinine has been adopted in some countries.

MEAT EXTRACTS (1934–1935)

		1	2	3	4	5	6	7	8
Water,	per cent.	20·8	17·9	28·1	34·6	23·3	42·9	17·3	15·2
Organic matter,	,,	58·2	58·5	51·6	47·2	48·2	41·2	62·7	64·2
Mineral matter,	,,	21·0	23·6	20·3	18·2	28·5	15·9	20·0	20·7
Salt,	,,	4·6	4·8	10·4	11·1	18·6	10·9	3·9	4·2
Total nitrogen,	,,	8·9	8·9	6·2	7·1	5·8	5·5	9·6	9·7
Meat fibrin,	,,	1·3	1·9	5·3	6·0	8·6	0·6	—	—
Albumoses and peptones,	,,	0·5	1·0	0·4	22·4	13·3	17·5	0·7	0·9
Gelatin,	,,	0·2	0·3	0·5	0·5	0·4	8·7	trace	trace
Non-nitrogenous extractives,	,,		traces only			10·0	7·0	trace	trace
Creatine and creatinine,	,,	8·5	8·1	5·5	6·1	6·3	1·4	4·7	4·6

Analyses of 12 brands of meat cubes (not "soup cubes") examined recently are given. The results show that the suggestion, that their content of meat extract may be determined by regarding meat extract as containing 6 per cent. of creatine and creatinine, must be taken with caution. All samples examined contained a binding material and added flavouring. The commonest binding material is gelatin, although starch is often used and occasionally lactose. The whole of the gelatin may be assumed to have been added as binder; usually it is from 7 to 15 per cent. The salt-content varies from 20 to 30 per cent., or even more, and most of it is a direct addition. Carbohydrate, which is always found, is due to vegetable tissue and starch binder; the latter may reach 20 per cent. Meat extract gives no reaction for carbohydrate, although traces may be found if liver has been used. The only analytical result upon which the estimation of meat extract can be founded is the content of creatine and creatinine, and estimation on this basis can be only an approximation. In estimating these bases care must be taken to avoid including sugars present as such or formed by hydrolysis. A recent form of adulteration is the enrichment of meat extracts and cubes with soya bean. Its presence may sometimes be inferred from microscopical examination, but its exact estimation depends upon the determination of glutamic acid. This is complicated by the fact that glutamic acid occurs naturally in small amounts in some beef extracts. The desirability of a standard for these products needs to be considered.

MEAT CUBES (1934–1935)

		1	2	3	4	5	6	7	8	9	10	11	12
Water,	per cent.	13·0	13·6	12·7	14·7	6·5	10·0	13·7	10·8	13·4	10·0	11·8	8·1
Organic matter,	,,	48·7	57·9	50·7	54·2	34·4	60·6	55·5	51·9	50·5	62·6	51·5	66·2
Mineral matter,	,,	38·3	28·5	36·6	31·1	59·1	29·4	30·8	37·3	36·1	27·4	36·7	25·7
Salt,	,,	31·5	23·0	29·5	27·2	50·3	22·7	25·2	34·7	30·1	20·8	30·0	18·8
Fat,	,,	2·4	4·4	0·9	—	—	—	—	—	—	—	—	—
Carbohydrates,	,,	19·3	3·4	29·1	29·0	9·5	12·0	12·3	40·4	29·7	11·5	25·0	33·2
Total nitrogen,	,,	4·2	8·0	3·3	3·9	3·9	7·8	6·8	1·7	3·2	8·2	4·2	5·0
,, protein,	,,	26·4	50·2	20·8	24·2	24·4	48·6	42·5	10·9	19·8	51·4	25·9	31·1
Gelatin,	,,	traces	+	traces	+	—	—	+	—	—	+	—	+
Starch,	,,	+	—	+	—	+	+	—	+	+	—	+	—
Creatine and creatinine,	,,	1·6	2·0	1·5	0·9	0·8	1·6	1·9	0·7	1·6	1·8	1·6	1·8

A. O. J.

Analysis of Vinegar. W. Ruziczka. (*Chem. Ztg.*, 1936, **60**, 48–49.)—Although not related to any exclusively constitutional properties, the iodine value and oxidation value of Schmidt (*Z. Unters. Lebensm.*, 1935, **69**, 472; Abst., ANALYST, 1935, **60**, 705) give a numerical idea of the oxygen and iodine consumption by different constituents of vinegar (primarily aldehydes, higher alcohols, caramel, vitamins), and thus have a practical value for distinguishing between spirit vinegar and wood vinegar. The iodine values for these kinds of vinegar have been determined by three different methods used previously by the author in his investigation of urine (*Klin. Woch.*, 1935, **14**, 775–778). As applied to vinegar, the first method, which is a modification of the rapid method of Margosches, Hinner and Friedmann (*Z. angew. Chem.*, 1924, **37**, 334), is as follows:—Twenty-five ml. of vinegar are shaken for a short time with 20 to 25 ml. of alcoholic $N/5$ iodine solution and 200 ml. of water in a stoppered flask. After 5 minutes the

contents of the flask are titrated with $N/10$ sodium thiosulphate solution. A blank determination is carried out in the same way. By the second method, as used by Lieb and Lanyar (*Hoppe-Seyler's Z.*, 1929, **181**, 199) for the determination of homogentisic acid, 25 ml. of vinegar are diluted with 90 ml. of water, 2 ml. of 2 per cent. starch solution are added, and the mixture is neutralised with sodium bicarbonate. Standard iodine solution is then run in until the blue colour persists for ½ minute after adding a little more sodium bicarbonate. The third method, which is a modification of that described by Wüstenfeld (*Lehrbuch der Essigfabrikation*, 1930, Verlag P. Parey, Berlin; ANALYST, 1935, **60**, 706), is as follows:— Twenty-five ml. of vinegar are treated with 100 ml. of water, 40 ml. of $N/2$ sodium hydroxide and 10 ml. of $N/10$ iodine and potassium iodide solution. After 15 minutes the mixture is acidified and titrated back with sodium thiosulphate solution. The first method must be regarded as giving the iodine value in the strictest sense of the term. The second method gives the lowest results. The third method gives very much higher results, and it must be assumed that partial oxidation has taken place. The oxidation value was determined in the usual way, 50 ml. of vinegar being mixed with 10 ml. of 20 per cent. sulphuric acid and titrated with $N/10$ potassium permanganate solution until the pink colour persisted for 2 minutes. It was found that, if the solution were now boiled for a short time, a further quantity of the permanganate solution was required to impart a permanent colour, thus indicating that oxidation of acetic acid takes place, catalysed by the initial products of oxidation, as this additional consumption of permanganate does not occur with pure acetic acid. The saponification value of the vinegar is useful, as it indicates the ester-content. Wine vinegars give higher values than spirit or artificial vinegars. The values found for the acidity, saponification value, oxidation value and iodine value (by the three methods described) are given below.

Vinegar diluted ready for use	Acidity	Saponification value	Oxidation value	Iodine value		
				I	II	III
From glacial acetic acid	56·87	56·69	—	0·02	—	0·03
Spirit vinegar	56·23	57·12	0·35	0·05	0·03	0·35
Vinegar "essence" (artificial vinegar)	49·68	49·94	0·05	0·03	—	0·09
Wine vinegar I	52·93	55·84	1·72	0·21	0·11	2·67
Wine vinegar II	52·97	56·42	1·57	0·16	0·08	2·36

All values are expressed as ml. of $N/10$ solution per 10 ml. of vinegar.

A. O. J.

Absorption Spectra of Honey. L. H. Lampitt and P. Bilham. (*Chem. and Ind.*, 1936, **55**, 71–72.)—Schon and Abilgaard (*Z. Unters. Lebensm.*, 1934, **68**, 502) showed that the addition to honey of invert sugar prepared by acid hydrolysis could be detected spectroscopically by an absorption band with a head at 282·5$m\mu$ due to hydroxymethylfurfural, which is formed on hydrolysing sucrose with acid or heating fructose with acid. It may be formed when genuine honey is heated during factory operations. Such honey, although answering to the Fiehe test for hydroxymethylfurfural, does not show the absorption band, which, however, is given when as little as 2 per cent. of invert sugar prepared by acid hydrolysis is added to genuine honey. It is suggested that the Fiehe test is too delicate, and that only strongly positive results should be taken as evidence

of adulteration. The absorption spectrum appears to be a better criterion, but needs more study. Neither test can detect invert sugar prepared by enzymic action. A. O. J.

Monohydroxypalmitic Acid in Butter-fat. A. W. Bosworth and G. E. Helz. (*J. Biol. Chem.*, 1935, **112**, 489–492.)—An optically active monohydroxypalmitic acid has been separated from butter-fat. The lead soap of this acid is soluble in ether, and the barium soap is soluble in benzene. The purest specimen obtained had mol. equiv. 271 (theory 272), m.p. 17° C., iodine value (Hanus 30 minutes) 0·48, acetyl value 175 (theory 178). S. G. S.

Biochemical

Nutritive Value of Soya Bean Powder and Soya Bean Oil Treated with Methanol. S. Kajizuka. (*J. Soc. Chem. Ind. Japan*, 1935, **38**, 745–746B.)—Soya bean extracted with an azeotropic mixture of petroleum spirit (benzine) and methanol has been found, by experiments with rats, to produce a more nutritive meal and oil than those obtained by extraction without the methanol. The presence of methanol could not be detected either in the meal or in the oil, and since no fall in n_D occurred in the oil, it was concluded that no methylation had taken place. A slight increase in the proportion of methoxyl groups was noticed in the meal, but no physiological effect on the rats could be detected. Meal containing 0·1 and 1 per cent. of methanol, respectively, and oil containing 0·67 and 6·7 per cent., had no ill-effects on the rats. The addition of 0·1 mg. per rat per day of a vitamin *A* preparation to the oil extracted with benzine and methanol from the soya bean caused normal growth, so that soya bean oil is regarded as highly nutritive. The nutritive value of soya bean oil after methanolysis at 20° C. was found to be lower than that of the original oil. D. G. H.

Calcium and Magnesium-content of the Flesh of Different Animals. M. Takamatsu. (*Hoppe Seyler's Z. Phys. Chem.*, 1936, **238**, 99–100.)—The calcium and magnesium contents of the flesh of various animals have been determined. Blood-vessels, tendons, nerves and fat were removed from the fresh flesh, and the muscle tissue was chopped small, dried in an oven, and brought to constant weight in a desiccator. The dried material was then carefully charred and ashed in a porcelain crucible, the ash was dissolved in dilute hydrochloric acid, and the solution was diluted to a definite volume with water. An aliquot portion of the solution was used for the determination. Calcium was precipitated as oxalate, and the precipitate was ignited and weighed as calcium oxide. After the calcium oxalate precipitation, magnesium was precipitated from the filtrate as magnesium ammonium phosphate, which was ignited and weighed as magnesium pyrophosphate. The results are summarised in the following table. Attention is drawn to the high calcium-content of the flesh of the adder, snail, mussel, and crab, and also to the high magnesium-content of the flesh of the adder and snail. An interesting point is that the flesh of the adder, which in Japan is used as a tonic, has a high content of sulphate, calcium and magnesium.

Species	Content per 100 parts of fresh flesh		Content per 100 parts of dried flesh	
	CaO	MgO	CaO	MgO
I. VERTEBRATA				
(*a*) *Mammals*				
Dog	0·0437	0·2019	0·1977	0·9133
Ox	0·0209	0·0853	0·1312	0·5358
Rabbit	0·0303	0·1252	0·1432	0·5905
Whale	0·0273	0·1072	0·1041	0·4083
Pig	0·0251	0·0544	0·1655	0·3588
(*b*) *Birds*				
Hen	0·0216	0·1308	0·0940	0·5968
Wild duck	0·0229	0·0250	0·0837	0·0916
(*c*) *Reptiles*				
Adder	0·3171	0·6398	1·2524	2·5268
(*d*) *Amphibia*				
Toad	0·0196	0·0675	0·1048	0·3605
(*e*) *Fishes*				
Carp	0·0368	0·0707	0·2297	0·4414
Sea-bream	0·0139	0·0952	0·0796	0·5573
Shark	0·0325	0·0762	0·1592	0·3731
Sting-ray	0·0279	0·0657	0·1487	0·3498
Flat-fish	0·0242	0·0850	0·1424	0·5002
II. MOLLUSCA				
Octopus	0·0346	0·1911	0·1683	0·5297
Haliotus	0·0283	0·0855	0·1079	0·3259
Snail	0·1958	0·4103	1·3202	2·7673
Mussel	0·1679	0·0856	0·7819	0·3988
III. ARTHROPODA				
Lobster	0·0378	0·0360	0·1518	0·1445
Crab	0·1750	0·1946	0·8424	0·9368

S. G. S.

Micro-determination of Ferrocyanide in Muscle and Urine. J. G. Edwards and W. D. Langley. (*J. Biol. Chem.*, 1935, **112**, 469–475.)—The method is a modification of that suggested by Williams (*J. Soc. Chem. Ind.*, 1910, **29**, 319), in which the ferrocyanide is decomposed by sulphuric acid and the hydrocyanic acid is distilled off into a caustic soda solution. The apparatus used for the determination consists of a round-bottomed flask having a side arm, which is connected with a wash-bottle containing 20 per cent. caustic soda solution. The neck of the round-bottomed flask is connected with a reflux condenser which is joined to another condenser at their upper ends. The inner tube of the second condenser is filled with glass beads and glass wool in alternate layers, and its lower end dips into a 20 per cent. solution of caustic soda contained in another wash-bottle. The material under investigation is placed in the round-bottomed flask together with sufficient water to give a volume of about 100 ml., and 20 ml. of 30 per cent. sulphuric acid are added, together with a few quartz pebbles. Air is drawn through the apparatus, and the contents of the flask are heated under reflux for 20 minutes for urine, or 45 minutes for muscle tissue. The water in the first condenser is stopped, but boiling is continued until the first condenser becomes hot (10 to 15 minutes). The condensers are then disconnected, the second

one washed with water, and the contents of the receiver are transferred to a beaker and titrated with 0·01 *N* silver nitrate solution, a micro-burette being used with a spot-light focussed on the beaker. With amounts from 1 to 40 mg. of soluble ferrocyanide per 100 g. of material, 98 per cent. has been recovered from pure solutions, urine and blood, and about 90 per cent. from muscle. The method has not been applied to insoluble ferrocyanides. S. G. S.

Application of the Modified Phospho-18-Tungstic Acid Method for the Determination of Cysteine, Cystine and Ascorbic Acid in Urine. K. Shinohara and K. E. Padis. (*J. Biol. Chem.*, 1935, **112**, 709–721.)—If cysteine, cystine and ascorbic acid are present together in urine, they can be determined by the following methods. To determine the cysteine, a test solution is made with 10 ml. of 2*M* sodium acetate solution, 3 ml. of 2*M* acetic acid solution, 10 ml. of the urine, and 4 ml. of the phospho-18-tungstic acid reagent, the total volume being made up to 50 ml. A blank solution is similarly made, except that 6 ml. of 2*M* formaldehyde solution are added 2 minutes before the reagent. The colour intensities are determined after about 15 minutes. The colour standard is prepared by making two solutions, one containing 10 drops of bromthymol blue solution, $4 \times 10^{-4}M$ cysteine solution, 0·4*M* sodium acetate solution, 0·12*M* acetic acid and 4 ml. of the reagent per 50 ml.; and a second solution containing all the above substances except the bromthymol blue. The two solutions are mixed in a proportion that will give a colour in the mixture about the same as that of the test solution containing the urine. If the intensity of the test solution is symbolised by I_t and that of the blank by I_b, then

$$C_o\,(I_t - I_b) = C_{\text{R-S}} \quad .. \quad .. \quad .. \quad .. \quad (1)$$

where C_o is the molar concentration of cysteine in the standard solution (4×10^{-4}) and $C_{\text{R-S}}$ that of the test solution. For the determination of cystine, a test solution is made containing 10 ml. of acetate and 2 ml. of acetic acid solutions, 3 ml. of sodium bisulphite solution (1·0*M*), 10 ml. of the urine and 4 ml. of the reagent, the total volume being 50 ml. A blank solution is made in exactly the same way, except that 2 ml. of a 0·1*M* solution of mercuric chloride are added 2 minutes before the reagent. The colour intensities are determined after 20 minutes. If $I_{t'}$ is the intensity of the test solution and $I_{b'}$ that of the blank, then

$$\frac{C_o(I_{t'} - I_{b'})}{2} = C_{\text{R-S}} + C_{\text{R-S-S-R}} \quad .. \quad .. \quad .. \quad .. \quad (2)$$

where $C_{\text{R-S}}$ and $C_{\text{R-S-S-R}}$ are respective molar concentrations of cysteine and cystine in the test solution. The amount of ascorbic acid present (C_a) is given by

$$C_o(I_b/_2) = C_a$$

which is derived from equation (1). If a perceptible amount of extraneous reducing substances, such as creatinine, is present, the urine must be oxidised by bubbling air through it for 2 to 3 hours and a new I_b value ($_oI_b$) determined; then

$$\frac{C_o(I_b - {}_oI_b)}{2} = C_a$$

By this method from 0·0 to 0·9 mg. of cystine, from 1·1 to 4·8 mg. of cystine, and

from 3·2 to 26·8 mg. of ascorbic acid have been found in 100 ml. of fresh normal urine. The phospho-18-tungstic acid reagent is prepared by the Folin and Marenzi method, except that the addition of lithium salt is omitted.

S. G. S.

Determination of Thiol and Disulphide Compounds, with Special Reference to Cysteine and Cystine. K. Shinohara and K. E. Padis. Reactions of Ascorbic Acid and Glutathione with Phospho-18-Tungstic Acid Reagent. (*J. Biol. Chem.*, 1935, **112**, 697–708.)—Ascorbic acid can be differentiated from cysteine and cystine and also determined by the suggested procedure (see preceding abstract), provided that other extraneous reducing substances are absent. A solution of glutathione is hydrolysed, on standing, to give an intense colour with the phospho-18-tungstic acid reagent. The hydrolysis is catalysed by acids and bases. This peculiarity of glutathione limits the determination of cysteine and cystine, unless the glutathione is previously hydrolysed and determined as cysteine. S. G. S.

Isolation of Pectic Substances from Wood. E. Anderson. (*J. Biol. Chem.*, 1935, **112**, 531–539.)—Pectic substances have been isolated from the sapwood and from the cambium layer of black locust (*Robinia pseudacacia* L.) by means of the usual methods for this type of material. Some portions of these substances approximated closely to certain of the pectinic acids, whilst other portions were similar to the polygalacturonic acid obtained from commercial citrus pectin. The calcium pectate obtained from these materials appeared to be similar to that obtained from citrus pectin. Calcium pectate obtained both from the cambium and the sap-wood was found to contain *d*-galacturonic acid. Although the sugars present were not identified, methyl pentose sugars were apparently absent. The sap-wood probably contained less than 3 per cent., and the cambium layer less than 13 per cent. of pectic material. It is suggested that the pectic substances are deposited in the middle lamella and the primary cell wall in the early stages of cell development, and that they remain, even in the old wood. During later growth other materials appear to be deposited on the pectic substance and so protect it from the action of pectin solvents. Although most of the water-insoluble pectic material appeared to be a calcium salt, some of it may be combined with cellulose or with lignin. S. G. S.

Bixin Solutions as Colorimetric Standards for the Determination of Carotene. H. N. Holmes and W. H. Bromund. (*J. Biol. Chem.*, 1935, **112**, 437–441.)—The use of an aqueous solution of potassium dichromate as a standard in the colorimetric determination of carotene solutions by the Willstätter–Stoll method was found to be unsuitable when the carotene was dissolved in chloroform or benzene instead of in petroleum spirit. This was due to the high refractive indices of these solvents, which caused the colour of the solutions to be shifted towards the red end of the spectrum. It was found that a solution of bixin in benzene could be used as a colour standard for both chloroform and benzene solutions of carotene. Graphs, included in the paper, show the relationship

between three concentrations of bixin in benzene and three concentrations of carotene in benzene and in chloroform, respectively. In every instance these graphs are practically straight lines. S. G. S.

Colour Reactions of Vitamins *A*, *D*, *E*, and some Sterols. S. Ueno and Z. Ueda. (*J. Soc. Chem. Ind. Japan*, 1935, **34**, 742–744B.)—The sources used for vitamin *A* were a sample prepared in the authors' laboratory (I), a commercial preparation (II), and a medicinal cod-liver oil; of these, the cod-liver oil gave less intense reactions than the preparations. The cholesterol used had m.p. 148 to 148·5° C., the ergosterol melted at 162·5 to 163·5° C., and the crude sitosterol at 134 to 135·5° C.; the sample of vitamin *D* was dissolved in vegetable oil and the vitamin *E* was prepared from rice oil.

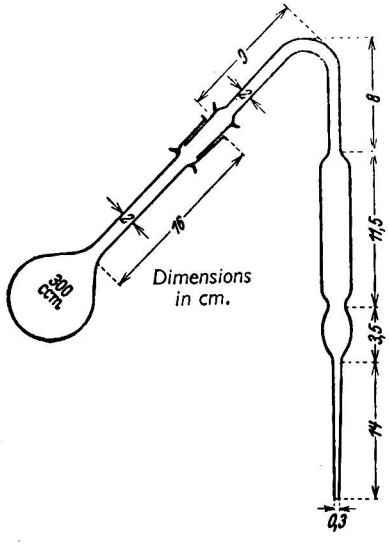
	Vitamin *A*		Cod-liver oil	Ergosterol	Vitamin *D*	Vitamin *E*
	I	II				
1. Conc. nitric acid	bluish-violet	bluish-violet	brown	pale yellow	——	brown
2. Phosphorus oxychloride	blue	blue	blue	pale yellow	pale yellow	pale yellow
3. Chlorosulphonic acid	bluish-violet, then brown	bluish-violet, then blue-reddish-brown	bluish-violet, then deep violet	pink, then deep greenish-blue	yellowish-brown, then brown	reddish-brown, then deep blue
4. Conc. hydrochloric acid with phenol	violet	violet	pink	violet	pink	brownish-yellow
5. Perchloric acid with phenol	violet	violet	pink	violet	pink	brownish-yellow
6. Cadmium chloride with phenol	bluish-violet	bluish-violet	bluish-violet	green	——	——
7. Cerium sulphate with phenol	green-blue	blue	blue	blue	——	——
8. Phosphomolybdic acid with phenol	bluish-violet	blue	indigo blue	blue	——	——
9. Phosphotungstic acid with phenol	blue	greenish-blue	blue	pale green	pale green	pale yellow
10. Acetyl chloride with phenol	violet	violet	reddish-purple	orange, then reddish-yellow	reddish-purple	reddish-yellow
11. Hydroxylamine hydrochloride with phenol	violet	violet	violet	violet	——	——
12. Phenylhydrazine hydrochloride with phenol	bluish-violet	bluish-violet	bluish-violet	bluish-violet	——	——
13. Sulphosalicylic acid with phenol	blue	blue, then grayish-green	blue, then green	reddish-purple, then greenish-blue	——	——
14. Thionyl chloride with phenol	greenish-violet	greenish-violet	greenish-violet	greenish-violet	reddish-purple	brownish-violet

Cholesterol and sitosterol each gave with chlorosulphonic acid a yellow colour changing to deep blue. As some samples were dissolved in olive and other oils,

blank tests were carried out with olive oil and a green colour was given in test 3, a slight blue in tests 8, 9 and 13, a brownish-violet colour with 14, but no colour with the other reagents. The colours are regarded as due either to a keto-enol form of re-arrangement of phenol or to the instantaneous dehydrating reaction in which the phenol accelerates the colour reaction of vitamin *A* with various reagents. Vitamins *D* and *E* show a close agreement in their reactions, and a natural connection between vitamin *D* and cholesterol in animal oils, and between vitamin *E* and sitosterol in vegetable oils is to be presumed, with a similarity in their respective biological rôles. The nitric acid reaction with vitamin *A* is regarded as an oxidising action, probably accompanied by dehydration. D. G. H.

Toxicological

Micro-determination of Arsenic in Must and Wine. J. Burkard and B. Wullhorst. (*Z. Unters. Lebensm.*, 1935, **70**, 308–315.)—The method depends upon the distillation of the arsenic as trichloride by the use of a modification of Engelson's apparatus (*Z. physiol. Chem.*, 1920, **111**, 201), and its subsequent colorimetric determination by Zinzadse's method (*Z. Pflanzenernährung, Bodenkunde u. Düngung*, 1930, **16**, 129).* Fifty (or 100 ml.) of the wine are evaporated in a Kjeldahl flask, heated 3 times with 3 ml. of hydrogen peroxide (perhydrol) and with concentrated nitric acid, and boiled with 20 ml. of concentrated (arsenic-free) sulphuric acid until colourless, and then for 10 minutes longer, after which the liquid is diluted with 30 ml. of water and boiled to remove all nitrous vapours. After cooling, it is treated with 1 g. of hydrazine sulphate for 30 minutes, shaken with 50 ml. of arsenic-free concentrated hydrochloric acid (sp.gr. 1·19), and distilled in the apparatus (shown in Fig. 1), the stopper of which has been moistened with a few drops of concentrated sulphuric acid. The end of the condensing tube dips to a depth of 1 cm. in 120 to 150 ml. of water contained in a 250-ml. Erlenmeyer flask, which is immersed to its neck in running water. The Kjeldahl flask is



* *Zinzadse's Reagent.*—This is made by heating 50 ml. of pure arsenic-free conc. sulphuric acid (sp.gr. 1·84) until white fumes appear; 3 g. of pure powdered MoO_3 are added to it, and the mixture is boiled for 5 to 10 minutes. When solution of the molybdenum trioxide is complete, the liquid is cooled completely and poured into 50 ml. of water (not *vice-versa*). While the solution is still hot, 0·15 g. of powdered metallic molybdenum is added as a reducing agent, and the mixture is boiled for 3 to 5 minutes. The reaction is finished and the reagent ready when 2·5 ml. of the reagent are required to decolorise 0·2 ml. of *N* permanganate solution. After standing for 10 to 20 minutes the blue liquid is decanted from the residue of metallic molybdenum. If the molybdenum-blue solution has been over-reduced, a solution of standard strength may be obtained by adding to it the calculated amount of the original solution (3 g. of MoO_3 + 50 ml. of conc. sulphuric acid + 50 ml. of water).

heated over a free flame so that its contents are vigorously boiling in 2½ to 3 minutes, and 10 minutes later the distillation is stopped. The distillate is treated with a few drops of 10 per cent. sulphuric acid, then with 20 ml. of concentrated nitric acid, and evaporated in a porcelain basin on the water-bath. The residue is heated until fumes of sulphuric acid appear, and, when cold, is taken up with a few ml. of hot water, and the solution is neutralised with 10 per cent. sodium hydroxide solution, which must be free from silica, β-dinitrophenol being used as indicator. For the colorimetric determination, 1·4 ml. of Zinzadse's molybdenum blue solution (*loc. cit.*) is added from a micro-burette, and the mixture is transferred to a graduated 100-ml. flask, cooled, and made up to 20 ml., and its colour-intensity is measured by means of a Zeiss photometer or a colorimeter; with the former the results agree within ± 5 per cent. of the theoretical values. Zinzadse's molybdenum blue reagent develops with arsenic acid a blue colour, the depth of which, within certain limits (0·01 to 0·8 mg.), is strictly proportional to the amount of arsenic pentoxide.*

The following typical results (in mg. per l.) were obtained with a series of new wines:—No. 15, 4·73, 4·87; No. 19, 6·19, 6·33, 6·04, 6·38; No. 41, 4·04, 3·66, 3·81, 3·84. A must contained:—(*a*) 3·91, 4·00; (*b*) 4·04, 4·16, 4·20; (*c*) 4·10, 4·30, 4·30 mg. per l. The method can also be used for making a series of rapid determinations of arsenic in foodstuffs and other substances.

Toxicity of Methanol. S. Kajizuka. (*J. Soc. Chem. Ind. Japan*, 1935, **38**, 746–747B.)—Albino rats fed on a diet containing 10 per cent. of methanol showed normal growth, except for a slight turbidity of urine and an increase in excreted formic acid (one died of indigestion). Two pigeons were fed daily with doses of 1 ml. of 50 per cent. methanol for about 80 days (an increase of 1·5 ml. caused death by indigestion within 16 to 17 days). No loss of eyesight was observed. Methanol thus appears to be somewhat toxic, giving a local stimulus to the mucous membrane, but not acutely affecting the nervous system. D. G. H.

Bacteriological

Apparatus for the Determination of the Fermenting Powers of Aerobic and Anaerobic Micro-Organisms. A. P. Struyk. (*Chem. Weekblad*, 1936, **33**, 44–45.)—The apparatus in its usual form consists of a test-tube 10 cm. long (capacity 10 or 15 ml.), at the top-end of which is a ground-glass joint, 2 cm. long, in which is inserted a tube (5 cm. long) open at both ends; the top-end has the same diameter as the test-tube, whilst the bottom is drawn out to a jet, the end of which is about 3 cm. from the base of the test-tube. The top tube is plugged with cotton-wool, the apparatus is sterilised, and the test-tube is filled with the medium, the top portion being replaced so as to exclude air-bells. The apparatus may then be sterilised (under pressure if required) and the organism added through the top tube. Advantages over the Einhorn and Durham tubes are that there is

* Zinzadse's molybdenum reagent of the correct composition is prepared by the firm of Schering–Kahlbaum, Berlin.

no loss of gas into the air, and that the growth of the organisms is not affected by the accumulation of gases. In addition, if the evolution of gas is slow, very little gas is lost, as it cannot readily escape through the jet, and it accumulates in the dead space under the joint. The presence of gas is easily seen, and reagents (*e.g.* sodium hydroxide solution for carbon dioxide) may be added through the top tube to absorb one constituent of a mixture. If the evolution of gas is very slow, the top tube is removed and a little liquid is taken out and replaced by air; then, by shaking the tube the bubbles of gas may be made to rise through the liquid to the air-space. The apparatus may be used for anaerobic or aerobic organisms. For the former, air should be removed from the liquid in the bottom tube by boiling in the presence of a little sterile sand and placing a sterile glass ball in the top tube so as to seal it at the constriction; for the latter, the top tube is used. Both tubes may be used simultaneously, and no vacuum desiccator is required. For use in water analysis or with a larger volume of liquid the test-tube is replaced by a bottle of 40 ml. capacity, and the jet is shorter; the sample is then added as described above, and is followed by sufficient sterile water to fill the bottle.

J. G.

Quantitative Studies on Yeast Suspensions by Turbidometric and other Methods. R. S. W. Thorne and L. R. Bishop. (*J. Inst. Brew.*, 1936, **42**, 15–26.)—Methods of estimating the "quantity of yeast" in a suspension are compared. *Determination of dry weight.*—The suspension (*e.g.* 20 ml.) is evaporated with 2 ml. of acetone, and the residue is weighed after 2 hours at 100° C. in a current of dry air. The acetone checks enzyme action and ensures that the yeast dries in a friable state and so loses moisture readily. If the suspension is in wort or beer, it is filtered on a Jena 1G4 sintered glass crucible, the residue being washed twice by sucking up about 5 ml. of water through the base of the crucible; the final residue is transferred to a weighed dish and dried and weighed. *Cell counts.*—The suspension is diluted so as to give about 5 cells per unit square (*i.e.* 0·00025 cubic mm.) in a Thoma-ruled haemocytometer, and cell-counts of 40 unit squares are made on each of 8 drops, the mean number of cells per ml. and the standard error of the mean being then calculated; boundary cases are included in one column of 20 squares and ignored in the other. "Semi" dark-ground illumination lessens the eye-strain. The above two methods are regarded as "standard," but they are tedious and lengthy and the following may be used for rapid and less accurate work:—*Centrifuge method.*—The suspension is centrifuged in a weighed tube for 10 minutes at 2500 r.p.m. in the presence of 25 per cent. of alcohol and 0·01 per cent. of haemoglobin which, by partly dehydrating it and removing protein, causes it to form a compact deposit. The liquid is poured off, the walls of the tube are wiped dry, and the tube and yeast are weighed; the standard error of duplicates (based on the mean) should be ±2 per cent. Measurement of the volume of yeast is unsatisfactory, owing to the presence of carbon dioxide and variations in the closeness of packing of the cells. The results obtained show that the centrifuged weight is proportional to the dry weight, with an error of ±4 per cent., but that the factor of proportionality varies for different yeasts, and even for the same yeast on different occasions, over a range

of 4 to 6·5. *Photo-electric method.*—(*Cf.* Awtonomowa and Stessel, *Biochem. Z.*, 1934, **274**, 220.) Light from a 6-volt, 6-watt lamp mounted in a parabolic mirror, passes through a sheet of glass rendered translucent by means of a suitable coating of cellulose lacquer, and then falls on a layer of the suspension (30 mm. thick) in a glass cell or flat-sided glass bottle. The intensity of the transmitted light is measured by means of a "Photronic" photo-electric cell which is used in conjunction with an ammeter having a range of 0 to 250 microamps., the current varying almost linearly with the intensity of illumination. The intensity of the source may be varied by means of a resistance, and its constancy is ensured by operating it from an accumulator which is being continuously recharged from the mains. The reading is taken first with water (A) and then with the suspension (B) in the cell, and it is convenient to adjust the apparatus so that A is 250; then $P = (A/B - 1)$ varies approximately directly as the concentration, independently of the intensity of the source. Departure from strict linearity in the presence of relatively large quantities of yeast is due to the complicating effect of scattered light as distinct from transmitted light. It is found that P is approximately proportional to the concentration of dry yeast, and that the relationship is more precise if a small correction for the size of the yeast cells is applied. For all yeasts, P is related more closely to the dry weight (D in g. per l.) than to the cell count (N per l.); the relation between D and the cell weight ($D/10^9N$) is linear for various values of P so long as the yeast cells are spherical. Statistical analysis of the results showed that a regression equation of the type $D = a + bP + cP^2 + dN$ fits the data with an error of about ±5 per cent., but there is no reason why the quantity P itself should not be taken as an estimate of the quantity of yeast. The colour and turbidity of the medium in which the yeast is suspended do not appreciably affect the results if the initial reading is determined for the medium alone, instead of for water; this reading may be made after removal of the yeast by filtration, but with very turbid worts a slight error sometimes arises thereby, owing to adsorption of some of the suspended matter by the yeast. The method is suitable for the estimation of yeasts during growth without sampling if special culture vessels with vertical sides are used. *Nephelometric method.*—The principle of the Zeiss–Pulfrich photometer with a nephelometer attachment is described. The drum reading (T) varies almost linearly with the dry weight for 0 to 0·15 g. per l. of yeast, and over this range $D = 0{\cdot}00027T$ with an error, for brewery yeasts, of ±5 per cent. With higher concentrations T increases rather less rapidly than the concentration. When the medium is coloured or turbid a correction is applied in the same way as already described, and for estimations of yeast-growth it is convenient to dilute 1 ml. of culture to 50 ml. J. G.

Agricultural

Comparative Quantities of Sulphur and Phosphorus in Plants Cultivated in the same Soil. G. Bertrand and L. Silberstein. (*Compt. rend.*, 1935, **201**, 1449–1453.)—A large number of agricultural plants (some of which are listed below) were grown on equal areas of a plot of ground considered to be of homogeneous composition and which had not been manured for several years.

The aerial parts of the annual plants were analysed just as flowering was beginning, and those of the biennials at an earlier stage.

	Dry matter Per Cent.	Sulphur in dry matter Per Cent.	Phosphorus in dry matter Per Cent.	Ratio S/P
Spinach ..	10·78	0·306	0·812	0·377
Rye	17·54	0·257	0·492	0·522
Pea	13·20	0·286	0·511	0·560
Wheat ..	27·72	0·260	0·410	0·633
Maize	19·25	0·149	0·234	0·636
Barley ..	22·09	0·284	0·362	0·785
Soja	26·18	0·230	0·258	0·892
Beetroot ..	10·49	0·534	0·489	1·092
Carrot	18·93	0·628	0·363	1·729
Turnip.. ..	10·76	1·494	0·848	1·762
Chicory ..	11·96	1·089	0·502	2·170
Radish ..	12·08	1·175	0·538	2·183
White mustard	12·84	1·314	0·545	2·410
Cabbage ..	11·25	1·919	0·477	4·021

The results emphasise the great variation in the plant requirements of sulphur, as contrasted with the much more constant proportion of phosphorus found. The physiological needs of the plant determine the probable intake of sulphur to a much greater degree than any variations in the sulphur-content of the soil.

D. G. H.

Boron Requirement and Boron-content of Cultivated Plants. M. P. Löhnis. (*Chem. Weekblad*, 1936, **33**, 59–61.)—The conclusion of previous workers (*e.g.* Brenchley and Warington, *Ann. Botany*, 1923, **37**; *id.*, 1927, **41**; Brandenburg, *Z. angew. Bot.*, 1932, **14**), that boron is necessary for the growth of plants, is confirmed, but it is concluded that plants show their sensitiveness to the absence of boron in different ways, and that varying quantities are involved, according to the nature of the plant. These conclusions are based on experiments in which 0·5 mg. of boron (in the form of borax, boric acid or boron citrate) per l. was added (with traces of manganese, copper, iodine and aluminium) to the nutrient solution in which the plant was grown (Crone's medium), salts of analytical quality being used. If boron was present, growth occurred whether tap-water or distilled water was used, but if it was absent, growth was obtained only in tap-water, and this is attributed to traces of boron in the water; addition of boron to distilled water produced the same effect as tap-water. The tourmaline in sand contains about 8 per cent. of boron in a finely-divided state, and, although this is usually considered to be insoluble, addition of tourmaline to the nutrient medium is as effective as boric acid in aiding growth. Comparison of the boron-content with the results of the growth-experiments on the plants suggests that the former is to some extent a measure of the boron requirement, and that lack of boron makes itself felt principally in the later stages of growth; there is also reason to believe that a relation exists between the boron and calcium contents. The method of determination used was that of Bertrand and Agulhon (*Compt. rend.*, 1913, **157**, 1433; abst., ANALYST, 1914, **39**, 96), which is satisfactory for 0·1 to

0·0005 mg. of boron. The results for the plants (in mg. of boron from 3 g. of dry material) may be classified in 3 groups as follows, the figures in brackets referring to the amounts found in the seeds:—Group I (0·1 to 0·05 mg.).—Lucerne (0·04), vetch (0·005), pea (0·007), tomato (0·01), white clover (0·04), sugar beet (0·01), and *Iberis umbellata*. Group II (0·05 to 0·005 mg.).—Pea, tobacco, red clover (0·09), wheat (0·001), oats (0·001), barley (0·002), and rye (0·001). Group III (below 0·005 mg.).—Oats and barley. Further data show that the boron-content may vary according to the year, the soil in which the plant is grown and the particular variety of the species under consideration. As a rule, the boron-contents of old and young portions of the same plant do not differ greatly, although in the former they tend to be less; an exception to both statements is the beet, the values for the old and young parts being 0·05 and 0·005, respectively. J. G.

Water

Influence of Chlorides on the Colorimetric Determination of Nitrates in Waters. R. Danet. (*J. Pharm. Chim.*, 1936, **23**, 34–36.)—In the official colorimetric method of the Laboratoire du Conseil Superieure d'Hygiene for the rapid determination of nitrates in waters (*cf.* Gros, *J. Pharm. Chim.*, 1935, **23**, 224–246; Abst., ANALYST, 1935, **60**, 774), the presence of chlorides lessens the intensity of the yellow colour formed. To the control test there should be added a quantity of sodium chloride (as 0·1 per cent. solution), equal to that in the sample tested. When the Gros standard-scale is used, the variable state of hydration of the phenol-sulphuric acid reagent is also a source of error. E. B. D.

Organic

Differential Reduction of the Nitro-Group by means of Glucose. G. Bacharach and R. Weinstein. (*Rec. Trav. Chim. Pay-Bas*, 1935, **54**, 931–933.)—In the presence of alkali, glucose solutions can be used to reduce the nitro-group (Wacker, *Ber.*, 1902, **35**, 62; Jansen, *Z. Farbenind.*, 1913, **12**, 181; cf. *Chem. Abs.*, 1934, **28**, 7254). The present authors show that it can be used to reduce *p*-nitrobenzoic acid to its azoxy-derivative or to its azo-derivative and that, provided the amount of reagent is sufficient to carry the reduction to the azo-stage, the temperature and concentration determine the nature of the end-product. To reduce *p*-nitrobenzoic acid to *p*-azoxybenzoic acid, the procedure is as follows:—Thirteen g. of *p*-nitrobenzoic acid are dissolved in 50 ml. of a solution containing 50 g. of sodium hydroxide, the solution is warmed slowly to 50° C., after which 150 ml. of 60 per cent. glucose solution (also at 50° C.) are added to it. After a rapid and violent reaction a yellow precipitate of the sodium salt of *p*-azoxybenzoic acid forms. The mixture is diluted and filtered, and the yellow precipitate is boiled with glacial acetic acid, filtered off, washed with cold water, then with 95 per cent. alcohol and dried. The yield is 95 per cent. of the theoretical yield. *p*-Azoxybenzoic acid is insoluble in cold organic solvents and in water; it is very sparingly soluble in hot alcohol and in hot glacial acetic acid. It decomposes without melting at 355° C., whilst *p*-aminobenzoic acid melts at 187° C., and is

soluble in the usual organic solvents. It does not yield the carbylamine reaction and cannot be diazotised. Determination of the nitrogen-content by a micro-Dumas method and titration of the carboxyl groups gave results agreeing substantially with the formula $C_{12}H_8ON_2(COOH)_2$. To reduce *p*-nitrobenzoic acid to *p*-azobenzoic acid, the procedure is as follows:—Thirteen g. of *p*-nitrobenzoic acid are dissolved in 200 ml. of a solution containing 40 g. of sodium hydroxide, the solution is warmed to 75° C., and 285 ml. of a 35 per cent. solution of glucose (also at 75° C.) are added. After the violent reaction has subsided the temperature is maintained at 75° C. until a red precipitate of the sodium salt of azobenzoic acid is formed. The reaction mixture is acidified with acetic acid, and the heavy precipitate is filtered off, dried, and purified by solution in sodium hydroxide solution and re-precipitation with acetic acid. On drying it assumes a red colour. *p*-Azobenzoic acid is insoluble in water and in the usual organic solvents, but is very sparingly soluble in hot alcohol and in hot glacial acetic acid. It decomposes above 300° C. without melting. It gives neither the carbylamine nor the diazo reaction. Determination of the nitrogen-content and titration of the carboxyl groups gave values in substantial agreement with the formula $C_{12}H_8N_2(COOH)_2$.

A. O. J.

The Presence of Butyric Acid in Commercial Acetic Acid. L. Kling. (*Ann. Chim. anal.*, 1936, **18**, 6–9.)—Acetic acids from different sources were tested for butyric acid, with the following results:

Sample	Butyric acid (mg. in 100 g. of sample)
Synthetic Acids	
Rhone–Poulenc; pure glacial	Nil
Lonza, Basle ; pure, 100 per cent.	Nil
,, ; head-fractions	Nil
Fermentation Acids	
Mihajlovic, Krestmac (Yugoslavia); 80 per cent. acid from maize	250
Kansky, Ljubljana (Yugoslavia); acid I	276
,, ,, ,, ; 80 per cent. acid from vinegar	280
Pyroligneous Acids	
E. Merck, Darmstadt; pure, 100 per cent.	Nil
Hiag, Liesing b. Wien (Austria); head-fractions, 24 per cent. ..	600
,, ,, ; tail-fractions, 88 per cent. ..	9,000
,, ,, ; crude acid, 82 per cent. ..	1,940
,, ,, ; pure, 98·2 per cent.	Nil
Lambiotte Frères, Prémery (Nièvre); glacial, 99·8 per cent. ..	660
,, ,, ; tail-fractions, 90 per cent.	6,300
Teslic, Osjek (Yugoslavia); pure, 100 per cent.	113
,, ,, ; head-fractions, 24 per cent. ..	380
,, ,, ; main-fraction, 99 per cent. ..	140
,, ,, ; tail-fractions, 91 per cent. ..	1,520
,, ,, ; commercial, 40–80 per cent. ..	600
Guttmann, Belisce (Yugoslavia); dark crude, 30 per cent. ..	9,700

The author's previously described method (*Biochem. Z.*, 1934, **273**, 1) was used for determining the butyric acid; this is based on oxidising the acid by means of hydrogen peroxide to acetone, which is distilled into the Scott–Wilson reagent

(alkaline mercury cyanide solution). Various aldehydes, which may also be formed in the oxidation process, give a similar precipitate with the Scott–Wilson reagent, but this may be distinguished from that given by acetone by distilling the used reagent with hydrogen peroxide; this destroys the aldehyde compound, but regenerates acetone, which may be absorbed in a fresh portion of the reagent and determined iodimetrically (*Bull. Soc. Chim. biol.*, 1932, **14**, 885). From the results obtained, given in the table, it was concluded that synthetic acetic acids are free from butyric acid. Fermentation acids contain butyric acid, but "pure" pyroligneous acids were free therefrom, with the exception of those of Yugoslavian origin and the first and last fractions of an Austrian product. S. G. C.

Fractional Distillation of Saturated Fatty Acids of Completely Hydrogenated Beef Tallow, Lard and Horse Fat. S. Ueno and T. Takeuchi. (*J. Soc. Chem. Ind. Japan*, 1935, **34**, 740–742B.)—The hydrogenated beef tallow (*a*), lard (*b*) and horse fat (*c*) were saponified, the mixed fatty acids were analysed and fractionally distilled (62, 60 and 39 fractions, respectively), and the m.p., neutralisation value and the iodine values of all the fractions were determined, and from these data the proportion of saturated fatty acids in the order of the carbon numbers were calculated. The composition was found to be comparatively simple, the acids consisting chiefly of C_{16} and C_{18} fatty acids with a very small quantity of C_{14} acid.

	a	*b*	*c*
M.p. °C.	58·7–61·2	58·8–60·6	61·2–62·2
Saponification value ..	196·7	195·0	196·4
Iodine value (Wijs) ..	0·57	0·24	0·31
Fatty acids			
M.p °C.	60·3–61·7	61·3–62·4	60·1–61·4
Neutralisation value ..	205·3	203·1	204·5
Iodine value (Wijs) ..	0·81	0·37	0·50
Titer (°C.)	59·1	60·4	58·9
Saturated fatty acids, per cent.			
C_{14}	2·0	—	1·4
C_{16}	30·3	23·7	34·8
C_{18}	64·5	73·3	60·7

D. G. H.

The Highly Unsaturated Acid of *Telfairia occidentalis*. E. H. Farmer and E. S. Paice. (*J. Chem. Soc.*, 1935, 1630–1632.)—*Telfairia occidentalis* oil closely resembles tung oil in appearance, and, on keeping, deposits a white precipitate (shown to be β-elaeostearin). An oil expressed some 7 months previously was saponified, and about 10 per cent. of a highly unsaturated acid of m.p. 70° C. was separated by crystallisation from petroleum spirit. The acid was found to possess the constitution of elaeostearic acid and was identical with authentic β-elaeostearic acid obtained by irradiation and subsequent saponification of tung oil. In view of the great improbability of this acid being originally present in the β-form, the oil was extracted from fresh samples of *Telfairia* seeds; no β-elaeostearic acid was found in this oil, but, after exposure to dim diffused light for some weeks, a small yield of that acid was obtained, and after exposure to ultra-violet radiation

for some hours, a larger yield. Fractional crystallisation of the acids separated after saponification of the fresh oil yielded pure α-elaeostearic acid, m.p. 48° C. Telfairic acid appears to be quite distinct from the highly unsaturated acid in the oil from *T. occidentalis*, but, since the glyceride of α-elaeostearic acid is a component of *T. occidentalis* oil, as it is of *Parinarium macrophyllum* (Brown and Farmer, *J. Chem. Soc.*, 1935, 761) and of China wood oils, the glyceride can constitute, though not exclusively so, the triene component of the kernel oils of the *Euphorbiaceae, Rosaceae,* and *Cucurbitaceae* groups. D. G. H.

Determination of Asphalt in Oils. P. Woog, J. Givaudon, F. Dayan and A. Bidet. (*Bull. Soc. Chim.*, 1936, **3**, 97–102.)—The asphalt-content of oils and petroleum derivatives increases through oxidation in air, the rate of increase being influenced by light and temperature. The usual method of determination should be standardised as follows:—The oil sample is kept in the dark and (unless the determination is made immediately after sampling) *in vacuo* or in an inert atmosphere. The temperature of the sample, and of the petroleum spirit used for precipitating the asphalt, is kept constant, preferably at 0° C., for 1 hour before precipitation, and the precipitate remains at this temperature during a fixed time of standing (16 to 24 hours), and during subsequent filtration. E. B .D.

Inorganic

Critical Studies of the Analytical Application of Organic Reagents. J. B. Ficklen, I. L. Newell and N. R. Pike. (*Z. anal. Chem.*, 1936, **104**, 30–34.)—A series of investigations into the sensitiveness and specificity of organic compounds proposed for analytical purposes. The first paper (*Ind. Eng. Chem., Anal. Ed.*, 1935, **7**, 26) dealt with kakothelin as a reagent for tin. The present paper criticises cinchonine iodide as a reagent for bismuth (Léger, *Bull. Soc. Chim.*, 1888, **50**, 91). The conclusion is reached that the reagent is very sensitive (limit, 1 : 1,000,000), but not specific; hence the bismuth should be previously isolated by standard separation methods. W. R. S.

New Organic Reagent for Cadmium. A. W. Scott and E. G. Adams. (*J. Amer. Chem. Soc.*, 1935, **57**, 2541–2542.)—1-(2-Quinolyl)-4-allylthiosemicarbazide, added as a saturated solution in 50 per cent. ethyl alcohol (0·05 g. dissolves in 100 ml.) to a cadmium solution containing potassium iodide, gives a bright greenish-yellow precipitate. In a test in which 1 ml. of 50 per cent. alcohol saturated with the reagent and with potassium iodide was added to 10 ml. of cadmium nitrate solution, it was found that 1 part of cadmium in 200,000 parts of solution gave a filterable precipitate, whilst 1 part per million gave an opalescence forming into a precipitate in 2 minutes. Cadmium may be detected in presence of copper by first adding potassium iodide; without filtering off the cuprous iodide, the reagent solution is added, giving the yellow precipitate; on subsequently adding ammonia, the copper precipitate dissolves to a blue solution, leaving the yellow precipitate undissolved. Ammonia must not be present prior to adding the reagent, and the following ions interfere: sulphate, zinc, nickel and cobalt. The

reagent was prepared "from 10 ml. of allyl isothiocyanate and 16 g. of crystalline 2-quinolylhydrazine in ether; yield 20 g." Slow recrystallisation from ether gave colourless crystals, m.p. 158° C.; the material is fairly soluble in ether, alcohol and benzene, and slightly soluble in cold water. S. G. C.

Determination of Tin in Minerals and Alloys by Means of Potassium Bromate. L. Deutsch. (*Ann. Chim. anal.*, 1936, **18**, 10.)—*Minerals.*—A mixture of the finely-ground sample with 2 to 3 times its weight of sodium peroxide is placed in an iron crucible, covered with a layer of sodium peroxide, and fused. The mass is extracted with water, and the solution is boiled, acidified with hydrochloric acid, cooled and diluted to 500 ml. About 5 g. of reduced iron powder are added, and the whole is shaken occasionally for half-an-hour. An aliquot part of the solution (100 ml.) is filtered off into a 500-ml. conical flask, and 1 g. of aluminium foil and 30 ml. of hydrochloric acid are added. The flask is closed with the valve attachment (Fig. 1), containing saturated sodium bicarbonate solution. A further addition of 60 ml. of hydrochloric acid is made when the tin has precipitated; the solution is heated to dissolve the metal and then cooled, more sodium bicarbonate solution being poured into the valve, if necessary. The stopper is finally removed, a piece of marble is added, and the solution is titrated to a blue colour with $N/10$ potassium bromate solution (1 ml. ≡ 0·00593 g. of tin), 15 ml. of starch and zinc iodide indicator solution being used (4 g. of starch dissolved in 100 ml. of water, with 20 g. of zinc chloride and 2 g. of zinc iodide added).

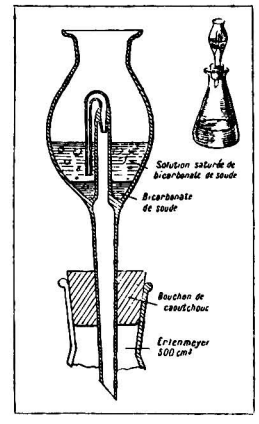


Fig. 1

Alloys (bearing metal, solder, etc.).—Two g. are dissolved by heating in 20 ml. of sulphuric acid containing 5 g. of sodium sulphate. After cooling, 100 ml. of water and 30 ml. of hydrochloric acid are added (antimony may now be determined, if necessary, by titrating the hot liquid with $N/10$ bromate solution, with methyl orange as indicator). The solution is diluted to 500 ml., reduced iron is added, and the process is continued as for tin in minerals.

[*Abstractor's Note.*—It would be advisable in this process to standardise the bromate solution against pure tin treated in a manner similar to the sample.]

S. G. C.

Titration of Thallous Salts with Potassium Iodate and other Reagents. E. H. Swift and C. S. Garner. (*J. Amer. Chem. Soc.*, 1936, **58**, 113–115.)—The titration of thallous salts with iodate, permanganate, and ceric sulphate solution has been studied. Thallous salts can be accurately titrated with iodate solutions as follows:—To the solution contained in a glass-stoppered bottle is added sufficient hydrochloric acid to render it from 3 to 5 N, together with 4 ml. of carbon tetrachloride and 5 ml. of iodine monochloride solution (prepared as described by Jamieson, "Volumetric Iodate Methods"). The solution is titrated with 0·1 N potassium iodate solution (standardised against arsenious acid by the same type of titration), with frequent shaking, until the disappearance of the iodine colour in

the carbon tetrachloride layer; the bottle should be kept cold by immersing it in water, and it is recommended that a small amount of water should be poured around the stopper before it is withdrawn, to avoid possible loss of iodine vapour. The result is calculated on the basis of the reaction having thallic salt and iodine monochloride as end-products. Quantitative results were obtained in test-experiments with 0·078 to 0·42 g. of thallium per 100 ml. of solution, and hydrochloric acid concentration from 1 to 5 *N*; below 3 *N* hydrochloric acid the end-point change was slow. Ceric sulphate solutions cannot be used for the titration with the iodine monochloride end-point; positive errors were found and the end-point change was slow. The titration of thallous salt in hydrochloric acid solution with permanganate, employing the iodine monochloride end-point is inaccurate owing to the catalysed oxidation of chloride by permanganate. Various modifications of Willm's method of titration were studied without conditions being found under which accurate titrations could be made. S. G. C.

Volumetric Determination of Molybdenum by means of Vanadate Solution. R. Lang and S. Gottlieb. (*Z. anal. Chem.*, 1936, **104**, 1–16.)—A solution of molybdate is reduced by stannous chloride at a definite acidity from Mo^{VI} to Mo^{V}. The excess of stannous salt is destroyed by bromine, and the excess of bromine by arsenite. The reduced molybdenum compound is then re-oxidised by vanadate, diphenylamine being used as indicator.

Reagents.—(1) Stannous chloride (25 g. crystals) dissolved in 170 ml. of strong hydrochloric acid and made up to 1 litre. (2) Bromine (16 g.) and potassium bromide (120 g.) made up to 1 litre. (2) Arsenic trioxide (15 g.) and sodium carbonate (10 g.) made up to 1 litre. (4) Ferrous sulphate (28 g. crystals) and 5 to 10 ml. of sulphuric acid, to 1 litre. (5) 0·1 *N* ammonium metavanadate solution: the salt (12 to 14 g.) is well rubbed in a mortar with 80 ml. of sulphuric acid (1 : 1), and the solution is made up to 1 litre. It is standardised against the ferrous sulphate solution. (6) Diphenylamine-*p*-sulphonic acid (1 per cent. solution), or 1 g. of the base in 100 ml. of syrupy phosphoric acid. (7) Primary standard: potassium dichromate, or pure molybdenum trioxide.

Procedure.—(*a*) *Indirect:* The molybdate solution (100 ml.), neutralised to methyl orange, is treated with 15 to 20 ml. of strong hydrochloric acid, and gradually with stannous chloride during agitation until a blackish-brown colour is no longer formed at the point of incidence. A small excess is then added, followed by about as much bromine solution as the stannous chloride used (the minimum being 5 ml.), then by an equal volume of arsenite reagent. After addition of 2 g. of sodium fluoride or ammonium bifluoride and 3 drops of indicator, the solution is titrated with vanadate until the brown colour becomes deep blue to violet, green and pale blue being intermediate tints. The excess of vanadate is finally titrated with ferrous sulphate to a sharp end-point (green solution).

The addition of fluoride causes smooth re-oxidation of the ferrous sulphate, without interference from molybdenum blue. The indicator correction is avoided by treatment of the 3 drops with 2 ml. of hydrochloric acid (1 : 1), vanadate solution (1 ml.), very little fluoride, and titration with ferrous sulphate to the colour change. The prepared indicator is added to the assay.

(*b*) *Direct.*—The procedure given under (*a*) is followed until the arsenite has been added. The solution is then titrated, without indicator, with vanadate, until almost pure blue. The prepared indicator is added, and when the solution has turned yellow after a short time, it is titrated, drop by drop, to the colour-change to reddish-violet, no fluoride being added. The indicator is prepared as follows:—Six drops of the diphenylaminesulphonic acid solution are treated with 2 ml. of 5 *N* sulphuric acid, 2 to 3 of 0·1 *N* dichromate solution, and 3 ml. of arsenite reagent. The dichromate is reduced after a few minutes, the liquid turning reddish-violet. Without this preliminary treatment, the indicator acts slowly with vanadate.

In presence of tungstate, the solution is treated with sodium fluoride (2 g.) and strong hydrochloric acid (20 ml.), and diluted to 120 ml. When clear, 15 to 20 ml. of stannous chloride reagent are added all at once, and the flask is allowed to stand for 25 minutes. The solution is treated with 2 to 3 drops of 0·5 *M* copper sulphate solution and 20 to 25 ml. of bromine reagent. After short agitation the bromine is removed with arsenite solution. Sodium fluoride (2 g.), strong hydrochloric acid (10 ml.) and 3 drops of indicator are added. The solution is then titrated with vanadate and ferrous sulphate. Tungsten blue is oxidised by bromine in presence of copper as a catalyst; the sodium fluoride prevents the precipitation of tungsten blue in a rather insoluble form. W. R. S.

Gravimetric Determination of Cerium with Ferrocyanide. P. Spacu. (*Z. anal. Chem.*, 1936, **104**, 28–30.)—The cold neutral solution (about 40 ml.) is treated, drop by drop, with excess of 0·1 *M* potassium ferrocyanide solution during continuous agitation. The crystalline precipitate is $Fe(CN)_6CeK.4H_2O$, containing 30·254 per cent. of cerium. After half-an-hour's standing the precipitate is collected in a porous porcelain crucible, washed with 50 per cent., then 96 per cent. alcohol, and finally with ether, dried *in vacuo* and weighed. The method is "applicable also in presence of erbium, but not of lanthanum and thorium," which also are precipitated.

[*Abstractor's Note.*—This brief sentence could be construed as meaning that a separation of cerium from erbium is practicable. Rare-earth metals other than the above are not mentioned.] W. R. S.

Determination of Chlorate and Perchlorate in Nitrates. E. S. Tomula. (*Z. anal. Chem.*, 1935, **103**, 427–430.)—*Perchlorate.*—The nitrate (10 g.) is dissolved in a little water in a glass basin. The solution is treated with formalin (3 to 5 ml.), 0·5 *N* ferric chloride solution (1 ml.), and 2 *N* nitric acid (2 ml.), heated for half-an-hour on a steam-bath for the reduction of the chlorate, and evaporated to dryness with 20 ml. of strong hydrochloric acid. The residue is dissolved in a little water, and the evaporation with hydrochloric acid is repeated. Two more evaporations are carried out—four in all. The resultant sodium chloride is precipitated with 50 ml. of strong hydrochloric acid, filtered off by suction, and washed several times with strong acid. The filtrate is evaporated to dryness, leaving a residue of about 0·5 g. containing the undecomposed perchlorate. It is dissolved in water, and the solution is made up to 100 ml. An aliquot part is titrated for chloride by Volhard's method, and the chloride is then precipitated in the bulk of the

solution by means of the calculated quantity of silver sulphate. The silver chloride is filtered off, and the filtrate is concentrated to 30 or 40 ml. and transferred to a flask, the short neck of which carries a ground-glass reflux condenser. Reduction of the perchlorate is brought about by addition of zinc (6 g.), iron powder (0·4 to 0·6 g.), and 3 per cent. cadmium sulphate solution (3 ml.). After 15 to 20 minutes 15 ml. of a solution of 10 g. of sodium titanate in 100 ml. of 1 : 4 sulphuric acid are added, and the condenser is inserted. The hydrogen evolution, which should be vigorous, is maintained by means of a small flame. The solution is allowed to boil gently for an hour. When cold, the solution is decanted from the zinc into a conical flask and oxidised with 10 ml. of 7 per cent. manganous sulphate solution, and *N* permanganate solution. The chloride-content of the slightly cloudy liquid is determined according to Volhard.

Chlorate.—In another 10 g.-portion the chlorate is reduced with zinc (0·5 to 1 g.), 2 *N* sulphuric acid (6 to 10 ml.), and 1 to 2 drops of copper sulphate solution. The reduced solution is titrated by Volhard's method, the operation giving the sum of the reduced chlorate and original chloride. A third portion is titrated for original chloride, and this amount is subtracted from the preceding. W. R. S.

Interference of Fluoride in the Precipitation of Phosphoric Acid by Molybdate. H. T. Bucherer and F. W. Meier. (*Z. anal. Chem.*, 1936, **104**, 23–28.)—Fluoride interferes in the molybdate method in that it retards the precipitation of the phosphomolybdate. In presence of substantial amounts of fluoride, the precipitation is incomplete: 5 g. of ammonium fluoride prevented precipitation altogether. The authors urge the necessity for a fluorine test previous to the determination of phosphoric acid, and evaporation with sulphuric acid if the test is positive. W. R. S.

Simultaneous Argentometric Titration of Cyanide and Halide. R. Ripan-Tilici. (*Z. anal. Chem.*, 1936, **104**, 16–22.)—The titration is carried out in two stages: (i) The cyanide is titrated according to Liebig until a slight cloudiness indicates complete conversion into argentocyanide; (ii) after addition of 2 drops of 2 per cent. alcoholic fluorescein solution, the halide and argentocyanide are titrated with the same silver solution until the red end-point is reached (Fajans, ANALYST, 1923, **48**, 401). The standard silver solution should be 0·1 to 0·05 *M*, the solution to be titrated, 0·05 to 0·01 *M*. During the first stage, vigorous stirring expedites solution of the silver cyanide; during the second stage, the flask should be rotated gently, so that clotting of the precipitate is minimised. When the indicator has been added, the flask should be protected against direct daylight, by black paper, as the precipitate is sensitised by the indicator, and its darkening obscures the end-point. Test determinations were made with cyanide solutions containing either chloride, or iodide, bromide, selenocyanate, thiocyanate, or cyanate. The last substance does not give as sharp an end-point as the 5 others, and a stronger silver solution (0·8*M*) is recommended for its determination. W. R. S.

Reversible Indicator for the Detection of Small Quantities of Hydrogen Sulphide in the Atmosphere. J. Bell and W. K. Hall. (*Chem. and Ind.*, 1936, **55**, 89–92.)—The minimum quantity of hydrogen sulphide detectable by its

odour is 0·13 p.p.m.; 50 to 100 p.p.m. affect the eyes after 1 hour; 500 to 700 p.p.m. are dangerous after 30 minutes, and 1000 to 2000 p.p.m. cause death in a few minutes (cf. *U.S. Bur. of Mines*, Paper No. 551, 1933). The standard iodine and lead acetate methods of determination are unsuitable for obtaining continuous results, and it is preferable to pass the gas through a dilute alkaline solution of sodium nitroprusside and to note the intensity of the red colour. The reagents contains sodium nitroprusside 0·05, sodium carbonate 0·37, and sodium bicarbonate 0·19 per cent., and it should be stored in a bottle wrapped in dark paper. For a given concentration of gas, a variation in carbonate-content has little effect, but an excess of bicarbonate reduces the intensity of colour. If air containing hydrogen sulphide is passed through 100 ml. of reagent at the rate of 100 l. per hour, the colours produced by 50, 100 and 500 p.p.m. of the latter gas are pink, mauve and deep violet, respectively. By altering the rate of flow to 1 l. and 5 l. per hour, and the corresponding volumes of reagent to 20 and 50 ml., the ranges 1 to 10 p.p.m. and 0·5 to 5·0 per cent., respectively, may be covered by a similar series of colour-changes. It is shown that the hydrogen sulphide probably reacts with the alkali to form sodium sulphide, and that this produces a red addition-product with the nitroprusside, which is subsequently oxidised by air to nitroprusside and free sulphur; this last reaction explains the effect of passing pure air through a solution containing the red compound, the colour being destroyed without the liberation of hydrogen sulphide. Since the gas is completely absorbed from the air by the first reaction, and since the partial pressure of the oxygen is constant, it can be shown that the intensity of the colour is directly proportional to the amount of gas passing in unit time. If the air is replaced by nitrogen, the colour increases progressively in intensity so long as hydrogen sulphide is present in the mixture; a brown colour indicates eventually that the reagent is exhausted. Apparatus for carrying out the test is described. In the simplest form an air-ejector draws samples through aluminium pipes (diam. 0·25 inch) into a train of absorbing bottles. Two types of apparatus for continuous measurements are also described (for 50 to 500 p.p.m. and for 2 to 10 p.p.m. of hydrogen sulphide, respectively). A special form of air-lift, operated by an ejector, ensures that the stream of gas to be analysed circulates through the solution, and also that no liquid which may absorb hydrogen sulphide from the inlet gases is trapped in the branch-tube. A filter consisting of two Soxhlet filter-papers, to remove any precipitated sulphur, is placed in series with the reservoir in which the colour of the reagent is observed. J. G.

Microchemical

Colorimetric Determination of Caffeine. G. Denigès. (*Mikrochem.*, 1936, **18**, 22–24.)—Weildal's reaction (*Bull. Trav. Pharm. Bordeaux*, 1934, **4**, 345) for xanthides is applied to the determination of 2 to 0·1 mg. of caffeine. The chloroform extract of caffeine is carefully evaporated to dryness in a porcelain crucible, and 6 drops of bromine water, saturated in the cold, and 1 drop of diluted hydrochloric acid (1 : 9) are added (*N*-hydrochloric acid is suitable). The crucible is rotated in the Bunsen flame while the contents evaporate and finally turn orange-red, when 10 ml. of water and 1 drop of 5 per cent. mercuric acetate in 2 per cent.

acetic acid are added, and after shaking, the coloured liquid is transferred to a 12 to 15 mm. comparison tube, and the colour is compared with that produced by a series of standard solutions of caffeine treated similarly. The colour is very stable. If the mercuric acetate is replaced by two drops of 2 per cent. zinc acetate solution, 1 drop of acetic acid, the resulting colour is yellow and is suitable for comparison in a colorimeter. J. W. M.

Spot-test for Caesium and its Application to Colorimetry. E. S. Burkser and M. L. Kutschment. (*Mikrochem.*, 1935, **18**, 18–21.)—The reagent is a mixture of gold and platinum bromides in the ratio of 2 mol. to 1 mol., and is used in concentrations containing 1 to 10 per cent. of gold. This forms a deep black precipitate with caesium salts, of the formula $Cs_2Au_2PtBr_{12}$. The more concentrated reagent is used when testing for very small amounts of caesium in the absence of rubidium; when rubidium is present the best results are obtained by the use of a solution containing 3 per cent. of gold and 1·5 per cent. of platinum. To detect caesium, a drop of the reagent is placed on paper and followed by a drop of the test chloride solution; when caesium is present, a black fleck (grey for very small amounts) appears; 0·25γ of caesium may be detected in a drop of 1 c.mm. Rubidium, potassium, sodium, lithium and ammonium chlorides do not interfere, with the exception of rubidium in high concentrations. Concentrations of rubidium exceeding 2 per cent. give a reaction analogous to caesium. The caesium in a 1 per cent. solution of a mixture of rubidium and caesium chlorides may be detected when the mixture consists of 3 per cent. or more of caesium, even in the presence of a mixture of the chlorides of potassium, ammonium, sodium and lithium. If a number of drops of reagent of equal size are placed on filter-paper, and on each of these, drops of solutions of caesium chloride of 2—1—0·9—0·8, etc., to 0·025 per cent. caesium ion content, a graduated scale of flecks ranging from black to grey is obtained. Such a scale is dried and will keep without changing. The test is carried out on paper in the same way, and the intensity of colour compared with the scale. The accuracy obtainable is about 5 to 10 per cent. J. W. M.

Reactions Common to Germanic Acid and Boric Acid. N. S. Poluektoff. (*Mikrochem.*, 1935, **18**, 48–50.)—Germanic acid, like boric acid, forms complex compounds with polyhydroxy alcohols, such as mannitol, glycerol and glucose. Hahn's test for boric acid (*Z. anal. Chem.*, 1934, **98**, 283) depends on the increase in acidity which occurs on adding mannitol or glycerol to boric acid, and this has been applied as a spot-test for the detection of germanic acid, as follows:— A drop of a slightly acid solution of sodium germanate is mixed with a drop of phenolphthalein solution and 0·01 *N* sodium hydroxide solution added until the indicator appears red. A little mannitol is then added and the red colour disappears or becomes much weaker. In this way 2·5γ germanium can be detected in 0·05 ml. (concentration limit 1 : 20,000). The hydroxyanthraquinone test for boric acid (Feigl, *Mikrochem., Pregl-Festscrift*, 1929, 77) is also applicable both to salts of germanic acid and to germanium dioxide. A drop of the acid or alkaline test solution (which must be free from chlorides and bromides, otherwise loss of germanium by vapourisation may occur) is evaporated to dryness in a porcelain

dish and then treated with 2 to 3 drops of a 0·01 per cent. solution of quinalizarin in concentrated sulphuric acid, and gently heated. In the presence of germanium the red-violet colour changes to blue. The *limit of identification* is 5γ of germanium, and the *concentration limit* 1 : 10,000. Fluorides may be removed beforehand by evaporation with concentrated sulphuric acid. The test for boric acid with the dyestuff *p*-nitrobenzene-azo-chromoteric acid (Komarovsky and Poluektoff, *Mikrochem.*, 1933–34, **14**, 317) is not very suitable as a test for germanium, as the colour change is not sharp, and the sensitivity still less than with quinalizarin.

J. W. M.

Physical Methods, Apparatus, etc.

Improvement of the Maquenne Block. R. P. Jacquemain. (*Bull. Soc. Chim.*, 1936, **3**, 142–143.)—In Maquenne's method for the determination of melting-points of organic compounds, the substance is heated on a brass parallelopiped. To prevent oxidation of the brass and consequent unsatisfactory results, the cleaned block should be chromium-plated (without preliminary nickel-plating), and subsequently polished. On the polished surface, the m.p. is observed instantaneously, and the method is considered more practical and more rapid than the capillary-tube method. The chromium plating costs little and can be repeated when necessary. E. B. D.

A Cycle of Ultra-violet Light Sources for Various Uses. L. Bendikson. (*Library J.*, Jan. 1st, 1936.)—A miniature form (called a "palimscope") of the ultra-violet lamp previously described (ANALYST, 1935, **60**, 61) has been constructed. It consists of a small compact spiral of quartz tubing having the dimensions of a reading glass and provided with a handle, so that small areas of documents may be inspected closely under a concentrated ultra-violet radiation. By means of a further modification in design the lamp has been made to serve as a ring-illuminator, and an apparatus is illustrated by means of which it is possible by the turn of a switch to take successive photographs of the same field in ordinary light (by vertical illumination) and in ultra-violet light. This was achieved by omitting the inside loops of the spiral and inserting a narrow cylinder, having the same diameter as the tube of the microscope, in the centre of the metal housing; the apparatus may also be used as a spot-light for photographic work, and 4 of them enable 8 × 10 inch plates to be used, this arrangement being advantageous when lenses of large diameters are preferred. Results obtained with a Latin breviary are described; every one of the 200-pages was a palimpsest (*i.e.* the leaves had previously been parts of other works, but the original writing had been erased with fine pumice). Actually, 8 entirely different manuscripts were recognisable under the lamp. J. G.

Use of Filtered Ultra-violet Radiation in the Examination of Hashish in the Pure State or Mixed with Various Drugs. J. Khouri. (*Ann. Falsif.*, 1935, **28**, 582–584.)—A whole fragment of fresh authentically-pure hashish had a brown fluorescence changing to a mahogany colour on exposure to filtered ultra-violet light, whilst that of powdered hashish was unchanged. An extract of the

powder in the common organic solvents, but preferably in cold petroleum spirit, has a clear green fluorescence and enables very small quantities of hashish to be detected in the presence of other drugs. The method is more sensitive than Beam's reaction, but the fluorescence of the solution disappears on prolonged exposure in diffused light, especially if the containing vessel is not completely filled. After treatment with animal charcoal the fluorescence is absent and Beam's test is negative, but both methods of detection are unaffected by a temperature of 100° C. for 30 minutes. The above results are in accordance with the known properties of cannabinol, the only specifically-active principle of hashish hitherto isolated; it distils above 100° C., but oxidises in air. Of the other substances usually associated with hashish, cinnamon, treacle, honey, black pepper, chocolate, mastic resin, cocoa, cantharides and coffee are non-fluorescent under the conditions described above; nutmeg and liquorice are yellow; tobacco powder, salmon pink; cloves, pale blue-green; ginger, pale violet; and ambergris, bright green. Other interfering substances may usually be eliminated by selective extraction with organic solvents. J. G.

Reviews

HOLLEMAN, LEHRBUCH DER ORGANISCHEN CHEMIE. Twentieth Edition. Enlarged and revised by F. RICHTER. Pp. xii + 546. Berlin and Leipzig: Verlag Walter de Gruyter & Co. 1935. Price, bound RM.14.

During the five years which have elapsed since the publication of the last edition of this widely used textbook, opportunity has been taken to incorporate the results of much recent work in a number of important branches of the subject.

The chapter on carbohydrates is particularly good, and embodies many new results due to English investigators in this extensive and complicated branch of organic chemistry.

Concise but clearly written sections give the main outlines of the following topics: free radicals, sterols, haemin, chlorophyll and anthocyanins.

A considerable amount of space is devoted to the more modern physical and physico-chemical investigations, and the help they have afforded in the elucidation of many difficult problems in which organic chemistry abounds. A notable omission is a section to deal with the parachor, consequent, perhaps, upon a still more remarkable omission, that of any discussion of the electronic theory of valency.

In contrast, the brief account of the Walden Inversion does not appear to have been sufficiently modernised; many chemists are beginning to appreciate that the studies of replacement reactions of optically active compounds, which are commonly collected under the heading of the Walden Inversion, are really of far-reaching importance because of the light they are throwing on the broad subject of substitution in saturated compounds. J. KENYON

TABELLEN ZUR QUALITATIVEN-CHEMISCHEN ANALYSE. By S. OEHLINGER. Pp. 34 + vii. Prague: Published by the Author. 1934. Price 100 Kronne.

Much information is compressed into this small and clearly printed book. The subject matter dealt with in the 36 pages of tables, each approximately 8 inches by 6 inches, is as follows: the elements and their properties, acids of importance in analysis, oxidation and reduction processes, concentrations of reagents and solutions, dry reactions, tests for anions, preparation of solution for analysis, coloured compounds, reactions of cations, group procedures for common cations, systematic identification of anions, solubilities, micro-reactions of cations and anions, reactions of rarer elements, reactions of organic substances, organic reagents for anions and cations, wave-lengths of emission spectra.

A list of errata is given, but more errors have passed undetected than have been noted. Some of the tables do not appear to be particularly germane to the practice of analysis. Theoretical principles and detailed descriptions are not included, and the author points out that the tables should be used in conjunction with a book containing this information.

The book is intended primarily for students, but, apart from the drawbacks just mentioned, it costs approximately eighteen shillings. A. M. WARD

FLUORESCENCE ANALYSIS IN ULTRA-VIOLET LIGHT. J. A. RADLEY and JULIUS GRANT. Second Edition. Pp. 326 + 23 luminograms. London: Chapman & Hall. 1935. Price 21s. net.

When a second edition of a book appears only two years after the first it may justifiably be enquired whether both were necessary. In the present instance it may be said at once that the answer is in the affirmative. The strides made in Fluorescence Analysis and the number of papers published have been sufficiently great to warrant a second survey and collation.

The new edition is constructed on similar lines to the former. The first and smaller part deals with Theory and Technique, and, although not exhaustive, is adequate. The second part, dealing with the Application of Fluorescence Analysis, is conveniently divided into nineteen parts, each dealing with a particular branch. Some overlapping unavoidably occurs, but cross-references are numerous. As before, the principle has been to collect the references dealing with a particular branch, to sub-divide these for discussion in the chapters, and to record all the references at the end. The added practical experience of the authors has enabled them to delete some work of a doubtful character previously recorded; but the field of fluorescence is so wide, and so many exaggerated claims have been made for it, that quite a number of references still cannot be taken, in a general sense, at their face value. It is one thing to make deductions from slight differences in fluorescence of substances of known origin and purity, but quite another to attempt to deduce anything when the history of a sample is unknown. The authors rightly insist at several places on the need for caution in interpretation, and emphasise the point that in most cases results must be taken in conjunction with other physical and chemical tests.

As far as can be judged, the available literature has been well surveyed, as is evident from the fact that there are over 1500 references. It is surprising, however,

to find no reference to the part played by fluorescence in determining the constitution of the anthocyanins, the pigments of flowers. Nor is mention made of the golden yellow fluorescence of aloes with borax solution, which is diagnostic for this drug, even in the presence of other emodin-bearing material. In the section on the testing for stains on garments there is no indication that pus and, to a less extent, perspiration show distinct fluorescences, indistinguishable from those given by traces of semen.

A few loose statements of significance have been noted. "Novococaine" (p. 99) refers to novocaine. "Vitamin *C* or substances similar to it in effect" (p. 119) are stated to be the seat of the luminescence of olive oil. The substance described as "artificial cream" (p. 133) is not the article legally defined as such in this country, but refers to synthetic cream made from fat other than milk-fat. "A false ripening, such as that of Camembert cheese . . ." (p. 134) might lead one to suppose that this cheese is not normally mould-ripened.

These are, however, relatively small defects in a book which covers such an immense amount of ground.

This volume is not a textbook in the sense of being a manual of instruction; it is a *vade mecum* to which workers may go for ideas and to explore possibilities, for it is quite certain that fluorescence analysis cannot be taken on trust, but must be applied by each worker for himself.

The authors are to be congratulated on bringing together and surveying the results of published work in a form convenient for the many workers in such a variety of diverse fields. J. R. NICHOLLS

FUNDAMENTALS OF BIOCHEMISTRY IN RELATION TO HUMAN PHYSIOLOGY. By T. R. PARSONS. Pp. xii + 453. Fifth Edition. Cambridge: W. Heffer & Sons, Ltd. 1935.

Confronted with a fifth edition of a book that was first published thirteen years ago, the reviewer may perhaps be forgiven for wondering exactly what he can say that shall in any way usefully supplement so clear and practical an expression of opinion by the book's actual and potential public.

In this particular instance he can at least make an apology. Reviewing the fourth edition of Dr. Parsons's book, he pointed out some minor errors (*Nature*, **1934**, **134**, 162), one of which existed only in the reviewer's imagination. Dr. Parsons's formula for the biuret-reaction giving protein group was correct then and is correct now.

It is pleasant, on the other hand, to realise that Dr. Parsons reads his reviews, and is as ready to correct inaccurate statements as he is to defend accurate ones. Thus one is glad to record that his reference to Hopkins's classical paper, published in 1912, leaves nothing further to be desired in the way of precise presentation and legitimate comment. The use of dots instead of line-links in all semi-structural formulae marks a further improvement in the author's and publisher's already unusually clear method for organic compounds.

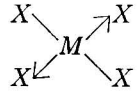
Dr. Parsons steers his readers with equal tact and encouragement over both old and new ground. Neither respiratory quotients nor oxidation-reduction potentials need have any terrors for the student guided by Dr. Parsons; he will

find himself equally at ease with hormones and vitamins, sugar isomerism, sterol and purine chemistry, the composition of glutathione, the relationship of the blood pigments; all of which will be to him but a pleasant preamble to the more serious climb into the heights of gas tensions and osmotic pressure, two sections of the concluding chapter on "Some Applications of Physical Chemistry."

Though it has been said before elsewhere (*loc. cit.*) by this reviewer, that shall not prevent its being once more recorded that, in his opinion, "it is still, without exception, the best elementary exposition of its subject" known to him.

A. L. BACHARACH

DIE ORGANISCHEN KATALYSTOREN UND IHRE BEZIEHUNGEN ZU DEN FERMENTEN. By W. LANGENBECK. Pp. 106. Berlin: At Grafswald, Springer. M.17.75.0.

This little manuscript of Professor Langenbeck contains material of great interest to those interested in catalytic processes in living organisms. The book is divided into six chapters: the first two are introductory, the third with information on the heavy metal bio-catalysts, especially those containing iron and copper in co-ordinated form, appeared to the reviewer to be the most interesting. It is somewhat remarkable that the organic oxidation catalysts can be written in a general form  where M may be a metal—iron, copper or magnesium, and X, nitrogen, oxygen or sulphur. In the later sections of the book the author discusses the production of synthetic organic catalysts containing no metals but possessing a structure akin to what is believed to be the structure of active groups in ferments. These sections are divided into catalysts for addition reactions, hydrolytic and lipoclastic processes. These are stimulating to read, but the mechanism suggested is difficult to accept in all cases for the catalytic processes described.

Thus benzoyl carbinol is found to be an effective lipoclastic and esterifying catalyst. It is possible in heterogeneous systems that the catalyst may function, at any rate partly, as a dispersing or emulsifying agent, thus increasing the rate of reaction. This consideration is evidently important and is worth examining in many of the experiments cited by the author. The subject of these chapters, *viz.* synthetic enzyme models, is too important to permit of any possible uncertainty in the interpretation of the results. The book is well documented and quite up to date.

E. K. RIDEAL

CHROMIUM STEELS. Department of Scientific and Industrial Research. RICHARD HENRY GREAVES, M.B.E., D.Sc., F.I.C. His Majesty's Stationery Office. Price 7s. 6d.

This is in no sense a textbook of metallurgy; it is essentially a book for the specialist, in which he will find correlated, in a condensed but very complete form, all the available information concerning a limited class of steels—those containing chromium alone as an alloying element; the author has also deliberately left aside the corrosion-resisting aspects of the stainless steels, as having been adequately

dealt with elsewhere. The book is divided into nine chapters, together with an appendix dealing with analysis; of these chapters, one treats of the history of chromium steels, three of the constitution of the various iron, chromium and carbon alloys, and one each of (*a*) heat treatment, (*b*) mechanical properties, (*c*) influence of manganese and silicon (which occur in all steels), (*d*) physical properties, and (*e*) uses.

It is interesting to note the very diverse uses recorded for plain chromium steels (which, by the way, like so much else in our modern world, owe their inception to Faraday) in spite of the fact that the still more important steels containing a fourth element do not fall within the scope of the book. The author has had unique advantages in writing a book of this kind, as much of the work mentioned has been carried out by him, or in the Research Department, Woolwich, under his direction; he is also in touch with many of the outside workers. In its condensation and very full bibliography (281 references) the book resembles some of the "Recent Advance" series which have been a feature of late years; to have handled this condensation within a compass of 298 pages, which include also upwards of 160 tables and 78 diagrams, without making the result unreadable, is a feat on which Dr. Greaves may well be congratulated. Printing and production are alike excellent, especially in view of its very moderate price, but there are a few proof-reading slips which should be corrected in a subsequent edition. B. S. EVANS

The Appendix (12 pp.) referred to above, and written by Dr. B. S. Evans, gives a brief but comprehensive survey of the methods available for the analysis of "straight" chromium steels. Lack of space prevented descriptions of the selected processes being given in full, but the numerous references indicate where the details may be found. This survey furnishes an authoritative guide to the selection of analytical methods in this field; the appendix is, therefore, a valuable feature, the more so as a similar survey is lacking elsewhere. S. G. CLARKE

Publications Received

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EMULSIONS AND THEIR TECHNICAL TREATMENT. By W. CLAYTON. Third Edition. Pp. ix + 458. J. & A. Churchill, Ltd. Price 25s.

PHYSICAL ASPECTS OF ORGANIC CHEMISTRY. By W. A. WATERS. Pp. 501. George Routledge & Sons, Ltd. Price 25s. net.

CHEMISTRY OF MILK. By W. L. DAVIES. Pp. xii + 522. Chapman & Hall, Ltd. Price 25s. net.

REACTIONS OF ORGANIC COMPOUNDS. By W. J. HICKINBOTTOM. Pp. x + 449. Longmans, Green & Co., Ltd. Price 16s. net.

TECHNOLOGIE DER TEXTILFASERN: KÜNSTLICHE ORGANISCHE FARBSTOFFE. By H. E. FIERZ-DAVID. Pp. 136. Berlin: Springer. Price RM.14.50.