THE ANALYST

THE ORGAN OF THE

Society of Public Analysts and other Analytical Chemists

A MONTHLY JOURNAL DEVOTED TO THE ADVANCEMENT OF ANALYTICAL CHEMISTRY

VOL. LIV.

1929

PUBLISHED FOR THE SOCIETY BY
W. HEFFER & SONS LTD.
PETTY CURY, CAMBRIDGE, ENGLAND
1929

JANUARY, 1929. Vol. LIV., No. 634.

THE ANALYST

PROCEEDINGS OF THE SOCIETY OF PUBLIC ANALYSTS AND OTHER ANALYTICAL CHEMISTS.

AN Ordinary Meeting of the Society was held in the Chemical Society's Rooms, Burlington House, on Wednesday, December 5th, the President, Mr. Edward Hinks, being in the chair.

Certificates were read for the first time in favour of William Bennett Adam, M.A., A.I.C., Alfred Louis Bacharach, B.A., F.I.C., Andrew Dargie, B.Sc., A.I.C., Wadie J. Itayim.

Certificates were read for the second time in favour of Edwin Herbert Bunce, A.I.C., Frederick O'Brien, M.Sc., F.I.C., William Macro Seaber, B.Sc., F.I.C., John Graham Sherratt, B.Sc., F.I.C.

The following were elected Members of the Society:—Charles Wesley Bayley, Harry Brindle, B.Sc., A.I.C., William George Burgess, George Leonard Clothier, Hector Ingram Downes, M.Sc., A.I.C., Alec Walter Greenhill, M.Sc., A.R.C.Sc., A.I.C., Donald R. Hayward, B.Sc., B. L. Khuller, M.Sc., A.I.C., James Donald Kidd, B.A., M.Sc., A.I.C., Herbert Drake Law, D.Sc., F.I.C., Sidney John Saint, B.Sc., A.I.C.

The following papers were read and discussed:—"The Natural Occurrence of Boron Compounds in Fruits and Vegetable Products," by A. Scott Dodd, B.Sc., F.I.C.; "Chemical Tests for Drunkenness: The Determination of Small Quantities of Alcohol in Urine," by John Evans, F.I.C., and A. O. Jones, M.A., F.I.C.; "The Analysis of Mixtures containing Acetone, Ethyl Alcohol and Isopropyl Alcohol," by C. A. Adams, B.Sc., F.I.C., and J. R. Nicholls, B.Sc., F.I.C.; "The Specific Gravities and Immersion Refractometer Readings of Dilute Mixtures of Acetone and Water," by J. R. Nicholls, B.Sc., F.I.C.; and "The Wijs Method as the Standard for Iodine Absorption," by J. J. A. Wijs, Ph.D.

Death.

WE deeply regret to have to record the death of Mr. J. H. B. Jenkins, on December 11th, 1928.

The Analysis of Mixtures containing Acetone, Ethyl Alcohol, and Isopropyl Alcohol.

By CHARLES AMBROSE ADAMS, B.Sc., A.I.C., and JOHN RALPH NICHOLLS, B.Sc., F.I.C.

(*Read at the Meeting, December* 5, 1928.)

I. Introduction.—Both acetone and isopropyl alcohol find application as substitutes for ethyl alcohol, and the analysis of a mixture of the three substances is occasionally required. The object of this paper is to give analytical methods which have proved satisfactory in this laboratory for the past three or four years.

Preliminary Remarks on the Determination of Ethyl Alcohol.—The usual method for the determination of ethyl alcohol is based upon the specific gravity of its aqueous solution. Many substances, which by their presence would invalidate this method of determination, can be removed by a petroleum spirit and brine separation (Thorpe and Holmes, *J. Chem. Soc.*, 1903, **83**, 314–7). Other substances commonly associated with ethyl alcohol in mixtures may, however, be only partly, if at all, removed by this preliminary treatment, and in this category appear acetone, methyl ethyl ketone, methyl alcohol, isopropyl alcohol, *n*-propyl alcohol, benzyl alcohol, etc. Hence the specific gravity of an aqueous solution of ethyl alcohol containing one or more of these ingredients affords no reliable criterion of the proportion of ethyl alcohol, and the presence of an interfering substance may not be anticipated. The purity of such aqueous alcohol solutions may be tested readily by means of the refractometer, the reading obtained for the test solution being compared with that given by a pure ethyl alcohol solution of the same specific gravity. Tables showing the readings given by the Immersion Refractometer for aqueous solutions of methyl, ethyl, isopropyl and *n*-propyl alcohols obtained in this laboratory by J. Holmes in 1911 have been published (see "Alcohol," by Simmonds; Macmillan & Co., 1919, pages 285 and 287). A similar table for acetone is given in the following paper. A comparative table interpolated from these results is given later in a form convenient for practical use. With the exception of methyl alcohol, solutions of the other substances referred to give higher refractions than solutions of ethyl alcohol of the same specific gravity. Methyl alcohol can be tested for by the Denigès method (Simmonds, Analyst, 1912, **37**, 16; Jones, *id.*, 1915, **40**, 221, etc.). It is to be noted that *n*-propyl, amyl and benzyl alcohols, when present in sufficient proportions, give positive results in this test, which is not, therefore, specific for methyl alcohol. The test is very valuable, however, for indicating the absence of methyl alcohol.

Only in those cases, therefore, where the refractometer reading of a solution, purified if necessary by the Thorpe and Holmes method, agrees with that given by a corresponding solution of ethyl alcohol, and where the Denigès test has given a

negative result, can the specific gravity of a solution be used with certainty for determining the ethyl alcohol.

In our opinion, the refractometer reading (or the determination of some other physical constant) should always be employed as a check upon a determination of ethyl alcohol made from the specific gravity of an aqueous solution, unless the absence of interfering substances is known with certainty.

SPECIFIC GRAVITIES AND REFRACTIONS OF AQUEOUS MIXTURES OF ACETONE AND THE LOWER ALCOHOLS.—In the case of an aqueous solution of acetone or of one of the lower alcohols neither the specific gravity nor the refraction is a linear function of the quantity in solution. When, however, the proportion present does not exceed about 10 per cent. by volume the function is very nearly linear. For purposes of calculation it is convenient to express the strengths of solutions in terms of a common unit, and for this purpose each liquid is regarded as though it were the "proof spirit" of the Official Specific Gravity Tables (H.M. Stationery Office, 1916). The following table shows the relation between percentages of apparent "proof spirit" and percentages by volume of some of the lower alcohols and of acetone. The ethyl alcohol figures are taken from the Official Specific Gravity Tables; the figures for methyl, *n*-propyl and isopropyl alcohols are interpolated from data obtained by Holmes, and those for acetone are interpolated from data obtained by one of us and given in the following paper:

TABLE I.

COMPARATIVE STRENGTHS (PERCENTAGE BY VOLUME).

Apparent proof spirit. Per Cent.	Methyl alcohol. Per Cent.	Ethyl alcohol. Per Cent.	*n*-Propyl alcohol. Per Cent.	Isopropyl alcohol. Per Cent.	Acetone. Per Cent.
1	0·60	0·57	0·62	0·60	0·76
2	1·20	1·15	1·26	1·20	1·53
3	1·81	1·72	1·90	1·80	2·32
4	2·42	2·29	2·53	2·40	3·11
5	3·01	2·86	3·17	3·00	3·89
6	3·61	3·43	3·84	3·60	4·66
7	4·19	4·01	4·50	4·21	5·41
8	4·76	4·58	5·16	4·83	6·15
9	5·31	5·17	5·80	5·44	6·91
10	5·85	5·73	6·50	6·08	7·69
11	6·42	6·30	7·19	6·69	8·42
12	6·99	6·87	7·87	7·33	9·20
13	7·57	7·45	8·55	7·97	9·94
14	8·13	8·01	9·20	8·59	10·65
15	8·68	8·58	9·89	9·20	11·36
16	9·33	9·16	10·55	9·83	12·05
17	9·88	9·74	11·22	10·47	12·75
Mean value for 1 per cent. proof spirit	0·585	0·573	0·655	0·612	0·760

The use of this mean value will give a percentage by volume not differing by more than about 0·1 per cent. from the correct value.

The following table gives the corresponding refractometer readings. All except the acetone figures, which are obtained from the following paper, are interpolated from data obtained by Holmes in this Laboratory:

TABLE II.

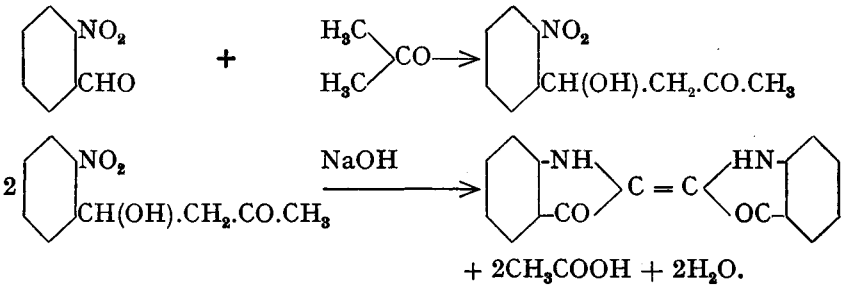
COMPARATIVE REFRACTOMETER READINGS.

Apparent proof spirit. Per Cent.	Immersion Refractometer reading at 60° F.				
	Methyl alcohol.	Ethyl alcohol.	*n*-Propyl alcohol.	Isopropyl alcohol.	Acetone.
0	15·4	15·4	15·4	15·4	15·4
1	15·7	16·1	16·5	16·5	16·6
2	16·0	16·8	17·7	17·6	17·9
3	16·2	17·5	18·9	18·6	19·1
4	16·5	18·2	20·1	19·6	20·3
5	16·8	18·9	21·3	20·6	21·6
6	17·1	19·7	22·6	21·7	22·8
7	17·4	20·5	23·9	22·8	24·0
8	17·7	21·2	25·2	23·9	25·3
9	17·9	22·0	26·6	25·0	26·5
10	18·2	22·8	28·0	26·2	27·8
11	18·5	23·6	29·4	27·4	29·0
12	18·8	24·4	30·8	28·7	30·2
13	19·1	25·3	32·2	29·9	31·4
14	19·3	26·1	33·6	31·2	32·6
15	19·6	26·9	35·0	32·4	33·7
16	19·9	27·8	36·4	33·7	34·9
17	20·2	28·7	37·8	34·9	36·0

We have repeatedly found that with aqueous mixtures of these substances, provided the total proportion does not exceed about 10 per cent. by volume, the apparent proof strength and the refraction of the mixture are practically the sum of those due to each of the ingredients. Hence in mixtures of the above substances it may be stated that up to a strength of about 17 per cent. of apparent proof spirit the specific gravities and refractions are, for practical purposes, additive factors. With two known substances alone present the quantities of each can be calculated by proportion from two determinations, *viz.* the specific gravity and the refractometer reading. With three known substances present, if one can be determined by any independent process, allowance can be made for it in the strength and refraction of the solution, and the proportions of the other two can then be calculated. Similarly, in an unknown mixture, any ingredients which can be separately determined can be allowed for, and the resulting figures give a measure of the undetermined ingredients. It is desirable always to have direct methods for the detection and determination of substances likely to be present, and the following methods are applicable to mixtures containing acetone, ethyl alcohol and isopropyl alcohol.

II. THE DETECTION AND COLORIMETRIC DETERMINATION OF ACETONE.—The test used by Penzoldt for the qualitative detection of acetone (*Deut. Arch. klin. Med.*, **34**, 127; referred to in *Z. anal. Chem.*, 1885, **24**, 149) appears to have escaped general notice. We have found this test satisfactory for the past four years, and have modified it to furnish a rapid quantitative colorimetric method for the determination of acetone.

When sodium hydroxide is added to a mixture of *o*-nitrobenzaldehyde and acetone, indigo is formed as a condensation product:



The indigo separates almost at once as a flocculent precipitate, and its appearance is conclusive evidence of the presence of acetone. With a very small concentration of acetone, however, precipitation of indigo may not at first be apparent, but the solution will develop a colour ranging from yellow through yellowish-green to greenish-blue, depending on the proportion of acetone present. By allowing the solution to stand for about 1 hour and filtering through a small white paper, a distinct indication of precipitated indigo can be obtained from 10 ml. of a 0·05 per cent. solution of acetone.

The yellow and yellowish-green colours produced in solutions containing not more than 0·2 per cent. of acetone are suitable for quantitative colorimetric determinations. Under the conditions described the colour reaches its maximum in 10 to 15 minutes, and a faint but perceptible colour is given by 1 mgrm. of acetone in 10 ml. (0·01 per cent. of acetone).

The *o*-nitrobenzaldehyde solution used tends to darken in colour on keeping. A recently-prepared solution should be used for quantitative work, and the blank should be colourless. The quantitative method is as follows:—

To an aliquot portion of the distillate to be tested (containing not more than 0·02 grm. acetone and diluted with water to 10 ml.) is added 1 ml. of a 1 per cent. solution of *o*-nitrobenzaldehyde in 50 per cent. ethyl alcohol (not methylated spirit, as this contains acetone). After mixing, 0·5 ml. of a 30 per cent. solution of sodium hydroxide is added, and the test solution allowed to stand for about 15 minutes, avoiding strong daylight. At the end of this time the colour is compared with the colour developed in a set of standard acetone solutions, containing from 0 to 20 mgrms. of acetone in 10 ml., which have been treated similarly at the same time. The range of the colours produced is very marked, and it is possible to have as many as twenty readily differentiated standards within the range suggested.

III. Detection and Colorimetric Determination of Isopropyl Alcohol. —Isopropyl alcohol can be oxidised readily to acetone, which can then be determined by the above method. We have found the most convenient oxidising agent for this purpose to be potassium dichromate and sulphuric acid under the conditions to be described later.

For the purpose of the routine testing of alcoholic distillates, however, it was desired to avoid the distillation necessary in the dichromate oxidation, and to use a method of oxidation which could be applied simultaneously to a large number of distillates. Various oxidising agents were tried. Dilute nitric acid, hydrogen peroxide and sodium peroxide, tested under various conditions, gave poor results. Potassium permanganate, either in acid or alkaline solution, gave good results, but the use of this reagent necessitated the filtration of the test solutions. Strong nitric acid was too vigorous in its action. Bromine water gave excellent results, and the conditions for the use of this reagent were therefore worked out.

For the detection of isopropyl alcohol in solutions containing ethyl alcohol the method is as follows:—

The test solutions (after appropriate purification) are diluted to a strength of approximately 10 per cent. by vol. of alcohol. To 10 ml. of each solution in a test-tube 5 ml. of saturated bromine water are added. The tubes are lightly corked and allowed to stand for about 3 to 6 hours in the cold, or even over-night in a cool dark cupboard. After the prescribed period of standing, 1 ml. of a 1 per cent. solution of *o*-nitrobenzaldehyde in 50 per cent. ethyl alcohol is added, and the solutions are mixed by gentle shaking. Finally, 2 ml. of a 30 per cent. solution of sodium hydroxide are added, and the solutions shaken once more. The tubes are then allowed to stand for about 15 minutes in diffused daylight, and the colours developed are compared with those produced in standard isopropyl alcohol solutions similarly treated. A convenient standard solution is one containing 2·5 ml. of pure isopropyl alcohol in 100 ml. of 10 per cent. ethyl alcohol. The standard colours are those produced by using 1·0, 1·5, 2·0 ml., etc., of this solution, made up to 10 ml. with 10 per cent. ethyl alcohol.

Under the conditions described, the test will detect with certainty the presence of 0·025 ml. of pure isopropyl alcohol in the 10 ml. of 10 per cent. alcoholic solution used. The method is not so delicate as when applied directly to acetone, owing to the complex nature of the action of bromine on isopropyl alcohol, but the proportion of acetone produced by this method from a given quantity of isopropyl alcohol in a given time under similar conditions appears to be constant.

In routine testing the value of the indications is not appreciably affected by the presence of small proportions of formaldehyde or methyl alcohol. Normal propyl alcohol in this test, however, gives a brownish colour on the addition of soda, probably due to the resinification of propyl aldehyde produced by the treatment with bromine. Although this colour gradually fades, it is liable to interfere with the shade of colour produced from isopropyl alcohol. In the presence of *n*-propyl alcohol a quantitative determination of isopropyl alcohol by this method should only be attempted when the proportion of *n*-propyl alcohol is known, so

that the equivalent quantity of *n*-propyl alcohol may be added to the standards before oxidation with bromine.

In this Laboratory the chief value of the test has been found in its giving a ready qualitative method of examination applicable to a large number of alcoholic solutions at a time.

Where a positive indication is obtained the test is repeated, omitting oxidation with bromine, in order to detect whether acetone is present. Quantitative determinations are then made as described above, if necessary both before and after oxidation. Alternatively, the dichromate oxidation process described later may be employed.

IV. Determination of Acetone, Ethyl Alcohol and Isopropyl Alcohol Mixtures.—For the determination of acetone the above-described colorimetric method is suitable, or the well-known Messenger process can be employed. For ethyl alcohol and isopropyl alcohol it was thought that if conditions could be found whereby these could be oxidised completely to acetic acid and acetone, respectively, then the determination of these products would enable the proportions of the alcohols to be calculated. Of the oxidising agents tried, potassium dichromate and sulphuric acid gave the most promising results, especially as it was found that the oxidation could be carried out at room temperature in a closed vessel, thus obviating loss. The behaviour of each of the three substances under various conditions of oxidation was studied.

Oxidation of Ethyl Alcohol.—The factor which is most pronounced in determining the rate of oxidation at room temperature is the proportion of sulphuric acid. With small proportions of sulphuric acid oxidation to acetic acid takes many hours, or even days, whilst with fairly large proportions it is complete in a few minutes. Unless the quantity of acid is extremely large, oxidation does not go beyond acetic acid. Under the conditions finally chosen the time required is less than 15 minutes. After reduction of the excess of dichromate the acetic acid can be steam-distilled and titrated.

Oxidation of Isopropyl Alcohol.—The proportion of sulphuric acid similarly determines the rate of oxidation, which proceeds regularly to acetone; with high proportions of acid the acetone formed is further oxidised. Under the selected conditions the oxidation is complete in 15 to 20 minutes. After reduction of the excess of dichromate the acetone produced can be distilled.

Oxidation of Acetone.—With moderate proportions of sulphuric acid oxidation is extremely slow, but the rate of oxidation increases with the proportion of acid. Under the selected conditions no appreciable action takes place in half-an-hour.

Method of Oxidation.—The concentration of sulphuric acid in the oxidising mixture which gives the best results is 12·5 ml. of concentrated acid per 100 ml. The proportion of potassium dichromate present is not so important, but about 5 grms. per 100 ml. is a suitable quantity. It is convenient to prepare a stock oxidising mixture of double strength to be mixed with an equal volume of the solution to be oxidised.

An aliquot part of the distillate to be tested, containing not more than 1·5 grm. ethyl alcohol or 3 grms. of isopropyl alcohol, is placed in a distillation flask of about 800 ml. capacity and diluted with water to approximately 100 ml. An equal volume of the stock oxidising mixture (10 grms. of potassium dichromate and 25 ml. of conc. sulphuric acid per 100 ml.) is added, and the flask is corked and allowed to stand for 25 to 30 minutes. An excess of ferrous sulphate is added, and the solution is steam-distilled, the contents of the flask being allowed to concentrate to about 100 ml. The concentration should not be carried too far, or the distillate becomes yellowish and gives an acidity not due to acetic acid. This occurs when evaporation has proceeded so far that iron compounds separate in the distilling flask and cause bumping. If the volume of liquid is not reduced below about 100 ml., this condition is completely avoided. The whole of the acetic acid should be in the distillate when about 500 to 600 ml. have been collected, but it is advisable to collect a further 100 ml. in a fresh receiver. The distillate is titrated with normal sodium hydroxide, solid phenolphthalein being used as indicator. The addition of 10 to 20 grms. of common salt makes the end point rather sharper. 1 ml. of *N* alkali=0·046 grm. of ethyl alcohol=0·058 ml. of ethyl alcohol=0·101 ml. of proof spirit. The neutralised solution is then redistilled, the distillate being made up to a known volume and the acetone present determined from the specific gravity of the solution (see following paper) or, if small in amount, colorimetrically. The acetone found represents that originally present, together with that produced from the isopropyl alcohol.

1 ml. acetone = 1·043 ml. isopropyl alcohol.

The following results were obtained by this method:—

Ethyl Alcohol:—10 ml. of a solution of specific gravity 0·97840 (=1·761 ml. ethyl alcohol) oxidised. Acetic acid titration=30·35 ml. of *N*/1 alkali. Ethyl alcohol=30·35×0·058=1·760 ml.

Isopropyl Alcohol:—25 ml. of a solution of specific gravity 0·98770 (=2·43 ml. isopropyl alcohol) oxidised. Acetic acid titration=0·2 ml. of *N*/1 alkali (equivalent to 0·01 ml. ethyl alcohol). The recovered acetone diluted to 100 ml. had specific gravity 0·99748=2·28 ml. acetone=2·38 ml. isopropyl alcohol.

Acetone:—3·98 grms. (=5·00 ml.) pure acetone treated. Acetic acid titration=0·1 ml. of *N*/1 alkali. The re-distilled acetone, diluted to 100 ml., had specific gravity 0·99467=4·96 ml. acetone.

V. NOTE WITH REGARD TO *N*-PROPYL ALCOHOL.—When *n*-propyl alcohol is present the above oxidation method can be used to determine isopropyl alcohol, but not ethyl alcohol. *n*-Propyl alcohol is oxidised by dichromate and sulphuric acid, and, as with ethyl alcohol, the proportion of acid determines the rate of oxidation. The product, however, is not solely propionic acid, but a mixture of that acid and acetic acid. The relative proportion of the two varies with the conditions, the ratio of acetic acid to propionic acid increasing with the proportion of acid. No conditions were found which at room temperature, would give solely

propionic or solely acetic acid. Under the above specified conditions of oxidation *n*-propyl alcohol gives about 70 per cent. propionic acid and 30 per cent. acetic acid. Although the product is a mixture, 1 mol. of *n*-propyl alcohol gives 1 mol. of acid, so that after steam distillation the mixed acid can be titrated; and with *n*-propyl alone present, 1 ml. of *N*/1 alkali=0·060 grm. *n*-propyl alcohol=0·0743 ml. *n*-propyl alcohol. With mixtures of ethyl and *n*-propyl alcohols the titration of the acids produced by oxidation may form a useful check of the proportions of the two alcohols, calculated from the strength and refractometer readings of their aqueous solution.

VI. SUMMARY.—1. Comparative Tables are given of the strengths and immersion refractometer readings of aqueous solutions of acetone and of some of the lower alcohols.

2. Provided not more than 10 per cent. by volume of these substances are present in aqueous mixtures, the specific gravities and refractometer readings are approximately additive factors.

3. A rapid method for the detection and colorimetric determination of acetone is given, with an adaptation of the process to the determination of isopropyl alcohol.

4. Conditions are given for the complete oxidation of ethyl alcohol and isopropyl alcohol, respectively, to acetic acid and acetone, with a method for the determination of these products.

The authors desire to thank the Government Chemist for permission to publish this work.

GOVERNMENT LABORATORY,
W.C.2.

The Specific Gravities and Immersion Refractometer Readings of Dilute Mixtures of Acetone and Water.

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

(*Read at the Meeting, December* 5, 1928.)

CAREFULLY fractionated commercial acetone was found to contain appreciable quantities of oxidisable material. It was therefore distilled twice from dichromate and sulphuric acid (the second operation showing no appreciable change of colour due to oxidation), the distillate being then shaken with anhydrous potassium carbonate and again distilled. The constant-boiling fraction was shaken with fused calcium chloride for several days, re-fractionated, and the process repeated until the specific gravity was constant. The final product boiled at 56·61–56·64° C. at 775·5 mm. (56·04–56·07° C. at 760 mm.) and had a specific gravity at 60° F./60° F. of 0·7960, and an immersion refractometer reading at 60° F. of 89·7 (N_D=1·36098).

Another sample of acetone was prepared from the sodium iodide compound and was similarly fractionated and dried. The product boiled at 55·65–55·70° C. at 749 mm. (56·05–56·10° C. at 760 mm.) and had a specific gravity at 60° F./60°F. of 0·7960, and an immersion refractometer reading of 89·5 at 60° F. (n_D=1·36092).

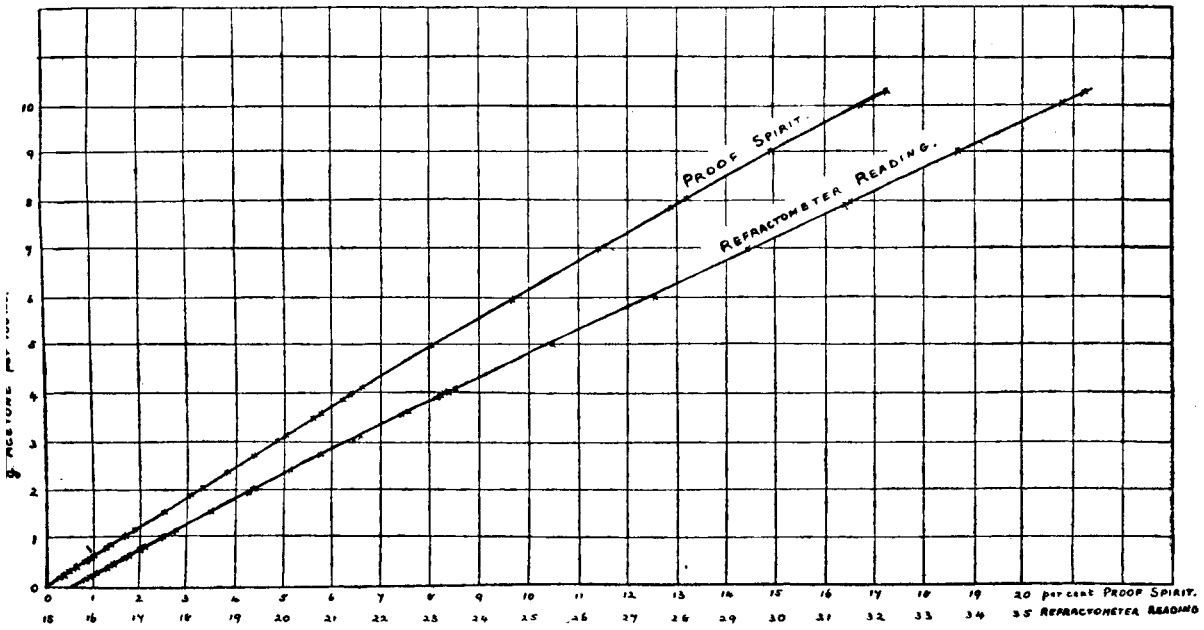
Weighed quantities of each of these products were diluted with water to 100 ml., and the specific gravities and refractometer readings taken. The following were the results obtained:—

ACETONE I.

Acetone, grms. per 100 ml.	Specific gravity $\frac{60° \text{ F.}}{60° \text{ F.}}$	Refractometer reading at 60° F.	Equivalent proof strength from Official Specific Gravity Tables.
0·174	0·99975	15·8	0·29
0·294	0·99958	16·05	0·48
0·476	0·99932	16·35	0·79
0·619	0·99914	16·65	1·00
0·781	0·99891	17·0	1·27
1·170	0·99837	17·75	1·90
1·565	0·99786	18·5	2·50
1·928	0·99737	19·25	3·08
2·379	0·99676	20·15	3·81
2·722	0·99632	20·8	4·35
3·137	0·99576	21·6	5·03
3·520	0·99525	22·4	5·66
3·587	0·99514	22·6	5·80
3·899	0·99478	23·2	6·26
3·964	0·99469	23·3	6·37
4·060	0·99451	23·5	6·60
7·922	0·98973	31·5	12·91

ACETONE II.

Acetone, grms. per 100 ml.	Specific gravity $\frac{60° \text{ F.}}{60° \text{ F.}}$	Refractometer reading at 60° F.	Equivalent proof strength.
0·205	0·99971	15·9	0·34
0·394	0·99943	16·3	0·66
0·570	0·99919	16·7	0·94
0·814	0·99887	17·1	1·31
1·005	0·99858	17·5	1·64
2·020	0·99719	19·5	3·29
3·004	0·99592	21·5	4·82
3·994	0·99464	23·5	6·43
4·972	0·99334	25·5	8·08
5·982	0·99203	27·6	9·78
6·997	0·99081	29·5	11·42
8·015	0·98954	31·6	13·17
9·007	0·98829	33·7	14·96
9·985	0·98706	35·8	16·76
10·238	0·98678	36·2	17·18



The following figures were interpolated from the plotted results:—

Acetone, grms. per 100 ml.	Equivalent proof strength.	Refractometer reading at 60° F.
0	0	15·5
1	1·63	17·45
2	3·24	19·4
3	4·83	21·35
4	6·45	23·4
5	8·12	25·5
6	9·80	27·65
7	11·44	29·6
8	13·15	31·55
9	14·93	33·7
10	16·73	35·75

GOVERNMENT LABORATORY,
W.C.2.

The Wijs Method as the Standard for Iodine Absorption.

By J. J. A. WIJS, Ph.D.

(*Read at the Meeting, December* 5, 1928.)

Among the subjects discussed at the ninth Conference of the International Union of Pure and Applied Chemistry at the Hague was the standardisation of the determination of the iodine value.

The discussion resulted in a resolution that the method devised by me should have the preference, especially in forensic cases, and that it should be accepted as the official method. In the literature on this subject there are two papers, the contents of which seem to be incompatible with this conclusion.

I. Schmidt-Nielsen, referring to the method in his *Vergleichende Untersuchungen*,* observed: "Besides the addition, a substitution also takes place. For this reason one can never obtain correct iodine values, and the method is altogether unsuitable for all scientific work."

Nielsen compares different methods of determining the iodine value by following the course of the iodine fixation. For this purpose he stops the reaction after 2, 6 and 24 hours, and represents the calculated iodine values by a graph. Then, from the results, he postulates that the curve, drawn through the three points, must show a horizontal portion, which would indicate a cessation in the iodine absorption. Not finding such a stoppage indicated in the line he drew for the Wijs reagent, he concludes that there must be substitution as well as addition. This conclusion is somewhat hazardous, but much more important is the fact, that the observation, the basis of the conclusion, is defective.

As long ago as 1899 I made and published (*Chem. Rev. Fett. Ind.*, 1899, **6**, 5; Analyst, 1899, **24**, 94) similar experiments. I reproduce here Table VII of that paper.

	Arachis oil.			Linseed oil.		
Time.	*N*/10 thiosulphate c.c.	Free halogen in terms of *N*/10 thiosulphate. c.c.	Velocity in terms of *N*/10 thiosulphate. c.c.	*N*/10 thiosulphate. c.c.	Free halogen in terms of *N*/10 thiosulphate. c.c.	Velocity in terms of *N*/10 thiosulphate. c.c.
0	36·25	—	—	37·89	—	—
1 min.	—	26·88	562·2	—	27·69	612·6
5 ,,	—	26·81	1·05	—	27·54	2·25
15 ,,	—	26·77	0·24	—	27·52	0·12
1 hour	—	26·77	0·00	—	27·50	0·02
2 ,,	—	26·77	0·00	—	27·50	0·00
6 ,,	36·25	26·75	0·005	—	27·47	0·01

* Edited by J. Dybwad, Kristiania, 1923.

It is evident that the stoppage required by Nielsen appears here very clearly; with arachis oil after a quarter of an hour, and with linseed oil within an hour. Nielsen, however, could not detect this stoppage, for the simple reason that he did not end his first experiment until after two hours.

It is hardly surprising that in such a mixture as the one in question a slow diminution of the free halogen will, in the long run, occur. If, however, during the short time of the experiment itself, these secondary reactions are so small as not to change perceptibly its result, the method is not affected. The table given above proves clearly that this is the case here.

In studying Nielsen's curves, one should bear in mind that only three points in each curve have been fixed by experiment. Nielsen, in drawing these curves, tacitly assumed that no stoppage or irregularity had occurred. As regards my reagent, my experiments clearly prove that this assumption does not agree with the facts.

II. The *Kritisch-experimentelle Untersuchungen* of Weiser and Donath (ANALYST, **1914**, **28**, 65) also appear not to support my method. These authors determined the iodine value of pure unsaturated fatty acids, and failed to obtain the theoretical values, when using the Wijs reagent.

I regret to have to say, however, that these experiments, so far as they relate to my method, involve a fundamental error. Weiser and Donath state explicitly that they prepared the reagent according to Lewkowitsch's formula, using 9·4 grms. of iodine trichloride and 7·2 grms. of iodine.

This is an unfortunate error in Lewkowitsch's excellent book, which was corrected in the next edition, but, unfortunately, passed in the French translation by Bontoux. To convert 9·4 grms. of iodine trichloride into monochloride 10·2 grms. of iodine are required.

Weiser and Donath made their experiments with a solution containing a large quantity of iodine trichloride, whereas it is known that even a small quantity makes the solution unstable and causes secondary reactions. The experiments of Weiser and Donath are therefore valueless as adverse criticism of my method.

In 1899 I determined the iodine values of erucic, brassidic and elaidic acids, and obtained the theoretical values. Analogous results can be obtained with the other fatty acids present in natural oils and fats, as is clearly shown in the interesting paper of Boeseken and Gelber (*J. Chem. Ind.*, 1927, *B*, 427), the study of which I warmly recommend to everyone interested in the determination of iodine values.

It may be useful to add some details to amplify the general directions for using the method, which were published in the Report of the Conference (*loc. cit.*). The best method of preparing the reagent is to dissolve about 9 grms. of iodine trichloride in one litre of glacial acetic acid of at least 99 per cent. strength. If the liquid is to be kept in a cold place, where it could crystallise, a mixture of 300 c.c.

carbon tetrachloride and 700 c.c. acetic acid may be used instead of one litre of acetic acid.

Exactly 5 c.c. of this solution are taken, and its halogen content determined by means of $N/10$ thiosulphate solution, after adding potassium iodide and water. The bulk of the solution is treated with 10 grms. of pulverised iodine and shaken to make it dissolve. When almost all the iodine is dissolved, exactly 5 c.c. are again taken, and the halogen content determined. As soon as this is found to be one-half more than that found by the first determination the solution is filtered into a bottle provided with a tightly fitting stopper. It is preferable to exceed slightly this limit of one-half more, as this ensures that no iodine trichloride remains in the finished reagent, the effect of which would be to make this preparation unstable and cause secondary reactions when it was used for the determination. If desired, the reagent thus prepared may be diluted with acetic acid to exactly $N/5$ strength.

The acetic acid and carbon tetrachloride must be absolutely free from oxidisable matter; this is controlled by warming one or two c.c. of each liquid with some concentrated sulphuric acid and a drop of a saturated dichromate solution. A green tinge should not be noticeable, even after prolonged standing. It is necessary to be very punctilious on this point, in order to get a stable solution; 0·46 per cent. of ethyl alcohol, oxidised to aldehyde, would be sufficient to effect complete reduction of all the halogen present in an $N/5$ solution. Kept in a well-closed bottle, in the dark, the solution remains in good condition for years.

It must be borne in mind that its coefficient of expansion by heat is rather high (0·00115); thus, for 25 c.c., a difference in temperature of 1° C. makes a difference in the titration with $N/10$ thiosulphate of 0·06 c.c.

The quantity of oil or fat to be taken for a test should be so measured that not more than 30 per cent. of the halogen present in the 25 c.c. of the solution added is absorbed. This quantity of oil is dissolved in a few c.c. of carbon tetrachloride before the 25 c.c. of the iodine solution are added.

After half-an-hour to two hours, according to the degree of unsaturation of the fatty substance, the non-absorbed halogen is titrated.

The Natural Occurrence of Boron Compounds in Fruits and Vegetable Products.

By A. SCOTT DODD, B.Sc., F.I.C., F.R.S.E.

(*Work done under the Analytical Investigation Scheme.*)

(*Read at the Meeting, December* 5, 1928.)

In a recent publication by the author (Analyst, 1927, **52**, 459) reference was made to the existence of boron compounds as normal constituents of various vegetable substances. It was then shown that boron compounds are present in cacao and cacao products to quite an appreciable extent. The present investigation was undertaken with a view to ascertain to what extent boron is present in other vegetable commodities, and to determine what quantitative margin should, in general, be allowed in differentiating between "natural" and "added" boron compounds in commercial vegetable products. G. Bertrand and H. Agulhon (*Compt. rend.*, 1914, **158**, 201) determined boron compounds in a large number of fruits and vegetables by means of a colorimetric method devised by themselves. In most cases, however, these results are much lower than those found by the author in the present investigation.

Method of Analysis.—The boric acid was, in each instance, determined by the author's modification of Thomson's method described in the previous publication (*loc. cit.*). As, however, the quantity of boric acid was very small, it was considered expedient to use about 40 grms. of the sample for each determination, and to carry out the titration by means of a 10 c.c. burette of narrow bore, so that the readings could readily be made to one hundredth of a c.c. In the case of those substances which contained little fat, ether extractions were dispensed with, while in every instance care was taken to ensure the presence of a distinct excess of alkali before igniting.

Selection of Samples.—Keeping in view the fact that "added" boron compounds constitute an infringement of the Public Health (Preservatives) Regulations, 1925, special attention was paid to those vegetable substances which form the constituents of manufactured products. Currants and raisins, for example, enter into the manufacture of cakes, buns, mince meat, wines, etc. Various fruits also enter into jams, cakes and sauces, while spices and flavourings, such as almond and coconut, were also regarded as likely sources of boron compounds. The field for such an investigation is, therefore, very wide; and though no claim to exhaustiveness can be made, this investigation covers a wide range of articles and indicates that the quantity of boron compounds occurring naturally

in vegetable products is comparatively small, and therefore not likely to be confused with those quantities, which would require to be added for preservative purposes.

CURRANTS AND RAISINS.

Variety.	Actual.			Dried.		
	Boric acid. Per Cent.	Moisture. Per Cent.	Ash. Per Cent.	Organic matter. Per Cent.	Ash. Per Cent.	Boric acid. Per Cent.
Vostizza currants	0·014	11·90	2·45	97·22	2·78	0·017
Australian currants	0·013	5·00	2·05	97·84	2·16	0·014
B.P. Patras currants	0·010	11·15	2·35	97·36	2·64	0·011
Spanish muscatels	0·015	20·70	1·80	97·73	2·27	0·020
Spanish muscatels	0·012	12·45	2·65	96·98	3·02	0·014
Smyrna sultanas	0·018	13·90	2·15	97·50	2·50	0·022
Australian sultanas	0·022	15·15	1·75	97·94	2·06	0·026
Smyrna sultanas	0·012	14·10	2·80	96·51	3·49	0·014
Valencia raisins	0·010	10·50	2·00	97·77	2·23	0·011
Californian raisins	0·013	14·85	2·40	97·18	2·82	0·016

These results show that both currants and raisins contain small, but appreciable, quantities of boron compounds. The amount in the dried fruits varies from 110 to 260 parts per million by weight, and, so far as these samples show, there is no evidence to prove that the Valencia raisins and the sultanas, which are subjected to "dipping" in process of preparation for the market, contain less boric acid than the currants and raisins which are not dipped. While the quantity of boric acid in each sample is admittedly too small to be of much use for the purpose of preservation, it is interesting to note that all the above currants and raisins contain more than 100 parts per million of boric acid.

DRIED FRUITS.

Variety.	Actual.			Dried.		
	Boric acid. Per Cent.	Moisture. Per Cent.	Ash. Per Cent.	Organic matter. Per Cent.	Ash. Per Cent.	Boric acid. Per Cent.
Australian apricots	0·022	22·60	3·75	95·16	4·84	0·029
Californian apricots	0·023	20·40	3·65	95·41	4·59	0·030
Persian dates	0·006	19·30	1·75	97·83	2·17	0·008
Crystallised cherries	0·014	17·85	1·25	98·48	1·52	0·018
Turkish figs	0·006	22·10	3·25	95·83	4·17	0·008
Australian peaches	0·025	16·35	3·15	96·24	3·76	0·030
Californian peaches	0·022	19·25	3·50	95·54	4·46	0·028
French prunes	0·003	15·85	1·80	97·86	2·14	0·004
Californian prunes	0·003	17·50	1·75	97·88	2·12	0·004
French prunes	0·003	16·40	1·90	97·73	2·27	0·004
Dried pears	0·008	19·00	1·45	98·21	1·79	0·011
Dried apple rings	0·006	18·40	1·25	98·47	1·53	0·008

In the above list of miscellaneous dried fruits the boric acid content of the dried sample is shown to vary from 40 parts per million, in prunes, to 300 parts per million, in apricots and peaches. It is noteworthy that the Californian prunes contain much less boric acid than the apricots and peaches from California, but little can be deduced from this fact, as the districts may be too far from one another to show definitely that the apricot and peach trees take up more boron compounds from the soil than the plum trees, although these results certainly suggest that such an occurrence is probable. The above results show, however, that quite appreciable quantities of boron compounds exist as natural constituents of apricots and peaches, and must therefore be allowed for in jams, tarts, and other products manufactured therefrom.

FRESH FRUITS.

Variety.	ACTUAL.			DRIED.		
	Boric acid. Per Cent.	Moisture. Per Cent.	Ash. Per Cent.	Organic matter. Per Cent.	Ash. Per Cent.	Boric acid. Per Cent.
Belgian black currants	0·005	81·50	0·55	97·03	2·97	0·030
Belgian red currants	0·005	85·10	0·90	93·96	6·04	0·033
German cherries	0·003	84·40	0·30	98·08	1·92	0·024
French cranberries	0·006	88·80	0·25	97·77	2·23	0·055
Belgian gooseberries	0·003	88·85	0·35	96·86	3·14	0·028
Dutch tomatoes	0·005	94·85	0·45	91·27	8·73	0·109
Danish cranberries	0·003	90·50	0·15	98·42	1·58	0·039

Although the actual amount of boron compounds found in these samples of fresh fruit is very small, namely, varying from 31 to 62 parts per million expressed as boric acid, it will be observed that fairly high figures are obtained when these results are calculated on the dry sample. The quantity of boric acid in the dry substance varies from 240 to 1090 parts per million; and although apparently of alarming magnitude, when considered with reference to the Public Health (Preservative) Regulations, 1925, is not likely to be nearly so large in any products prepared therefrom, as most of the latter contain a considerable proportion of water. On comparing the amounts of boric acid in the fresh fruits and dried fruits calculated on the dried samples, it will be observed that the fresh fruit results are, on the whole, much higher. This, however, may be largely, if not entirely, accounted for by a loss of boric acid occurring during the drying of acid fruits without the presence of an alkaline fixing agent.

A. Hebebrand (*Z. Unters. Nahr. Genussm.*, 1902, **5**, 1044–1049) found minute quantities of boric acid in orange juice and lemon juice, amounting to 4 and 6 mgrms., respectively, per litre. The Report of the Government Chemist for the year ending in March, 1928, shows that a number of oranges were tested to ascertain if boric acid were present in the edible portions of the fruit, and that mere traces only were found therein. These two investigations and results are of

especial interest, in view of the fact that Australian growers sterilise the skins of citrus fruits before export.

SPICES AND FLAVOURINGS.

Variety.	Actual.			Dried.		
	Boric acid. Per Cent.	Moisture. Per Cent.	Ash. Per Cent.	Organic matter. Per Cent.	Ash. Per Cent.	Boric acid. Per Cent.
Californian almonds	0·014	3·90	2·65	97·24	2·76	0·015
Dutch caraway seeds	0·012	11·50	7·25	91·81	8·19	0·014
Ground cinnamon	None	—	—	—	—	None
Ground coconut	0·001	3·35	1·30	98·65	1·35	0·001
Ground ginger	None	—	—	—	—	None
Black pepper	0·014	11·85	4·25	95·18	4·82	0·016
Mixed spice	0·008	9·50	5·00	94·48	5·52	0·010

With the exception of almonds and coconut, none of the above spices and flavourings occurs in large proportion in any manufactured product. The quantity of boric acid in no instance is high, and is so small (if present) as to be undetectable in cinnamon and ginger, while in coconut it is practically negligible. One would expect to find less boron compounds in the fibrous than in the more succulent portions of a plant, and the above results certainly tend to support this theory, and show that in the vegetable kingdom boron compounds are more plentiful in fruits than in stem structures.

VEGETABLE PRODUCTS.

Variety.	Actual.			Dried.		
	Boric acid. Per Cent.	Moisture. Per Cent.	Ash. Per Cent.	Organic matter. Per Cent.	Ash. Per Cent.	Boric acid. Per Cent.
Indian relish	0·001	67·50	1·60	95·08	4·92	0·006
Tomato ketchup (1)	0·003	63·40	3·90	89·34	10·66	0·008
Tomato ketchup (2)	0·001	64·50	4·10	88·45	11·55	0·005
Indian mango chutney	0·008	34·05	3·25	95·00	5·00	0·012
Fruit cake (1)	0·011	28·50	1·25	98·25	1·75	0·016
Mince meat	0·009	18·40	1·65	97·98	2·02	0·011
Green tomato chutney	0·003	75·40	0·40	98·38	1·62	0·015
Fruit cake (2)	0·013	25·70	1·45	98·05	1·95	0·017

The above results show the boric acid content of certain classes of manufactured vegetable products, which one would expect, from their concentrated nature, to contain a proportion of boron compounds approximating to the maximum. In no instance, however, was the quantity of boric acid found to be high; it varied from 19 parts per million in relish and ketchup to 130 parts per million in fruit cakes, which were largely composed of currants and other dried fruits and spices. Even allowing for the maximum limits of variation produced by different degrees of dryness, the amount of boric acid is still small, as the water-free product, which

would never be found in practice, is shown to contain only 170 parts per million of boric acid.

WINES AND VINE PRODUCTS.

Variety.	Boric acid. Per Cent.	Extract. Per Cent.	Ash. Per Cent.
British wine (1)	0·003	14·60	3·80
British wine (2)	0·002	15·15	4·30
Spanish grape juice	0·024	44·50	0·75
Greek grape juice	0·016	47·80	0·50
South African grape juice	0·018	70·80	0·60

For many years it has been known that boron compounds exist as natural constituents of wines and vine products. H. Jay and Dupasquir (*Compt. rend.*, 1895, 260) found Bordeaux and Burgundy wines to contain amounts of boric acid varying from 0·010 to 0·022 grm. per litre. The above results are expressed in parts by weight, and are similar, though somewhat higher, than those found by these investigators.

The vine is the main source of natural boric acid in the majority of wines, and the above results indicate that the quantity of boric acid existing naturally in wines is very small, and amounts merely to 10–30 parts per million. Grape juice, which is imported from various countries as a basis for the manufacture of British wines, is a concentrated article, and therefore contains quite an appreciable quantity of boric acid. The results given show considerable variation in the boric acid content, and the variation is even more pronounced when the figures are calculated to the same concentration of extract. It is, however, clearly shown that the quantity of natural boron compounds expressed as boric acid is much less than would be likely to be added to prevent the juice fermenting.

CONFIRMATORY TEST FOR BORIC ACID.—It was observed in carrying out the final titrations in the boric acid estimation (ANALYST, 1927, **52**, 464) that the neutralised solution, free from carbonic anhydride and phosphates, gave a pink coloration when mannitol was added (Sofnol Indicator No. 1 being present as the indicator). No pink colour was given, when boric acid was absent, and the depth or intensity of pink was greater or less according to the quantity of boric acid present.

In the fruits and samples in which minute quantities only of boron compounds were present the pink reaction was found to be very faint, whereas in other substances and fruits containing a larger quantity of boron compounds the pink reaction was very distinct.

Each of the substances under examination in this investigation was carefully tested by the sensitive turmeric paper test detailed by the author (ANALYST, 1927, **52**, 465). It was found that in each instance this qualitative test was satisfactorily confirmed in the course of the investigation by the intensity of the pink reaction, which acted as a useful confirmatory test.

GENERAL CONCLUSIONS.—In surveying the foregoing analytical results it will be observed that, in the actual samples tested, the amount of boron compounds, expressed as boric acid, varies from none to 0·025. Fuller investigation would require to be made before one could arrive at a definite maximum, but, so far as the results of this investigation go, it would appear that no product manufactured from mixtures of fruits and spices would be likely to contain more than 0·03 per cent. of boron compounds, expressed as boric acid. This amount is, as already shown, very much less than the minimum quantity of boric acid which would be added as a preservative, as no one would be likely to add less than 0·15 per cent. of boric acid with any hope of ensuring efficient keeping qualities. Even allowing a substantial margin for all possible variations, it would appear that if any article of food were found to contain more than 0·1 per cent. of boric acid, it would be reasonable to conclude that the article contained added boron compounds.

The wide geographical distribution of boron compounds is shown by the presence of boron in the various vegetable products mentioned in this and the previous investigations. It would, therefore, appear that boron compounds exist in practically all soils, and it is quite conceivable that in districts where it is very abundant the vegetation grown thereon will contain a larger amount than the vegetation grown on soils which contain minute traces only of boron compounds. Certain plants may, of course, possess greater powers of assimilating boron compounds than other plants, as researches in this subject have shown that, up to certain amounts, boron compounds assist growth and development, though larger quantities invariably retard the growth.

Special care has been taken throughout this investigation to ensure that the results obtained are reliable. It was found that, in undertaking the determination of such small quantities of boric acid, great care and strict adherence to technique was necessary in order to avoid the introduction of minute but nevertheless appreciable errors. The modified Thomson's process, though somewhat laborious, was adhered to throughout, and was found to give very satisfactory results.

In conclusion, the author would like to acknowledge his indebtedness to the Scottish Federation of Grocers and others who have kindly supplied him with reliable samples necessary for this investigation.

DISCUSSION.

The PRESIDENT said that he was a little sceptical about the accuracy or reality of these small quantities when they depended upon the titration method by $N/10$ sodium hydroxide solution. The method used was the modified "Thomson," which was really known as the Government Laboratory method, but he understood that the Government method was brought forward to deal with large quantities of boric acid. His experience had been that with many products there was always a slight blank attaching to this method, and the old Thomson method gave a similar blank, due to small quantities of phosphoric acid.

Dr. B. S. EVANS said that he thought that metallurgical analysis might throw a little light on the question. He had carried out many experimental determinations of boron in copper, and in this case, as it was impossible to burn off the copper, it had to be precipitated; this was done with sodium hydroxide,

which caused a considerable concentration of salts in the liquid. Under these conditions he found blanks varying from 0·3 to 1·0 c.c. of $N/10$ sodium hydroxide solution, and as the circumstances entirely precluded the possibility of the presence of phosphates, and rendered any appreciable amount of carbon dioxide unlikely (the acid solutions had been boiled for a considerable time under a reflux condenser), he had attributed the blank to silica. This was rendered more probable by the fact that sodium hydroxide dissolved in porelain gave a lower blank than that in glass, and in platinum lower still. He found the error in his determinations to be $\pm 0{\cdot}3$ mgrm. He did not know if glycerin was still used in the Thomson process, but he found mannitol greatly superior. He wondered whether the author had followed up the colour reaction quantitatively, as he believed a colorimetric method would be very valuable.

Mr. A. More agreed that there was a blank in the Thomson method, often amounting to 0·1 or 0·15 ml.; it was due to the carbonic acid not having been entirely removed by boiling. He doubted some of the figures given by the author, and wondered whether there was not some constant error both in these figures and in the author's previous results on cocoa. As an example of a titration error, he cited an instance where, some years ago, a sample of cocoa had been examined by this process and reported to contain a large amount of boric acid. The ash of the cocoa, however, did not give an indication of boric acid with turmeric, and contained no boric acid. The cause of the error was traced to the fact that calcium chloride had not been added in sufficient quantity before the neutralisation stage. Cocoa ash contained a very much larger proportion of magnesium than of calcium, and magnesium phosphate did not precipitate so completely as calcium phosphate. At the stage of the process where the phosphate should have been precipitated completely some remained soluble as magnesium phosphate, and caused a very pronounced difference between the neutral points to the two indicators, which was reported as boric acid. In most cases since then he had obtained a quantitative confirmation of the amount of boric acid by a colorimetric method before reporting the titrimetric result.

Mr. Chaston Chapman remarked that the figures given by the author appeared to refer only to land fruits. He might, however, remind the meeting that he had shown in a recent communication that traces of boric acid were present in agar and in other sea-weeds. He felt strongly that it was unwise to attach too great importance to very small titration numbers, and he himself would not feel satisfied unless he confirmed such results by some specific test. Probably the author had done so. He felt that the practical value of the communication was reduced to some extent by the fact that quantities such as the author had found could not properly be regarded as significant in connection with analyses made under the Sale of Food and Drugs Acts. He contended that the real object of those Acts was to protect the public without harassing the trader, and therefore there would be no necessity for the Public Analyst to report the presence of traces, even having full regard to existing Regulations.

Dr. Monier-Williams, while agreeing with the views expressed by the last speaker, said he was prepared to accept the figures published, but at the same time there was often some uncertainty in the case of imported fruits, *e.g.* oranges, as to whether they had been treated with boric acid in the country of origin. He could confirm the amount found in Australian sultanas, as he had examined a sample sent from Australia House, which was guaranteed not to have been treated.

Professor W. H. Roberts said that he could confirm the presence of small amounts of boric acid in currants, but that he had evidence that in some cases, at all events, it was due to the currants having been washed with boric acid.

Mr. A. E. PARKES said that the paper had cleared up a small mystery which had been bothering jam manufacturers for some time. Unaccountable traces of boric acid had frequently been found in jams and mincemeat. During the last year he had examined half-a-dozen samples of jam which contained 1–2 grains per pound of boric acid—an amount which roughly corresponded with that found by the author in fruits. During the past season he had examined several samples of mincemeat, all of which gave a strong boric acid reaction with turmeric paper

The PRESIDENT asked Dr. Cox if he would kindly reply in the absence of the author.

Dr. COX stated that much of the criticism was anticipated, and Mr. Scott Dodd had sent samples to him, and he had examined them for boric acid. With regard to the blank—he could state definitely that it did not exceed one drop from an ordinary burette. Mannitol was certainly very sensitive, and gave more definite results than glycerin. He was afraid he could not say what "Sofnol No. 1" was.

With regard to the point raised by Mr. More, it was expressly stated by Mr. Scott Dodd that excess of calcium chloride should be added before filtering off.

Two samples of apricots examined by Mr. Scott Dodd and Dr. Cox gave the following percentage results:

Mr. Scott Dodd	..	0·0222	Mr. Cox	.. 0·0186
		0·0236		0·0217

and in a number of other samples quantities were of the same order.

It was really essential, in dealing with small quantities, to burn off the carbon completely. When one considered how absorption carbons was made, it would be realised that exactly the same thing was done in this process, and therefore the carbon had absorptive power, and it was essential to burn it off. It was rather tedious in the case of the necessarily large sample of dry fruit, but to get accurate results it must be done. Neglect to do this might explain lower results.

Mr. Scott Dodd pointed out that qualitative confirmation of the presence of boric acid had been obtained.

ADDENDUM.—The point raised by Prof. W. H. Roberts is one of considerable importance, as the value of the results obtained in this investigation depends very largely upon the freedom of these samples from "added" boric acid. The subject of the treatment and preparation of dried fruits has been carefully investigated, and the Secretary of the Scottish Federation of Grocers, who supplied a large number of the samples, and has a very wide and intimate knowledge of these matters, stated emphatically that boric acid is never used in the preparation of dried fruits for the market. Sultanas, apricots, peaches and prunes are commonly treated with sulphur dioxide, but never with boric acid. Dried fruits are treated with preservative substances for two reasons only: to improve their appearance or to kill insects. Boric acid would do neither of these things, and there would would therefore be no sense or reason in treating dried fruits with boric acid solutions.

Special care was taken in the selection of the samples used in this investigation, and there is little or no doubt that all the boric acid found therein has been due to its natural occurrence in the plants from which they spring.

With regard to the suggestion that "Sofnol" might be used as the basis for a colorimetric determination of boric acid, it is doubtful if anything would be gained thereby. The mannitol and sofnol colour with boric acid solutions can only be of reliable depth when other acids and phosphates have been eliminated, so it is really more satisfactory to titrate the boric acid than to try to match coloured solutions.—A. SCOTT DODD.

A Method for the Determination of Traces of Antimony in Copper and its Alloys.*

S. G. CLARKE, B.Sc., A.I.C., AND B. S. EVANS, Ph.D., F.I.C.

EVANS published some time ago (ANALYST, 1922, **47**, 1) a method for the determination of small amounts of antimony in copper and brass; this method, whilst giving fairly good results, had certain objectionable features:

(*a*) The copper was removed as a sponge by treatment of the sulphate solution with sodium hypophosphite. This process worked well with samples of fairly pure copper, but led to complications and probable loss of antimony in presence of any considerable amount of tin. Moreover, we have latterly found that sodium hypophosphite is liable to be contaminated with an unidentified impurity, which leads to loss of antimony. Further, should the hypophosphite used contain chlorides, as did certain samples of this salt which we have examined, a precipitation of some antimony on the copper sponge would certainly take place.

(*b*) The arsenic had to be removed completely, owing to the antimony being finally determined colorimetrically as sulphide. This was effected by boiling the chloride solution, after removal of the copper, as mentioned, with sodium hypophosphite, and was quite satisfactory with copper samples; but, as one of us has recently found (Clarke, ANALYST, 1928, 373), arsenic cannot be separated quantitatively in presence of stannic salts by precipitation with hypophosphite. This fact would also render the procedure inaccurate when applied to a bronze containing any arsenic.

(*c*) The traces of copper, bismuth, etc., remaining in the solution obtained by stripping the deposited antimony from the copper after deposition by the Reinsch method, were removed by a tedious, and not too satisfactory, separation with zinc sulphide.

(*d*) The final colorimetric determination was carried out on a colloidal solution of antimony sulphide. The colour obtained was entirely ruined by any trace of copper, bismuth, etc., which had not been removed by the preceding operation, or rendered too intense by any arsenic which escaped precipitation by sodium hypophosphite.

In 1928, one of us published (Clarke, ANALYST, 1928, 373) a method of colorimetric determination of small quantities of antimony, which was a great improvement on the sulphide method, being unaffected by relatively large amounts of tin and arsenic.

Subsequent to the publication of the method for determining antimony in copper, one of us showed (Evans, ANALYST, 1923, **48**, 264) that, whilst copper in

* Communication from the Research Department, Woolwich.

the cupric state is fatal to the Reinsch reaction, cuprous copper is not. The line of attack, therefore, which led to the present process was:—

(*a*) Reduction of the copper to the cuprous state in hydrochloric-sulphuric acid solution. This was effected simply by adding sodium hypophosphite.

(*b*) Deposition of the antimony on copper by the Reinsch reaction.

(*c*) Determination of the antimony in the solution obtained by stripping the Reinsch film with sodium peroxide, by the method mentioned above.

The full method is as follows:—Five grms. of the sample are dissolved in 30 c.c. of dilute sulphuric acid (1 : 3) and 15 c.c. of concentrated nitric acid, and evaporated until the sulphuric acid fumes strongly. This may be accomplished safely and quickly by placing the beaker on an asbestos pad on a hot plate till fumes of sulphur trioxide begin to be evolved, and then removing it to the bare plate. The residue, after cooling, is dissolved in 150 c.c. of water, 150 c.c. of concentrated hydrochloric acid (sp. gr. 1·18) are added, followed by 10 grms. of sodium hypophosphite, and the solution is boiled for 10 minutes.

In the absence of any appreciable amount of arsenic (which is generally the case) the liquid may be treated at once by the Reinsch method, as below. If a brownish-black precipitate appears during this boiling (due to arsenic), the liquid must be boiled for a further 20 minutes, cooled somewhat, and well shaken after the addition of 20 c.c. of benzene to coagulate the precipitated arsenic. It is filtered through a wet filter paper, which is washed with the minimum of hot water and rejected. This treatment removes the greater part of the arsenic in the case of all metals not containing a large amount of tin; although arsenic does not interfere in the colorimetric method, its removal, when present in more than small amounts, is advantageous, inasmuch as arsenic depositing with antimony in the Reinsch test tends to weaken the film and cause it to become detached from the copper.*

In the case of a tin bronze, 10 grms. of oxalic acid are dissolved in the hydrochloric acid solution before adding the hypophosphite. In general, a white turbidity is produced which may be ignored; after about 30 minutes' boiling during the Reinsch process this turbidity disappears, yielding a perfectly bright solution.

A piece of pure electrolytic copper foil, about $20 \times 2{\cdot}5$ cm., is rolled into a flat spiral, as open as is consistent with its being dropped into the flask containing the solution under examination; it is cleaned by warming gently with nitric acid (sp. gr. 1·2), rinsed with water and dropped into the solution, which is then boiled gently for two hours. (It is desirable that the coil should stand upright, not lie on its side.) At the end of this time the coil is lifted out of the boiling liquid by means of a hooked glass rod; rinsed *quickly* by plunging into a beaker of water, and placed *without delay* into a small beaker of diameter only slightly greater than that of the coil; it is covered with distilled water, and about 1 grm. of sodium

* In practice, we have not met with tin bronzes containing notable amounts of arsenic; we have therefore not found it necessary to introduce any modification of the process to meet this case.

peroxide is at once added. After standing for five or ten minutes the beaker containing the coil is warmed gently until the coil becomes darkened with a layer of oxide. If too much time has not been allowed to elapse between withdrawing the coil from the boiling acid solution and adding the peroxide, the antimony should now be completely removed from the copper, together with any bismuth and arsenic that was present and a little copper. The liquid is now poured off into a small flask, and the coil and beaker rinsed in twice with distilled water; the coil should be immersed in dilute sulphuric acid, which removes the film of oxide and shows up any antimony which may have escaped stripping. In the rather *unlikely* event of antimony having been incompletely stripped, the stripping process with peroxide must be repeated, and the solution added to that obtained from the first stripping.

The solution containing the antimony is now treated with a rapid current of hydrogen sulphide for 15 seconds, and the flask allowed to stand on a water bath for about half-an-hour; the precipitated copper and bismuth sulphides are then filtered off through a small, close-pressed, pulp filter, and lightly washed with 1 per cent. ammonium nitrate solution; 5 to 6 c.c. of concentrated sulphuric acid are added to the filtrate, and it is evaporated until fumes of sulphur trioxide begin to be evolved, a few drops of nitric acid being added during the latter part of the evaporation. The sulphuric acid solution of antimony is taken up with 15 c.c. of water, heated just to boiling point and cooled; the antimony in this solution is determined colorimetrically as follows:—

Into a 10 c.c. Nessler glass are put reagents in the order named: 10 c.c. of 1 per cent. gum arabic, 5 c.c. of 20 per cent. potassium iodide, 1 c.c. of 10 per cent. aqueous pyridine, 1 c.c. of a dilute solution of sulphur dioxide (one tenth saturated), 60 c.c. of cold dilute (1: 3) sulphuric acid. The antimony solution, obtained as described above, is now added, after filtration if necessary, through a small filter paper, the beaker being rinsed in with not more than 5 c.c. of water; the whole is well stirred with a glass rod. Standard antimony solution (0·0001 grm. Sb. per c.c.) is run into another Nessler glass containing similar quantities of reagents (*except that* 80 *c.c. of* 1 *:* 3 *sulphuric acid are used instead of* 60 *c.c.*) until the colours match after the solution has been well stirred. A final adjustment is made just before the final colour-match, by adding a small quantity of water to make the volumes in the Nessler glasses equal. The colorimetric comparison is made by viewing the tubes vertically over a white tile inclined at an angle to act as a light reflector.

The standard antimony solution contains 0·2764 grm. of tartar emetic in 1 litre of 10 per cent. sulphuric acid.

If more than 10 c.c. of this standard solution have to be added the colour obtained is too deep for accurate comparison; if the amount of antimony present is still greater, a turbidity is produced. In this case 20 c.c. of the solution are withdrawn from the Nessler glass into another one, similar quantities of reagents are added as in the first instance; any turbidity thereupon disappears, and the colour is matched with fresh standard. It is preferable, however, in this colorimetric method that the amount of antimony should not exceed 0·0005 grm.; in this, as

in almost all other colorimetric work, accuracy begins to fall off when the colour developed becomes unduly deep. Few of the ordinary copper alloys contain more than the above amount in a sample weight of 5 grms., so that subdivision of the solution, as mentioned above, or reduction of the sample weight taken, is rarely necessary.

Trials of the above process made on samples of copper (with and without arsenic), bronze, brass and cupro-nickel, to which varying amounts of antimony had been added (as a standard solution, to the metal in a beaker before dissolving) gave the following results.

COPPER.

Copper taken.	Antimony added.	Antimony solution required.		Antimony found.	Antimony	
		Total.	Nett.		Added.	Found.
Grms.	Grm.	c.c.	c.c.	Grm.	Per Cent.	Per Cent.
5·0	Blank	0·1	—	—	—	—
5·0	0·00005	0·6	0·5	0·00005	0·001	0·001
5·0	0·00010	1·05	0·95	0·000095	0·002	0·0019
5·0	0·00030	3·0	2·9	0·00029	0·006	0·0058
5·0	0·00050	5·1	5·0	0·00050	0·010	0·010
5·0	0·0010	10·1	10·0	0·0010	0·020	0·020

ARSENICAL COPPER.

Copper taken.	Arsenic added.	Antimony added.	No. of c.c. required.		Antimony recovered.	Antimony.	
			Total.	Nett.		Added.	Found.
Grms.	Per Cent.	Grm.			Grm.	Per Cent.	Per Cent.
5·0	—	Blank	0·1	—	—	—	—
5·0	0·5	0·0003	3·05	2·95	0·00030	0·0060	0·0060
5·0	0·2	0·0003	2·90	2·80	0·00028	0·0060	0·0056
5·0	0·05	0·0003	2·85	2·75	0·00028	0·0060	0·0056
5·0	0·008	0·0003	3·00	2·90	0·00029	0·0060	0·0058
5·0	0·49	0·0005	4·70	4·60	0·00046	0·0100	0·0092
5·0	0·19	0·0005	4·90	4·80	0·00048	0·0100	0·0096
5·0	0·05	0·0005	4·80	4·70	0·00047	0·0100	0·0094
5·0	0·008	0·0005	4·90	4·80	0·00048	0·0100	0·0096

BRONZE 90: 10.

Copper taken.	Arsenic taken.	Antimony added.	No. of c.c. required.		Antimony recovered.	Antimony.	
			Total.	Nett.		Added.	Found.
Grms.	Grm.	Grm.			Grm.	Per Cent.	Per Cent.
4·5	0·5	Blank	0·10	—	—	—	
4·5	0·5	0·00005	0·60	0·50	0·00005	0·0010	0·0010
4·5	0·5	0·00010	1·1	1·0	0·00010	0·0020	0·0020
4·5	0·5	0·00025	2·4	2·3	0·00023	0·0050	0·0046
4·5	0·5	0·00050	5·0	4·9	0·00049	0·0100	0·0098
4·5	0·5	0·00075	7·4	7·3	0·00073	0·015	0·0146

CUPRO-NICKEL 80 : 20.

Taken.	Antimony added.	No. of c.c. required.		Antimony recovered.	Antimony.	
		Total.	Nett.		Added.	Found.
Grms.	Grm.			Grm.	Per Cent.	Per Cent.
5·0	Blank	0·40	—	—	—	—
5·0	0·0020	2·5	2·1	0·00021	0·0040	0·0042
5·0	0·0030	3·4	3·0	0·00030	0·0060	0·0060
5·0 (another sample Blank=0·2 c.c.)	0·0050	5·1	4·9	0·00049	0·010	0·0098

BRASS 70 : 30.

Taken.	Antimony added.	No. of c.c. required.		Antimony. recovered.	Antimony.	
		Total.	Nett.		Added.	Found.
Grms.	Grm.			Grm.	Per Cent.	Per Cent.
4·27	Blank	0·1	—	—	—	—
4·60	0·00025	2·4	2·3	0·00023	0·0054	0·0050
4·07	0·0004	4·0	3·9	0·00039	0·0098	0·0096

A point of paramount importance in this process is the acid strength necessary in the quantitative separation of antimony on copper in the presence of cuprous chloride. While it was known from the previous work of Evans that cuprous chloride had no fundamental inhibitive effect on the Reinsch reaction, yet very low results were obtained in the early stages of the work with the acid concentration usually used in the Reinsch test.

This was at first thought to be due to the action of atmospheric oxygen in preventing complete reduction of the copper in solution, but the results were not improved by excluding air and passing a stream of carbon dioxide into the flask during the boiling with copper.

Series of experiments were carried out with progressively increased hydrochloric acid concentration. With 50 c.c. of hydrochloric acid in 300 c.c. of solution at the commencement of the boiling with a copper coil, for example, results were obtained which were low to the extent of about 70 per cent.; the greater the amount of antimony taken, however, the greater was the percentage recovery. Using 150 c.c. of hydrochloric acid (sp. gr. 1·18) in 300 c.c. of solution, quantitative results were obtained. It would seem, therefore, that cuprous chloride renders some hydrochloric acid non-reactive in this connection, probably by withdrawing it into complex combination, so that, unless excess is provided, the amount of effective hydrochloric acid may fall below that necessary for promoting the Reinsch reaction.

Sodium hypophosphite is a most effective reducing agent for use in this process, as a slight excess is sufficient to keep all the copper in the cuprous state during the Reinsch reaction. Nitric acid is somewhat resistant to the reducing action of sodium hypophosphite, so that, if this acid is not removed by fuming with sulphuric acid, a green colour is produced in the solution during boiling, due

to cupric chloride, which is inimical to the quantitative precipitation of antimony. Thorough fuming with sulphuric acid is therefore necessary.

It may be remarked that the process described presents distinct advantage over those described in the two recent Continental papers (Tschernichof, *Z. anal. Chem.*, 1928, **73**, 265; Blumenthal, *id.*, 1928, **74**, 33). In both these methods the amount of antimony is finally arrived at by titration with $N/10$ potassium bromate solution; as 1 c.c. of this solution is equivalent to approximately 0·003 grm. of antimony, it is evident that a considerable sample weight is necessary when determining the amounts of antimony dealt with in the present paper. These processes were not tested, as it seemed that considerable expenditure of time would be necessary to obtain results of equal accuracy to our own.

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.

BORIC ACID IN ORANGES.

RECENTLY we had referred to us, by one of the authorities for whom we act as Public Analysts, seven samples of oranges; comprising three Californian, three South African, and one West Indian brand.

These samples were submitted to us because it was believed that oranges were being treated with an antiseptic.

Six of the seven samples were wrapped in papers, in which we found no antiseptic, nor did we find any upon the surface of the peels of any of the samples, but we found that in all the samples the peel and the pulp, which were analysed separately in each case, contained boric acid.

The results obtained were as follows:—

	PEEL.		PULP.	
	H_3BO_3 Per Cent.	Grains per lb.	H_3BO_3 Per Cent.	Grains per lb.
1.	0·033	2·31	0·006	0·42
2.	0·022	1·54	0·002	0·14
3.	0·012	0·84	0·002	0·14
4.	0·008	0·56	0·002	0·14
5.	0·017	1·19	0·004	0·28
6.	0·020	1·40	0·008	0·56
7.	0·005	0·035	0·004	0·28
	Average	1·12		0·28

The average weights of the peel and pulp were 30 and 120 grms. respectively.

Samples Nos. 4, 5 and 7 were Californian, Nos. 1, 3, and 6 South African, and No. 2 West Indian; all were wrapped with the exception of No. 7.

Boric acid has before been found to be a natural constituent of oranges, and of a large number of other vegetable substances, in quantities comparable with

those cited above; so that the detection of such quantities of boric acid affords no evidence that it has been purposely added.

The occurrence of boric acid as a widely distributed natural constituent of many food stuffs, animal and vegetable (as well as of the common salt used in their preservation), is of special interest in view of the fact that its use is now prohibited under the Public Health (Preservatives, etc., in Food) Regulations, 1925; and we consider it of sufficient importance to call attention to the matter.

J. T. DUNN.
H. CHARLES L. BLOXAM.

INSTABILITY OF PRECIPITIN ANTI-SERA IN THE TROPICS.

I HAVE read with great interest Mr. Bamford's note on this question (ANALYST, October, 1928, p. 531), and believe that it will help to throw light on a difficult problem.

Mr. Bamford suggests bacterial action as the cause of instability. This is a tempting hypothesis, and the first of which one thinks, although in this laboratory it has been found that *all* anti-human precipitin sera (in sealed ampoules) lose their potency in three months.

But is it not significant that many workers (including those in this laboratory) have obtained good reactions with putrid human blood, which must necessarily have undergone bacterial decomposition? It cannot be inferred that bacterially decomposed anti-sera will also react, but good reactions have been obtained in this laboratory from anti-sera which were apparently decomposed. It is, of course, impossible to use such anti-sera for tests to be quoted in the Courts. These considerations have suggested a doubt as to bacterial decomposition being the cause of instability of anti-sera.

Mr. Bamford mentions anti-sera for (apparently) other than the human species which have been proved stable (? albeit weakened), though submitted to variations of temperature (up to 15° C.) for five years. It would be interesting to know if any anti-human sera were included. Is it possible that anti-sera for species other than human are more stable than human anti-sera? Or is the cause of their (apparent) stability the powerful character of the original anti-sera?

Variability in temperature has seemed the only hypothesis left to account for instability of anti-sera. It is, perhaps, possible that anti-sera would lose their potency with comparative rapidity if maintained at the blood temperature of the rabbit or other species used as reservoir. Mr. Bamford mentions various anti-sera which had been subjected to temperatures of 30–35° C. for twelve days before they were tested. At that time they were satisfactory, but for what length of time would they have remained stable?

From a study of the literature and from some experience the picture one has formed of these anti-sera is that of unstable compounds, produced at the blood temperature (probably of the rabbit), which are always tending to revert to their original molecular constitutions, particularly at the temperature of the blood of the rabbit or other animal used for their preparation. Anti-sera made from a bird might possibly be more stable than anti-sera made from a mammal.

The subject is obscure, and all chemists interested in anti-serum tests will welcome contributions thereto.

HERBERT S. SHREWSBURY
(*Government Analyst, Trinidad and Tobago, B.W.I.*).

THE PRODUCTION OF UNIFORM STAINS IN THE GUTZEIT TEST FOR ARSENIC.

LERRIGO (ANALYST, 1928, **53**, 90) has called attention to the occasional difficulty experienced in removing mercuric chloride papers which have been attached with seccotine to the top of the tube of the B.P. 1914 (Appendix VI) apparatus.

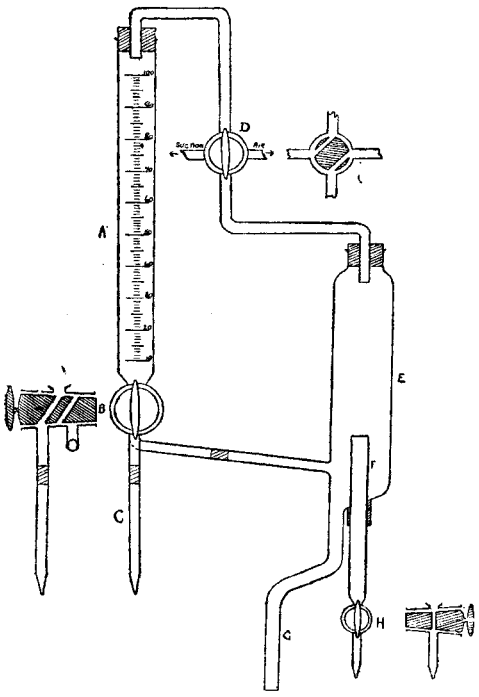
I find that tearing of the paper is avoided by the use of "Gloy"; a square (length of side, 18 mm.) of mercuric chloride paper is laid on a clean surface on the table and the rim of the glass tube, previously treated with "Gloy," firmly pressed upon the paper. The attached rubber cork is then re-inserted in the bottle. The paper is afterwards removed with greater ease than would be the case with a disc cut to the exact size of the top of the tube.

Apart from the 20 c.c. tap funnel the apparatus conforms to the requirements laid down in the B.P. 1914 (Appendix VI).

C. H. MANLEY.

AN IMPROVED MISCOMETER.

THE miscometer described in the ANALYST (1926, **51**, 453) has been replaced by an alternative form of apparatus. In the new apparatus the measuring device is more satisfactory, and the measuring chamber has been lengthened and narrowed. The apparatus is simpler to manipulate and is cheaper.



It will be seen from the figure that the new measuring device consists of a burette instead of a hollow stopper. The hollow stopper proved expensive to make and difficult to graduate accurately. The burette may be made quite accurate by grinding down the top to the required dimension. Different burettes may be fitted to deliver quantities of different volumes.

The new miscometer is used in the following manner :—With the suction pump operating and connected with the measuring chamber (A), the samples to be made composite are drawn in turn into the measuring chamber by opening the stopcock (B), which is so left that the inlet tube (C) drains. The samples are mixed by turning the stopcock (B) so that it connects the two chambers. Air is thus drawn through the mixture. When mixing is complete the stopcock (D) is turned through an angle of 90°. The composite sample is thus drawn into the second chamber (E). During this operation the burette (F) fills up, and at the end of the operation the remainder of the composite sample flows out (by gravity) through the outlet tube (G) into any suitable receptacle. Stopcocks (B) and (D) are then closed, and the measured quantity is drawn off by opening stopcock (H).

JOHN HOUSTON.

CHEMICAL RESEARCH DIVISION,
QUEEN'S UNIVERSITY, BELFAST.

Notes from the Reports of Public Analysts.

The Editor would be glad to receive the Annual or other Reports of Public Analysts containing matter of special interest to the Society. Notes made from such Reports would be submitted to the Publication Committee.

CITY OF BIRMINGHAM.

REPORT OF THE CITY ANALYST FOR THE THIRD QUARTER, 1928.

DURING this quarter 1260 samples were analysed, of which 1098 were under the Food and Drugs Acts. Of these, 1024 were bought informally (51 adulterated), and 74 were formal samples (9 adulterated).

CREAM.—The whole of the 18 samples were free from boric acid, but one was composed of artificial cream and the vendor was cautioned. Another was a sample of tinned cream which contained 23 per cent. of fat and was marked: "Pure Thick Cream," "Highly Concentrated." This was a false label, as the sample contained only about half the proportion of fat present in a good "thick" cream.

VERMICELLI.—Four informal samples sold as vermicelli contained from 11·0 to 12·9 per cent. of protein. One sample was described as "——'s Egg Vermicelli," for which there is no standard. The label, however, stated that if four ounces of *egg* vermicelli were taken to make vermicelli omelet, only one egg would be necessary, instead of two eggs if ordinary vermicelli were used. This was a false label, as it claimed that four ounces of the egg vermicelli were equal to four ounces of vermicelli and an egg. If this had been true, about 100 grains of fat and 300 grains of protein should have been present in that quantity, instead of the 9 grains of fat and 230 grains of protein which were actually present. The vendor was cautioned.

BORAX HONEY.—Five of the 13 informal samples were of incorrect composition; 10 per cent. of borax should be present in this preparation, but in three samples the amounts present were 2·9, 4·4 and 8·2 per cent., respectively.

Two samples from one vendor were labelled "Guaranteed to conform with the requirements of the British Pharmacopoeia," but had been prepared with artificial honey instead of the genuine article. The vendors of the five samples were cautioned.

SODIUM CITRATE TABLETS.—Samples containing two and three grains were of full strength, and broke down very well in water. In each case about 6 per cent. of talc was present. This seems to be an undesirable constituent for tablets which are often used for babies. The manufacturer undertook not to use talc in these tablets in the future.

TINCTURE OF MYRRH.—Eleven of the 13 informal samples varied in specific gravity from 0·851 to 0·864, and contained from 5·3 per cent. to 7·8 per cent. of total solids. Two samples were condemned, as they only contained 4·3 per cent. and 4·2 per cent. of total solids, respectively, and the vendors were cautioned.

The question arose as to whether the low solids in the last two samples were the result of unsatisfactory keeping, and experiments were made on this point. About 2 oz. of tincture of myrrh were kept in a 4 oz. bottle with a narrow neck,

and the bottle was left unclosed for 49 days. At the beginning of the time the total solids amounted to 5·5 per cent., and at the end of that period to 8·7 per cent. The sample remained quite clear, and so it was evident that the only change taking place was concentration due to evaporation of the spirit, and that enough spirit remained to keep the dissolved matter in solution.

The effect of keeping, therefore, was to increase the proportion of solid matter, and not to diminish it.

J. F. LIVERSEEGE.

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.

ARTIFICIAL VINEGAR: LIABILITY OF RETAILER.

HIGH COURT APPEAL CASE. PRESTON *v.* JACKSON.

ON November 23 an appeal was heard in the High Court, King's Bench Division, by the Lord Chief Justice, sitting with Mr. Justice Avory and Mr. Justice Acton, against a decision of the Atherstone (Warwickshire) magistrates, not to convict the respondent, a grocer, for selling vinegar not of the nature, substance and quality demanded.

The appellant, an inspector under the Food and Drugs Acts, had asked for "table vinegar," and had been supplied with an article which, on analysis, was found to consist of 100 per cent. of artificial vinegar. The justices, said counsel, seemed to have come to the conclusion that, as there was no standard of quality for vinegar, it was for the prosecution to prove that the respondent had some guilty knowledge when he sold the vinegar. But under section 6 of the Act the question of guilty knowledge did not arise. If goods were sold that contravened the Act an offence had been committed. The respondent was a village grocer, and he had in his shop a cask containing, according to the label, "Finest table vinegar, wholesome." The inspector bought some, and, when analysed, it was found to be artificial.

The Lord Chief Justice asked: "What is artificial vinegar?"

Mr. Bartley replied that it was not vinegar at all, not having been made from cereals and without phosphates.

Mr. Justice Avory: "A synthetic vinegar. Merely diluted acid with a little colour."

Mr. Bartley, continuing, said that there was evidence before the justices that malt vinegar was sold by wholesalers at 8s. 6d. for six gallons, whilst artificial vinegar cost about 4s. for the same quantity. The appellant had been charged for malt vinegar. For the respondent it has been urged that there was no fixed standard for vinegar, and that therefore he had not been guilty of an offence, although he might have sold as "table vinegar" an artificial vinegar that had not been produced by fermentation or acetification, but no evidence had been called for the respondent. The justices had refused to convict, considering that no

question of law arose, and that, if anyone ought to be punished, it should be the wholesalers who sold the liquid to the respondent, and charged 8s. 6d. for stuff that was worth only 4s.

Mr. Justice Avory asked whether these justices were Members of Parliament, since they seemed to think that they could alter the law instead of carrying it out.

Mr. Sandlands, for the respondent, contended that the justices meant to deal with the case under the Probation of Offenders Act, but the Lord Chief Justice pointed out that to have dealt with the case under that Act, the justices must first have found that an offence had been committed. This they had not done. They had found, indeed, that the offence had not been committed by the respondent, for they said: "The inspector went to the shop, asked for table vinegar, and got it, according to the label on the cask." Upon the uncontradicted evidence of the analyst the only course open to the justices was to convict the respondent. Therefore the case would go back to them with an intimation that the only true conclusion in law was that the charge had been proved. The appeal succeeded, with costs.

The other Justices concurred.

PARAFFIN WAX IN DRIPPING.

ON November 16 a firm of manufacturers was charged, at the Birkenhead Police Court, with consigning beef dripping containing paraffin wax to a stallholder in the Birkenhead market, on October 15.

Mr. H. E. Davies, Public Analyst, said that he had analysed a sample of the dripping, and had found it to contain 8·4 per cent. of paraffin wax. He stated that the wax was not only an adulterant, but might set up indigestion in anyone consuming the dripping.

The solicitor for the defence said that the firm had bought the business about 18 months before, and, unfortunately, had carried on the old methods of dealing with the dripping. On making enquiries, they had found that the man responsible for making the dripping used paraffin wax for stiffening purposes.

A director of the company said that they had no knowledge that any harmful adulteration was taking place. The wax had been supplied to them as lard wax; instructions had been given that its use was to be discontinued.

The Chairman of the bench said that it was the duty of manufacturers to make themselves acquainted with the materials they used. A fine of £10 with £1 11s. 6d. costs was imposed.

International Standard Measurements for Mineral Water Analysis.

THE International Society of Medical Hydrology was founded seven years ago by Dr. R. Fortescue Fox, its first President, and still the Chairman of its Council. It is a medical society, having for its chief object the advancement of scientific and systematic conceptions in the use of mineral waters for the treatment of disease and the promotion of health. It is obvious that this object demands as one of its foundation stones some systematic scheme of analysis of the waters

concerned, and with this object in view it appointed, on the occasion of its Annual Meeting in Rome, in October, 1927, an International Standard Measurements Committee to draw up recommendations. This Committee includes:—Professor Chassevant (France), Sir John Flett (England), Dr. Fitch (U.S.A.), Dr. Fresenius (Germany), Dr. S. Judd Lewis (London), Professor Nasini (Italy), Mr. Race (Buxton), Mr. Woodmansey (Harrogate), Dr. Zörkendörfer (Czechoslovakia).

In October this year, the Society held its Annual Meeting in London, and adopted the following resolutions:—

1. That the analysis be expressed in ions, whether it be expressed also in salines or other terms or not.

2. That the quantities be expressed in parts per million; either milligrams per litre or milligrams per kilogram.

3. That the specific gravity refer to water at the same temperature, preferably at 15° or 20° C.

4. That the quantities be limited to four significant figures or to one decimal point, except where the quantity is less than ten, when two decimal figures may be used.

5. That the analysis may be expressed also in terms of salines, the salines being calculated according to an internationally approved arbitrary formula, which has not yet been settled.

6. That the analysis be expressed also in terms of milli-normality ($N/1000$), both in the case of ions and salines.

7. That the analysis, together with other data, be reported according to a definite scheme now under consideration, one feature of which will be that the analytical table will embody four columns:—(*a*) Name of the ion or saline; (*b*) proportions in terms of the international standard, namely, parts per million; (*c*) the same in terms customary in the country where the spring is found; for example, parts per 100,000 in England, grms. per kilogram in Japan; (*d*) the same in terms of $N/1000$.

8. A National Committee is to be appointed in each country to regularise the publication of analytical and other data.

It should be emphasised that neither the Society nor the Committee has any intention of prescribing methods of determination, except in such cases where uniformity is desirable, as in the specific gravity and the arbitrary formula for calculating salines, and possibly in two or three other cases.

The whole scheme is as yet in embryo, and much has to be done in organising the National Committees, and also in deciding upon a formula for the calculation of the salines from the ions, such as will be generally acceptable by chemists throughout the world, and upon other matters.

It is desirable that all concerned in the analysis and other data relative to mineral springs should keep these resolutions in mind, so that when the scheme is complete, analytical or other information derived in the meantime, may fall into place without difficulty.

S. JUDD LEWIS

(*Chairman of the Committee*).

Department of Scientific and Industrial Research.

REPORT OF THE FOOD INVESTIGATION BOARD FOR THE YEAR 1927.*

THE Director's Report embodies the results of work done under a very large number of sections, only a few of which can be mentioned.

SECTION A: MEAT.—The transport of chilled beef, conditioning of beef, freezing of tissue, and coagulation of muscles plasma all receive attention. Experiments to determine the best use for surplus pig products of the Dominions suggest that, although bacon cannot be kept for long in the chilled condition, if properly frozen, at a temperature of −10° C. for fresh, and −15° C. for mild-cured bacon, successful results may be obtained.

SECTION B: FRUIT AND VEGETABLES.—Keeping properties of apples may be predicted to a certain extent, since long life is related to low respiratory activity, small size and to small and delayed climacteric. Also, the trees with the largest fruit, as a rule, bear apples with the highest respiratory activity and the shortest life. No marked relation was found between the chemical composition of apples and their keeping properties in cold storage, but bad-keeping apples appear to lose acid and sugar in respiration more rapidly than those that keep better. Oranges stored at various constant temperatures and under two conditions of atmospheric humidity for each temperature, showed two forms of breakdown—a collapse and browning of small scattered areas which was most prevalent at 5° C., and a discoloration of larger areas, with an appearance of water-logged flesh and disagreeable odour, which were more frequent at 1° C. In 60–80 per cent. of the specimens fungal disease developed at the button area of the orange. At temperatures of 25° C., 15° C., and 10° C. damp atmospheres, with 95–98 per cent. relative humidity, on the whole, increased liability to invasion by fungal rot organisms. A study of the water relationships of fruit rotting fungus, particularly of *Alternaria citri, Colletotrichum glososporioides, Cephalothecium roseum* and *Fusarium fructigenum,* showed that germination could take place in saturated and sub-saturated atmospheres with no other source of water, and the nearer the temperature to the optimum, the lower is the humidity at which germination is possible. The various relationships are illustrated by graphs. Specimens of certain types of wastage of imported fruit have been examined, and a preliminary summary is given in two tables for grape fruits and oranges from different sources, showing the fungi isolated, with remarks on them. In addition, tomatoes from Teneriffe and Jersey showed *Phoma destructiva, Diplodina lycopersici, Pleospora* (*Macrosporia*) sp., and *Fusarium* sp., from rots starting mainly at the stem end; egg fruit from S. Africa, *Alternaria* sp., *Cladasporium herbarum, Botrytis cinerea,* and *Macrosporum* sp. (soft spotting); melons from S. Africa *Alternaria* sp. (spotting); peaches from Georgia, U.S.A., *Sclerotinia americana*; and plums from Algeria showed *Rhizopus nigricans,* and from Spain, *Monilia cinerea.*

SECTION C is concerned with large scale storage and transport investigation.

SECTION D: FISH BY-PRODUCTS.—The nutritive value of various fish-meals was investigated on pigs. A comparison between sea bream meal (*Pagellus*

* Obtainable at Adastral House, Kingsway, W.C.2. Price 4s. net.

centrodontus), best white fish meal, and blood-meal and bone-flour, showed that pigs fed on sea bream meal grew faster, and the growth was at the expense of less food. Further, the animals were in better condition, and the curing tests showed that there was not the slightest trace of taint in the flesh. It is shown that the present valuation of fish-meal, which depends on the nitrogen and calcium phosphate content, is not a true criterion of feeding value, and feeding experiments are necessary for evaluation.

Gelatin.—In work on gelatin it is shown that the mode of preparation has a marked effect on its emulsifying power. Treatment with alkalis only is always more efficient, perhaps because the precursor undergoes molecular rearrangement during treatment with acid or alkalis, and the percentage of free amino-groups is greater from an alkali-treated precursor. Thus for electrically purified gelatins the percentages of free amino-nitrogen were:—From calf-skin, SO_2 treated, 3·50, NaOH treated, 4·12; ossein, NaOH, 4·12, HCl, 3·70; fish skin SO_2, 3·95, NaOH, 4·87; isinglass, 3·75.

The Sterols from the Muscular Tissue of Marine Animals.—The available data on the isolation and identification of sterols from various organisms indicates the presence of very large numbers of isomers of cholesterol of obscure origin, but of biological significance. If derived from plant sterols, the process is complicated. In the case of marine organisms and inferior forms of life the problem is specially complicated, partly owing to the presence of other complex alcohols and sometimes hydrocarbons. Dorée concluded that ordinary cholesterol is obtained from cold-blooded vertebrates and is present as such in invertebrates or as a substitute with similar properties. He showed many points of resemblance between the sterol from *Asterias rubens* and cholesterol, but also that they differed, and later the sterol of *Asterias auranticus* was found to contain a cholesterol isomer. In the present investigation the sterols from cod, skate, octopus, sepia, lobster, oyster, starfish, dogfish, and porpoise brains were extracted by Gardner and Fox's method (*Proc. Roy. Soc.*, 1921, (*B*), **92**, 358); in the case of lobster, oyster and starfish saponification with sodium ethylate was used. The different esters were prepared, fractionally recrystallised, and the properties of the various fractions from which the different isomers were obtained were compared.

It is concluded that two new sterols have been isolated from oysters and starfish, and that the sterol from the muscular tissue of *Octopus vulgaris* is probably different from true cholesterol.

Bleaching of Fish Oils has given satisfactory results on a commercial scale. All free fatty acids are removed by addition of a slight excess of caustic soda, and from the resulting oil emulsion the aqueous alkaline layer may be separated and used for production of cheap soap, whilst the emulsion and any separated oil is washed with boiling water until the wash water comes out colourless; the treatment is continued with a weak solution of sulphurous acid, and after a final water washing the oil is filtered.

Cod muscle protein.—The composition of cod muscle protein was as follows per 100 parts dry protein: Tryptophane 2·2; tyrosine, gravimetric 2·3, colorimetric 3·9; cystine (from S-content) 4·1; leucine 11·7; valine, 3·0; aspartic acid, 4·8; glutamic acid, 7·8; phenylalanine, 1·1 (part only); proline, —; lysine, 7·6; arginine, 11·5; histidine, 1·5; ammonia, 1·3; total, 60·5.

SECTION E is concerned with engineering practice.

SECTION F deals with researches for the Director by the Imperial College of Science and Technology in connection with fruit. In the factors affecting the

Origin.	Kahlenberg ($AsCl_3$).	Salkowski-Whitby's modification. (a)	Salkowski-Whitby's modification. (b)	Liebermann-Burchard.	Tschugajeff.	Whitby's Reaction C.
Cod, m.pt. 144°C.	Cherry red, mauve on boiling, muddy green on cooling	(1) Cherry red (2) Brown, with green fluorescence	Bright blue	Reddish purple	Colourless, pink on heating	Blue
Skate, m.pt. 144° C.	Purple, blue to green on boiling, muddy green on cooling	(1) do. (2) do.	do.	Reddish purple, with green fluorescence	do.	do.
Octopus, m.pt. 146° C.	Pink, remained so on boiling, blue-green on cooling.	(1) do. (2) do.	do.	do.	do.	Colourless
Sepia, m.pt. 147° C.	Pink, purple on boiling	(1) do. (2) do.	do.	Dark rose, with brown tinge	Yellowish pink, intensified on heating	Blue
Lobster, m.pt. 143° C.	Purple, blue-green on boiling	(1) do. (2) do.	do.	Reddish purple, green fluorescence	Colourless, pink on heating	do.
Oyster, m.pt. 115° C.	Reddish brown, dark muddy green on boiling	(1) Pale reddish yellow (2) Yellow red	Colourless	Dark reddish brown	Yellow green, intensified on heating	Greenish-yellow
Starfish, m.pt. 142° C.	Light brown, dark muddy-green on boiling	(1) Pale yellow (2) Yellow red	Pale yellow	Dark amber	Yellow green, intensified on heating, with blue fluorescence	Reddish-brown
Dog-fish brain, m.pt. 146° C.	Purple, blue-green on boiling, muddy-green on cooling	(1) Cherry red (2) Brown with green fluorescence	Bright blue	Reddish purple, green fluorescence	Colourless, pink on heating	Blue
Porpoise brain, m.pt. 146° C.	Cherry red, intensified on boiling	(1) do. (2) do.	do.	do.	do.	do.

internal resistance of apples to fungal disease it was found that: (*a*) Each individual apple in a sample has a characteristic resistance; (*b*) the rate of invasion in an apple of a given variety does not remain constant, but may rise or fall in time according to the variety used and the experimental conditions; (*c*) a comparison of *Fusarium* strains on various apples has shown that the virulence of the strains falls into an order which is independent of the variety of apple and the seasonal changes, but is affected by the temperature of storage. All the saltants examined exhibited a lower virulence than the original strains from which they were derived; (*d*) locality of origin influences resistance and differences can be correlated with differences in chemical composition; (*e*) resistance is complex, as *e.g.* low water content, high acidity and potash content and low nitrogen content are associated with high resistance and *vice-versa*.

SECTION G: INVESTIGATION OF SQUALENE.—Researches conducted by the University of Liverpool include an investigation into squalene. The formation of squalene can be regarded as the result of "head and tail" linking of 6 isoprene nuclei. It is probably synthesised in the living organism by aldol condensation of the aliphatic plant terpene aldehydes such as citral, citronellal or farnesal, followed by reduction in the animal liver. The formula of batyl alcohol is given as $C_{21}H_{44}O_3$; it is a saturated dihydric alcohol characterised by means of its phenylurethane and *p*-nitrobenzoate. The higher fatty alcohols found in many Elasmobranch fish are definitely related to those found in sperm-whale oil, for octadecyl, oleyl and cetyl alcohols, occurring in the latter, are present in the liver oils combined with glycerol, and selachyl and chimyl alcohols are also found. The occurrence of these glycerides in nature, forming a link between the fats and waxes, is of outstanding biological interest, and they are probably common constituents of other fish oils. They have no direct connection with vitamin *A*.

Cholesterol and Vitamin D.—The work on cholesterol and vitamin *D* has given fairly definite evidence that vitamin *D* is related to an absorption band with maximum at 247$\mu\mu$, and a detailed spectrographic examination of various cholesterol derivatives proves that selective absorption is only shown when at least 2 ethenoid linkages are present in the sterol molecule. The absorption spectrum of cholesterilene bears a close resemblance to that given by the pro-vitamin, and it is probable that of the 3 double bonds in ergosterol, 2 occupy the same positions as in cholesterilene. The absorption spectrum of cholesterione shows 2 bands at 312 and 242$\mu\mu$, the latter being very similar to the vitamin *D* band at 247$\mu\mu$. The passage of cholesterol to acholestenone is the fundamental reaction taking place whenever the former is decomposed by heat.

D. G. H.

Standard Method of Prussiate Analysis.*

IN the modern practice of buying chemical products on the analytical basis it is necessary that some particular and standardised method should be agreed upon between buyer and seller which is to be used in case of dispute. This has been done long since in most branches of the industry, but there are still some chemical products on which no general agreement has been reached.

* At a recent meeting of the prussiate makers of the world a standard method of analysis was agreed upon, and it was arranged that this standard method of analysis should be published in the various countries by the different workers. The method has been submitted to the ANALYST as one of the two journals for the English communication.—EDITOR.

In the prussiate industry a number of analytical methods, more or less accurate and convenient, are recognised. Some of these methods, while accurate for the estimation of pure ferrocyanides, are unreliable when used on the commercial product, because of the possible presence of certain impurities which render the method inaccurate; unless special precautions are taken to eliminate or render them inoperative.

Until quite recently no attempts have been made to judge of the merits of the various analytical methods for the estimation of ferrocyanides from the point of view of its use for the analysis of the commercial article and not of the chemically pure product.

For this purpose the method selected should be both accurate and rapid. It must not be influenced by any impurity likely to be present in the commercial article, and it should not require a very high order of skill or practice.

The various methods of ferrocyanide analysis in general use may be roughly classed under the following heads:

1. Conversion into hydrocyanic acid and estimation as sodium cyanide by standard silver nitrate solution.
2. Oxidation methods.
4. Titration with a salt of a heavy metal or other body capable of forming an insoluble ferrocyanide.

Under the first head the cyanogen of the ferrocyanide is distilled off as hydrocyanic acid either by the method of W. Feld (*J. Soc. Chem. Ind.*, 1903, 1068), in which the ferrocyanogen radicle is broken up by boiling with mercuric chloride and the cyanogen converted into mercuric cyanide, which is then distilled with an acid; or by the method of H. E. Williams (*J. Soc. Chem. Ind.*, 1912, 315), in which the ferrocyanide and acid is distilled with a small quantity of cuprous chloride, the latter acting catalytically in breaking up the ferrocyanide and converting the cyanogen contents into hydrocyanic acid.

The hydrocyanic acid evolved is absorbed into dilute caustic soda solution, and the resulting cyanide solution titrated with standard silver nitrate solution after the addition of a few drops of a 10 per cent. solution of potassium iodide.

Both these methods are accurate if performed with care, but the results are liable to be somewhat low, particularly the former, owing to the fact that slight losses may occur in the different operations, which are accumulative and appreciable when estimating material for 100 per cent. purity.

They have the advantage that the cyanogen content is finally titrated by a solution which may be accurately standardised by known and accepted methods, instead of by standardisation with what may or may not be pure ferrocyanide. On the other hand, while the cuprous chloride method is comparatively simple, both methods require considerable skill and practice, and are therefore not suitable for general analytical practice.

Under the second heading, titration of the ferrocyanide solution with a standard solution of potassium permanganate is the most general method, and the only one that need be considered here. The method is simple, the standard solution of permanganate is generally found in any analytical laboratory, and it is easy to standardise. Unfortunately, however, the method is liable to very considerable inaccuracy, owing to the possible presence in the ferrocyanide of oxidisable impurities, both organic and inorganic, which render the results obtained much higher than the truth. The oxidisable impurities likely to be present are sulphides, thiosulphates, thiocyanates, organic matter, and in the case of synthetic ferrocyanides, formates. This method, therefore, should not be used for the estimation of commercial ferrocyanides.

In the third class of methods, the ferrocyanide solution is exactly precipitated with a solution of a heavy metal salt which has been standardised under exactly similar conditions with a solution of a known weight of pure ferrocyanide. A number of metallic salts have been recommended for this purpose, including the soluble salts of copper, nickel, lead, iron, zinc, etc. Of these, only solutions of copper and zinc have come into general practice. When a solution of a copper salt is added to an acidified solution of the ferrocyanide, cupric ferrocyanides are precipitated which vary in composition according to the conditions of the precipitation and the amount and nature of the alkali salts present. Errors also arise if sulphides, thiosulphates, thiocyanates, etc., are present with the ferrocyanide.

These objections to the use of a solution of a copper salt for the titration of ferrocyanides, however, do not apply to the use of zinc sulphate. With this salt the composition of the zinc ferrocyanide is constant, and the impurities likely to be associated with the ferrocyanide have no effect on the composition of the precipitate or the amount of zinc used. Thus sulphides, thiosulphates, thiocyanates, or formates have no effect on the titration.

The main objection against these methods of titration is that the metallic salt solution is standardised against a ferrocyanide which may or may not be pure and dry. When zinc sulphate is used as the precipitating agent this objection has been overcome by the writer, who has found that the composition of the precipitate under the conditions of the titration when potassium ferrocyanide is used is constant and agrees with the formula $Zn_3K_2(Fe(CN)_6)_2$. The zinc sulphate solution, therefore, may be made up, its zinc contents estimated, and the precipitating equivalent of potassium ferrocyanide for each c.c. calculated, or the solution adjusted until 1 c.c. will precipitate 0·01 grms. or 0·05 grms. as desired, of potassium ferrocyanide, according to the equation:

$$3ZnSO_4 + 2K_4Fe(CN)_6 = Zn_3K_2(Fe(CN)_6)_2 + 3K_2SO_4.$$

The standardisation of the solution is thus independent of a ferrocyanide.

When sodium ferrocyanide is precipitated by a soluble zinc salt, the composition of the precipitate closely approximates to the formula: $Zn_3Na_2(Fe(CN)_6)_2$. But if potassium chloride solution is added (20 c.c. of 20 per cent. solution) before titration, a precipitate of constant composition is obtained, agreeing with the formula: $Zn_3K_2(Fe(CN)_6)_2$, so that either potassium or sodium ferrocyanide may be titrated with equal accuracy, provided that in the case of the latter salt a solution of potassium chloride or sulphate is added before titration.

For the analysis of commercially pure sodium ferrocyanide, the zinc sulphate solution may, if preferred, be standardised with chemically pure sodium ferrocyanide.

The determination of the impurities of a ferrocyanide is sometimes a question of importance, and the usual laboratory methods cannot be applied without a preliminary treatment. The impurities that may possibly be met with in ferrocyanide are chloride, sulphate, thiosulphate, thiocyanate, formate, carbonate, cyanide, mechanical impurities in the form of insoluble dirt and dust, etc.

Below is given an outline of methods for the estimation of the most probable and important impurities of ferrocyanides worked out by the writer.

Take 50 grms. of the ferrocyanide, dissolve in about 250–300 c.c. of distilled water and precipitate with a slight excess of pure recrystallised zinc acetate, dilute to 1000 c.c., shake well and filter.

Determination of Chloride.—Take 100 c.c. of the clear filtrate in a 300 c.c. beaker, add a few drops of potassium chromate solution, and titrate with $N/10$

or $N/100$ silver nitrate solution in the usual manner. The end-point is sharp and distinct.

Determination of Sulphate.—Acidify 100 c.c. of the clear filtrate with pure hydrochloric acid, boil, and then add an excess of boiling barium chloride solution, filter, ignite, and weigh as barium sulphate.

Determination of Formate.—Take 100 c.c. of the clear filtrate, add an excess of mercuric chloride solution together with a few drops of acetic acid, boil for half-an-hour, filter on a tared filter paper, wash, dry and weigh.

$$\text{Weight of HgCl} \times 0{\cdot}178 \times 10 \times 2 = \text{KHCO}_2 \text{ per cent.}$$
$$\text{Weight of HgCl} \times 0{\cdot}144 \times 10 \times 2 = \text{NaHCO}_2 \text{ per cent.}$$

At a recent conference of the chemists of the European Prussiate Manufacturers, the various methods of ferrocyanic estimation were critically examined, and the zinc sulphate titration method finally accepted by them as the standard method of ferrocyanide estimation for the settlement of disputes between manufacturer and customer.

A translation of the findings and the method as agreed to by the chemists of the European Prussiate manufacturers is as follows:

1. It was generally agreed that the permanganate method could not be used, as the presence of oxidisable impurities would give false results.
2. The method for the determination of ferrocyanide by distilling as hydrocyanic acid was agreed by all to be good. Doubts were raised against this method, however, as lower results were frequently obtained by it than by other methods, and it did not seem practicable to accept it as a general standard method which was to be made available for public laboratories. The method in which mercuric chloride was used was preferred to that in which cuprous chloride was used.
3. The copper sulphate method was generally condemned, as it had been found that the results were not reliable, variable compounds being readily formed.
4. The copper sulphate method of converting the ferrocyanides into ferricyanides was agreed to be good so long as no sulphur compounds were present. Sulphur compounds caused the precipitation of copper, so that for this reason the method was unreliable under certain conditions.
5. The zinc sulphate method was agreed by all to be the best method.

Method of Procedure.

Strength of Solutions.

Zinc Sulphate Solution.—About 1/5th normal solution (28·7 grms. to a litre) is standardised against Kahlbaum's potassium ferrocyanide and the factor thus determined.

Ferrocyanide Solution.—Ten grms. of the ferrocyanide are dissolved in water, and the solution diluted to 500 c.c. Fifty c.c. of this solution are taken for analysis. They are diluted with water, and acidified slightly with dilute sulphuric acid (about 10 c.c. of 1/10th normal pure (iron free) sulphuric acid). A 15 per cent. solution of pure iron alum is used as indicator. Titration is carried out at a temperature of 15–20° C. The paper used for the determination of the end of the titration must be iron-free and, in particular, as ash-free as possible. The

titration is carried out by adding the zinc sulphate solution to the acidified potassium ferrocyanide solution. The end-point is determined by placing 2–3 drops of the titrated solution on a filter paper by means of a glass rod and leaving the paper for a few minutes to let the drops spread well. Then one or more drops of the indicator solution are placed on the paper at a distance of 1–1½ cm. The end of the titration is shown when no trace of the blue colour appears at the junction of the two liquids. Special care must be taken to see whether any blue colour appears about 2–3 minutes after the two liquids come into contact. It is best, when placing the drops on the paper, to make a dent in it with the glass rod, as by this means the precipitate is kept back more easily and not carried to the outside edge. It is necessary to do this, for a blue colour will always appear when the iron salt comes into contact with the precipitated zinc ferrocyanide.

If such a quantity of the liquid to be titrated is taken for the drop tests that it considerably influences the final result, the titration must be repeated, and the zinc sulphate solution allowed to flow into the liquid to be titrated until just before the termination with a minimum of drop reaction tests, *i.e.* with a minimum loss of solution. It may be stated here that a drop taken on a thin glass rod represents about 1/10th c.c. of the titration solution.

MOISTURE.—A determination of the moisture contents should follow. This is determined by placing a weighed quantity in a drying oven at a temperature of 125° C. to drive off the moisture and water of crystallisation, until constant weight is obtained. The moisture content is calculated by subtracting the calculated water of crystallisation from the difference between the original weight taken and the dry weight.

SAMPLING.—As a special method of sampling it is recommended that a sample should be taken from each cask, so that for each ton of goods a kilo. sample is taken. This sample is to be divided into four containers and sealed. The sampling is to be done by a sworn sampler from the Chamber of Commerce or a similar authority.

Parliamentary Notes.

EXPIRING LAWS CONTINUANCE ACT, 1928.

THIS Act continues—"So far as it authorises the making or revoking in whole or in part, of Part III of the Sale of Food Order, 1921, and provides for the enforcement and imposes penalties for the breach thereof."*

* Part III, Sale of Food Order, 1921, deals with the labelling of certain imported produce.—EDITOR.

ABSTRACTS OF PAPERS PUBLISHED IN OTHER JOURNALS.

Food and Drugs Analysis.

New Carbohydrate in Rye Flour and Detection of Rye Flour in Wheat and other Flours. J. Tillmans. (*Z. Unters. Lebensm.*, 1928, **56**, 26–32.)—Trifructosan or trifructose anhydride, $C_{18}H_{30}O_{15}$, is a new carbohydrate isolated from rye flour, and may be identical with the secalose of Schulze and Frankfurt (*Ber.*, 1894, **27**, 626, 3525). When pure, it is a white, crystalline, slightly sweet powder, soluble in dilute acids, but insoluble in strong alcohol, optical rotation at 20° C., –43·93° (–92·70° after inversion). Since it is found only in rye flour, the presence of 10 per cent. or more in wheat or other flours may be detected. The sample (5 grms.) is centrifuged with 20 c.c. of 70 per cent. alcohol for 15 minutes, the mixture maintained at –3° C. for 10 minutes, then well stirred, and again centrifuged for.5 minutes. The extract is decanted, filtered clear if necessary, and 10 c.c. added to 0·5 c.c. of a *N* solution of sodium hydroxide in 70 per cent. alcohol. Pure wheat flour yields, at the most, a white turbidity, but if rye flour is present, a distinct precipitate of the sodium salt of trifructosan is produced. J. G.

Determination of Reducing Sugars, especially Dextrose, in Presence of Hydrocyanic Acid by means of Alkaline Copper Solutions. H. Hérissey and A. Chalmeta. (*J. Pharm. Chim.*, 1928, **8**, 393–406.)—When applied to the determination of dextrose in presence of hydrocyanic acid, Bertrand's method gives low results, the precipitation of cuprous oxide being lessened or, sometimes, prevented entirely owing (1) to the interaction of the sugar and hydrocyanic acid, according to Kiliani's reaction, (2) to precipitation of cuprous cyanide, and (3) to solution of the cuprous oxide by the hydrocyanic acid. On the other hand, sensibly exact results are obtained by a volumetric method in which the end of the reaction is judged by the decolorisation of the alkaline copper solution. Hydrocyanic acid may be eliminated from pure sugar solutions by evaporating the liquid to dryness. When the products of the enzymic hydrolysis of amygdalin are to be tested, the effect of the hydrocyanic acid may be overcome by evaporating 20 c.c. of the solution to 10 c.c. on a boiling water-bath, shaking with 1 c.c. of lead acetate solution and 2 c.c. of saturated sodium sulphate solution, making up to 30 c.c. with water, again shaking and filtering. The hydrocyanic acid may be eliminated also by subjecting the solution to the prolonged action of a current of air, or by precipitating with silver nitrate, excess of this being removed by precipitation with sodium chloride. T. H. P.

Detection and Determination of Sucrose by the Ammonium Molybdate Method. N. W. Matthews. (*Chemist Analyst*, 1928, **17**, 8.)—Solutions of sucrose of the order of 1 in 1000 to 1 in 40,000 may be determined by the addition to 5 c.c. of sample of 3 drops of concentrated hydrochloric acid and 3 c.c. of a 4 per cent.

solution of ammonium molybdate. The mixture is heated in boiling water for 6 minutes, and the blue colour produced is matched against that of a suitable standard prepared under the same conditions. The standards, which are not permanent, may be substituted by Fehling's solution or blue-black ink, suitably diluted. J. G.

Iodine Value of Spanish Paprika Oil. L. C. Mitchell and S. Alfend. (*J. Assoc. Off. Agric. Chem.*, 1928, **11**, 523–527.)—Eleven authentic samples of paprika pods, grown in various districts in Spain in 1927, were separated into shells, seeds, and placentae, these being dried and, as is the commercial practice, mixed in varying proportions according to the grade or quality required, and ground. The procedure adopted in the extraction of the oil and in the determination of its iodine value was as follows: To 10 grms. of the ground sample, in a 200 c.c. stoppered flask, were added 100 c.c. of chloroform, the flask being rotated during the introduction of the first 50 c.c. After being left for an hour, the flask was shaken and the contents filtered through a 12·5 cm. fluted filter. From 10 c.c. of the filtrate, the solvent was evaporated in a weighed crystallising dish, 50 × 35 mm., which was afterwards kept at 100° C. for an hour, cooled in the air, and weighed. Another 10 c.c. portion was treated, in a suitable glass-stoppered flask or bottle, with 30 c.c. of Hanus solution, and the iodine value determined by the official method of the Assoc. Off. Agric. Chem. (*Methods of Analysis*, 1925, p. 287).

The limiting (and average) numbers obtained for the iodine values are: For mixtures containing shells (53·9–60·8 per cent.), seeds (34·6–41·9 per cent.), and placentae (4·0–8·8 per cent.) in their natural proportions, 134·0–138·9 (136·5); for mixtures containing 70 per cent. of shells and 30 per cent. of seeds and placentae, 133·6–139·7 (136·5); and for mixtures containing 45 per cent. of shells and 55 per cent. of seeds and placentae, 133·0–136·4 (134·5). Thus, the lowest iodine values were obtained in the series containing the largest proportion of seeds and, therefore, of oil. The range of iodine values specified by the official U.S. Standards, namely, 125–136, is not applicable to the results obtained by the chloroform extraction method. T. H. P.

Crab Liver Oil. M. Tsujimoto. (*J. Soc. Chem. Ind., Japan*), 1928, **31**, 279B.)—The liver oil of the Japanese crab, "Tarabakani" (*Paralithodes Camtsehatica*, Tilesius), is a dark brown liquid with a characteristic and unpleasant smell. The colour reaction with sulphuric acid is slightly greenish dirty brown. On saponification a brownish-red precipitate is formed which contains nitrogen and sulphur. The sample examined had the following constants:—Sp. gr. at 15°/4°, 0·9456; acid value, 97·9; saponification value, 155·6; iodine value, 163·6; n_D^{25}, about 1·475, and unsaponifiable matter, 6·39 per cent. *Fatty acids.*—M.pt., below 15° C.; neutralisation value, 185·5; iodine value, 183·3; bromides insoluble in petroleum spirit, 116·7 per cent.; insoluble in ether, 56·7 per cent.; bromine content of bromides, 71·5 per cent.; liquid fatty acids, 85·6 per cent.; unsaturated acids, 33·6 per cent.

The iodine value of the unsaturated acids was 320·9. The fatty acids consist

mainly of C_{18}, C_{20} and C_{22} acids. The unsaponifiable matter is viscous and orange in colour, dissolving readily in methyl alcohol (hydrocarbons absent). With acetic anhydride and sulphuric acid the colour is violet red, changing to bluish and finally dark green. With antimony chloride in chloroform there is no remarkable coloration, indicating the possible absence of vitamin. The unsaponifiable matter contains 22·67 per cent. of cholesterol, also batyl, selachyl and possibly chimyl alcohols. The authors also report the presence of a new unsaturated liquid alcohol with a formula probably of $C_{11}H_{20}O_2$, of which the acetyl derivative distilled below 200° (5 mm.); after hydrogenation the product ($C_{11}H_{22}O_2$) remains liquid at 0°. R. F. I.

Purity of Ether for Analytical Use. G. Middleton. (*Quart. J. Pharm.*, 1928, **1**, 319–326.)—Commercial ether may contain impurities, and the most reactive of these is ethoxyethyl hydrogen peroxide, although other substances, such as aldehyde or alcohol, also exert deleterious effects. Acetaldehyde and the organic peroxide are found in ether stored under unfavourable conditions, especially if the ether is initially impure, and numerous examples are cited in ordinary analytical procedure where the ether has introduced a considerable factor of error. If old ether is used for extraction of a substance subsequently recovered by evaporation of the ether, the residue attains constant weight very slowly, the final weight being higher than if pure ether had been used, and the residue is chemically altered, as shown by a difference in appearance, solubility, iodine value and titration. D. G. H.

Determination of Iodine in Organic Combinations, especially in Thyroid Gland. W. Smith. (*Quart. J. Pharm.*, 1928, **1**, 372–377.)—Many methods for determining iodine in organic combination are criticised and the following adopted. If the iodine content of the material to be analysed is fairly high (about 0·2 per cent.) Hunter's method is used (*J. Biol. Chem.*, 1910, **1**, 321), whereby the substance is fused with sodium and potassium carbonates and potassium nitrate, the melt dissolved in water, and the iodide oxidised to iodate in phosphoric acid solution by a slight excess of sodium hypochlorite solution. Excess chlorine is boiled off, the removal being controlled by starch iodide paper until no reaction occurs, and boiling is then continued for 15 minutes. Potassium iodide is then added, and the iodine titrated with 0·005 *N* thiosulphate solution. This method, very slightly modified, is that of the U.S.P. (X.) for the assay of iodine in thyroid gland. For small quantities of iodine a modification of Kendall's method is used, whereby 1 grm. of material is gently heated with powdered sodium hydroxide in a nickel crucible, practically covered, and subsequently the heating increased until the organic matter is oxidised. After being extracted with water and filtered through cotton wool, the mixture is neutralised with syrupy phosphoric acid (bromphenol blue as indicator), excess bromine added, followed by excess of 2 c.c. phosphoric acid. After boiling to half the volume the remaining traces of bromine are eliminated by addition of salicylic acid, and the iodine is titrated with 0·005 *N* thiosulphate solution. D. G. H.

Determination of Camphor in Pharmaceutical Preparations. J. Bougault and Bl. Leroy. (*Ann. Falsific.*, 1928, **21**, 456–460.)—To 0·5 grm. of camphor, dissolved in 5 c.c. of 90 per cent. alcohol, are added 1 grm. of hydroxylamine hydrochloride in 5 c.c. of water and 2 c.c. of 20 per cent. sodium hydroxide solution, and the tube sealed and placed for 2 hours in a boiling water bath. After cooling, the contents of the tube are washed out with 10 times diluted sodium hydroxide, and 20 c.c. of water added, when a cloudiness forms, due to precipitation of part of the camphoroxime. The cloudiness is redissolved by the addition of 3 c.c. of sodium hydroxide solution, but if it still persists is due to camphene or borneol, which are filtered off. The liquid is exactly neutralised with hydrochloric acid, and the camphoroxime removed by 20 c.c. of ether; the ethereal solution is washed with 5 c.c. of water and decanted into a weighed dish of 7 cm. diameter, and the aqueous solution again extracted with three further portions of ether. The solution is left to evaporate in the air for 12 hours, and the residue dried for 12 hours over calcium chloride and weighed, 4 per cent. being added to the result to compensate for evaporation of camphoroxime. Then the weight of camphor in the sample is given by multiplying the result by 152 and dividing by 167, the molecular weights, respectively, of camphor and camphoroxime. This method may be applied directly to the determination of camphors in tinctures; for camphorated oils the camphor must be separated from the oil, by distilling slowly for 2 hours a mixture of 50 c.c. of water to 20 grms. of camphorated oil, with fragments of pumice in the presence of 30 c.c. of 95 per cent. alcohol added from a dropping funnel, and the distillate is made up to 100 c.c. Synthetic camphor may contain camphene, borneol or isofenchone, but that produced in France rivals the best Japanese camphor in purity. D. G. H.

Compound Tincture of Benzoin. T. T. Cocking. (*Quart. J. Pharm.*, 1928, **1**, 337–346.)—The complete analysis or evaluation of compound tincture of benzoin requires the determination of total solids, acid, ester, and saponification values, and free, combined, and total balsamic acids. The total solids are found by drying *in vacuo* over sulphuric acid at laboratory temperature. For the acid value 20 c.c. of the tincture are diluted with 50 c.c. of neutral alcohol and titrated with *N* alcoholic potash, and the ester determined in the neutralised liquid. Free balsamic acids are determined in 20 c.c. of tincture in which 2 grms. of light magnesium oxide have been diffused, 100 c.c. of water and 20 c.c. of xylene added, and the whole boiled for 1 hour. After filtering, the separated liquid is boiled again with 100 c.c. of water, separated, and after a third boiling, the combined liquids are washed once with ether. The balsamic acids are then liberated by hydrochloric acid, extracted with ether, which is evaporated, and, after drying, weighed. The total balsamic acids are found by saponification, evaporation, solution of the residue in water, acidification, addition of magnesia and xylene, separation of the aqueous liquid after boiling as before, and weighing of the acids. Tables are given showing the maximum variations in compound tinctures of benzoin, together with figures for commercial samples. D. G. H.

Microchemical Reactions of Homatropine. M. Wagenaar. (*Pharm. Weekblad*, 1928, **65**, 1213–1216.)—Homatropine, $C_{16}H_{21}NO_3$, a very strong base, crystallises in colourless, hygroscopic, *d*-rotatory prisms (m.pt. 98° C., refractive index 1·56–1·62), which are insoluble in water, easily soluble in alcohol or chloroform, and partly soluble in ether or benzene. Solutions of the strengths indicated give characteristic crystalline precipitates, which may be recrystallised from alcohol, with solutions of gold chloride, picrolonic and picric acids (1 : 250), 0·1 *N* iodine solution (1 : 500), and with Eder's reagent (a solution of 1 part of bromine, and 2 parts of potassium bromide in 20 parts of water) (1 : 1000). The iodine and Eder reagents are unaffected by mineral acids and yield feathery and block-shaped crystals, respectively. J. G.

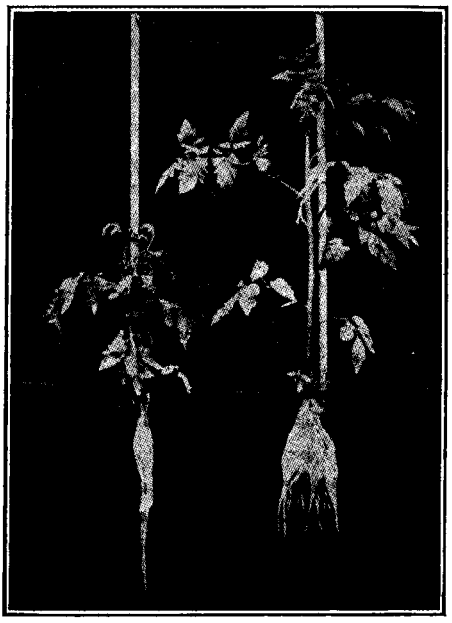
Formula for Calculating Composition of Mixtures of Mydriatic Alkaloids. J. C. Munch and G. S. Gittinger. (*J. Assoc. Off. Agric. Chem.*, 1928, **11**, 521–523.)—Examination of a number of solutions containing atropine mixed with either hyoscyamine, or cocaine, or homatropine, shows that the resultant physiological effect of each such solution is equal to the summation of the separate physiological effects of the two constituents. If, therefore, the concentration necessary to cause just perceptible mydriosis in the cat's eye (threshold value) is known for each of the two alkaloids present and for any given solution containing both, a simple calculation gives the proportions of the two in this solution. T. H. P.

Biochemical.

Note on Quantitative Methods of Measurement of the Nutritive Value of Proteins. H. H. Mitchell. (*Biochem. J.*, 1928, **22**, 1323–1326.)—In a study of the nutritive value of the protein tuberin, Kon (*Biochem. J.*, 1928, **22**, 261) has compared the method of the author (*J. Biol. Chem.*, 1924, **58**, 873) with that devised by Osborne, Mendel and Ferry (*J. Biol. Chem.*, 1919, **37**, 223). The author takes exception to Kon's statement that : " A specific error seems to be inherent in the method . . . namely, the tendency to give higher biological values in the periods immediately following the standardising nitrogen-free or low-nitrogen periods . . .," and to his explanation of the differences in the nutritive ratings of proteins as obtained by the two methods. In the first method, the nutritive value of the protein is taken as the total nitrogen retention of the experimental animal in percentage of the total absorbed nitrogen ; this percentage is called the " biological value " of the protein, a term introduced by Thomas (*Arch. anat. Physiol.*, 1909, 219) ; in the second method the nutritive value of the protein is measured by the gain in body-weight of the experimental animal per grm. of protein consumed. With regard to the first point at issue, the author states that the question is a statistical one, and cannot be settled by citing isolated instances in which experimental data *appear* to indicate an increased biological value following nitrogen underfeeding. He gives a table which summarises all published data on the effect of a period of low-nitrogen feeding on the biological value of protein obtained in an

immediately following period, and states that as the method is applied in the author's laboratory, there seems no reason to believe that a period of low-nitrogen feeding exerts any but anything inappreciable effect upon the utilisation of protein in a subsequent period. Regarding the differences in nutritive ratings of proteins, he states that the biological value of a protein measures that fraction of the absorbed protein nitrogen that is being used in all of the anabolic reactions of the body, whilst the other method takes notice only of that fraction of protein intake that is being used for growth, so that this latter method of protein value is subject to variation caused by variable food intakes, *i.e.* there is no reference to size of animals used, or to amounts of food consumed. " It is a hazardous undertaking to compare gains per grm. of protein consumed obtained in different experiments without reference to the size of animals employed, or to the amounts of food consumed, as Kon and others before him have done." Differences in sex of rats and digestibility of proteins also affect the results of the numerical method of Osborne, Mendel and Ferry. The author concludes with the statement that the methods are not equivalent, and the results obtained need not parallel each other.

P. H. P.



(*a*) (*b*)

(*a*) Tomato plant grown without boric acid.
(*b*) ,, ,, ,, with boric acid.

Importance of Boron in Plant Growth. E. S. Johnston. (*J. Chem. Educ.*, 1928, **5**, 1235–1242.)—The addition of 0·55 parts per million of boron compounds, expressed as boron, to the nutrient solution in which tomato plants were being grown restored growth, which had been very feeble, to a normal state, and

it was found that the addition of similar quantities of boron to potato plants grown without it greatly improved top growth and doubled the weight of tubers. It was found that potato plants grown in new glazed earthenware jars were able for some time to obtain sufficient boron from the glaze, and similarly tap water proved a sufficient source of supply for tobacco plants. Analytical data of tomato plants grown with and without boron, as percentages of total dry matter per stem and leaves are as follows :—

	Leaves.		Stems.	
Amount of boron added to the nutrient solution (p.p.m.) ..	0·00	0·55	0·00	0·55
Starch	12·03	8·41	5·42	1·45
Reducing sugars (hexoses) ..	8·18	3·83	5·32	8·70
Sucrose	3·31	1·34	3·13	2·89
Total sugars	11·50	5·16	8·45	11·59

A deficiency of boron affects the growing point (and thus the general growth direction) and meristematic tissues, and the conducting system breaks down. This is confirmed by both microscopical and chemical examination. Starch and total sugars are more abundant in leaves of boron-deficient plants than in normal plants, owing to the plants being able to manufacture these substances, but not to distribute them. In boron-deficient potato plants the leaves became thick and rolled, as in potato leaf-roll disease, and starch was abundantly present, as when phloem necrosis occurs. Addition of as little as 0·5 p.p.m. to the nutrient solution corrects deficiencies, but a concentration of only 5 p.p.m. is extremely toxic. Boron should be added to the list of elements necessary for plant growth.

D. G. H.

Valuation of Insecticides. C. H. Peet. (*Ind. Eng. Chem.*, 1928, **20**, 1104–1165.)—Many variables have to be considered in testing insecticides biologically ; of these, temperature, time, concentration, and humidity may be controlled, but other variables, such as degree of agitation of the air in the chamber and condition of the insect, are more difficult to regulate. For use in experimental work, the author breeds flies (*Musca domestica*) in quantity ; although less robust than wild flies, they are of fairly uniform strength, and the age of each batch is, of course, known definitely. Repeated tests have to be made before any definite conclusion as to the efficiency of an insecticide is obtained, and the immediate result of a test is not necessarily the final one. Certain compounds have almost no effect when first applied, but produce high mortality after a longer period of contact ; others produce almost instantaneous narcosis from which the insects recover completely after some time.

W. P. S.

Use of Piperazine in the Analysis of Urine and Blood. R. Gros. (*J. Pharm. Chim.*, 1928, **120**, 313–316.)—In analysing urines for xanthic bodies and uric acid by the Haycraft-Denigès method 100 c.c. of the well mixed sample are made up to 110 c.c. with an aqueous 10 per cent. solution of piperazine and shaken till clear, and 25 c.c. of Deniges' solution A added. After filtration through a folded filter 108 c.c. are collected, and the original procedure followed. The

addition of the piperazine introduced no source of error, and gave a clear solution where an abundant deposit of uric acid or urates was originally present. In the determination of uric acid in blood by Grigaut's method (modification of the methods of Folin and Wu) the solution of the uric acid in the presence of the mixture of mono and disodium phosphates, may be rapidly brought about by the addition of 10 c.c. of an aqueous 10 per cent. solution of the piperazine. D. G. H.

Glucose in Normal Urine. A. Hassan. (*Biochem. J.*, 1928, **22**, 1332–1340.)—A trustworthy method is described for the preparation of the osazones of the urine sugars. The urine is treated with adsorbent charcoal before application of the phenylhydrazine test; in this way the substances in urine which interfere with the crystallisation and identification of the osazones are removed on the charcoal. With this method glucosazone is obtained in aqueous solutions of glucose, and in solutions of glucose in urine from which the normal sugars have been removed, in concentrations as low as 1 mgrm. per 100 c.c. In suitable cases glucose added to ordinary urines can be detected when the amount added is as low as 2·5 mgrms. per 100 c.c. Normal urines give numerous types of crystal mixtures, formed of a mixture of 2 simple osazones; one of these is identical with glucosazone, and the other appears to correspond with the *iso*-maltosazone of Baisch (*Z. physiol. Chem.*, 1895, **20**, 249), as regards melting point, but differs markedly in crystalline appearance. The urines of over 700 Egyptians have been examined under different conditions, and it has been found that typical glucosazone crystals are given by 20 to 30 per cent. of the urines voided 1 to 2 hours after an ordinary meal; the percentage drops to 12 to 15 in urines passed 4 to 5 hours after meals, and to about 7 per cent. after a 12 hours' fast. An examination of the tolerance of 28 students to 50 grms. glucose has shown that, in most cases, this is not due to an abnormal carbohydrate metabolism. The excretion of glucose after 50 grms. have been taken seems to be less than that which follows an ordinary mixed meal. It is shown that some of the " physiological osazones " of Höst (*J. Metab. Res.*, 1923, **4**, 315) are impure crystal mixtures, and possibly the same applies to some of the osazones of Geelmuyden (*Norsk. Mag. Laegevidenskaben*, 1915, **13**, 985). The results presented leave little doubt that the old view that glucose in small quantities is a constituent of normal urine is confirmed. Photographs of some of the crystalline forms obtained of the osazones are reproduced. P. H. P.

Colorimetric Method for Determination of Lipoidal Phosphorus in Blood. S. L. Leiboff. (*J. Biol. Chem.*, 1928, **80**, 211–214.)—A method is described for the determination of lipoidal phosphorus in small amounts of blood. The lipoidal material is extracted from oxalated blood with the alcohol-ether mixture of Bloor (*J. Biol. Chem.*, 1918, **36**, 33), which was found to give the best results out of the various solvents tried. The alcohol-ether is then evaporated on the water bath, and the organic matter is destroyed with concentrated sulphuric acid and hydrogen peroxide. The excess acid is neutralised with ammonia and the liquid reacidified with acetic acid. The phosphate is then precipitated with uranium acetate and

determined colorimetrically by a procedure recently described by Leiboff (*J. Biol. Chem.*, 1928, **79**, 611; ANALYST, 1928, **53**, 663). Sulphuric acid followed by hydrogen peroxide was found to be more satisfactory for the digestion of lipoidal material than the sulphuric-nitric acid mixture used by some other workers. Although 0·1 c.c. of concentrated sulphuric acid destroys completely all the lipoidal extract from 0·5 c.c. of blood, yet 0·3 c.c. is used because it shortens the heating time, and more completely covers the sides of the tube (where lipoidal matter has been left during evaporation of the alcohol-ether mixture) during rotation. No superheating and no loss of phosphorus occur in this method. Some details of the method, and the preparation of reagents are omitted, as they have been described in the previous publication. It is well known that the phosphorus content of an alcohol-ether extract is not a true measure of the lipoidal content, since other substances containing phosphorus, not lipoid, are extracted along with the lipoids, but this is the only means available, since isolation of lipoids in a pure state is a matter of great difficulty, and impossible in small amounts of tissue.

P. H. P.

Note on Volatile Sulphide from Muscle. W. A. Osborne. (*Biochem. J.*, 1928, **22**, 1312.)—When the leg muscles of a well-nourished, recently killed guinea-pig were cut into small pieces and boiled immediately in water in a distillation flask, the delivery tube of which dipped into lead acetate solution, no volatile sulphide could be detected; if, however, about 24 hours had elapsed between the killing of the animal and the removal of the muscles, the presence of sulphide in the distillate was obvious, even when various procedures were adopted to remove the possibility of bacterial decomposition. Apparently, therefore, autolysis makes a change in the muscle proteins whereby loosely bound sulphur is produced or liberated. Sulphide was readily detectable in muscle from a recently killed guinea-pig which had been starved for 48 hours; the flesh of sheep in poor condition also emitted sulphide readily on boiling, but the same was not found with beef. Probably the muscle of different animals varies considerably in this reaction. P. H. P.

Determination of Carnosine. W. M. Clifford and V. H. Mottram. (*Biochem. J.*, 1928, **22**, 1246–1252.)—The method of Clifford (*Biochem. J.*, 1921, **15**, 400; ANALYST, 1921, **46**, 507) for the determination of carnosine, based on the colour produced when carnosine reacts with diazobenzene-sulphonic acid, and the application of the method to the chromogenic substances in muscle (*Biochem. J.*, 1921, **15**, 725; 1922, **16**, 341; 1922, **16**, 792; 1923, **17**, 549; ANALYST, 1922, **47**, 266; 1922, **47**, 443; 1923, **48**, 184) have been severely criticised by Hunter (*Biochem. J.*, 1921, **15**, 689; 1922, **16**, 640; 1924, **18**, 408; ANALYST, 1922, **47**, 266; 1923, **48**, 34) and by Mitsuda (*Biochem. J.*, 1923, **17**, 630). The method has therefore been reinvestigated, and the experimental work has been shared, so that one author has handed over clear, colourless solutions to the other for determination, the source, previous history and carnosine content of which were quite unknown to the receiver. Results show that the original carnosine upon which the work was based was chemically pure, and not, as has been suggested,

only 43 per cent. pure, the stock solutions of dyes once standardised were satisfactory, as they keep over a number of years, and that the method of determination has a high order of accuracy ; by this method the carnosine content of a solution of pure carnosine in water can be determined with an error of less than 1 per cent. With the use of this method to determine the chromogenic substances in samples of muscle, it has been shown that they vary in amount directly as the amount of muscle taken, no matter what skeletal muscle is used, nor if it is from a different animal of the same species, *i.e.* for the skeletal muscle of a species the carnosine content is a constant. This constant varies from species to species, but not from member to member of that species. A series of determinations of the chromogenic substances has been made on 13 different muscles or groups of muscle in the ox, and 9 different muscles and 4 groups of muscle in the cat. This work confirms the previous observations of Clifford on the muscles of rats, rabbits, calves, sheep and lambs. Hunter and Mitsuda, however, claim to have found differences in carnosine content between muscle and muscle and cat and cat. P. H. P.

Determination of Silica in Tissues. E. J. King. (*J. Biol. Chem.*, 1928, **80**, 25–31.)—A micro method is described for the determination of silica in animal tissues, which depends on the intense yellow colour of silicomolybdic acid, produced when a silicate solution is treated with ammonium molybdate and sulphuric acid. The method is quick and easily applied, and the proportionality of colour produced in test solutions prepared from sodium silicate and from ashed tissue holds over a wide range of concentration. The yellow colour is of the same tint as that of a dilute solution of picric acid, and thus an artificial standard is possible ; it is also preferable, since a sodium silicate standard tends to deposit silica, and is difficult to standardise. The picric acid standard remains unchanged for at least 2 months, and can be made from any good brand of C.P. picric acid without recrystallisation. Where the phosphorus content is low, the silica present may be determined fairly reliably without precipitation of the phosphate, but for an accurate determination the removal of phosphate is necessary, and is accomplished by the addition of magnesia mixture to the solution of the ash, and the filtering off of the magnesium ammonium phosphate. It is shown that the silica is not partly precipitated with the phosphate, but is quantitatively determined by this method. An amount of tissue 0·2 to 1·0 grm. is a convenient quantity for a determination, and Hehner tubes, or graduated cylinders with stop-cocks near the bottom, mounted over white cardboard, form an inexpensive colorimeter. A table shows the silica content of some tissues which have been determined. P. H. P.

Irradiation of Ergosterol. T. A. Webster and R. B. Bourdillon. (*Biochem. J.*, 1928, **22**, 1223–1230.)—The work recorded by Rosenheim and Webster (*Lancet*, 1927, **213**, 622 ; ANALYST, 1927, **52**, 652) has been continued in search of a possible means of isolation of vitamin *D*. The effect was studied of the use of " filtered " light (obtained by means of an alcoholic cobalt chloride filter) at various temperatures, in an attempt to produce more concentrated preparations of vitamin *D*. It seems reasonable to conclude from biological tests of the antirachitic activity

that the exclusion of wave-lengths shorter than 265$\mu\mu$ (which were cut out by the filter) does not seriously alter the ratio of rates of production and destruction of vitamin *D*. This suggests, but does not prove, that vitamin *D* either has strong absorption for wave-lengths longer than 265$\mu\mu$, or has not great absorption for wave-lengths of 230 to 250$\mu\mu$, as suggested by previous workers. From the lack of marked effect of changes of temperature between +78° C. and – 18° C. on the equilibrium, the temperature coefficients of the changes which cause destruction and production are not widely different, and the moderate effect of lowering the temperature to –180° C. suggests that the temperature coefficient of both reactions is very small ; hence both reactions are directly photochemical in nature. After the removal of unchanged ergosterol (as ergosterol digitonide) from solutions irradiated for short periods only, the products obtained formed a transparent, glassy hard solid of indefinite m.pt. beginning about 30° C., at times colourless, but often contaminated with a yellow pigment. In contrast to the small (0·2 per cent.) solubility of ergosterol in alcohol, the product is soluble in its own weight of alcohol at 30° C. It shows high antirachitic activity, which varies considerably in different samples, as does the absorption spectrum ; the presence of traces of yellow pigment showed strong absorption between 300 and 400$\mu\mu$. From numerous attempts to find a quantitative relation between the magnitude of the absorption coefficients of the products described and their antirachitic activity, the following conclusions are reached :—If a 0·1 per cent. solution of ergosterol in alcohol or ether is irradiated at room temperature as described, products which have an antirachitic activity and absorption of the type described by the authors are present to a small extent after 30 seconds' irradiation, to a marked extent after 1 minute's irradiation, and to about 10 times this value after 10 minutes' irradiation, whilst for irradiation periods of between 10 and 60 minutes both properties increase to about 2 or 4 times their value after 10 minutes' irradiation. Therefore both antirachitic activity and absorption are produced at approximately the same rate and both begin in the earliest stages. Both decrease if the products of irradiation after removal of ergosterol are exposed to ultra-violet light, and disappear almost entirely after between 3 and 5 hours' irradiation under the above conditions. No product showing either antirachitic activity without the type of absorption described, or *vice versa*, has yet been obtained. A type of absorption which may be a property of vitamin *D* is shown in a curve. As the most probable explanation of the observed phenomena, it is suggested that the irradiation of ergosterol produces two substances in succession, of which the first has an absorption maximum at about 280 or 290$\mu\mu$, and the second a maximum at about 230$\mu\mu$, and that the former is vitamin *D*. Plates of the photographed absorption spectra are reproduced. P. H. P.

Antirachitic Substances. VIII. Studies on Highly Purified Ergosterol and its Esters. C. E. Bills and E. M. Honeywell. (*J. Biol. Chem.*, 1928, **80**, 15–23.)—A procedure was designed to prepare ergosterol of the greatest possible purity for research on antirachitic activation. Crude yeast sterol was obtained

from the unsaponifiable fraction of the fat of the common yeast, *Saccharomyces cerevisiae*, and it has been shown on examination to be a mixture of at least three sterols, separable with difficulty. Ergosterol itself is the main component, another is dextrorotatory and of low m.pt., and possibly identical with the zymosterol of MacLean (*Biochem. J.*, 1928, **22**, 22), and the third, named cerevisterol, is laevorotatory, and melts above 240° C. The ergosterol of yeast is identical with that of ergot obtained by Tanret (*Ann. chim. et phys.*, 1908, **15**, 313), and has been isolated in pure form. Previous workers have failed to get satisfactory purification, but purification has now been accomplished in two ways—by recrystallisation from an exceptionally effective solvent mixture (alcohol-benzene, 3 : 1), and by saponification of purified ergosteryl isobutyrate. Pure ergosterol exhibits in chloroform $[\alpha]_D^{20} = -132°$ and $[\alpha]_{5461}^{20} = -171°$. The high specific rotation is not due to admixture with the two contaminant sterols, for the specific rotation of the contaminants is low. The melting point is of little significance as an index of purity, for it varies from 166–183° C., according to the hydration of the sample. It would seem that the recent discoveries in the relation of ergosterol to vitamin *D* have all been made with ergosterol of questionable purity, since until now only Tanret had prepared ergosterol free from contamination. However, it has been found that the spectrographic and physiological properties associated with ergosterol of ordinary purity are exhibited by ergosterol free from contaminants. Pure ergosterol, unlike zymosterol and cerevisterol, is highly activatable, and shows the absorption bands found for ordinary ergosterol. More properties of pure ergosterol not given by Tanret in his report are described. Three new esters, ergosteryl isobutyrate, isovalerate, and cinnamate have been prepared and purified.

P. H. P.

Conditions of Formation and Destruction of Vitamin *D* on the Irradiation of Ergosterol. D. Van Stolk, E. Dureuil and Heudebert. (*Comptes. rend.*, 1928, **187**, 854–856.)—Elimination of waves of shorter length than 2550 Å during the irradiation of ergosterol did not stop the decomposition of the vitamin *D* formed, but irradiation of the alcoholic solution in an atmosphere of nitrogen considerably retarded the process, so that the final destruction of the vitamin is looked upon as due to oxidation and not to any destructive radiations. The absorption bands noted for the pure ergosterol had their maxima at 2932, 2815, 2700, and 2600 Å. The first three disappeared during irradiation, whilst the fourth became augmented, and two new bands at 2503 and 2405 Å also appeared, these latter being characteristic of vitamin *D*.

D. G. H.

Vitamin *A* Content of the Unsaponifiable Matter of Liver Oils. I. S. Meno, M. Yamashita and Y. Ota. (*J. Soc. Chem. Ind., Japan*), 1928, **31**, 281B.)—The authors report a large amount of vitamin *A* in the liver oil of "ishinagi" fish, *Stereolepis ischinagi*" (Hilgendorf). After saponification the unsaponifiable portion from 100 grms. of the oil was extracted from the aqueous medium with ether, washed with water, and final traces of potash removed by precipitation with carbon dioxide. For applying the vitamin tests the purified dry product was dissolved in 100 grms. of olive oil. Growth curves are given for

albino rats fed on a basal diet of 69 parts rice starch, 5 parts cane sugar, 10 parts fat-free horse-flesh, 5 parts McCollum and Simmonds' salt mixture 185, 1 part " oryzanin " powder (vitamin *B*), and 10 parts of sample oil. It is shown that the vitamin *A* content of " ishinagi " liver oil is several hundred times that of cod-liver oil. The most rapid and reliable colour test for vitamin *A* is to add dried Japanese acid clay to 1 grm. of the oil dissolved in 10 c.c. of benzene. A violet coloration indicates the presence of vitamin *A* (see Abstract, p. 65). R. F. I.

Synthesis of Vitamin *B* in the Rumen of the Cow. S. I. Bechdel, H. E. Honeywell, R. A. Dutcher and M. H. Knutsen. (*J. Biol. Chem.*, 1928, **80**, 231–238.)—It has been shown by Bechdel, Eckles and Palmer (*J. Dairy Sc.*, 1926, **9**, 409) that a calf will grow normally to maturity and produce normal offspring on a ration that carries an insufficient amount of the vitamin *B* complex to support growth and well-being in rats ; also, by Bechdel and Honeywell (*J. Agric. Research*, 1927, **35**, 283) that vitamin *B* in milk is not dependent on the presence of this vitamin in the ration of the cow. It thus appeared that cattle, and possibly all other ruminants, can synthesise vitamin *B*, and so experiments were designed to determine whether the micro-organisms present in the rumen of an experimental cow were responsible for the synthesis of vitamin *B* complex. Investigations were conducted on the fermented rumen contents of a Holstein cow, representative of a group of 17 animals that were grown to maturity on a ration highly deficient in vitamin *B* complex. A permanent fistula about $3\frac{1}{2}$ inches in diameter was made in the rumen of the experimental heifer through the left side ; it was kept tightly closed in order that normal conditions could be maintained within, and with this means the rumen contents could be sampled easily. Alcoholic extracts of the fermented rumen contents were proved potent in the vitamin *B* complex through rat-feeding trials ; therefore vitamin *B* must have been synthesised by bacteria or other micro-organisms. One bacterium of the genus *Flavobacterium* was found to predominate to the extent of about 90 per cent. in the rumen microflora which were next examined. As no account of it was found in the literature, it is described, and is called *Flavobacterium vitarumen.* This organism was grown in large quantities on vitamin *B*-free media and given to rats to the extent of about 12 per cent. of dried bacterial cells in a synthetic vitamin *B*-free ration. The bacterial cells were proved to be highly potent in the vitamin *B* complex. It is therefore concluded that the vitamin *B* complex was produced in the rumen of the experimental cow by bacterial fermentation ; this offers a satisfactory explanation as to why cattle, unlike any other species of animal yet studied, can grow to maturity, produce normal offspring, and produce milk of normal dietary composition, on a ration that has not sufficient vitamin *B* complex to support growth and well-being in rats. P. H. P.

Toxicological and Forensic.

Toxicological Study of Bismuth. R. Fabre and M. Picon. (*J. Pharm. Chim.*, 1928, **120**, 297–308.)—Oil solutions of bismuth camphocarbonate were injected by the int a-muscular or intravenous method into rabbits and dogs, and it was found that the bismuth was retained in important quantities by the liver

and kidneys, but only in minimum quantities by the brain. The kidneys retained more bismuth than the liver, and death was probably, at least partly, due to alteration in the renal functions. Elimination was by means of the kidneys, but salivary excretion and excretion by the skin also play a very important part.

D. G. H.

Arsenic Test of the German Pharmacopoeia. G. Frerichs. (*Pharm. J.*, 1928, **121**, 383.)—The solution of sodium hypophosphite in fuming hydrochloric acid, which in the German Pharmacopoeia VI has replaced Bettendorf's reagent for testing for arsenic, is incorrectly named, as it contains, not hypophosphite, but free hypophosphorous acid with a small quantity of sodium chloride. The author proposes to name this reagent " Thiele's hypophosphite solution," and regards it as superior to stannous chloride solution, except perhaps for the examination of iron preparations, where the colour interferes, owing to incomplete reduction of the ferric oxide. This difficulty may be overcome by addition of a sufficiently large quantity of stannous chloride solution, and the test then admits of the detection of 0·00001 grm. of arsenic in 1 c.c. of ferric chloride solution. Potassium iodide may also be used for decolorising ferric solutions. In testing *sulphur sublimatum* and *praecipitatum* the troublesome treatment with nitric acid may be avoided by following the slightly modified test of the German Pharmacopoeia V : 2 grms. of the sulphur are shaken vigorously in a flask with 10 c.c. of ammonia solution and heated on a water-bath at 40° to 50° C. for about five minutes, 5 c.c. of the filtered liquid being then evaporated to dryness. The residue is dissolved in three or four drops of sodium hydroxide solution and 5 c.c. of hydrogen peroxide solution, the liquid being again evaporated to dryness and the residue dissolved in about 1 c.c. of dilute hydrochloric acid and heated with 3 c.c. of the hypophosphite solution in a boiling water-bath for fifteen minutes ; the liquid should remain clear and colourless.

T. H. P.

Detection of Arsenic. Dauvé. (*Ann. Chim. analyt.*, 1928, **10**, 320–321.)—It is recommended that in using Gatehouse's method for detecting arsenic, as described in Wurtz's Dictionary (Supplement, p. 240), mercuric chloride should replace the silver nitrate in making the test paper, since silicon hydride is formed under the conditions of experiment from the silica present in the aluminium, and whilst this blackens silver paper it has no effect on the mercury solution. The solution to be tested is placed in a bottle in the neck of which is suspended a glass tube, lightly closed at the end with cotton wool, and with a hole at the side for the entrance of the hydrogen arsenide, and in which is suspended the test paper, moistened with only one drop of mercuric chloride, to give a contrast of colour.

D. G. H.

Bacteriological.

Determination of the Number of Organisms in Water. W. Plücker and W. Bartels. (*Z. Unters. Lebensm.*, 1928, **56**, 51–60.)—The current (German) methods for the determination of the number of organisms in water are discussed

in the light of the recommendations of numerous workers. Prall's medium, which is a meat extract and peptone-salt mixture, containing 5 per cent. of gelatin and 0·75 per cent. of agar, is recommended in addition to those ordinarily used. A P_H value of 7·0 to 7·1 is suitable for the harmless water bacteria, as well as for *B. coli*, *B. alcaligenes*, *Staphylococcus pyogenes* and *Paratyphus*, though *B. typhi* requires P_H 7·2 to 7·9. An incubation period of 48 hours at 20 to 22° C. is recommended, and Brudny's automatic apparatus (*Zentr. f. Bakt.*, 1911, **57**, 478) has been found useful for counting the colonies. The microscopic method of Hesse–Niedner (*Z. Hyg.*, 1904, **20**, 119) is more sensitive, the number of colonies per c.c. of water being given by the formula D^2vm/d^2, where D is the diameter of the Petri dish (90 to 94 mm.), v the dilution of the water, and m the number of colonies in the field of vision of the microscope (diameter d mm.). The thickness of the layer of medium should not exceed 1·5 mm., corresponding with 9 c.c. of liquid. J. G.

Gas Production in the Making of Sauerkraut. L. M. Preuss, W. H. Peterson and E. B. Fred. (*Ind. Eng. Chem.*, 1928, **20**, 1187, 1190.)—The gas evolved during the formation of sauerkraut consists of almost pure carbon dioxide, and most of the gas is given off within forty to one hundred and sixty hours after the cabbage has been packed in the container. The fermentation is more rapid at higher temperatures (25° to 28° C.) than at lower temperatures, and there is a close relation between the volume of gas, numbers of bacteria, and acidity, denoting that the gas production is due to bacterial action and not to yeast growth.

W. P. S.

Organic Analysis.

New Oxidation Reactions of Aldehydes. J. B. Conant and J. G. Aston. (*J. Amer. Chem. Soc.*, 1928, **50**, 2783–2798.)—Oxidation of isobutaldehyde by alkaline potassium ferricyanide solution at 80° C. gives 2:2:5:5-tetramethyldihydropyrazine and 2:2:5:5-tetramethyl-3:6-dicyanopiperazine, but no isobutyric acid ; under similar conditions, methyl isopropyl ketone is oxidised to hexamethyldihydropyrazine. When oxidised in acid solution by either ceric sulphate at 80° C., or potassium permanganate at 80–90° C., or cobaltic sulphate at 0° C., isobutyraldehyde yields acetone, α-hydroxyisobutaldehyde, and isobutyric acid, whilst chloranil in presence of palladium black slowly oxidises the aldehyde to α-hydroxyisobutaldehyde. In acid solution at 80° C., potassium dichromate oxidises isobutaldehyde in the α-position, giving acetone to the extent of 40 per cent. in solutions of high dilution, which favours the oxidation ; *n*-butaldehyde also is oxidised in the α-position, carbon dioxide being formed. Potassium permanganate oxidises acetaldehyde in acid solution at 80° C., giving carbon dioxide as well as acetic acid if the reactants are kept very dilute and the aldehyde is in excess.

T. H. P.

New Procedure for the Separation of Alcohols and Phenols from Oil Mixtures. H. Schmidt. (*Chem. Ztg.*, 1928, **52**, 898.)—The oil mixture is warmed at 80 to 100° C. in a distillation-flask with the approximate amount of boric acid

required to form the triboric ester $B(OR)_3$, and the water produced is distilled over in a low vacuum. The vacuum is then raised till the oil has distilled off, the residue saponified with sodium hydroxide solution, and the liberated phenol or alcohol removed by steam distillation. Solid borates, which often denote the presence of cyclic alcohols, may be recrystallised. Boric anhydride, triaceto-boric acid $B(O.COCH_3)_3$, arsenic, antimonic or phosphoric acids may also be used, and the method is preferable to the benzoylation or acid ester methods, in that it is simple, inexpensive, rapid and quantitative. Since primary, secondary and tertiary alcohols are esterified with progressive difficulty, they may be separated by fractional esterification with determined amounts of boric acid. Borneol and dihydroterpineol were isolated from American pine oil (*Ber.*, 1928, **60**, 1372), whilst menthol and menthone (*id.*, 1926, **59**, 2306), and geraniol and citral were among other substances separated from one another. J. G.

Use of Iron Reagents in the Detection and Differentiation of Phenols. A. H. Ware. (*Quart. J. Pharm.*, 1928, **1**, 377–387.)—The phenols are divided into 3 classes: Class A, consisting generally of phenols giving only one definite colour reaction with ferric salts; Class B, possessing two or more hydroxyl groups in contiguity and giving colour changes with ferric chloride largely determined by P_H concentration, but in a series of five colours; and Class C, containing pyrone and quinonoid phenols, and giving with ferrous salts and weak alkali an intense brown colour. In practical work Mitchell's reagent (0·1 grm. ferrous sulphate and 0·5 grm. of Rochelle salt in 100 c.c. of water) is used with decreasing P_H, and ferric chloride with increasing P_H. Mitchell's reagent (ANALYST, 1923, **48**, 2) is more satisfactory than ordinary ferrous salts, because the P_H is very near 7; ferrous or ferric hydroxides, basic ferric acetate or phenol-iron-complexes may be eliminated, but the controlled precipitation in bulky filterable form of certain complexes may be brought about when necessary; very little ferric iron is present, and maximum intensities of colour for Classes B and C may be obtained by appropriate adjustment of the P_H. The reagent is added until no further darkening occurs, the P_H adjusted with very dilute ammonia or sodium bicarbonate solution, and, if a negative result is obtained, the phenol is placed provisionally in Class A; a violet colour indicates Class B, and a deep brown Class C. Potassium acetate is then added and the mixture boiled, when tannin is precipitated. Haemotoxylin is partly precipitated (blue), and also maclurin and many anthroxanthins, as brown complexes. For the ferric chloride test, a 1 per cent. so-called neutral solution is added, drop by drop, to the solution of the phenol; the results with a large number of phenols are described. Special tests are as follows:—One or two drops of liquid phenol or a pinch of solid is dissolved in 5–10 c.c. of water together with a little ferrous sulphate; 1 drop of 10 volume hydrogen peroxide is then added, and the mixture shaken until a marked degree of green, red or brown colour is given, whichever appears first. The mixture is then shaken with from 0·2–0·5 grm. of sodium sulphite.

Name of Phenol.	Result before adding sodium sulphite.	Result after adding sodium sulphite.
Carbolic acid, the cresols (including B.P. cresol)	Deep green	Blue, violet or purple
Guaiacol and B.P. creosote	Brownish green	,, ,, ,,
Salicylic acid and other salicyl bodies	Purple	Brown
Thymol and eugenol (in alcoholic solution)	Not distinctive	Not distinctive
Hydroquinol	Reddish colour	Blue, violet or purple
Resorcinol and orcinol	Brown	,, ,, ,,
Phloroglucinol	Yellow, greenish or brown	Brown or yellow
Aloin and phloridzin	Brown or reddish	Brown

Test for isocarbon.—To a pinch of aloes dissolved in 5–10 c.c. of water and filtered, is added one drop of hydrogen peroxide, and, after shaking, 1 per cent. ferric chloride solution, drop by drop, with shaking after each drop, when the colour changes are: green —> brown —> ruby red —> reddish purple. A precipitation test for certain phenols is carried out in 5 steps with Mitchell's reagent, or a solution of citrate of iron and ammonia, with subsidiary reagents Rochelle salt, ammonium hydroxide, 35 per cent. acetic acid, and a 35–40 per cent. solution of formaldehyde. Details and results are tabulated. Clearly-defined stages are present at which typical pure gallo-tannins, phlobatannins and phenols of Class B are precipitated and pyrogallol tannins are distinguished from all other plant principles. D. G. H.

Determination of Pyruvic Acid. B. H. R. Krishna and M. Sreenivasaya. (*Biochem. J.*, 1928, **22**, 1169–1177.)—The methods used for the determination of pyruvic acid are largely based upon the reaction of its carbonyl group with phenylhydrazine; its determination in complex biological fluids where there are other compounds which react similarly is therefore difficult, particularly when the pyruvic acid is present only in small quantities. The authors have made a study of some of the existing methods, but found none suitable without modifications for their work, *i.e.* for determinations of small quantities of the acid in solutions of very low concentration. They have developed a new method, more specific when applied to biological fluids, which is a modification of the technique of Lieben (*Biochem. Z.*, 1923, **135**, 240), and is based upon the reactions:

(1) $CH_3.CO.COOH+2H=CH_3.CHOH.COOH$ and

(2) $CH_3.CHOH.COOH+O=CH_3.CHO+CO_2+H_2O$.

It is shown that the reduction of pyruvic to lactic acid with the use of a zinc-copper couple in sulphuric acid solution, oxidation of lactic acid by a slight modification of the recent method of Friedmann, Cotonio and Schaffer (*J. Biol. Chem.*, 1927, **73**, 335; ANALYST, 1927, **52**, 418–419), with the use of a simpler apparatus, and titration of the bound aldehyde according to the method of Clausen (*J. Biol. Chem.*, 1922, **52**, 263; ANALYST, 1922, **47**, 363) gave quite constant results over a large range of pyruvic acid concentrations. Although the aldehyde yield was small,

the error was quite regular, as shown by tables of results. By means of an empirical factor, *i.e.* 1 c.c. $N/10$ iodine represents 5·5 mgrms. of pyruvic acid—the exact amount of pyruvic acid can be computed. The following technique is advised when the method is applied to biological fluids :—The solution should not contain more than 15 mgrms. of pyruvic acid during its reduction to lactic acid ; about 2 to 5 c.c. of an approximately 0·05 per cent. solution is used for protein separation either by ether extraction or alcoholic precipitation, and the whole of the filtrate is taken up for subsequent processes. The filtrate is rendered neutral to litmus and evaporated under diminished pressure at 40 to 50° C. The substance is then transferred to an extractor with a small quantity of saturated ammonium sulphate solution, rendered slightly acid, and extracted with ether. The ether extract is evaporated to dryness, shaken up with excess sodium bisulphite, and again extracted. The residue is transferred to a 100 c.c. flask, the pyruvic acid reduced to lactic acid by sulphuric acid and zinc with a trace of copper, and the lactic acid is then determined. The probable error of a single determination has been found to be about 1·4 per cent. P. H. P.

Lehmann's Method for the Determination of Aniline. A. V. Pamfilov and V. E. Kisseleva. (*Z. anal. Chem.*, 1928, **75**, 87–92.)—The method described in " Chemisch-technische Untersuchungsmethoden," 1921, p. 657 (G. Lunge and E. Berl) has been examined and modified. The aniline is absorbed in 10 per cent. sulphuric acid, and 20 c.c. of 0·8 to 0·0006 N sodium hypobromite solution (prepared by the addition of sodium hydroxide solution to 0·4 per cent. bromine water till the yellow colour disappears), and 1 to 2 grms. of potassium bromide are added. After 3 minutes in a stoppered bottle the solution is back-titrated with 0·1 to 0·001 N sodium thiosulphate solution in the presence of potassium iodide, with starch as indicator. Satisfactory results were obtained for 0·05 M aniline sulphate solutions. For more dilute solutions (0·005 to 0·00005 M) the weaker reagents are used with indigo carmine as indicator, but the results are progressively high as the concentration decreases. J. G.

Reactions of Dyestuffs with Nitrous Acid. J. V. Dubský and A. Okáč. (*Z. anal. Chem.*, 1928, **75**, 92–111.)—The use of the coloration produced by diazotisation, with or without subsequent coupling with suitable reagents, for the colorimetric detection of nitrous acid, has been tested for about 100 dyestuffs of varied constitution. A sensitiveness of $1:10^6$ to $1:10^7$ was usually found, and a table shows the colours of the solutions before and after treatment with nitrous acid, and also after coupling with one or more of 23 suitable compounds, together with the sensitiveness of the reaction concerned. As a rule, α-compounds with a free para-position gave a faster reaction and a deeper colour, and were about 10 times more sensitive than β-compounds with a free ortho-position (*cf.* Vaubel, ANALYST, 1928, **53**, 674). J. G.

Chemistry of Jaffe's Reaction for Creatinine. V. Isolation of the Red Compound. I. Greenwald. (*J. Biol. Chem.*, 1928, **80**, 103–106.)—The new dicreatinine compound described by Greenwald (*J. Biol. Chem.*, 1928, **77**, 539 ;

ANALYST, 1928, **53**, 400–401) was proved not to be the substance responsible for Jaffe's reaction. It had previously been shown by Greenwald and Gross (*J. Biol. Chem.*, 1924, **59**, 601 ; ANALYST, 1924, **49**, 346) that although only 1 molecule of picric acid entered into the reaction, the maximum colour in Jaffe's reaction was not obtained unless at least 2 molecules of picric acid were present. It was therefore thought of interest to ascertain what might be precipitated when an alkaline mixture containing 2 molecules of picric acid for each molecule of creatinine was run into alcohol. The first preparation yielded a red precipitate which seemed to be a mixture of the dicreatinine compound and a new one containing 1 molecule of creatinine, 1 of picric acid and 2 of sodium hydroxide. With the use of a little more picric acid (2·5 or 3 molecules), and not too great an excess of sodium hydroxide, the new compound was obtained nearly pure. After drying *in vacuo*, the substance forms a brilliant red, hygroscopic powder. When dissolved and diluted in water to contain 10 mgrms. of creatinine per 500 c.c., and compared with 0·5 *N* potassium dichromate, the colour obtained corresponds to only about 20 per cent. creatinine instead of the calculated 26·8 per cent. ; this is probably due to dissociation, for, if a mixture of 15 c.c. of 1 per cent. picric acid and 5 c.c. of 10 per cent. sodium hydroxide is added before dilution, the full colour is obtained immediately. Therefore it is the formation of this compound that is responsible for the red colour of Jaffe's reaction. This does not altogether contradict the previous view that the formation of the red tautomer of creatinine picrate gives the colour, for the new compound may be regarded as a compound of the red tautomer with 2 molecules of sodium hydroxide. Hydrochloric acid precipitates the red tautomer from fairly concentrated solutions of the new compound. A solution of the new compound gives a red precipitate with basic lead acetate solution. When filtered, washed, and dried over sulphuric acid, the composition of the precipitate agrees closely with that calculated for a compound of 1 molecule of creatinine, 1 of picric acid, 2 of lead hydroxide, and 2 of water. As with the dicreatinine compounds, both the sodium and the lead compound contain more base than the formulae would require. With the high equivalent weight of lead, this results in a decided effect on the creatinine and picric acid content. Differences in the behaviour of both the creatinine and the picric acid show that the nature of the combination between picric acid and creatinine in the new compounds is quite different from that in the dicreatinine compounds. P. H. P.

Determination of Sulphur in Rubber by the Perchloric Acid Method. E. Wolesensky. (*Ind. Eng. Chem.*, 1928, **20**, 1234–1238.)—The following procedure is recommended : One grm. of the finely-divided sample is heated for two minutes in a 500 c.c. flask with 10 c.c. of 41 per cent. nitric acid ; 10 c.c. of concentrated nitric acid are then added, and the heating is continued for fifteen minutes, or until the rubber has dissolved. Five c.c. of 60 per cent. perchloric acid are added, and the mixture is boiled until white fumes appear. It may be necessary to add more perchloric acid if free carbon is present, or, in the absence of other precipitates, the carbon may be removed by filtration. The clear solution

is then treated with 5 c.c. of concentrated hydrochloric acid, again heated, cooled, diluted, and the sulphate precipitated with barium chloride. If barium sulphate is present in the rubber, it is separated from the oxidised solution previous to the precipitation with barium chloride. W. P. S.

Determination of Iron Carbonyl. R. H. Griffith and G. C. Holliday. (*J. Soc. Chem. Ind.*, 1928, **47**, 311–312.)—The formation of iron carbonyl in water gas or coal gas stored under pressure in steel cylinders suggested the possible occurrence of the carbonyl in the mixture of carbon monoxide and hydrogen used for the synthesis of methyl alcohol. Such occurrence is actually observed, and catalyst which has been used in this synthesis is found to be stained with a brown deposit of iron. To determine iron carbonyl in a gas, this is passed through cotton wool and washed with sulphuric acid, which absorbs the carbonyl quantitatively. The acid is then evaporated to dryness, and the residue dissolved in hydrochloric acid and tested with ferrocyanide, the colour density of the uncoagulated Prussian blue formed being measured under standard conditions. For this purpose use is made of a modified Sanger–Shepherd density meter with a red light filter to convert the colour to neutral grey. A cell, 10 mm. wide inside, may be used to contain the solution, and a 100-watt Fullolite lamp as the source of light. The density wedge is first calibrated by means of a solution containing 0·1 grm. of iron, dissolved in slight excess of hydrochloric acid, per litre. Various volumes (between 0·5 and 10 c.c.) of this solution are placed in 50 c.c. graduated cylinders, 1 c.c. of concentrated hydrochloric acid (10 *N*) being added and the volume made up to 25 c.c. with water. One c.c. of 10-volume hydrogen peroxide is added to oxidise any ferrous iron, and the solution made up to 48 c.c. ; 2 c.c. of 0·1 M potassium ferrocyanide solution are added, and the whole shaken. After the lapse of 10 mins., the cell is charged and the density reading taken. A table is given showing, for solutions containing quantities of iron varying from 0·00005 to 0·001 grm. per 50 c.c., the corresponding wedge readings and the densities of the " blue." The wedge reading may be determined to within 0·02, equivalent to 0·0002 mgrm. of iron. The conditions of the calibration must be adhered to strictly during the determination ; the readings are affected considerably by increasing additions of acid or the presence of salts, and slightly by curtailing the time during which the liquid stands before the reading is taken. Concentrated sulphuric acid may be used in place of hydrochloric acid, provided that not more than 0·5 c.c. is taken.

The amounts of iron carbonyl, in grms. per 1000 litres, found in carbon monoxide from various sources, are : from the active-iron reaction tube, 1·26 ; from storage holders, compressed, 0·1853 ; stored at 80–100 atmos. for 6 months in a cylinder which had been in use for about 20 years, 0·00245 ; freshly made in Tantiron pots from sulphuric and anhydrous formic acids, 0·00844. Town gas contained 0·0003 grm. of the carbonyl per 1000 litres ; in this case the absorption was effected by means of B.D.H. charcoal, since only a limited amount of this gas can be treated with sulphuric acid, owing to absorption of unsaturated compounds.

Methyl alcohol (either once distilled or " pure "), absolute alcohol (fermentation or synthetic), and nitric, sulphuric, and hydrochloric acids contain iron in small proportions. T. H. P.

β-Methyl-umbelliferone as a Fluorescent Indicator. C. Bülow and W. Dick. (*Z. anal. Chem.*, 1928, **75**, 81–86.)—About 2 drops of a 0·3 per cent. alcoholic solution of β-methyl umbelliferone (Pechmann and Duisberg, *Ber.*, 1883, **16**, 2122) appear colourless in acid solutions, but give a strong blue fluorescence in alkaline solutions, which is visible against a background of black glazed paper without the aid of a quartz lamp. The end-point, which is masked in dark brown, but not in yellow, red or blue solutions, occurs at P_H 6 to 7, and the indicator is therefore best suited for the titration of strong bases against strong acids. For weak acids, however, back-titration may be used. J. G.

Inorganic Analysis.

Rapid Method for the Determination of Selenium. E. Benesch. (*Chem. Ztg.*, 1928, **52**, 878–879.)—The solution of selenium is reduced (*e.g.* by an acid solution of sodium bisulphite) and the amorphous selenium (0·2 to 0·3 grm.) filtered off, washed, and dissolved in 100 c.c. of a cold saturated solution of sodium sulphate. The red coloured solution and filter paper are placed in a flask, 150 c.c. of water added, and the whole titrated with a 0·1 *N* potassium cyanide solution (1 c.c.=0·0079 grm. Se), which is standardised under the same conditions against a known weight of selenium. A change of colour to bright yellow, similar to that of methyl orange, indicates the end-point of the reaction Se+KCN=KCNSe. J. G.

Determination of Palladium by 6-Nitroquinoline. S. C. Ogburn and A. H. Riesmeyer. (*J. Amer. Chem. Soc.*, 1928, **50**, 3018–3022.)—The chloride solution is heated to boiling and treated with a hot saturated aqueous solution of the reagent. The mixture is stirred and boiled for a few minutes, and tested for complete precipitation by the addition of more reagent. The flocculent yellow precipitate is collected after 15 minutes, washed with water, dried, ignited carefully, reduced in hydrogen, and cooled in carbon dioxide. In presence of other platinum metals, the results show a negative error. W. R. S.

Detection of Copper in Presence of Iron. L. Szebellédy. (*Z. anal. Chem.*, 1928, **75**, 167–168.)—The addition of ammonium fluoride (1 grm.) to a solution of ferric iron (0·1 grm.) prevents the formation of the blue ferrocyanide precipitate, a white precipitate being formed. The precipitation of copper ferrocyanide is not impeded, hence a fraction of a mgrm. of copper imparts a pink tinge to the white iron precipitate. W. R. S.

Modification of Low's Short Iodide Method for Copper. H. F. Bradley. (*Chemist Analyst*, 1928, **17**, 14.)—Low's method (*Technical Methods of Ore Analysis*, p. 85) gives good results for ores of high manganese content if modified as follows :— To 0·5 grm. of ore are added a few drops of water, a little potassium chlorate,

5 c.c. of nitric acid, and a drop of hydrochloric acid, and the mixture heated for 5 minutes. Any brown oxides of manganese are dissolved in a few drops of a mixture of hydrogen peroxide and hydrochloric acid, 7 c.c. of concentrated sulphuric acid added, and when copious fumes are evolved the solution is cooled, diluted to about 40 c.c., and again boiled. The cooled solution is then almost neutralised with 13 c.c. of ammonia and a strong solution of ammonium acetate added till the red ferric acetate appears. After the addition of a further 5 c.c. of the acetate solution, the red colour is dispersed with sodium fluoride (0·5 grm.), and the solution titrated in the usual way in the presence of 3 grms. of potassium iodide. J. G.

Analytical Chemistry of Gallium. (Part I.) L. Moser and A. Brukl. (*Monatsh. Chem.*, 1928, **50**, 657–668.)—Gallia is a much weaker base than alumina, from which it differs by yielding a ferrocyanide precipitate in strongly acid solution. Though not precipitated by hydrogen sulphide from acid solution, gallium is adsorbed by the sulphide precipitates of the heavy metals. Gallium is weighed as white sesquioxide, obtained by strong ignition of the hydroxide, nitrate, or sulphate in porcelain or silica, but not platinum (partial reduction through diffusion). The ignited oxide is hygroscopic, and should be weighed with dispatch. The ammonia precipitate is gelatinous like aluminium hydroxide, and soluble in caustic alkali; it is more soluble in ammonia than the aluminium precipitate, and the presence of ammonium salts increases the solubility. The precipitation of basic gallium acetate is quite incomplete; a large excess of ammonium acetate may prevent the precipitation altogether. The authors recommend tannin as the best and most sensitive precipitant for gallium (sensitiveness, 1:5,000,000). The boiling, weakly acid acetate solution containing 2 per cent. of ammonium nitrate, is stirred and treated, drop by drop, with a 10 per cent. solution of tannin till precipitation is complete: 10 parts of tannin suffice as a rule, but for minute amounts of gallium the tannin should be not less than 0·5 grm., otherwise the precipitate does not deposit readily. It is very bulky, hence with more than 0·1 grm. Ga_2O_3 it is of inconvenient size; if a large amount of gallium is to be precipitated, the bulk may be obtained as basic acetate, and the balance in the filtrate by tannin. The precipitate is washed with hot water containing a little ammonium nitrate and a few drops of acetic acid. Filter and precipitate are dried and ignited in porcelain to Ga_2O_3, which is weighed. Ammonium chloride should not be used in the washing, as gallium chloride is volatile. The above procedure permits of the accurate separation of gallium from zinc (its most important mineral associate), nickel, cobalt, manganese, cadmium, beryllium, and thallium. The weakly acid (one per cent. acetic) solution is treated with ammonium acetate and two per cent. of ammonium nitrate, boiled, and precipitated with tannin as before. The precipitate is dissolved in hot dilute hydrochloric acid, and the precipitation repeated. Directions are given for recovering each of the other metals from the combined filtrates: zinc, cadmium, cobalt, nickel, and manganese, by hydrogen sulphide; beryllium, by tannin and ammonia (ANALYST, 1928, 402); and thallium, by

destruction of the tannin with fuming nitric acid, followed by precipitation of the chromate (*id.*, 1928, 459). (Separation of gallium from iron, *id.*, 1928, 558.)

W. R. S.

Oxalate Method for Separating Calcium and Magnesium. W. T. Hall. (*J. Amer. Chem. Soc.*, 1928, **50**, 2704–2707.)—The results of the author's experiments confirm the view that excess of ammonium oxalate is required for the precipitation of calcium oxalate in presence of magnesium, and show that, if this excess is properly regulated, it is possible to precipitate pure calcium oxalate. If, however, a very large quantity of ammonium oxalate is present, the precipitation of magnesium ammonium phosphate is incomplete, even after long standing. For the precipitation of 0·3 grm. of calcium ions in a volume of 500 c.c., 75 c.c. of 0·5 *N* ammonium oxalate is sufficient, whereas the same quantity of this reagent is necessary for only 0·02 grm. of calcium in presence of a considerable amount (0·12 grm.) of magnesium. T. H. P.

Detection of Potassium in Presence of Ammonium Salts. R. D. Reed and J. R. Withrow. (*J. Amer. Chem. Soc.*, 1928, **50**, 2985–2987.)—A solution of zirconium sulphate (0·1131 grm. per c.c.; slightly acid) was found to detect 0·00048 grm. or more of potassium in 2 c.c. of solution in presence of a large amount of ammonium sulphate. In all other wet tests for potassium, ammonium is also precipitated. The procedure is the same as that used for the detection of potassium in presence of sodium (ANALYST, 1928, 456). W. R. S.

Synthesis of Japanese Acid Clay. N. Kameyama and S. Oka. (*J. Soc. Chem. Ind.* (*Japan*), 1928, **31**, 269B.)—Analysis of Japanese acid clay shows it to contain 49·9 to 68·4 per cent. of silica, 9·8 to 20·9 per cent. alumina, and traces of iron, calcium, magnesium, and alkali metals. Preparations were made of pure silica gels dried under varying conditions, silica gels containing alumina in the proportion of 1 mol. Al_2O_3 per 6 mols. SiO_2, and silica gels containing alumina and ferric oxide. In every case these preparations showed similar characteristics as Japanese acid clay, except one. They turned blue litmus red, liberated acid from potassium chloride solution, inverted cane sugar, absorbed methyl violet from solution, absorbed moisture from the atmosphere, and were coloured blue by contact with liver oil. Furthermore, heat was produced by wetting the powder with turpentine oil. But the preparations all lacked the power of oxidising an aqueous solution of benzidine base. R. F. I.

Physical Methods, Apparatus, etc.

Determination of the Heat Value of Coal in Nickel-lined Bombs. A. E. Stoppel and E. P. Harding. (*Ind. Eng. Chem.*, 1928, **20**, 1214–1218.)—To determine the amount of nickel dissolved from the bomb by the action of the nitric and sulphuric acids resulting from the combustion, the acidity of the bomb washings is titrated with 0·1 sodium hydroxide solution, methyl red being used as indicator.

The solution is then boiled and the titration continued, phenolphthalein being used as indicator. The number of c.c. of alkali solution used in the first titration is multiplied by 1·45 to give the correction, in calories, for the free acid ; the quantity of alkali solution required in the second titration is multiplied by 4·50 to obtain the correction for the combined acid. To the sum of these two values are added 14 calories for each cgrm. of sulphur in the coal burned. W. P. S.

Carbon and Hydrogen Determinations with the Use of a Metal Tube. S. Avery. (*Ind. Eng. Chem.*, 1928, **20**, 1232–1234.)—The tube described consists of a copper tube of about 15 mm. internal diameter, each end of which extends for about 3 inches from a thin, tightly fitting outer jacket of nickel tube. The two ends of the copper tube are provided with water-jackets through which a current of water is conducted, whereby the temperature is maintained between 60° and 80° C. The tube is durable, and the inner coating of copper oxide which forms is an advantage. Nickel tubes, or nickel-copper alloy tubes, would also appear to be suitable if they could be obtained free from carbon, but this does not seem to be possible. W. P. S.

Reviews.

LUNGE AND KEANE'S TECHNICAL METHODS OF CHEMICAL ANALYSIS. Edited by CHARLES A. KEANE and P. C. L. THORNE. Second Edition. Vol. II. Pp. xix+644. 1928. London: Gurney & Jackson. Price £3 3s.

In this volume of the revised edition of Lunge and Keane's well-known manual of analysis a considerable departure has been made in the arrangement of the subject matter, as compared with the first edition. Correlated industries have been grouped together in order to make the volume more self-contained, and the material has been divided into six sections dealing, respectively, with Iron and Steel (93 pp.), Non-Ferrous Metals (263 pp.), Metallic Salts (41 pp.), Potassium Salts (30 pp.), Paints and Pigments (134 pp.), and Paint Vehicles, Japans and Varnishes (60 pp.).

In the seventeen years which have elapsed since the first edition of this volume was published great advances have been made in analytical chemistry, and one would naturally expect to find evidence of these advances in such a classic as this book is now considered to be. In this respect the reader will be somewhat disappointed, for although much recent work is included, there are many obsolete methods described which might have been omitted to make room for fuller descriptions of more modern procedures. Especially is this noticeable in the section dealing with the non-ferrous metals which, in the reviewer's opinion, should have been entirely re-written and not merely revised by the addition of a few new methods and the omission of a paragraph or section here and there.

The section on Iron and Steel by Prof. C. O. Bannister contains methods for the analysis of iron and manganese ores, pig iron, malleable iron, steel, fluxes used in smelting and the various slags and by-products obtained in working up iron ores into the finished steel product. All the well-recognised standard methods are given, including procedures for the analysis of the numerous special steels, the introduction of which constitutes one of the greatest advances made in the metallurgy of steel during the present century. New features of this edition, therefore, supply details for the determination of cobalt titanium, zirconium, uranium, cerium, boron, and nitrogen in steel; Pickard's method of determining oxygen in steel is also given, but no reference is made to the work of Oberhoffer and his collaborators on the determination of the various oxide and slag inclusions.

Mr. G. Patchin has collaborated with Prof. Bannister in writing the section on the non-ferrous metals. This follows closely the corresponding section in the first edition, except that the chapters on tantalum and thorium have been omitted. Among the many obsolete methods which still find a place here are those of Deville and Debray and of the St. Petersburg Mint for the analysis of platinum ores, which are now of historic interest only, while those of Hess and Miller are far too unreliable now that extreme accuracy is required, owing to the high price of the metals. The methods given for the analysis of iridium-platinum alloys, metallic platinum and dental alloys, however, are quite modern, incorporating as they do the latest work of the American Bureau of Standards.

In the mercury chapter no reference is made to the determination as mercuric sulphide or as mercurous chloride or to the volumetric thiocyanate method, the separation of arsenic, antimony and tin by hydrogen sulphide in hydrochloric acid of different concentrations receives only a brief mention, and no really reliable process for separating selenium and tellurium is described, the cyanide method being, as stated, approximate only. The tin chapter has been enlarged by the addition of descriptions of the gravimetric determination of tin as stannic oxide, and of the zinc-zinc oxide and lime methods of decomposing cassiterite. The removal of ilmenite from tin ores by digestion with sulphuric acid is tedious, and a much more efficient cleaning of the ore can be obtained by fusion with bisulphate. Now that aluminium is one of the most important of the non-ferrous metals, it seems strange that the omission of any reference to the analysis of bauxite from the first edition has not been rectified in this volume. The analysis of aluminium alloys is, however, treated fairly fully and much better than in the first edition. The copper section is on the whole quite good, but surely it is time that the stannous chloride volumetric method disappeared from text-books on analysis.

The tungsten and molybdenum chapters are inadequate; an antiquated form of the *aqua regia* method of determining tungstic acid in ores is given, but Bullnheimer's method is given in detail, although it is questionable whether this clumsy method of doubtful accuracy is ever used nowadays. The addition of Cremer's method (persulphate fusion followed by precipitation with cinchonine) is hardly an improvement, especially as the meagre details given are actually misleading.

The description of Arnold's method for the analysis of tungsten powder is also too brief to be of much value. The procedures described for the determination of molybdenum in ores are all based on the same principle, namely, precipitation of the trisulphide by acidification of a thiomolybdate solution followed by ignition to the disulphide; neither the trioxide nor the lead molybdate method is mentioned, nor is pressure precipitation with hydrogen sulphide in acid solution as a means of separation from vanadium, which frequently is associated with wulfenite.

Dr. Schoeller has written the section on Metallic Salts which, though brief, contains adequate information on the properties and methods of analysing almost all the important inorganic salts which have an industrial use, with the exception of sodium sulphate, which was dealt with in volume I, and potassium salts which are discussed by Dr. J. T. Dunn in the succeeding section. This section, which is the shortest in the book, deals quite adequately with natural potash deposits and the various products obtained therefrom.

The fifth section is written by Dr. R. S. Morrell and Mr. W. E. Wornum. They classify the pigments by colours: white, grey, yellow, red, blue, violet, green, brown, black and bronze. The analysis of all the chief pigments of every colour is described, and special attention is paid to the detection of adulteration. Notes on the preparation and the fastness to light of most pigments are given. Among the newer pigments described are titanium white, "Timonox," and cadmium scarlets. One error only was noted in this section—the description of tungsten bronze as "sodium para-tungstate."

The final section by Dr. Morrell provides an account of the analysis of drying oils, thinners including recently introduced compounds such as hexalin, tetralin, etc., natural and synthetic balsams and resins, oil varnishes, black bituminous paints and varnishes, and cellulose ester varnishes and enamels.

The book will, no doubt, appeal to a large circle of chemists, but it is questionable whether it fills a real gap in chemical literature, for there are many text-books on the market covering in just as good, or a better manner, the ground dealt with in the individual sections, and it is a debatable point whether the grouping of all these subjects between one pair of covers is sufficiently attractive to the average chemist to justify him in paying the rather high price demanded, especially in these days of intensive specialisation.

A. R. POWELL.

COLLOID SYMPOSIUM MONOGRAPH. No. 6. Pp. 346. New York: The Chemical Catalog Company, Inc. 1928. Price $6.50.

The appearance of the volume collecting the papers read at the annual Colloid Symposium in America is a matter of great interest to all advanced students of colloid chemistry. The present volume collects the papers read at the Sixth Colloid Symposium held at the University of Toronto, June 14, 15 and 16, 1928.

Twenty-five papers have been published, edited by Professor H. B. Weiser.

The first is the address given by Sir W. B. Hardy, who was the guest of honour. He outlines deep problems in his discussion on "Living Matter." The other papers are definite contributions to colloid chemistry based on experimental investigations.

Professor W. D. Harkins and his co-workers extend their survey of surface tension and adsorption phenomena. Special attention is paid to the ring method of determining surface tensions. A valuable inquiry is also made concerning the familiar Antonow rule, that "the interfacial tension between two liquids, mutually saturated with each other, is equal or very approximately equal to the difference between the surface tension of the two phases, each in contact with the vapour of the other phase." Practically all books on surface phenomena regard this rule as valid. Harkins now points out its limitations, and his results are of much importance.

Briggs follows with a paper on "Surface Conductance," showing that in aqueous solutions of low specific conductance present in the interstices of a diaphragm material, the electrical conductance through the inferface phase is much greater than that through an equal volume of the liquid in bulk. Such conductance is not a function of the ζ potential. Possibly the method described may be utilised to obtain values of the relative specific surface areas, *i.e.* colloidality of materials.

Several authors deal with adsorption. Professor McBain and his associates discuss the "Adsorption of Sodium Oleate at the Air-Water Interface." The results are contrary to the prediction of the Gibbs theorem, and suggest that "the surface of a solution may be covered with a monomolecular film of adsorbed solute, but may also exhibit a high concentration of solute in the neighbourhood of the surface."

Professor Burton continues his work on the effect of temperature on the coagulation of copper solutions, whilst Stamm outlines 4 dynamic physical methods for revealing the structure of soft woods. There is practical value in this work.

Medical aspects are reflected in papers dealing with the fractionation of diphtheria antitoxic plasmas; the cataphoresis of blood cells and inert particles in sols and gels; methods of studying the surfaces of living cells and their relation to the phagocytosis of bacteria; and the rôle of haemoglobin in the blood.

Two accounts are given of investigations on emulsions: (1) The Effect of Emulsification in the Peptic Synthesis of Protein. (2) Emulsions and the Effect of Hydrogen-Ion Concentration upon their Stability. In the latter paper Krantz and Gordon support Fischer's hydrate theory in emulsions stabilised with gum tragacanth. Their use of the Donnan pipette for determining interfacial tensions is, however, open to serious objection. There follow two papers on rubber, one on organophilic colloids, and others dealing with gelatin systems, the technology of smokeless powder manufacture, catalysts, and photographic problems. Nichols gives a most important and highly interesting account of the development

by Svedberg and his pupils of the Ultra-centrifuge, and outlines the field of research opened by such an instrument.

The whole volume reflects the versatility of the colloid chemist, and indicates the rapid strides being made in *quantitative* research. The former purely descriptive aspect of colloid chemistry has now given way before the advances possible because of refined technique.

The Sixth Colloid Symposium Monograph is a credit to all concerned, including the publishers. It is a necessary addition to the bookshelf of the serious student of physical chemistry.

WILLIAM CLAYTON.

PRACTICAL PHYSIOLOGICAL CHEMISTRY. By S. W. COLE, M.A. Eighth edition. Pp. xii+481. Cambridge: W. Heffer & Sons. 1928. Price 16s.

When a book of this type reaches eight editions in twenty-four years, the present appearing only two years after the seventh, it is evident that it is accepted generally by the physiologist and the student as more than an ordinary standard text-book. The volume contains a large amount of theoretical instruction in chemistry, as well as practical exercises, since it is intended primarily for the medical student rather than for the chemist.

The analyst, who deals occasionally with blood or urine analysis, is almost sure to find all that he requires on these subjects, combined with an authoritative opinion as to the value of many of the tests described, and (at the same time) he will regret that it has not been possible to find room for a chapter on the examination of faeces. There are only one or two minor points on which he might join issue with Mr. Cole, as he may wonder why it is necessary in the determination of chlorides to ash gastric juice, but neither blood nor urine, where the titration is made direct. It is doubtful whether the benzidine test is the best for blood in urine, the reduced phenolphthalein test is surely more delicate; and, although lactose may appear in the urine of pregnant women, it should be mentioned that while a positive result probably indicates pregnancy, the absence of lactose does not exclude the possibility.

It is to be regretted that the calomel cell is considered so inaccurate and out of date that it is no longer worth a description, while a short account of the glass electrode would be useful.

Regarding the book as a whole, it is a pity that Mr. Cole did not take to heart the suggestion in the review of the seventh edition (ANALYST, 1926, **51**, 273) that the proof reading should have been more thorough, as an otherwise excellent book is spoiled by misprints and misleading statements. On page 111 the student is instructed to dry a solution on a filter paper heated with boiling water, and on page 323 mention is made of "scraps of the precipitate," instead of "scrape off the precipitate." On p. 120 it would surely be advisable to clip off the vacuum pump before letting in air to the distillation flask *via* the capillary, and for the same

experiment Mr. Cole gives two different figures, using the same series of letters in each figure, which is confusing. During the various editions of the book the author appears to have changed his allegiance from Duroglass to Pyrex ware, with the result that on p. 384 it is necessary to use a Duroglass flask for a Kjeldahl digestion, and two pages later one is advised to use a Pyrex flask for the same purpose. It would also appear unnecessary in these days to tell the student twice on a page, and about fifty times in the book, that the apparatus can be obtained from one particular firm of suppliers.

Referring to the estimation of acetone bodies in urine by Goldblatt's method, Mr. Cole tells us that it has only just been published, and that he has not had time to try it, but it is surely unnecessary to say this three years after its publication.

His bibliographical references are inconsistent, in that we get *Biochem. Journ.*, **19**, 626 (1925)" and *Biochem. Journal*, VIII, p. 134"; sometimes they are included in the text in brackets and sometimes as footnotes.

In describing the degradation of starch the author appears to have attempted to combine old and new theories, with the result that he refers to amylo-amylose and amylo-dextrin, erythro-amylose and erythro-dextrin, and achroo-amylose and achroo-dextrin as separate entities.

The publishers have done well to issue a new edition of such a book at a reasonable figure.

T. McLachlan.

THE PROTAMINES AND HISTONES. By A. KOSSEL. Pp. 108. London: Longmans, Green & Co. 1928. Price 9s. net.

The late Professor Kossel devoted his scientific career to a study of the protamines and the histones. He was a pioneer in the investigation of these two groups and remained throughout his life the pre-eminent authority in the field of work. The results of a life-time of research were gathered together in this volume, which was completed only a few days before his death, and his book, a model of lucid handling of a complex subject, will undoubtedly remain for many years the standard text book of the subject with which it deals.

Kossel started his investigations of the protamines and histones from the biological aspect. He was first led to study the evolutionary changes which proteins undergo in the differentiation of the tissues. Those studies opened up an entirely new field of chemical investigation, and new methods of analysis had to be developed for the investigation of these newly recognised proteins. The general methods developed for the analysis of the material are firstly, the separation of the protamines from alcoholic solution as insoluble sulphates; secondly, the acid hydrolysis of the purified material and the separation of the basic from the non-basic constituents. The basic constituents are investigated by transformation into insoluble salts, and the non-basic by differential solubilities in various alcohols. The basic constituents of both protamines and histones consist entirely of one or

more of the di-amino acids—arginine, lysine or histidine; the non-basic constituents of mono-amino acids. In the endeavour to discover the methods by which the different units are linked into the molecule, considerable use has been made of the differential action of the proteolytic enzymes as an analytical tool.

The great interest of Kossel's work lies in the evidence which he brings forward that the proteins of the animal body are, in the biologically active tissues such as gland cells and generative cells, rebuilt to form histones and protamines, the latter group showing the greatest extent of reconstruction. In this reconstruction the mono-amino acids, found to such a large extent in the ordinary proteins of the cytoplasm, have been partially eliminated. These comparatively simple amino acids, which are without any strongly marked chemical individuality, doubtless assist in the building up of the colloidal character of the protein molecule, giving it a high degree of sensitiveness to physical changes in the environment. In cells showing great metabolic activity, such as gland cells and generative cells, maximum chemical acitivity and economy of space is obtained by the elimination of these amino acids and their replacement by others which possess highly individual chemical groupings, such as arginine with its guanidine grouping, histidine with the iminazole ring, and lysine with its terminal amino group. (It is interesting that cystine and tryptophane, both with a highly specialised structure, appear to be absent from both histones and protamines.) The result of this condensation is the production of bodies of a highly basic character, the basicity being due in some cases to the guanidine groups of arginine, in others to the terminal amino groups of lysine. In the living cell, the protamines and histones exist only in combination with nucleic acid, another body with a complex but precise constitution that is probably the index of a specialised chemical activity. It must remain for the present a subject of speculation as to how far the basic character of the protamines has been called into being by the acidic character of nucleic acid and *vice versa*, the biological functions of the active chemical groupings of both constituents of the nucleo-proteins remaining at present entirely unknown. Professor Kossel's book forms an indispensable guide to the methods that are available for further chemical investigation of the basic constituents.

D. JORDAN LLOYD.