See Special Notices on p. 200

Volume 69 June, 1944
No. 819, Pages 169—200

THE ANALYST

The Journal of The Society of Public Analysts and other Analytical Chemists

Published for the Society by
W. HEFFER & SONS LTD., Cambridge, England

ENTERED AS SECOND CLASS AT NEW YORK, U.S.A., POST OFFICE
PRINTED IN ENGLAND

THE ANALYST

PROCEEDINGS OF THE SOCIETY OF PUBLIC ANALYSTS AND OTHER ANALYTICAL CHEMISTS

AN Ordinary Meeting of the Society was held at 5 p.m. on Wednesday, May 3rd, at The Chemical Society's Rooms, Burlington House, London, W.1, with the President, Mr. S. Ernest Melling, in the chair. The following papers were presented and discussed: "The Detection and Determination of Auxins in Organic Manures. Part II—Extraction of Auxins from Manures, and Applications of the Perchloric Acid Test for β-Indolylacetic Acid and of the Went Pea Test"; with a short introduction on Auxins, by J. Hubert Hamence, M.Sc., Ph.D., F.R.I.C.; "The Rapid Photometric Determination of Tellurium in Tellurium-Copper Alloys," by P. B. Crossley, F.R.I.C. (read by M. S. Naik, B.Sc.).

NEW MEMBERS

Leonard Alfred John Balchin; William Richard Ernest Clark, B.Sc. (Lond.), A.R.I.C.; Percy Roy Clemow, B.Sc. (Lond.), F.R.I.C.; Miss Mary Corner, B.Sc. (Lond.), A.R.I.C.; Arthur Thomas Cox, B.Sc. (Lond.), A.R.I.C.; Cecil Alfred Maunder Foster, M.Sc. Ph.D. (Liv.), F.R.I.C.; Henry Hamilton Green, D.Sc. (Glasgow), Hon. D.Sc.Agric. (Univ. of S. Africa); Robert Bruce Harley, B.Sc. (St. Andrews), A.R.I.C.; Harold Hunter, D.Sc. (Lond.), F.R.I.C., A.M.I.Chem.E.; William Roderick McLean, B.Sc. (Lond.); Charles William Mellon, B.A., M.Sc. (T.C.D.), A.R.I.C.*; Capt. Gudipaty Narasimhamurthy, B.A. (Andhra), M.Sc. (Benares), F.R.I.C.; John Richard Percival O'Brien, B.Sc., M.A. (Oxon.); Frank Smith, Assoc. Fuel (Sheff.); William Frank Stephenson, B.Sc. (Liv.), F.R.I.C.; Thomas Swinden, D.Met. (Sheff.); Ernest James Vaughan, M.Sc. (Lond.), A.R.C.S., D.I.C., F.R.I.C.; Percy George Ward, B.Sc. (Lond.), A.R.C.S.

DEATHS

WITH great regret we record the deaths of the following members:

John Simpson Ford. Samuel Gordon Liversedge. Ernest John Parry.

The Volumetric Determination of Tin in Brasses and Bronzes after Separation of Copper as Oxalate

BY F. H. EDWARDS, B.Sc., AND J. W. GAILER, B.Sc.

(*Read at the Meeting, April 5, 1944*)

INTRODUCTION—The chemical methods for the determination of tin in routine samples of brasses and bronzes have always been attended by some difficulty and, unless elaborate precautions are taken, do not yield results of great accuracy or reproducibility. Gravimetric procedures for the determination of tin normally give a precipitate contaminated with other elements, while with bronzes of high tin and phosphorus contents solution of the sample in nitric acid is difficult to effect. The usual volumetric procedures for tin in these alloys, involving the separation of tin by such reagents as ammonium phosphate or acetate, suffer from the disadvantage that the precipitate is difficult to filter and wash. It therefore seemed desirable to find a method for the preliminary separation of the copper which would leave the tin in a condition suitable for reduction and subsequent titration.

PRELIMINARY CONSIDERATIONS—In the early part of the work the use of sodium hypophosphite or red phosphorus as precipitant was studied. When, however, the separation was followed by the iodimetric titration of tin, difficulty was experienced in obtaining a stable end-point. Several expedients were tried to overcome this defect, and in the course of the investigation it was found that copper could be precipitated quantitatively by oxalic acid.

* Through the Scottish Section.

A subsequent search of the literature revealed that Bournemann,[1] Peters,[2] and Gooch and Ward[3] had used oxalic acid to precipitate copper about forty years ago, and it is perhaps surprising that the separation is not more generally known or applied in modern analytical methods. We have confirmed their findings. In particular, oxalic acid appeared to satisfy the requirements specified in the introduction, since tin is known to form a stable and soluble complex in dilute acid solution.

OXALATE SEPARATION APPLIED TO ANALYSIS OF COPPER-TIN ALLOYS—In dilute mineral acid solution copper is pptd. by oxalic acid except for the last traces (2 or 3 mg); this is sufficiently good for the subsequent determination of tin by the Evans procedure.[4] For instance, a sample of a copper-tin alloy was dissolved in dil. hydrochloric acid with the aid of hydrogen peroxide and the copper was pptd. as oxalate and filtered off. The filtrate was reduced by boiling in an inert atmosphere with sodium hypophosphite and a little mercuric chloride. Careful work gave quite good results by this method. The effect of traces of copper upon the end-point was overcome by introducing ammonium thiocyanate immediately before titration. From a routine aspect, however, the method had three disadvantages, *viz.*, (1) possible loss of stannic chloride during removal of hydrogen peroxide; (2) necessity for filtration and washing of the copper oxalate; (3) the Evans finish which, although accurate, is rather awkward for large batches of samples.

A revised method was, therefore, substituted by opening the analysis with sulphuric acid and hydrogen peroxide and performing the reduction of the tin with a nickel spiral. After pptn. of the copper, the filtrate was diluted to a known bulk and an aliquot portion was decanted off for the determination of the tin. Satisfactory results were obtained for alloys containing up to 10% of tin, but with samples of higher content the tin tended to be pptd. in the early stages. The addition of a small amount of hydrochloric acid after initial solution of the material in sulphuric acid and hydrogen peroxide overcame this difficulty.

METHODS—(1) BRONZES (Time required 2½ hours)—Treat 1·0 g of fine drillings with 15 ml of 50% sulphuric acid and 10–15 ml of hydrogen peroxide (100 vol.) in a 400-ml conical beaker. Warm until dissolved and evaporate to low bulk. Dilute with 10 ml of water and 10 ml of 50% hydrochloric acid. Disregard any ppt. which may appear in the sulphuric soln., as this redissolves on warming with the hydrochloric acid. Add 100 ml of 8% oxalic acid soln. and boil gently for 15 min. Cool quickly, make up to 250 ml in a measuring flask, return to the original beaker and allow the ppt. to settle. Decant the supernatant liquid through a No. 40 Whatman paper (dry), transfer by means of a pipette 50 ml into a flat-bottomed flask, and add 50 ml of hydrochloric acid (sp.gr. 1·16), 150 ml of water and 8 g of sodium chloride. Boil briskly with a nickel spiral for 20 min. Cool under carbon dioxide, add 20 ml of 1% soluble starch soln., and titrate as rapidly as possible to a full blue colour, using standard iodine or potassium iodate soln.

Standard solutions—Stock Iodine Solution: 10·69 g of iodine with 20 g of potassium iodide per litre. Standardise against $N/10$ permanganate, using $N/10$ thiosulphate as intermediary soln. Stock Iodate Solution: 3 g of potassium iodate, 33 g of potassium iodide and 3 g of potassium hydroxide per litre. Both solutions are diluted to 1/5 strength for use and, with each, 1 ml ≡ 0·10% of tin on 1 g.

Results on Synthetic Samples—The volumetric figures for tin (Table I) are in close agreement with the quantities introduced into the synthetic samples. It is also apparent that the added elements, representing normal alloying constituents or impurities associated with bronzes, have no appreciable effect on the tin estimation.

Results on Routine Samples of Bronzes—In Table II the tin results obtained by the volumetric method are compared with gravimetric figures and with those in which tin was estimated by difference, *viz.*, copper determined electrolytically after removal of the tin by treatment with brominated hydrochloric acid, phosphorus and the other elements being determined separately.

(2) BRASSES (Time required 2 hours)—Dissolve 2 g of fine drillings in 20 ml of 50% sulphuric acid with the aid of 20 ml of hydrogen peroxide (100 vol.).* The temperature should not exceed 40° C. during this operation and solution should be complete within 10 min. Remove the excess of peroxide by boiling, cool slightly and add 10 ml of 50% hydrochloric acid. Warm until any ppt. dissolves. Dilute with 150 ml of 10% oxalic acid, boil for 15 min.,

* If difficulty is experienced in dissolving the sample, nitric acid (sp.gr. 1·42) may be "spotted in" to assist solution. Excess of nitric acid should be removed by evaporation until fumes of sulphur trioxide appear.

cool quickly and make up to 250 ml. Decant 125 ml through a No. 40 Whatman paper (dry), and add 50 ml of hydrochloric acid (sp.gr. 1·16), 8 g of sodium chloride and 75 ml of water. Boil with a nickel spiral for 20 min. Cool under carbon dioxide and titrate with a standard iodate soln.

TABLE I

Composition of synthetic samples, in g						Tin found, g	
Sn	Cu	Fe	Sb	Zn	Pb	Iodine	Iodate
0·10	—	—	—	—	—	0·0998	0·1003
						0·0990	0·1000
						0·0995	0·1000
						0·0998	0·0998
0·10	0·90	—	—	—	—	—	0·1010
							0·1008
0·10	0·90	0·005	—	—	—	—	0·1020
							0·1020
							0·1013
0·10	0·90	—	0·005	—	—	—	0·1008
							0·1005
							0·0995
0·10	0·90	—	—	0·04	—	—	0·1010
							0·1013
							0·0995
0·10	0·90	—	—	—	0·04	—	0·1010
							0·1010
							0·1005
0·10	0·90	0·005	0·005	0·04	0·04	—	0·1005
							0·1005
							0·1010

TABLE II

Sample	Tin, % (grav.)	Tin, % (diff.)	Tin, % (volumetric, iodine)	Tin, % (volumetric, iodate)
Phos-Bronze (P 0·79%)	12·91	12·92	12·75	12·85
	12·91		12·78	12·90
			12·85	12·75
			12·80	
			12·88	
Phos-Bronze (P 0·07%)	12·17	12·22	12·20	12·35
	12·25		12·15	12·23
			12·23	
			12·25	
			12·20	
Phos-Bronze (P 0·72%)	10·69	10·86	10·88	10·85
	10·73		10·80	10·80
			10·85	
			10·70	
			10·78	
			10·85	
Phos-Bronze (P 0·18%)	5·70	5·75	5·75	5·80
	5·68		5·75	5·75
			5·75	
			5·70	
			5·75	
			5·80	
Complex Bronze (Pb 2·65%, Zn 5·58%, Fe 0·18%, Ni 0·39%, Sb 0·19%, Cu and Sn remainder)	6·07	—	—	6·08
				6·05

Standard Solutions—Stock Iodate Solution: 1·80 g of potassium iodate, 20 g of potassium iodide and 2 g of potassium hydroxide per litre. Dilute to 1/10 strength. **1 ml ≡ 0·0003 g** of tin, ≡ 0·03% of tin on 1 g of sample, as fractionated in the above method.

Results on Synthetic Samples—To mixtures (60 : 40) of pure copper and zinc known amounts of tin were added from a standard solution. The results are given in Table III.

TABLE III

Tin added, %	Tin found, %
0·10	0·10, 0·12
0·70	0·69, 0·69
1·00	1·00, 1·04
1·50	1·48, 1·49

Results on Routine Samples—A comparison of some results by the volumetric and the gravimetric sulphide methods is given in Table IV for a variety of brass samples.

TABLE IV

Composition of brass sample (% element)							Tin, %	
Cu	Pb	Fe	Mn	Al	Ni	Zn	Grav. sulphide	Vol. iodate
58·8	1·6	—	—	—	—	remainder	0·16 0·16	0·16 0·16
56·8	0·9	0·8	0·9	1·4	—	,,	0·17 0·18	0·17 0·17
55·7	1·0	1·0	1·9	0·2	2·0	,,	0·52 0·52	0·51 0·54
64·4	0·1	—	—	—	—	,,	0·83	0·81
57·8	0·9	—	1·4	—	—	,,	0·95 1·00	1·06 1·08
64·2	—	—	—	—	—	,,	0·95	0·98
59·9	—	1·0	1·3	—	—	,,	1·16 1·18	1·17 1·19
56·6	—	2·1	0·9	—	—	,,	1·20 1·21	1·24 1·21

DISCUSSION OF THE RESULTS AND CONCLUSIONS—The tin figures quoted in the tables represent results by several analysts and show the reproducibility obtained by the volumetric determination. There is little to choose between the accuracy of the iodine and the iodate finish, but the latter is preferred, as the end-point of the titration is much sharper.

For routine work with bronzes the volumetric method has the advantage that it gives an accurate determination of the tin content, whereas the tin oxide of the gravimetric method (1) may be contaminated with other elements of the alloy (in routine work an empirical deduction is usually made), and (2) is assumed to contain all the phosphorus of the alloy as P_2O_5. The corresponding deduction for P_2O_5 depends upon the accuracy of a separate phosphorus determination and, moreover, in samples of high tin and phosphorus contents instances occur in which all the phosphorus is not co-precipitated with the tin.

The gravimetric figures quoted in Table II were obtained as the result of the full gravimetric procedure, whereby the amount of contaminating oxides is determined chemically. The consistency of the volumetric method for tin in a variety of brasses is exemplified in Table IV. In this laboratory the method has been found much more reliable than the gravimetric sulphide procedure. With the strength of standard iodate solution used, the volumetric determination is also preferable for brasses of low tin content.

We wish to thank the Director of Scientific Research, Admiralty, for permission to publish this paper and the Superintending Scientist, Bragg Laboratory, for facilities granted. The help and co-operation of the Staff of this laboratory are gratefully acknowledged.

REFERENCES

1. Bornemann, G., *Chem.-Ztg.*, 1899, **23**, 565.
2. Peters, C. A., *Amer. J. Sci.*, 1900, [IV], **10**, 359.
3. Gooch, F. A., and Ward, H. L., *id.*, 1909, [IV], **27**, 448.
4. Evans, B. S., ANALYST, 1931, **56**, 171.

BRAGG LABORATORY, BIRMINGHAM SECTION. *February*, 1944

DISCUSSION

Mr. F. L. OKELL said that separation of the chief interfering substance by precipitation, leaving tin in a solution suitable for reduction by nickel, was an innovation in bronze analysis and an outstanding

contribution to the subject that would be much appreciated by those with experience of the determination. Nickel reduction would appear to be an important part of the method, since it would remove remaining sources of interference, *e.g.*, antimony, before the iodimetric finish.

Mr. GAILER, in reply to a question about the solution of tin bronzes in 1 : 1 nitric acid to give a clear solution which upon boiling yielded a ppt. of metastannic acid, said that the explanation of this depended upon the fact that the metastannic acid, originally formed, was soluble in the 1 : 1 nitric acid. Upon heating, however, transformation to the beta form took place, and this was insoluble in the nitric acid.

In reply to the question of what happened to lead and zinc in the oxalate separation, he was of opinion that the bulk of the lead was precipitated with the copper and the small amount of lead remaining in solution did not affect the subsequent determination of the tin. As regards zinc, it had been found that the oxalate was not pptd. under those conditions or even in presence of small amounts of acetic acid when copper was absent. In presence of copper, however, a certain amount of zinc was probably co-pptd. with the copper oxalate. Their tests showed that most of the zinc remained in solution even under such conditions.

As regards the analysis of tellurium-copper alloys, they had applied the oxalate procedure to separate the copper. An aliquot portion of the solution, after the separation of the copper, was evaporated, treated with phosphoric acid and gum arabic, reduced with sodium hypophosphite and finally titrated with standard iodine soln. The procedure after the copper separation was essentially that of Evans (ANALYST, 1942, **67**, 346), who had recently published a method for selenium, tellurium and arsenic in copper. The present authors would like, however, to stress the point that the method was only tentative as yet, although quite good results had been obtained in preliminary work. The interference of selenium had not been considered so far as they have gone, but the complete Evans procedure would eliminate any effect of that element.

The Analysis of Sour Milk

BY F. J. MACDONALD

LITTLE improvement has been effected in the chemical examination of sour milks since the maceration process was first devised by Dr. James Bell and discussed by Thorpe[1] (1905) and by Richmond and Miller[2] (1906).

The actual maceration process for the separation of the fat and the solids-not-fat is somewhat tedious, and methods for the determination of the decomposition products seem capable of improvement. Determination of ammonia by the official method involves the preparation of fresh standards on each occasion, whereas by the use of a suitable technique the determination may be made in a B.D.H. Nessleriser with permanent colour discs. The separation of alcohol from the milk by the usual distillation method normally requires considerable attention, and is, on occasion, a rather speculative procedure. The evaporation method for the determination of volatile acids is accurate only when the volatile acidity lies between 0·1 and 0·2% as acetic acid; moreover, the end-point of the titration is inclined to be indefinite.

The present work represents an attempt to overcome some of these difficulties, and is put forward in the hope that it may suggest further investigation of this problem. The method is suitable for all milks except those in which considerable proportions of butyric acid have been formed; it requires about 15 ml of milk for a complete analysis.

DETERMINATION OF FAT AND UNCORRECTED S.N.F.—Dry a 150-ml conical flask, A, and a plug of non-absorbent cotton wool (*ca.* 0·4 g) for 1 hr. at 98°/100° C., cool in a desiccator, and weigh. Set the plug aside, re-weigh the flask, introduce 10·3 ± 0·05 g of the milk, and re-weigh. Add 50 ml of *n*-heptane (b.p. 99°/100° C.) and fit the flask to a Dean and Stark receiver and condenser. Fill the receiver, which is graduated up to 10 ml and fitted with a tap, with the same solvent. Boil the mixture for 2 hr. on a hot-plate, at the end of which time the moisture, volatile acids and alcohol will have collected in the receiver and practically the whole of the fat will be dissolved in the *n*-heptane. Set the receiver aside for the subsequent determination of volatile acids and alcohol. Decant the solution of fat from flask A into a previously dried and tared flask, B, and recover the solvent by distillation. Next, by grinding the remaining solids in flask A with 6–8 successive 25-ml portions of light petroleum (b.p. 40/60° C., dried over $CaCl_2$), which are filtered through the cotton plug fitted in a 4-cm glass funnel, transfer the remainder of the fat to flask B and rinse the funnel and plug with a little light petroleum; remove the light petroleum on the water-bath, and dry the flask B and fat to constant weight at 98°/100° C. Remove any solids adhering to the glass rod used for completing the separation of the fat by wiping with the cotton plug moistened

with a few drops of 50% alcohol, put the plug into flask A with the solids-not-fat, and dry at 98°/100° C. until constant in weight.

By this process efficient separation of the fat and solids-not-fat is effected. No re-extraction of the separated fat is necessary. Although there is slight browning of the solids, this does not appear to affect the accuracy of the method.

DETERMINATION OF VOLATILE ACIDS—Add water to the aqueous distillate in the receiver to bring the vol. to 10 ml and, after mixing, run the watery layer into a dry test-tube. Titrate 5 ml with $N/10$ sodium hydroxide, using 5 drops of 0·5% phenolphthalein soln. as indicator. Calculate the result as % of acetic acid.

This is applicable to the bulk of sour milks encountered, but, as mentioned above, is not suitable for those in which appreciable proportions of butyric acid have developed. Acetic and propionic acids are recovered quantitatively when present in the milk in total proportions up to 1%, whether singly or together. For concentrations of total volatile acid up to 0·2%, the maximum error involved in the ultimate S.N.F. correction by the assumption that all of the acid is acetic would be of the order of 0·1%. With concns. higher than this, advantage may be taken of the oxidation method of Fyleman[3] for the determination of the higher member of the series, the lower being obtained by difference.

DETERMINATION OF ALCOHOL[4]—Allow a further portion of the aqueous distillate to react at room temp. with a nitric acid soln. of dichromate, and measure in a Lovibond Tintometer the resultant decrease in yellow units caused by the reduction of the dichromate. The presence of acetic or propionic acid in concns. up to 1% does not interfere under the experimental conditions. Pipette 1 ml of the aqueous distillate into a stoppered test-tube graduated at 10 ml. Add 2 ml of a soln. of potassium dichromate in nitric acid (0·25% in HNO_3 1 : 2 v/v), mix and leave for 24 hr. at laboratory temp. Allow a further 1-ml of the aq. distillate to react with 2 ml of potassium dichromate soln. (1·25% in HNO_3 1 : 2 v/v) in a 50-ml graduated flask under the same conditions as above. After the reaction is complete (24 hr.) make the solns. up to vol. and record the yellow units in a 1-cm cell in a Lovibond Tintometer. By the use of these two concns. of dichromate from 0 to 1 mg of alcohol per ml and 1 to 5 mg of alcohol per ml may be estimated respectively, the proportions of alcohol normally encountered being within these limits.

The graphs relating alcohol to yellow units are quite smooth, and pass through the following points:

Potassium dichromate 0·25%						
Alcohol in mg ..	0	0·2	0·4	0·6	0·8	1·0
Lovibond yellow units	7·0	5·3	4·0	2·9	2·0	1·4
Potassium dichromate 1·25%						
Alcohol in mg ..	0	1	2	3	4	5
Lovibond yellow units	7·0	5·3	3·8	2·5	1·5	0·7

Satisfactory recoveries of alcohol, acetic acid and propionic acid were obtained by the above methods from mixtures of the three in various proportions, both from aqueous solution and from fresh milk to which additions had been made.

DETERMINATION OF AMMONIA—Conway's[5] method provides a useful means of determining ammonia in the proportions normally encountered in sour milks. Pipette 1 ml of N hydrochloric acid into the central chamber of a Conway unit, whose rim has been smeared with vaseline, and introduce 2 ml of milk into the outer chamber. Add 1 ml of a sat. soln. of potassium carbonate, quickly replace the lid, and leave the unit for 2 hr. at laboratory temp. Then by means of a fine-pointed pipette transfer the acid to a 100-ml graduated vessel, rinse the central chamber 3 or 4 times with ammonia-free water, and add the washings to the same flask. Make up the mixture to volume, dilute a suitable aliquot portion to 50 ml in a Nessler cylinder, add 2 ml of Nessler reagent, and match the colour against a permanent colour disc in a B.D.H. Nessleriser.

CORRECTIONS OF S.N.F. FOR DECOMPOSITION PRODUCTS[6]—*Alcohol*—For each 184 parts of alcohol add 342 parts.

Ammonia—For each part of ammonia add 5·2 parts.

Volatile Acids—For each 60 parts of acetic acid add (*a*) 60 parts to correct for removal of acetic acid from solids during distillation; (*b*) 25·5 parts to correct for decomposition of lactose during souring.

For each 74 parts of propionic acid add (*a*) 74 parts to correct for removal of propionic acid from solids during distillation; (*b*) 68·5 parts to correct for decomposition of lactose during souring.

Table I gives the results of the analysis of 20 samples of sour milk, corrected according to the above scheme. Nos. 7, 8, 13 and 14 were homogenised milk; Nos. 10 and 16 watered milk. When No. 16 was opened it possessed a distinct smell of alcohol, and a suitably diluted portion of the distillate was used for the determination of this constituent.

TABLE I

Sample No.	Fat, %			S.N.F., %		Alcohol		Volatile acids, %			Ammonia		Sour S.N.F., %		
	Original	Sour	Difference	Original	Sour (uncorrected)	%	Correction	Acetic	Propionic	Correction	%	Correction	(corrected)	Difference	Age in days
1	3·41	3·28	−0·13	8·85	8·51	0·065	0·12	0·114	—	0·16	0·031	0·16	8·95	+0·10	28
2	3·45	3·44	−0·01	8·82	8·63	0·034	0·06	0·102	—	0·14	0·036	0·19	9·02	+0·20	28
3	2·79	2·71	−0·08	8·90	8·52	0·055	0·10	0·114	—	0·16	0·012	0·07	8·85	−0·05	34
4	3·38	3·28	−0·10	9·10	7·81	0·055	0·10	0·212	0·310	0·90	0·013	0·07	8·88	−0·24	34
5	3·43	3·39	−0·04	8·79	8·67	0·048	0·09	0·108	—	0·16	0·008	0·04	8·96	+0·17	36
6	3·72	3·75	+0·03	9·12	8·79	0·066	0·12	0·120	—	0·17	0·004	0·02	9·10	−0·02	54
7	3·54	3·43	−0·11	8·65	8·19	0·072	0·13	0·120	—	0·17	0·008	0·04	8·53	−0·12	54
8	3·46	3·40	−0·06	9·17	8·86	0·072	0·13	0·144	—	0·20	0·010	0·05	9·24	+0·07	54
9	3·31	3·26	−0·05	8·94	8·48	0·071	0·13	0·108	—	0·16	0·008	0·04	8·81	−0·13	28
10	2·78	2·71	−0·07	8·01	7·61	0·073	0·14	0·120	—	0·17	0·020	0·10	8·02	+0·01	28
11	4·06	3·97	−0·09	9·17	8·97	0·015	0·03	0·120	—	0·17	0·012	0·06	9·27	+0·10	28
12	4·13	3·95	−0·18	9·19	8·91	0·063	0·12	0·108	—	0·16	0·006	0·03	9·22	+0·03	33
13	3·13	3·08	−0·05	9·00	8·47	0·069	0·13	0·108	—	0·16	0·012	0·06	8·82	−0·18	32
14	3·21	3·14	−0·07	8·85	8·47	0·020	0·04	0·102	—	0·14	0·015	0·08	8·73	−0·12	38
15	3·31	3·17	−0·14	8·82	8·13	0·063	0·13	0·226	0·150	0·61	0·010	0·05	8·92	+0·10	40
16	2·73	2·68	−0·05	7·36	4·69	1·23	2·23	0·084	—	0·12	0·013	0·07	7·11	−0·25	48
17	2·08	1·95	−0·13	8·92	8·54	0·085	0·15	0·120	—	0·17	0·002	0·01	8·87	−0·05	41
18	2·54	2·55	+0·01	9·19	8·78	0·053	0·10	0·096	—	0·14	0·001	—	9·02	−0·17	41
19	3·32	3·11	−0·21	8·92	8·61	0·044	0·08	0·108	—	0·16	0·003	0·02	8·87	−0·05	38
20	3·22	3·10	−0·12	9·01	8·84	0·056	0·10	0·060	—	0·09	0·008	0·04	9·07	+0·06	38

The fat (on the fresh milks) was determined by the Röse–Gottlieb method, and the total solids by the method described by Elsdon and Walker,[6] the S.N.F. being obtained by difference. Estimation of the fat of fresh milks by the new method described above was not completely satisfactory in every instance, repeated extractions with light petroleum being necessary to obtain complete separation. It would appear that the protein associated with the fat globules in the fresh milk tends to prevent free penetration by the solvent under the experimental conditions, but that it is sufficiently denatured during the process of souring to lose this property.

TABLE II

Sample No.	Fresh milk			Sour milk						Differences	
	Sp.gr. at 15·5° C.	Fat (Gerber) %	S.N.F., %	3 *N* Ammonia %	Sp.gr. at 15·5°C.	Corr. Sp.gr. at 15·5°C.	Fat (Gerber) %	Fat (corr.) %	S.N.F., %	Fat	S.N.F.
1	32·1	3·40	8·84	5·7	29·1	32·2	3·20	3·40	8·87	nil	+0·03
2	32·0	3·45	8·83	4·8	29·5	32·1	3·25	3·40	8·84	−0·05	+0·01
3	32·8	2·80	8·90	5·2	30·4	33·2	2·55	2·70	8·98	−0·10	+0·08
4	33·2	3·40	9·12	4·8	30·1	32·7	3·25	3·40	8·99	nil	−0·13
5	31·8	3·45	8·78	5·2	29·1	32·0	3·25	3·41	8·82	−0·05	+0·04
6	32·9	3·70	9·10	4·8	30·0	32·6	3·40	3·60	9·01	−0·10	−0·09
7	31·3	3·55	8·68	5·7	28·4	31·5	3·20	3·40	8·70	−0·15	+0·02
8	33·3	3·45	9·15	5·7	29·2	32·3	3·20	3·40	8·99	−0·05	−0·16
9	32·6	3·30	8·95	4·8	29·7	32·3	3·15	3·30	8·87	nil	−0·08
10	29·1	2·95	8·00	4·8	26·2	28·8	2·70	2·85	7·91	−0·10	−0·09
11	32·6	4·35	9·16	5·7	29·6	32·7	3·85	4·05	9·12	−0·30	−0·04
12	32·8	4·15	9·17	4·8	29·2	31·8	3·80	4·00	8·89	−0·15	−0·28
13	32·9	3·15	8·99	6·6	28·2	31·8	3·00	3·20	8·73	+0·05	−0·26
14	32·3	3·20	8·85	5·7	29·0	32·1	3·00	3·20	8·80	nil	−0·05
15	32·1	3·30	8·82	5·2	29·0	31·9	3·05	3·20	8·76	−0·10	−0·06
17	33·5	2·10	8·93	4·8	30·1	32·7	1·90	2·00	8·71	−0·10	−0·22
18	34·2	2·55	9·20	4·8	30·9	33·5	2·50	2·60	9·03	+0·05	−0·17
19	32·5	3·30	8·92	3·8	29·8	31·9	3·15	3·30	8·78	nil	−0·14
20	32·9	3·20	9·00	5·2	30·1	33·0	3·00	3·15	9·02	−0·05	+0·02

DETERMINATION OF FAT AND S.N.F. BY TREATMENT WITH AMMONIA—For routine purposes where the complete determination of decomposition products is not required we

have found the following process to be of considerable use, and it might be of interest to outline the method adopted in our laboratories. Add to the whole sample sufficient of a measured volume of 3 *N* ammonia to provide a slight excess as judged by the odour. Mix well, but gently, avoiding the formation of an emulsion, and leave overnight at room temp. As a rule the milk is then in a suitable condition for examination, but with more obstinate samples resort may be had to gentle warming. Measure the vol. of the mixture and calculate the % by vol. of 3 *N* ammonia.

Take the sp.gr. of the mixture by means of a lactometer or Sprengel tube and apply a correction of +0·55 of a lactometer degree for each 1% of 3 *N* ammonia present. In most samples the fat may be determined by the Gerber method, but where much butyric acid has developed the Werner–Schmid process may be employed; multiply the result thus obtained by the appropriate factor to correct for the dilution of the original sample by the added ammonia. Calculate the S.N.F. figure by the Richmond formula.

The results for 19 of the 20 samples examined above by the proposed gravimetric method are shown in Table II and are typical of some hundred samples so examined over a period of several years.

SUMMARY—(1) A method is described whereby the fat and S.N.F. (uncorrected) of sour milk may be separated and determined, while volatile acids and alcohol are recovered in the form of an aqueous solution.

(2) Methods are described for the determination of the volatile acids, alcohol and ammonia, required for the application of corrections to the S.N.F. obtained in (1).

(3) The routine determination of fat and S.N.F. of sour milk by treatment with ammonia is described.

I wish to thank Mr. B. Crowhurst for assistance in this work, and the Governing Director of the Express Dairy Co., Ltd., for permission to publish.

REFERENCES

1. Thorpe, T. E., ANALYST, 1905, **30**, 197.
2. Richmond, H. D., and Miller, E. H., *Id.*, 1906, **31**, 317.
3. Fyleman, E., *J. Soc. Chem. Ind.*, 1924, **43**, 142T; Abst., ANALYST, 1924, **49**, 396.
4. Snell, F. D. and C. T., "*Colorimetric Methods of Analysis*," Van Nostrand, New York, 1937.
5. Conway, E. J., *Biochem. J.*, 1933, **27**, 419.
6. "*Richmond's Dairy Chemistry*," Griffin, London, 1942.

EXPRESS DAIRY CO., LTD.
CLAREMONT ROAD, LONDON, N.W.2

February, 1944

The Softening-point of Fats

BY C. R. BARNICOAT, M.SC., PH.D., F.R.I.C.

OWING to the wide range of constituent fatty acids in butter fat, its melting-point is one of the most difficult to determine with consistent results (particularly between different workers).

In connection with a series of investigations on New Zealand butter fat, it was necessary to determine the m.p. as a routine procedure, and considerable attention was given to available methods—capillary tube, "slip" point, floating pellet on mercury, setting-point (modified Dalican's titre test), and Wiley's standard A.O.A.C. method.[1] All except Wiley's method appeared to be unreliable owing to their doubtful end-points. Wiley's method gave very reproducible results but was very tedious. The softening-point determination (based on the ring and ball method for bitumen products[2]) has given satisfactory results in this Institute for some years past, yielding values close to those obtained with the standard Wiley technique. The method is simple, the results are reproducible and several determinations can be carried out simultaneously.

METHOD—Put 0·5 ml of mercury in a thin-walled (5 × 1 cm external diam.) lipped test-tube, and cool the tube and contents for some minutes in finely-cracked ice and water. Above the mercury introduce 1 ml of melted fat; this rapidly solidifies, but leave the tube in the ice-water for about 30 min. longer, and then, preferably in a refrigerator, overnight.

For the actual softening-point determination, all that is required is a large beaker of water, a thermometer, to which is attached a metal plate with a number of holes (we use 8), in which the tubes of fat are suspended by the lips. Place a $\frac{1}{8}$ in. ball-bearing in each tube on the fat, in the depression which always forms when the fat cools. The fat column must be

level with the thermometer bulb. Immerse the tubes in water at about 20° C. for 30 min., and then raise the bath temp. at the rate of 0·5° C. per min., the water in the bath being stirred vigorously by means of an air current. Within a degree or two of the softening-point the fat begins to clear, and the temperature is recorded when the ball-bearing has fallen half-way through the fat column. At this temp. the fat will not be quite clear. Actually, the temperature-range from the beginning to end of the passage of the ball-bearing through the fat-layer is less than 1° C. Between workers, independently determined duplicate tests should not vary by more than ±0·2° C., and the agreement is usually closer.

Many experiments were made to test the relative importance of different stages of the manipulation on the accuracy and reproducibility of the results. The main factors considered were:

(1) *Rate of Heating*—The results for 2 samples of butter-fat are shown in Table I. The higher value recorded for the most rapid heating is due to lag, and emphasises the necessity of keeping closely to standard conditions in raising the temperature of the water-bath.

TABLE I

Rate of heating per min., °C. ..	0·2°	0·5°	0·8°
Softening pt. of butter-fat, No. 1 ..	32·0°	32·4°	32·6°
,, ,, ,, No. 2 ..	30·9°	31·3°	31·5°

(2) *Temperature for melting fat samples*—A series of results with different samples of butter fat heated in the oven at temps. from 40° to 100° C. showed that the temp. at which the fat is melted is not important. A temperature of *ca.* 60° C. has been found convenient for melting, "oiling-off," and filtering butter fat.

(3) *Storage of prepared tubes*—Holding the solidified fats in their tubes in a refrigerator for long periods has no effect on the results.

(4) *Oxidation of the fat*—Oxidation of butter fat tends to lower its softening-point. Thus two butter fats with initial softening points 32·4° and 31·3° C. gave, after oxidation to the bleached "tallowy" stage, 32·1° and 31·0° C., respectively.

(5) *Rate of cooling.*—As was to be expected this is of major importance (see Table II).

TABLE II

Conditions of cooling	Softening pt., °C. No. 1	Softening pt., °C. No. 2	Conditions of cooling	Softening pt., °C. No. 1	Softening pt., °C. No. 2
As recommended	32·4	31·3	Ice, 10 min.	32·5	31·3
In air (*ca.* 15° C.) 7 hr. ..	32·4	31·2	,, 2 hr.	32·5	31·3
,, ,, 24 hr. ..	32·4	31·2	,, 7 hr.	32·4	31·3
Refrigerator (0°–5° C.) 7 hr.	32·4		Ice and salt, 10 min. ..	31·8	30·9*
,, ,, 24 hr.	32·4		,, ,, 1 hr. + 6 hr. refrigeration	31·9	30·8*

Conditions of cooling	No. 3	No. 4	No. 5
As recommended	33·1	32·0	32·7
Cooling very slowly (45°–13° C.) overnight ..	28·6	29·0	28·5

* Fats had glassy appearance.

Too rapid or too slow cooling thus tends to give low softening-points.

The foregoing results give the impression that a wide range of cooling conditions has no effect on the result, but in practice, anomalous values were occasionally found (due, apparently, to slow cooling caused by higher air temperatures than those recorded in Table II) before the standard procedure of cooling in ice and water was adopted.

While the method was not satisfactory with a "sharply-melting" fatty ester (methyl stearate, m.p. 38° C.), it has proved useful for certain other fats and hydrogenated products and also for tropical butter-spreads with unusually high melting-points. The method is suitable more particularly for those fats with wide melting-ranges and indefinite melting points, which are difficult to determine by previous methods.

I am grateful to Mr. G. A. Cox, M.Sc., of this Institute, for his helpful criticism.

REFERENCES

1. Wiley, "*A.O.A.C. Methods of Analysis*," 4th Ed. (1935), p. 407.
2. I.P.T. Serial Designation A.20.

DAIRY RESEARCH INSTITUTE (N.Z.)
DEPARTMENT OF SCIENTIFIC AND INDUSTRIAL RESEARCH
PALMERSTON NORTH, NEW ZEALAND

March, 1943

Discussion

Mr. K. A. Williams has sent the following communication: The method is an ingenious development from the ring and ball method used for bitumen products and bears some resemblance to a method due to Jean and described, without specific reference to the literature, by Lewkowitsch (*Chem. Tech. and Anal. of Oils, Fats and Waxes*, 6th Ed., Vol. I, p. 327). It is straightforward and simple to carry out, and should give the reproducible results the author claims. It will give as much information as the Wiley standard A.O.A.C. method, or the Ubbelohde method, standardised in the B.P. and by the B.S.I., but it gives no more information, as it determines only a single temperature at which the fat acquires a definite degree of softness. Melting-points are of most use in arriving at some idea of the manner in which a fat melts, that is, the range of temperature over which melting is spread, and the degree of softening at each point in this range. Neither of these factors is necessarily related to a specific "softening-point," even with a single fat such as butter-fat. It would therefore appear that the author has allowed the needs of reproducibility in routine analysis to dominate those for the fullest possible information about a sample, and has thereby limited the value of his test to that of the two standard tests already mentioned. Nevertheless it has certain advantages over them in ease of application or in avoiding the need for special apparatus. The author rightly emphasises the need for great care in preparing the sample for the determination, and, while the points to which he draws attention are not new, they can hardly be stressed enough.

A Cell for the Performance of Amperometric Titrations

By M. A. FILL and J. T. STOCK, B.Sc., A.R.I.C.

(*Read at the Meeting, April* 5, 1944)

During recent years Kolthoff *et al.*[1] have developed the technique of *amperometric titration*, based on polarographic technique. Although the number of examples so far studied is not large, the procedure appears promising, being very suitable for dilute solutions. Further, the only expensive piece of apparatus required is a suitable galvanometer or other means of measuring small currents.

Since polarographic methods measure *concentrations* and not total quantities, the use of small volumes renders possible the estimation of very small amounts of substances. In most of the amperometric titrations so far described vols. of *ca.* 40–50 ml have been used. The cell here described permits the ready titration of 5-ml portions and has certain other advantages, simplicity of construction being prominent among them.

The cell is shown in Fig. 1. Its construction is self-evident. The outer portion is made

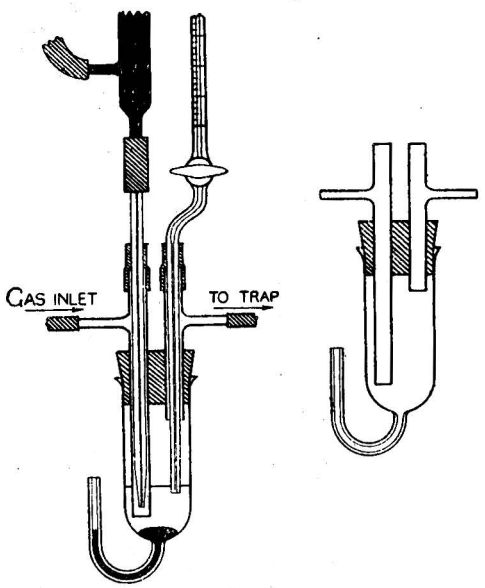


Fig. 1

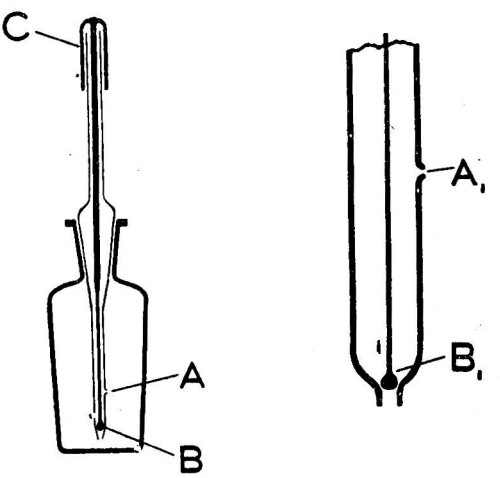


Fig. 2

from ordinary 1-in. diam. Pyrex boiling tube. The straight-through portions of the T-pieces are so constructed that the dropping mercury electrode and the jet of the micro-burette have a clearance of about 0·5 mm within them. The annular spaces thus formed are ample

for the entry and egress of the stream of nitrogen or hydrogen used initially to remove dissolved oxygen and subsequently to stir the solution after each addition of reagent. The longer T-piece effectively protects the tip of the dropping electrode from accidental damage. Short jointing sleeves of rubber tubing[2] permit ready removal of the dropping electrode and burette when desired.

A mercury pool is used as quiet electrode. It was shown by Majer[3] that, in presence of halide ions, the pool remains at constant potential, provided that its area is at least 1 cm². In this cell a small amount of mercury (1–1·5 ml) gives a pool of ample area. It is our practice to measure the requisite quantity of mercury into the cell by means of the suctionless pipette shown in Fig. 2, a device we have found useful for handling other (particularly corrosive) liquids; using a pool of definite size facilitates positioning of the dropping electrode, etc., to permit titration of small volumes.

Removal of dissolved oxygen is rapid. With 5 ml of soln., passage of gas at the rate of 2 bubbles per sec. for 5 min. is sufficient. The titration is then carried out as described by Kolthoff and Lingane.[1] When the gas stream is passing, the agitation considerably increases the flow of current through the soln., with consequent risk of damage to the galvanometer. In the apparatus described, the soln. is forced away from the dropping electrode during the passage of the gas, thus automatically breaking the circuit. Small impulses of current due to the falling of the charged mercury drops are all that is observed. When the stream of gas is stopped the soln. rises to cover the tip of the electrode.

Titrations of copper with α-benzoinoxime, as described by Langer,[4] and of nickel with dimethylglyoxime reported by Neuberger[5] and investigated much more fully by Kolthoff and Langer,[6] were carried out with the apparatus.

Solutions (0·01 *M*) of copper sulphate and nickel ammonium sulphate were standardised gravimetrically. The supporting electrolyte solns. were examined polarographically for reducible impurities.

Solutions of 0·01 *M* α-benzoinoxime in 50% alcohol and of 0·02 *M* dimethylglyoxime in 95% alcohol were prepared and standardised by amperometric titration of the copper and nickel solutions respectively, the larger scale apparatus described by Kolthoff and Langer[6] being used.

The characteristics of the capillary used in this work (measured in 0·1 *M* potassium chloride at 25° C.) were $m = 0{\cdot}917$ mg sec.$^{-1}$, $t = 3{\cdot}39$ sec. The symbols are those of Kolthoff and Lingane.[1]

The copper or nickel soln. was measured by means of a 1-ml micro-burette into 5-ml portions of supporting electrolyte contained in the cell. After removal of dissolved oxygen by passing a stream of hydrogen the titration was carried out with the use of a similar burette. The current was measured by a calibrated Tinsley galvanometer* with enclosed lamp and scale and was corrected for the diluting effect of the titrant.[1] Some of the results appear in Tables I and II.

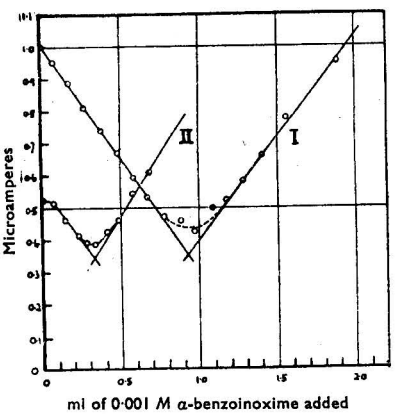


Fig. 3

TABLE I

Copper solution added to 5·00 ml of supporting solution of 0·1 *M* ammonium chloride —0·02 *M* ammonia containing 0·03% gelatin. Titrated with 0·01 *M* α-benzoinoxime solution. Applied voltage, −1·7. T = 16–17°

Copper taken, mg. ..	0·631	0·316	0·195	0·064	0·022
,, found, ,, ..	0·626	0·317	0·180	0·061	0·022
Error	−0·005	+0·001	−0·015	−0·003	0·000

TABLE II

Nickel solution added to 5·00 ml of supporting solution of 0·5 *M* ammonia —0·1 *M* ammonium chloride containing 0·03% gelatin. Titrated with 0·02 *M* dimethylglyoxime solution. Applied voltage, −1·6. T = 15–16°.

Nickel taken, mg ..	0·583	0·291	0·145	0·059	0·018
,, found, ,, ..	0·588	0·296	0·140	0·061	0·020
Error	+0·005	+0·005	−0·005	+0·002	+0·002

* This had the following characteristics: Max. sensitivity, 2·03 × 10^{-9} amps. per mm; period, 3 sec.; sensitivity used for most readings, 1/10th; effective resistance of galvanometer and shunt at sensitivity 1/10th, 780 ohms. The means of the galvanometer oscillations were used in calculating the current flowing.

Langer[4] titrated copper in 20 ml of soln. with 0·001 *M* α-benzoinoxime soln., but found that the experimental points were scattered. We found that, when titrating 5-ml portions of the soln. with reagent of this strength, quite reasonable results were obtained, considering the very small amounts of copper (below 0·1 mg) being determined. Fig. 3 shows two of the titration curves actually obtained.

A few experiments, using 0·002 *M* dimethylglyoxime soln., to titrate similar amounts of nickel, were not so successful.

References

1. Kolthoff, I. M., and Lingane, J. J., *"Polarography,"* Inter-science Publishers, Inc., New York, 1941, p. 447.
2. Furman, N. H., *"Standard Methods of Chemical Analysis,"* D. van Nostrand Co., Inc., New York, 1939, 2310.
3. Majer, V., *Collection Czechoslov. Chem. Commun.*, 1935, **7**, 146, 215; 1937, **9**, 360.
4. Langer, A., *Ind. Eng. Chem., Anal. Ed.*, 1942, **14**, 283.
5. Neuberger, A., *Z. anal. Chem.*, 1939, **116**, 1.
6. Kolthoff, I. M., and Langer, A., *J. Amer. Chem. Soc.*, 1940, **62**, 211.

CHEMISTRY DEPARTMENT, UNIVERSITY COLLEGE
SOUTHAMPTON

December, 1943

Notes

A MICROMETER-CONTROLLED BURETTE

IN a review of micro-volumetric apparatus[1] reference was made to a micro-burette in use in this laboratory, in which are incorporated the advantages of several models, but which is based mainly upon the micro-pipette devised by Rosebury and Heyningen.[2] The latter was used for the delivery of accurately known, small volumes of "heavy water" for sp.gr. determinations by the falling-drop method, and was, in turn, a modification of a similar pipette described by Keston, Rittenberg and Schoenheimer.[3]

The present micro-burette (see Fig. 1) was made in the railway workshops; internal parts are of steel,

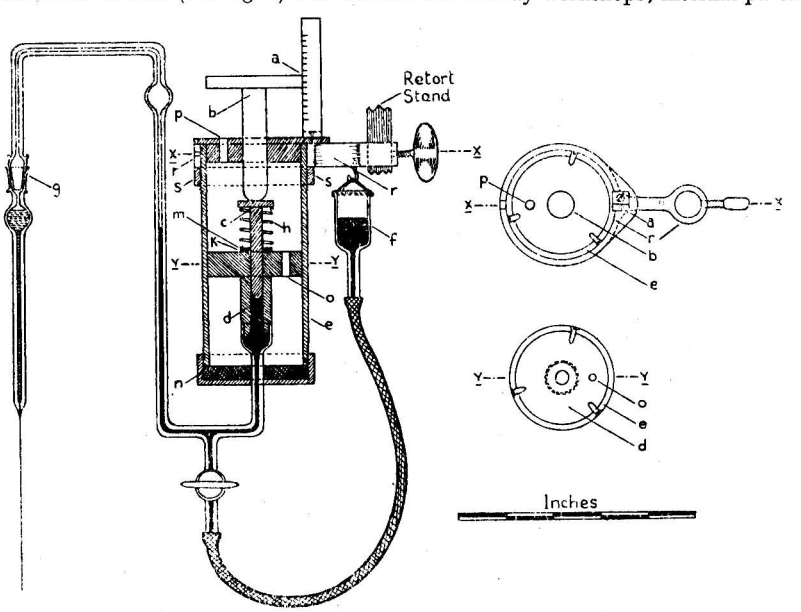


Fig. 1

external parts of stainless steel. It consists essentially of a piston *c* working in a cylinder *d* without lubricant. The cylinder *d* and the capillary glass tube to which it is cemented at the lower end are filled with mercury, which is displaced by the piston. The standard solution is contained in another capillary tube attached to the former by an interchangeable ground joint *g*; its delivery is controlled by the movement of the mercury. Burette readings are observed on the micrometer head *a*, the head being divided into 50 parts and the vertical scale indicating the number of complete turns of the screw *b* in the usual manner. The piston *c* is kept in contact with the micrometer screw by means of a light steel spring *h* (13 turns of No. 18 S.W.G. wire). The joints are made leak-proof in a manner similar to that described by Rosebury and Heyningen[2]: (1) The piston passes through a well-fitting rubber washer* *k* which rests upon the head of

* After 6 months' use the mercury had become fouled through attack by the sulphur contained in the rubber; this could be replaced advantageously by Neoprene.

the cylinder; this washer is protected from movements of the spring by a loose steel washer *m*. (2) The cylinder is cemented to the glass capillary and the joint covered with self-adhering rubber tape*; the joint was made with a paste of plaster of Paris and a chloroform solution of gum benzoin. (3) The lower cap is screwed on to the outer casing *e* and bears on a 1/4-in. thick rubber washer* *n* fitting the capillary tightly. Before being assembled the rubber washers are lubricated at the centre with glycerin. (4) Final sealing of the joints, particularly that at *k*, is effected by filling the outer case *e* with mercury, a hole *o* being provided for its passage through the head of the cylinder *d* and another hole *p* serving as a "breather" in the top of the outer case. The instrument is supported by a ring *r* carrying a thumb-screw for attachment to a retort stand; the ring *r* is secured by a locking ring *s* and has a hole to give access to the screws fixing the top to the case *e*. The bulbs shown in the capillary tube and detachable jet serve to adjust their volumes to that traversed by the piston during its full travel.

The micro-burette is assembled and filled in the following order: (1) Seal the expanded end to the capillary, with the lower cap of the case *e* and the associated washer *in situ*; alternatively, a join may be made in the capillary below the cap. (2) Cement the cylinder *d* in the capillary. (3) After the cement has set fix the cylinder in position and screw up the lower cap. (4) By means of the reservoir *f* fill the capillary and cylinder *d* with mercury to the top of the latter. (5) Place in position the piston *c*, together with the associated spring and washers. (6) Fix the top (carrying the micrometer) to the case and screw down the micrometer until contact with the piston is indicated by movement of the mercury meniscus in the capillary. (7) Almost fill the outer case with mercury.

To fill the burette with solution prior to titration: (1) Raise the piston *c* by means of the micrometer screw to the upper limit of its effective travel. (2) Raise the reservoir *f* (tap open) until the mercury meniscus is near the top of the bulb in the left-hand branch of the capillary. (3) Raise a beaker of the solution so that the tip of the detachable jet is just immersed. (4) Lower *f* until the solution just fills the bulb below *g*. (5) Close the tap communicating with *f*. During the titration the burette jet is immersed in the soln. being titrated; the micrometer screw is turned only in the downward direction, to avoid drawing partly titrated solution into the jet and to avoid possible "backlash" in the screw. The titration should not be performed too rapidly or the air contained in the capillary and bulbs is compressed and delivery of soln. continues after the micrometer screw has stopped turning. Since standard solution does not enter the main part of the instrument, routine cleaning is restricted to the detachable jet; since mercury does not enter this part it cannot react with the standard solution.

The range and sensitivity of the micro-burette obviously depend upon the dimensions chosen for the piston *c* and for the micrometer head respectively. In the instrument here described the micrometer screw has a pitch of 1/40-in. and the piston has a diameter of 0·2476 in., it travels over 3/4-in. The capacity is approx. 0·6 ml and the volume delivered per division of the micrometer head is 0·000395 ml; fractions of a division may be estimated if desired. The accuracy of the burette is dependent upon that of the cutting of the micrometer screw and upon the constant diameter of the piston over its working length, both of which may be guaranteed to approx. ± 0·0002 in.; calibration by the usual methods is therefore unnecessary, the volumes being calculated. Assuming the thermal coefficient of linear expansion of steel to be 11×10^{-6}, a part of the piston having a volume of 0·6 ml at 15° C. has at 25° C. a volume of 0·600198 ml, *i.e.*, the change corresponds to half a division in the micrometer head, and for ordinary purposes no correction is necessary. The air separating the mercury from the standard solution is, however, more highly susceptible to temperature changes, and it is important that these should not be large during the course of a titration; it is recommended that the instrument should be used in a glass case (the micrometer head projecting through an opening in the top) to protect it from heat radiated from the operator. To minimise errors due to a changing hydrostatic pressure, which would alter the volume of this air during titration, only solution contained in the bulb is delivered from the burette.†

L.M.S. Railway Co., Research Department
Chemical Laboratory, Stonebridge Park

G. H. Wyatt
December, 1943

References

1. Wyatt, G. H., Analyst, 1944, **69**, 81.
2. Rosebury, F., and van Heyningen, H. E., *Ind. Eng. Chem., Anal. Ed.*, 1942, **14**, 363.
3. Keston, A. S., Rittenberg, D., and Schoenheimer, R., *J. Biol. Chem.*, 1937, **122**, 227.

Privy Council

STATUTORY RULES AND ORDERS

1944—No. 65. Order, adding Regulation 55G to, and amending the Third Schedule to, the Defence (General) Regulations, 1939. January 20, 1944.§

This Order prohibits, except with the consent of the Minister of Food, the sale by retail (as defined), in any area in England specified in an order made by him, of milk which is not "Tuberculin Tested" or "Pasteurised," or otherwise heat treated, and, in any area in Scotland, so specified, of milk which is not "Certified," "Tuberculin Tested" or "Pasteurised," or otherwise heat treated. There is an exception, in England, for "Accredited Milk" and, in Scotland, for "Standard Milk," derived from a single herd. The Order also contains certain ancillary provisions, notably requirements as to labelling.

---

* See footnote, p. 180.

† Owing to an arithmetical error, this effect is greater than was at first supposed. It may be overcome readily by moving the ground joint *g* (see Fig.) to the upper horizontal portion of the capillary tube and replacing the jet by one made without a bulb from 3 mm-bore tubing, only the fine capillary being vertical.

§ H.M. Stationery Office. Italics signify changed wording.

In its application to Scotland references to the Minister of Health shall be replaced by references to the Secretary of State, any reference to Sec. 68 of the Food and Drugs Act, 1938, by a reference to Sec. 16 of the Food and Drugs (Adulteration) Act, 1928, and any reference to the Milk (Special Designations) Order, 1936, by a reference to the Milk (Special Designations) Order (Scotland), 1936.

1944—No. 66. Order adding Regulation 57AA to the Defence (General) Regulations, 1939. January 20, 1944. Price of Nos. 65-66 together, 2d.

The purpose of this Order is to suspend the operation of Sec. 9 (1) of Gas Undertakings Act, 1920, and to allow an undertaker for the purposes of Sec. 14 of the Gas Undertakings Act, 1934, to average the calorific value of his gas, whatever the amount of the deficiency, over the quarter in which the deficiency occurred.

Ministry of Health

STATUTORY RULES AND ORDERS

— **No. 349. The Heat-Treated Milk (Prescribed Tests) Order, 1944, dated March 23, 1944, made by the Minister of Health under Regulation 55G of the Defence (General) Regulations, 1939.** Price 1d.

This Order restricts the use of such terms as "heat-treated milk," "pasteurised milk," "sterilised milk" or the like calculated to deceive unless (inter alia) the milk has been treated by heat and otherwise, and is in such condition as to satisfy prescribed tests. These tests (a phosphatase test and a methylene blue test) shall be carried out as prescribed in Part I and II of the Schedule respectively. The phosphatase test shall be deemed to be satisfied by milk giving a reading of 2·3 Lovibond blue units or less. In the methylene blue test the milk must fail to decolorise methylene blue in 30 min. under specified conditions.

Ministry of Food

STATUTORY RULES AND ORDERS

— **No. 296. Order, dated March 18, 1944, amending the Feeding Stuffs (Regulation of Manufacture) Order, 1943.** Price 1d.

This Order allows a manufacturer to regard the two quarters Jan./March and April/June as a single period for the purpose of calculating his permitted production of compounds, concentrates, or livestock mixtures. It alters the composition of National Cattle Foods Nos. 1 and 3 by reducing the min. quantity of wheat by-products and "D" flour (*to 20% by weight*) and increasing the min. (*to 15%*) and max. (*to 25%*) quantities of cereals (*including barley*) and dried potato products.

— **No. 297. The Starch Food Powders (Control) Order, 1944. Dated March 18, 1944.** Price 1d.

In this Order, which revokes and re-enacts the Starch Food Powders (Control) Order, 1941, "Cornflour" means starch in powder form suitable for use in the preparation of human food but does not include arrowroot.

Art. 5 prohibits the removal of, addition to, alteration of, defacing or rendering illegible label, ticket or marking on or attached to the wrapper or container of any cornflour, custard powder or blancmange powder, but it shall be a good defence in any proceedings to prove (a) that the article was in the defendant's possession at the time of infringement otherwise than for sale; or (b) that he acted without intent to deceive.

Ministry of Food Press Notice. P.N. 3145a, April 3, 1944, Vinegar.

The Ministry of Food is considering making a maximum prices Order for vinegar. As it will be necessary to define various types of vinegar the Inter-departmental Committee on Food Standards has recommended certain definitions.

The definitions of distilled vinegar and malt vinegar conform to current practice, the Ministry having in 1942 sanctioned the addition of sugar (not exceeding 15% of the sugars present in the infusion) to the wort in the manufacture of malt vinegar.

"Artificial Vinegar" or "Non-Brewed Vinegar" means a solution of acetic acid of edible quality, with or without added colour and/or flavouring matter containing not less than 4% w/v and not more than 8% w/v of acetic acid, $CH_3.COOH$, the acid not being wholly produced by a process of acetous fermentation; but does not include such a solution containing neither colouring nor flavouring matter unless so sold or described as to lead an intending purchaser to believe that he is purchasing a description of vinegar.

"Concentrated Artificial Vinegar" or "Concentrated Non-Brewed Vinegar" means a solution of acetic acid of edible quality with or without added colouring and/or flavouring matter containing not less than 50% and not more than 60% w/v of acetic acid, $CH_3.COOH$, the acid being not wholly produced by acetous fermentation; but does not include such a solution containing neither colouring nor flavouring matter unless so sold or described as to lead an intending purchaser to believe that he is purchasing a description of concentrated vinegar.

"Spirit Vinegar" means the product, with or without colouring matter and containing not less than 4% and not more than 15% w/v of acetic acid, $CH_3.COOH$, obtained by the acetous fermentation of a distilled alcoholic liquid.

"Distilled Vinegar" means the product, with or without added colouring matter and containing not less than 4% w/v of acetic acid, $CH_3.COOH$, obtained by the distillation of malt vinegar.

The Committee state that if the product artificial vinegar were now to be marketed for the first time, they would have regarded "artificial" as a more appropriate description and better calculated to convey to the public the idea of a product which embodies some, but not all, of the properties of malt vinegar. The Committee felt bound to take cognisance of the fact that the description "non-brewed" has been in use for several years, that no instance of a successful Food and Drugs Act prosecution was brought to their notice, and that the description did not excite any adverse comment in the County of London Sessions Appeal Case in 1937 (*cf.* ANALYST, 1937, **62**, 605). They were of opinion that the description "non-brewed" probably conveys no more to the general public than that the product is something different from malt vinegar. In recommending the above definitions they therefore adopted the alternative description "artificial vinegar or non-brewed vinegar."

Similarly for the concentrated products requiring dilution before use the Committee adopted the description "Concentrated Artificial Vinegar or Concentrated Non-Brewed Vinegar." The Committee also recommend that as a safety precaution all such products should be required to be sold at the same strength and be labelled with directions for dilution. It is also suggested that consideration be given to prohibiting their sale in small bottles.

Ministry of Supply

NATIONAL COMPOUND FERTILISERS

Circular No. 29, May 5, 1944.

For the 1944/45 *season a change is made in the standard for National Compound Fertiliser No.* 1 *to* Nitrogen, 7%; P_2O_5, 7%; K_2O, 12%. *The standards for Nos.* 2 *and* 3 *remain as in Circular No.* 37, *June* 22, 1943, *viz.*:

		Nitrogen	P_2O_5	K_2O
National Compound Fertiliser, %	No. 2	9	7·5	4·5
,, ,, ,,	No. 3	6	12	—

Notes from the Reports of Public Analysts

The Editor would be glad to receive reports containing matter of special interest

CITY OF MADRAS: REPORT OF THE PUBLIC ANALYST FOR 1942

THE number of samples analysed under the Madras Prevention of Adulteration Act, 1918, was 1775, as compared with 1990 in the previous year. This falling off was due to the large exodus of the city population during the months of April, May and June, 1942, with the result that sampling was confined to the very few shops that remained open. The figures indicate an improvement in milk, butter and ghee, and adulteration of edible oils continues to be small. The adulterated ghee samples from shops were only 7·2% of those taken, but the corresponding figure for the samples from hawkers was, as usual, 100%. Higher fines were imposed in adulteration cases, but they are still far from deterrent. There was a sudden increase in the adulteration of coffee and coffee tablets, 49 of the 129 samples examined being adulterated.

COFFEE AND COFFEE TABLETS—According to the Act, mixtures of coffee and chicory may be sold, provided that they are labelled with the % of chicory. No other substance may be present even with a declaratory label. Probably owing to the high price of coffee and the unavailability of chicory, prohibited adulterants, such as Bengal gram and mahua flowers, were used. Coffee pericarp was another new adulterant met with during the year. One sample was adulterated with a mixture of coffee pericarp and mahua flowers.

Coffee berry pericarp—This may be identified under the microscope by the characteristic round and oval parenchyma, with spiral bodies from the fibres and bundles of criss-crossed sclerenchyma from the parchment. An estimate of the quantity present may be made by determining the light petroleum extract. Coffee seed gives from 12 to 13% of extract and pericarp less than 2%. The pericarp was almost always associated with immature coffee berries, and it seems probable that the fraud may be due to the use of rejected and immature whole coffee berries for the preparation of the powder rather than to a separate admixture of pericarp.

Mahua flowers—Mahua (*Bassia*) flowers may be identified microscopically by (1) the rounded parenchyma of the soft petals; (2) characteristic spiral vessels; (3) long hairs with one end tapering and a bend at the other; (4) characteristic pollen grains.

Roasted and ground mahua flowers gave the following results (dry basis). Aqueous extract, 67%; total ash, 6·4%; alkalinity of ash ($N/10$ HCl per 5 g), 11·0 ml; light petroleum extract, 1·6%. The flowers thus closely resemble chicory in their chemical characteristics; there is also a pronounced resemblance in taste. An estimate of the quantity present in admixture with coffee may be made from (1) aqueous extract; (2) light petroleum extract; (3) alkalinity of ash; (4) caffeine. For routine analysis, the aqueous extract is sufficient if mahua flowers are the only adulterant present.

PRESUMPTIVE STANDARD FOR BUTTER AND GHEE—An important decision was given by the Madras High Court in a case of butter adulteration. The Court held that since low Reichert–Wollny values,

low saponification values, etc., are possible in abnormal cases, and since no statutory value for Reichert–Wollny value had been fixed by the Madras Government, the presence of an adulterant should be directly proved to sustain a conviction. For these reasons the Court gave the benefit of the doubt to the accused.

As a result of this decision the Madras Government has fixed a statutory presumptive minimum Reichert–Wollny value of 28 for genuine butter and ghee. V. VENKATACHALAM

Legal Notes

The Editor would be glad to receive particulars of cases with points of special legal or chemical interest

"SUBSEQUENT OFFENCE." FUNCTION OF EXPERT WITNESSES

CONCENTRATED FOODS LTD. *v.* CHAMP

ON January 17th, 1944, in the King's Bench Divisional Court, Concentrated Foods Ltd. appealed against a conviction by Justices at Maidenhead on October 13th, 1942, under Sec. 83 (3) of the Food and Drugs Act, 1938, for giving with an article of food a label which was calculated to mislead as to its nature, substance and quality, contrary to Sec. 6 (1) of the Act. The appellants, on May 27th, 1941, had sold an article labelled "Concentrated Cordial Essence: Orangette," with directions as to use, to a wholesaler who on June 4th, 1941, had sold it to a retailer, who in his turn had sold it to a sampling officer on September 4th, 1942. The article consisted of a solution of phosphoric acid flavoured with a little orange oil.

In his judgment, Mr. Justice Laurence said that three points had been argued. First, the appellants had submitted the label to a Public Analyst, Mr. F. W. Edwards, who had approved it. Mr. Edwards was called as a witness before the Magistrates; another Public Analyst, Mr. T. McLachlan, was also called (by the prosecution) and gave it as his opinion that the label was objectionable. The appellants had argued that, as they had taken the opinion of Mr. Edwards before the label was used, the magistrates could not find that they had not discharged the onus of showing that they did not know and could not with reasonable diligence have ascertained that the label was of a character calculated to mislead. No person from the appellant company gave evidence before the magistrates, who had said that they preferred the evidence of Mr. McLachlan to that of Mr. Edwards where such evidence conflicted. The magistrates had, it was evident, addressed their minds to the two questions of fact which were before them, namely, the character of the label and the onus resting upon the appellants. The magistrates had evidently thought that it was material for them to have heard evidence of some officer of the company. It appeared to him therefore that there was ample evidence upon which the magistrates could find as a fact that the appellants had not discharged the onus.

The second question was whether the prosecution was out of time. It appeared to him that it was clear from the words of Sec. 83 (3) that the offence with which the appellants were charged was committed on September 4th, 1942 (*i.e.*, the retail sale) and was not the offence which they actually committed on May 27th, 1941 (the prosecution was not out of time in respect of the offence of September 4th, 1942).

The third point was: was this a "subsequent offence"? This point had apparently already been decided by a Divisional Court, but the case had not yet appeared in the ordinary Reports though mentioned in the *Justice of the Peace* for October 30th, 1943. That case, if properly reported, would be binding upon them, but in any event in his opinion the offence of which the appellants had been found guilty was committed on September 4th, 1942, and that was at a time after the various convictions of the appellants referred to in the case. This was thus a subsequent offence and the amount of the fine was competent to the Justices.

For all these reasons the appeal should be dismissed.

Mr. Justice Lewis concurred and said that in an appeal by way of Case Stated this Court cannot criticise and reverse a finding of fact by the magistrates when they had directed their minds to the proper question, and he was satisfied they had in this case. He referred to *Collins Arden Products Ltd.* v. *Barking Corporation* (1943, 1 King's Bench, p. 419), in which the Court had reversed the decision of the magistrates on the point of law; he distinguished this from the present case, for it was actually a decision under Sec. 3 sub-sec. 1, and the magistrates had not addressed their minds to the defence against the charge under Sec. 6, with which they never dealt properly, or indeed at all.

Mr. Justice Wrottesley said: "I agree. I have only this to add. I notice that considerable argument was based on some evidence which, contrary to the usual practice, found its way into this case—the evidence of a Public Analyst who is said to have been consulted by the limited company, the defendants here, and who is said to have approved the label and to have told the company that the contents of that label were unobjectionable; the argument was therefore this: What then could the company do? That is, it was put, 'reasonable diligence' referred to in Sec. 6. That evidence, having regard to the nature of the label which used words with which we are all familiar, seems to be both irrelevant and inadmissible, anyhow in examination-in-chief. But that question as to whether the label was objectionable meant, of course, whether it offended against the Act. That was a matter not for any expert witness but for the Court. This kind of argument is in my judgment founded on a misconception of the true scope and function of expert witnesses. Experts in these cases are of course chemists. They are there to inform the Court of the composition of the subject matter of the charge. In addition they are, like all expert witnesses, entitled, if asked, to give their opinion based on their examination of the subject matter, provided it is within the scope of their special knowledge. The question whether a label is calculated to mislead means calculated to mislead the public, not the qualified chemist, and so it is a question not for qualified chemists but for the justices, for the Court. Unfortunately, of course, in these cases we find that manufacturers are very prone to use phrases such as 'concentrate,' 'extract,' 'essence,' 'cordial'—phrases which may very well have special meaning to the chemist. It is not surprising, therefore, that a chemist should tell the Court that an article is not his idea of an essence or an extract. But that is not the test. The test is: what does the ordinary man understand by the language? Was he misled? Not whether the chemist was misled but whether the ordinary man was misled. That was pointed out in the case of *Collins Arden Products Ltd.* v. *Barking Corporation*; it was pointed out by Mr. Justice Stable who used the phrase 'the ordinary person.' Nor is

such an enquiry, that is to say, of the analyst, in a case of this kind primarily, in my judgment at any rate, the kind of reasonable diligence referred to or the inquiry referred to in Sec. 6. If, for instance, this firm had chosen to put up something which they called first class phosphoric acid, it might have been a reasonable inquiry of a chemist as to whether this stuff could be called first-class phosphoric acid. That is not the case before these justices. I think in this case these justices took the true meaning of this proviso, that the seller 'did not know, and could not with reasonable diligence have ascertained, that the label was of such a character as aforesaid,' and I am not surprised at their finding that anyone having knowledge of the contents of the bottle in question could have had no doubt that the label was calculated to mislead. That finding of fact disposes of all the defences in this case."

Bibliography on Metals in Foods and Biological Materials

(Supplementing the series published in THE ANALYST up to 1933, **58**, 340, and bringing the Bibliography up to date)

VIII. THALLIUM

ARASHIMA. Influence of Chronic Thallium Poisoning on Function of Female Sex Organs of Rats. *Japan. J. Med. Sci.*, 1933; Abst., *B.C.A.*, 1936, 1148A.

BALTACEĂNU, STANESCU and IOANID. Thallium Poisoning. *Bull. Soc. Méd. Hôp. Bucarest*, 1938, **20**, 261; Abst., *B.C.A.*, 1939, 509AIII.

BERGE and PATZSCH. Poisoning of Partridges with "Zelio" Wheat and Detection of Thallium. *Deut. tierärtzl. Wochschr.*, 1935, **43**, 20; Abst., *C.A.*, 1936, **30**, 5247.

BOHMER. New Observations on Thallium Poisoning. *Deut. Z. ges. gericht. Med.*, 1939, **30**, 270; Abst., *C.A.*, 1939, **33**, 8805.

BORZINI. Influence of Thallium Ions on the Germination of Seeds and on the Initial Growth of Young Plants. *Boll. Staz. patol. vegetale.* [*N.S.*], 1935, **15**, 200; Abst., *C.A.*, 1935, **29**, 5476.

BOSCO. Effect of Thallium on Glutathione in Blood and Tissues. *Riv. Patol. sper.*, 1935, **14**, 385; Abst., *C.A.*, 1936, **30**, 7206. (Application of Tl acetate to skin caused variations in glutathione content of blood.)

BUSCHKE. Biological Action of Thallium. *Dermatol. Z.*, 1938, **77**, 186; Abst., *C.A.*, 1938, **32**, 9282.

BUSCHKE and KONHEIM. Actions of A.T. 10 and of Thallium. *Schweiz. med. Wschr.*, 1939, **69**, 701; Abst., *B.C.A.*, 1939, 1088AIII. (Growth of young rats stunted if treated with Tl.)

BUZZO and GANDOLFO. Poisoning by Thallium Acetate. *Rev. Assoc. Med. Argentina*, 1932, **46**, no. 313, 97; Abst., *C.A.*, 1935, **29**, 1886.

CARBONESCHI. Thallium. *Semana Méd.*, 1940, II, 42; Abst., *C.A.*, 1940, **34**, 6188. (Chemistry, toxicology and method of detctn. discussed.)

CIARAVINO. Toxicity of Thallium. *Arch. di Antrop. Crimin.*, 1936, **56**, 177; Abst., *C.A.*, 1938, **32**, 9276. (Rabbit weighing 3200 g died 12 hours after oral administration of 1 g TlAc.; liver contained 1972 p.p.m., spleen 528, brain 101, intestine 471, muscle 23.)

CRAFTS. Effects of Thallium on Soils. *Hilgardia*, 1936, **10**, 377; Abst., *C.A.*, 1937, **31**, 6790. (Concns. of 100 p.p.m. toxic in most soils.)

DONOVAN. Determination of Thallous Sulphate in Ant Poisons. *J.A.O.A.C.*, 1939, **22**, 411; ANALYST, 1939, **64**, 620.

DOTY. Rat Control. *Rept. Hawaian Sugar Planters' Assoc. Expt. Sta.*, 1940, **60**, 92; Abst., *C.A.*, 1941, **35**, 7060. (Expts. with Tl-poisoned oats described.)

DUMITRESCU-MANTE and CIORAPCIU. Thallium Poisoning. *Med. Klin.*, 1940, **36**, 1091; Abst., *C.A.*, 1941, **35**, 8095.

ESSER. Clinical, Anatomical and Spectrographic Investigations of Central Nervous System in Acute Metal Poisoning with Particular Consideration of their Importance for Forensic Medicine and Tissue Pathology. I. *Deut. Z. ges. gericht. Med.*, 1936, **25**, 239; Abst., *C.A.*, 1936, **30**, 3008. (Tl discussed.)

FREY and SCHLECTER. Thallium Excreted in Various Body Fluids. *Arch. exptl. Path. Pharmakol.*, 1939, **193**, 530; Abst., *C.A.*, 1940, **34**, 3823.

GERBASI and TRAINA. Inorganic Phosphorus and Calcium in Blood Serum of Children Treated with Therapeutic Doses of Thallium Acetate. *Pediatria Riv.*, 1937, **45**, 385; Abst., *C.A.*, 1939, **33**, 4678.

GJERTZ. Case of Thallium Poisoning. *Acta Med. Scand.*, 1935, **85**, 531; Abst., *C.A.*, 1935, **29**, 8142.

HADDOCK. Determination of Traces of Thallium in Presence of other Metals. ANALYST, 1935, **60**, 394. (Methods suitable for application to food and biological material.)

HORN, WARD, MUNCH, and GARLOUGH. Effect of Thallium on Plant Growth. *U.S. Dept. Agr. Circ.*, 1936, **409**; Abst., *C.A.*, 1937, **31**, 2337.

ILJIN. Selective Thallium Moult in Sheep. *Nature*, 1938, **141**, 162; Abst. *Nutr. Abstr. Rev.*, 1938–9, **8**, 251.

ILJIN. Effects of Thallium with Varying Methods of Administration. *Arch. int. Pharmacodyn.*, 1938, **60**, 377; Abst., *B.C.A.*, 1939, 414AIII. (Tl has same depilatory effect in rabbits when given orally, hypodermically, intravenously or applied locally.)

ILJIN. Artificial Thallium Moult in Sheep. *J. Genetics*, 1936, **33**, 305; Abst., *C.A.*, 1938, **32**, 2218.

KLUGE. Toxicological Detection of Thallium. *Z. Unters. Lebensm.*, 1938, **76**, 156; ANALYST, 1938, **63**, 748.

KRSEK. Two Cases of Murder by Thallium. *Cas. lek. cesk.*, 1934, 40; Abst., *C.A.*, 1935, **29**, 4834.

KÜNKELE. Thallium Poisoning. *Chem.-Ztg.*, 1938, **62**, 49; Abst., *B.C.A.*, 1938, 362AIII.

LANGSTEINER. Thallium Poisoning. *Med. Klin.*, 1939, **35**, 980; Abst., *B.C.A.*, 1939, 1088AIII.
LEVKOVICH. Influence of External Temperature, Diet and Emotion on Pharmacological Action of Thallium. *Bull. biol. méd. exptl. U.S.S.R.*, 1938, **5**, 271; Abst., *C.A.*, 1940, **34**, 2461.
MARQUEZ, LAMI and KRAUSS. Thallium Intoxication. *Rev. Med. Cienc. Afines.* (*Buenos Aires*), 1939, 1, No. 8, 8; Abst., *C.A.*, 1940, **34**, 2457.
MUNCH. Human Thallotoxicosis. *J. Amer. Med. Assoc.*, 1934, **102**, 1929; Abst., *C.A.*, 1935, **29**, 3400. (6% of 778 cases of Tl poisoning died.)
OHEIM. Case of Thallium Poisoning. *Deut. Z. ges. gerichtl. Med.*, 1937, **29**, 95; Abst., *C.A.*, 1938, **32**, 3026. (Physiol. effects from ingestion of a prepn. containing 0·7 g Tl_2SO_4.)
PALESTINE. Annual Report of Government Analyst, 1933. Thallium Poisoning. ANALYST, 1934, **59**, 752.
REDFIELD. Thallium Metallicum. *J. Amer. Inst. Homeopathy*, 1937, **30**, 22; Abst., *C.A.*, 1937, **31**, 1496. (Description of physiological and toxic actions of Tl.)
RICHARDS. Analysis of Growth as Illustrated by Yeast. *Cold Spring Harbor Symposia*, 1934, **2**, 157; Abst., *C.A.*, 1935, **29**, 8057. (Traces of Tl decreased rate of sugar utilisation, increased crop growth.)
SAPINO. Thallium XI. Weight and Water Content of Thymus and Thyroid in Chronic Thallium Poisoning. *Arch. intern. Pharmacodyn.*, 1936, **53**, 14; Abst., *C.A.*, 1936, **30**, 7695.
SCHENK. Thallium Poisoning. *Deut. med. Wschr.*, 1939, **65**, 643; Abst., *B.C.A.*, 1939, 616AIII. (Report of a Case.)
SCHNELLER. Toxicological Communications. *Deut. Z. ges. gericht. Med.*, 1935, **25**, 223; Abst., *C.A.*, 1936, **30**, 2524. (Spectrographic detection of Tl in cadavers.)
SCHRADER and KNORR. Thallium Poisoning. *Deut. Z. ges. gericht. Med.*, 1935, **25**, 61; Abst., *C.A.*, 1935, **29**, 8136. (Symptoms attributed to administration of Tl in coffee described.)
SCHWARTE. Poisoning by Thallium Dust. *Munch. med. Wochschr.*, 1939, **86**, 1299; Abst., *C.A.*, 1940, **34**, 1770.
SPENCER. Frenching of Tobacco and Thallium Toxicity. *Amer. J. Bot.*, 1937, **24**, 16; Abst., *B.C.A.*, 1937, 286AIII. (Of 33 toxic elements examined, only Tl caused chlorosis of young leaves.)
SPRADO. Thallium Poisoning. *Deut. med. Wchschr.*, 1938, **64**, 537; Abst., *C.A.*, 1938, **32**, 5499. (Review of clinical manifestations.)
STEIDLE. Toxicology of Thallium. *Med. Welt.*, 1939, **13**, 1557; Abst., *C.A.*, 1940, **34**, 4157.
STIEFLER. Thallium Poisoning. *Wien. klin. Wochschr.*, 1936, **49**, 429; Abst., *C.A.*, 1936, **30**, 7684.
SWAB. Ocular Lesions Resulting from Thallium Acetate Poisoning. *Arch. Ophthalmol.*, 1934, **12**, 547; Abst., *B.C.A.*, 1937, 29AIII.
TESTONI. Thallium VI. Reduced Glutathione in Thallium Acetate Poisoning. VII. Influence of Thallium Salts on Fragility of Erythrocytes for Alkali and Saponins. *Arch. intern. Pharmacodyn.*, 1934, **48**, 1; Abst., *C.A.*, 1935, **29**, 3040.
TESTONI. Thallium VIII. Influence of Thallous Acetate on the Cholesterol Content of the Blood and Suprarenal Glands. IX. Lecithin in Liver, Heart and Kidneys, in Thallous Acetate Poisoning. *Arch. intern. Pharmacodyn.*, 1934, **49**, 144, *Ibid.* 165; Abst., *C.A.*, 1935, **29**, 7492.
TESTONI. Thallium X. Weight and Weight Content of Organs in Chronic Poisoning. *Arch. intern. Pharmacodyn.*, 1935, **51**, 15; Abst., *C.A.*, 1936, **30**, 768.
TESTONI. Effect of Thallium and Lead on Water Exchange of Hair. *Arch. intern. Pharmacodyn.*, 1937, **56**, 195; Abst., *C.A.*, 1937, **31**, 8693.
ULLOA. Pharmacological Action of Metallic Compounds on Haematopoiesis. *Kitasato Arch. Exptl. Med.*, 1936, **13**, **70**. (Fe, Cu, Y, Sn, Hg, Zn, Al, Tl, Sb have only mild effect.)
URABE. Effect of Thallium on Teeth. *Acta Dermatol.*, 1936, **27**, 54; Abst., *Nutr. Abstr. Rev.*, 1936–7, **6**, 732.
URABE. Effect of Thallium on Chemical Composition of Teeth. *Acta dermatol.*, 1936, **27**, 87; Abst., *Nutr., Abstr. Res.*, 1936–7, **6**, 1038. (Tl caused increase in proportion of organic matter in teeth.)
URABE. Action of Thallium on Teeth of Mother and Young in Lactating Rats. *Acta Dermatol.*, 1937, **28**, 18; Abst., *Nutr. Abstr. Rev.*, 1937–8, **7**, 132.
URABE. Effect of Thallium on Teeth. *Acta Dermatol.*, 1937, **30**, 34; Abst., *Nutr. Abstr. Rev.*, 1937–8, **7**, 1024. (Summary of findings and literature.)
USPENSKAYA. Rôle of Thyroid Gland in Thallium Poisoning. *Vertnik Venerol. Dermatol.*, 1939, No. 2–3, 78; Abst., *C.A.*, 1941, **35**, 5569.
USPENSKAYA. Thallium Distribution in Organism. *Bull. biol. méd. exptl., U.S.S.R.*, 1940, **9**, 494. Abst., *C.A.*, 1942, **36**, 162. (Administration of 16 mg/kg body wt. orally produced toxic symptoms.)
WARD. Thallium XIII. Symptoms and Systemic Action on Cattle. *J. Amer. Pharm. Assoc.*, 1936, **25**, 687; Abst., *C.A.*, 1936, **30**, 7221.

J. H. SINGER

Drinking Water Standards in U.S.A.*

THE standards are not rigid and the acceptance of a supply which falls below standard in some respect is left to the discretion of the certifying authority.

COLIFORM ORGANISMS—The alternative methods specified for the determination of *coliform* organisms are those described in *Standard Methods for the Examination of Water and Sewage*, 8th Ed., 1936, as the *Completed test*, the *Confirmed test* (using 2% Brilliant Green bile lactose broth), and the *Confirmed test* (using Crystal Violet lactose broth, Fuchsin lactose broth, or formate ricinoleate broth). Incubation is for 48 hr. at 37° C. If five 10-ml portions of the sample are examined, not more than 10% of the total number of tests per month may give positive results. Also, not more than 5% of positives are allowed if

* *U.S. Publ. Health Repts.*, 1943, **58**, 69–111.

3 or more tubes are positive in one test. If five 100-ml portions of the sample are tested on each occasion, the total number of positive tubes per month must be less than 60% of the total, or not more than 20% if all 5 tubes are positive in one test.

BACTERIOLOGICAL SAMPLING—The frequency of sampling for bacteriological examination, based on a population scale, is to be as follows:

Population served (millions):	0·0025	0·01	0·025	0·1	1·0	2·0	5·0.
No. of samples per month:	1	7	25	100	300	390	500.

TURBIDITY is limited to 10 p.p.m. (silica scale).

COLOUR is limited to 20 (standard cobalt scale).

LIMITS FOR INORGANIC IONS.—Lead 0·1, fluoride 1·0, arsenic 0·05, selenium 0·05, copper 3·0, iron and manganese together 0·3, magnesium 125, zinc 15, chloride 250, sulphate (as SO_4'') 250, total solids preferably 500 and not more than 1000, all as p.p.m. by wt.

PHENOLS should not exceed 0·001 p.p.m. (as $C_6H_5.OH$). These substances (and the inorganic ions) are to be determined by the methods described in *Standard Methods for the Examination of Water and Sewage* or by the A.O.A.C. methods. Alternative acceptable methods are: arsenic as determined by Jacobs and Nagler (*Ind. Eng. Chem. Anal. Ed.*, 1942, **14**, 442; ANALYST, 1942, **67**, 403); fluorides as approved by an American Water Works Committee (*cf. J. Amer. Water Works Assoc.*, 1941, **33**, 1965–2017); and selenium by the method of Robinson *et al.* (*Ind. Eng. Chem., Anal. Ed.*, 1934, **6**, 274). Barium, hexavalent chromium, heavy metal glycosides, and other definitely toxic substances are specifically excluded.

ALKALINITY—The total alkalinity (*T.A.*) of a lime-soda softened water may not exceed the hardness by more than 35 p.p.m. (as $CaCO_3$). In a chemically treated water the caustic alkalinity (as $CaCO_3$) must not exceed $15 + 0{\cdot}4 \times T.A.$, the effect of which is to limit the *p*H to 10·6 approx.

DISCUSSION OF THE STANDARDS—It is suggested that soluble polyphosphates should preferably not exceed 10 p.p.m., and that free chlorine and chloramine might be limited to 1 and 2 p.p.m., respectively. The formulae of Moore (*J. Amer. Water Works Assoc.*, 1939, **31**, 51) have been used to compute the hydroxyl, carbonate, bicarbonate and free carbon dioxide concentrations corresponding with various values for *T.A.* and *p*H; these are shown on a triangular diagram. In addition to various technological matters concerning the protection of sources of supply, treatment and delivery, suggestions for the routine laboratory control of chlorination are made. The minimum residuals suggested are: free chlorine 0·2 p.p.m. for at least 10 min.; chloramine (*o*-tolidine method) 0·4 p.p.m. for 2 hr., or with breakpoint chlorination 0·05–0·10 p.p.m. of chlorine should be found at all points in the distribution system. D. D.

ABSTRACTS OF PAPERS PUBLISHED IN OTHER JOURNALS

Food and Drugs

Determination of Soluble Pectin and Pectic Acid by Electrodeposition. K. T. Williams and C. M. Johnson (*Ind. Eng. Chem., Anal. Ed.*, 1944, **16**, 23–25)—Since soluble pectinous bodies are negatively charged colloids, the possibility of collecting the pectin on the anode of a suitably arranged electrolytic system was investigated. It was found that pectin can be quantitatively determined by electrodeposition provided that the concn. of electrolyte in the soln. is low. Ion-exchange resins can be used when it is necessary to remove electrolytes before electrodeposition. The apparatus consists of a mercury cathode cell constructed from a 250-ml beaker to which a mercury-filled side arm is fitted and a platinum wire is fused into the side. Clean mercury, completely covering the platinum wire in the vessel, serves as the cathode, and the anode is a disc of 45-mesh platinum gauze of 2·5 in. diam. edged with 0·03 in. platinum wire to give rigidity and supported by a 6 in. piece of 0·05 in. platinum wire attached to the centre. This arrangement of mercury cathode with disc anode makes it possible to have all the soln. in the field without stirring. Ion-exchange resin columns are prepared from 24 × 190 mm glass tubing constricted to a 6 mm outlet, and 10 g (drained wt.) of resin supported on a glass-wool mat is used in each tube. The upper cation-exchange column is mounted so that the effluent drips directly into the lower acid-absorbing column. For deposition of pectin from solns. of low ash the procedure is as follows—Dilute an aliquot portion of the soln., containing 5–50 mg of pectin to 58 ml with water in the electrolytic vessel and add 42 ml of 95% alcohol with stirring. The concn. of alcohol is important. Pptn. of pectin at this stage indicates that the concn. of electrolyte is too high and that electrolyte must be removed. During electrolysis immerse the vessel in water and ice to prevent undue rise of temp. After applying a current of 220 volt (5–20 milliamp.) withdraw the anode, immerse it in 99% alcohol for 3 min. and then in anhydrous ether for 3 min. Finally dry the anode at 105° C. for 1 hr. and weigh the deposit. For the removal of electrolytes from solns. having a high ash two commercial resins may be used. The IR 100 resin removes the cations by exchange with hydrogen ion, and the IR 4 resin removes the acids which would interfere by conductance with the electrodeposition. The capacity and regeneration of these resins are described by the manufacturer (Resinous Products and Chemical Co., Philadelphia. "The Amberlites"). Solns. containing 0·4–1·0 mg of pectin per ml are treated as follows—Rinse the columns with two 10- to 15-ml portions of the soln. with thorough drainage between the rinsings. Discard the rinsings and allow the soln. to percolate through the columns at the rate of *ca.* 3 ml per min. Mineral matter is thus reduced to 0·5–4 mg per 100 ml. To determine pectin in fruit (*e.g.*, grape fruit, apple, orange), disintegrate the fruit in a Waring Blendor with addition of enough water and hydrochloric acid to make a thin paste of *p*H 1·7–2·0. Heat the product on the water bath at 85° C. for 30 min., filter through Cellite and treat the residual pulp twice in the same manner. Adjust the combined extracts to a pectin content of *ca.* 1 mg per ml. Treat the soln. with ion-exchange resins, if necessary, and determine the pectin by electrodeposition. None of the extracts

used for investigation contained starch or dextrin, but it is probable that amylolytic treatment before treatment with resins would prevent interference by these substances. Analyses by electrodeposition gave results from 9 to 13% higher than those obtained by the method of Emmet and Carré (*Biochem. J.*, 1926, **20**, 6; ANALYST, 1926, **51**, 307). If, however, fruit extracts were treated with resins and then subjected to the preliminary acetone purification and hydrolysis of the process of Emmet and Carré, the agreement was closer. A. O. J.

Biochemical

Determination of the Iodine Value of Whole Phospholipid. P. L. MacLachlan (*J. Biol. Chem.*, 1944, **152**, 97–102)—The determination of the iodine value of whole phospholipid by the method of Yasuda (*J. Biol. Chem.*, 1931–2, **94**, 401), in which chloroform is used to dissolve phospholipids pptd. with acetone and alcoholic magnesium chloride soln., leads to erratic and variable results, apparently owing to some of the magnesium chloride, often exceeding the phospholipid in amount, being taken up by the chloroform soln. It has now been found that accurate and reproducible results can be obtained when a 1 : 1 mixture of chloroform and ether is used; this mixture (likewise a 2 : 1 or 3 : 1 mixture) does not, however, dissolve the phospholipid as completely as does chloroform. Satisfactory results can also be obtained when the chloroform extract is evaporated to dryness and the residue is re-dissolved in chloroform. The following procedure is therefore recommended. Precipitate the phospholipid (0·3 to 2·0 mg in 1 ml of light petroleum) with 7 ml of acetone and 0·1 ml of a 30% soln. of magnesium chloride in 95% alcohol. Dissolve the phospholipid in two successive portions, 7- and 3-ml each, of chloroform and, after centrifuging, evaporate a suitable aliquot part of the chloroform soln. to dryness and redissolve the phospholipid in chloroform for the determination of the iodine value. F. A. R.

Fat of Sow's Milk. P. B. D. de la Mare and F. B. Shorland (*Nature*, 1944, **153**, 380)—Analytical values for a sample of Berkshire sow's milk (130 ml containing 7·0% of fat, obtained by hand-milking over a period of 1 week) and for pig outer back fat were, respectively: Steam-volatile fatty acids, 2·4, —; myristic acid, 1·8, 1·0; palmitic acid, 28·3, 28·8; stearic acid, 6·1, 11·5; hexadecenoic acid, 8·8, 5·8; oleic acid, 35·0, 46·0; octadecadienoic acid, 14·0, 5·7; C_{20-22} acids, 3·6, 1·2 mols.-% (*cf.* Laxa, ANALYST, 1931, **56**, 321). The palmitic acid content is within the range (28–30 mols.-%) considered to be typical of animal depot fats, as found by other workers for buffalo and camel milk fats, but not for sheep, goat or cow milk fats. In comparison with butter fat it seems likely that sow's milk fat contains more octadecadienoic acid, the assimilation of which from the diet of the suckling pigs is a reasonable explanation of the increase in the iodine val. of pig depot fat from birth until weaning. It is unlikely that the large amount of polyethenoid C_{18} acids present was assimilated by the sow from a maize and barley meal diet, since the C_{18} acid from these consists mainly of linolic acid, of which evidence only of traces was obtained. The high unsaturation of the C_{18} fractions may arise from pasture lipids which are known to contain an octadecadienoic acid isomeric with linolic acid (*cf.* Smith and Chibnall, *Biochem. J.*, 1932, **26**, 218). However, linolenic acid, a major constituent of pasture lipids, was not detectable in the sow's milk fat. J. G.

Use of *Streptococcus lactis* R for the Measurement of Folic Acid. T. D. Luckey, G. M. Briggs and C. A. Elvehjem (*J. Biol. Chem.*, 1944, **152**, 157–167)—The following method has proved reliable for the estimation of folic acid. Prepare inocula of *Streptococcus lactis* R directly from a stab culture into a sterile inoculum medium, consisting of the basal medium plus 200μg of "solubilised liver" per 10 ml, and incubate for 8–30 hr. at 30° C. Prepare a standard curve using graded amounts of from 0 to 300μg of "solubilised liver." Where it is necessary to liberate combined folic acid in natural materials, put 1 g of the finely-ground sample into 8 ml of 1% sodium acetate soln. at *p*H 4·5–4·7 and add 20 mg of takadiastase in 1 ml of water. Incubate the mixture under toluene for 24 hr. at 37° C., neutralise, autoclave for 15 min. at 15 lbs. and dilute as required. Put 5 ml of the soln. to be tested into test-tubes, add 5 ml of the basal medium and sterilise for 15 min. at 15 lbs. Cool, add 1 drop of inoculum and incubate at 30° C. for 16 hr. It is unnecessary to plug the tubes with cotton-wool, provided that a cover is put over each rack during sterilisation and incubation, for possible contamination is of little importance in view of the rate at which the organism grows. Measure the response with a photoelectric turbidimeter and calculate the amount of folic acid in the unknown from the standard curve in terms of the standard of "solubilised liver." If the material is available, the activity can be expressed as units in terms of the standard liver extract described by Mitchell and Snell (*Univ. Texas Pub.*, 1941, No. 4137, 36), which is given a potency of 1; folic acid is assumed to have a potency of 40,000. When the samples are coloured or turbid the amount of growth can be estimated by titration.

Basal Medium for Streptococcus lactis *R in 16 hour "Folic Acid" Assay.*

Constituent	Amount per tube
Sodium acetate	20 mg
Glucose	100 ,,
Casein (acid-hydrolysed)	50 ,,
l-Cystine	1 ,,
l-Tryptophan	3 ,,
Adenine sulphate	0·1 ,,
Guanine hydrochloride	0·1 ,,
Xanthine	0·1 ,,
Thiamine hydrochloride	2 μg
Riboflavin	2 ,,
Nicotinic acid	6 ,,
Pyridoxine hydrochloride	12 ,,
Calcium pantothenate	4 ,,
Biotin (concentrate)	4 mμg
K_2HPO_4	50 mg
Water to make	5 ml

Abstractor's Note—The "solubilised liver" used by American workers is not available in this country, and supplies of material standardised in the U.S.A. are strictly limited. It is hoped that folic acid preparations made and standardised in this country may soon become available. F. A. R.

Microbiological Determination of Amino Acids. I. Valine and Arginine. J. R. McMahan and E. E. Snell (*J. Biol. Chem.*, 1944, **152**, 83–95)—Stock cultures of the organisms *Lactobacillus casei* and *L. arabinosus* 17–5, are carried as stab

cultures on yeast extract-glucose-agar (1% glucose, 1% yeast extract, 1·5% agar), and duplicate tubes of each organism are incubated for 24–48 hr. at 30–37° C. and stored in the refrigerator until required. One of each is used for preparing inocula and the other for preparing further stab cultures at fortnightly intervals. Put 10-ml portions of a medium containing, per litre, 5 g of Bacto-peptone, 1 g of Bacto-yeast extract, 10 g of sodium acetate, 10 g of glucose and 5 ml each of inorganic salt solns A and B (*cf.* footnote, ANALYST, 1944, **69**, 157) into tubes, plug with cotton-wool and sterilise for 15 min. at 15 lbs. pressure. Inoculate these tubes from the stock-cultures 18–24 hrs. before the inoculum is required and incubate at 32° C. The basal medium for the assay of amino acids is as follows:

Basal Medium for Assay of Amino Acids—Items I to V, mixed in the amounts designated, form the medium for 80 assay tubes.

I. Solution 1, 80 ml

Adenine sulphate	10 mg.
Guanine	10 ,,
Xanthine	10 ,,
Uracil	10 ,,
l(−)-Cystine	100 ,,
Glycine	100 ,,
l(−)-Hydroxyproline	100 ,,
dl-Isoleucine	200 ,,
dl-Methionine	200 ,,
dl-Norleucine	200 ,,
dl-Norvaline	200 ,,
dl-Phenylalanine	200 ,,
l(−)-Tyrosine	100 ,,
Ammonium sulphate	3·0 g.
Sodium acetate (anhydrous)	6·0 ,,
Salts B‡	5·0 ml

Dissolve by heating with water, cool, add NaOH to *p*H 6·8–7·0, dilute to 400 ml.

II. Salts A,‡ 2 ml

III. Glucose, 2 g

IV. Solution 2, 10 ml

p-Aminobenzoic acid	600 μg
Biotin	0·4 μg
Calcium pantothenate	200 μg
Nicotinic acid	200 ,,
Pyridoxine hydrochloride	600 ,,
Riboflavin	200 ,,
Thiamine chloride	200 ,,
Folic acid*	800 mg units
Choline chloride	5·0 mg
Inositol	5·0 ,,
Water to make	100 ml

V. Solution 3, 10 ml

dl-Alanine	200 mg
l(+)-Arginine monohydrochloride†	100 ,,
l(+)-Aspartic acid	400 ,,
l(+)-Glutamic acid	1000 ,,
l(−)-Histidine	100 ,,
l(−)-Leucine	100 ,,
l(+)-Lysine	100 ,,
l(−)-Proline	100 ,,
dl-Serine	200 ,,
dl-Threonine	200 ,,
l(−)-Tryptophan	100 ,,
dl-Valine†	200 ,,

Dissolve in 30 ml of water, add NaOH to *p*H 6·8–7·0, dilute to 50 ml.

Add suitable aliquot portions of a standard soln. of *l*(+)arginine (50μg per ml) or *dl*-valine (200μg per ml) or of the protein hydrolysate under test to Pyrex test-tubes. To construct a standard curve for valine, use amounts ranging from 0 to 16μg per tube with increments of 2μg; for arginine use amounts ranging from 0 to 8μg per tube with increments of 1μg. Use the protein hydrolysates in amounts sufficient to give at least 6 points on the standard curve. The standard solns. and hydrolysates can either be diluted so that ordinary 1- or 2-ml pipettes can be used, or can be used undiluted with special micro-pipettes; the advantage of the latter procedure is that the vol. of the added sample, being small, can be disregarded. Prepare the basal medium as described above, adjust to *p*H 6·8–7·0 and dilute with an equal vol. of water. Add 2·5 ml of this diluted medium to each tube, plug with cotton-wool, sterilise at 15 lbs. for 15 min. and cool to room temp. Centrifuge the inoculum and re-suspend the cells in 10 ml of sterile saline, add 0·2–0·5 ml of this suspension to another 10-ml of sterile saline and use 1 drop of this suspension to inoculate each assay tube. Incubate in a water-bath at 32° ± 0·1° C. for 72 hr. The amount of growth can be estimated by titration, by measurement of the *p*H or turbidimetrically; the last procedure is generally used. In any event, the valine or arginine content is calculated from the standard curve obtained by the particular procedure used. The most convenient method of hydrolysing proteins is to seal them in tubes with 10% hydrochloric acid and autoclave at 15 lbs. pressure for 10 hr. The method gave reproducible results and the recovery of arginine from casein and gelatin hydrolysates was 92–109% of the theoretical. The results with arginine were in close agreement with those obtained by chemical methods. F. A. R.

Estimation of Inositol in Animal Tissues. B. S. Platt and G. E. Glock (*Biochem. J.*, 1943, **37**, 709–712)—Remove the tissue from the animal, cut into sections on a freezing microtome and dry them in Petri dishes in a cold room over silica gel *in vacuo*. Complete the drying in an oven at 110° C., powder the material and preserve in the cold until required. Mix an amount of dried tissue containing 0·5–1·0 mg of inositol (1–2 g of dry muscle powder) with 50 ml of water, heat to boiling and filter through a small plug of cotton-wool. Rinse the vessel twice with water (5 ml) and filter, repeat the process twice and evaporate the combined extracts to about 50 ml. Add acetone to give a final concn. of 70%, filter and remove the acetone from the filtrate by distillation. Evaporate the extract to about 25 ml and extract with ether. Wash the ethereal extract once with 20 ml of water, add the washings to the aq. residue, and concentrate to 30 ml. If combined, as well as free, inositol is to be estimated, divide the soln. into two portions and remove the glucose from one portion by yeast fermentation, as follows. Shake 1 part by weight of pressed baker's yeast with 5 parts of water and centrifuge; repeat the process four times and dilute the washed suspension to the original vol. Check the potency of the yeast, which should be prepared fresh each day, by incubating 2 ml with 25 ml of a soln. containing

* Any concentrate of "potency" 1000 or greater is suitable (Mitchell, Snell and Williams (*J. Amer. Chem. Soc.*, 1941, **63**, 2284). Material of potency 3000 was used in the present investigation. 800 mg units of this material weigh 800 ÷ 3000 = 0·27 mg.

† When the medium is used for valine assay, valine is left out of this soln.; when it is used for arginine assay, arginine is left out.

‡ See footnote, ANALYST, 1944, **69**, 157.

5 mg of glucose for 10 min. at 37° C., and estimate the reducing power of the supernatant liquid by Hagedorn and Jensen's ferricyanide method. Incubate the tissue extract at 37° C. with 2 ml of the yeast suspension for 10 min. and then centrifuge. Remove the supernatant liquid and wash the yeast twice by centrifuging with 10-ml portions of water, retaining the washings separately. Next remove interfering substances, such as creatine and creatinine, by means of carbonaceous zeolite* for cation exchange and "M.P.D. Resin"* for anion exchange; treatment with a mixture of the two adsorbents is more efficient than successive treatments with the two materials separately, but the adsorbents cannot then be regenerated. Shake 250 g of the carbonaceous zeolite at intervals with 2 vols. of 2 *N* hydrochloric acid, filter and wash with about 5 litres of water. Dry at 100° C. and sieve so that the particles are 20/40 mesh size. Treat the "M.P.D. Resin" in the same way with 2 *N* ammonia, washing with at least 1 litre of hot water. Mix equal parts of the two adsorbents and put 15 g of the mixture into a tube 20 × 7/16 in. narrowed at the bottom to approx. 3/16 in., and fitted with a small piece of rubber tubing carrying a glass tip and provided with a screw clip. Wash the adsorbent with 100 ml of water or more until the final washings give no blank when allowed to react with 5 ml of 0·01 *M* periodic acid for 10 min. at room temp. Pour the supernatant liquid from the yeast treatment into the tube and allow it to run through the column so that the whole of the liquid passes through in about 90 min. Add the first 10-ml portion of the yeast washings and adjust the rate of flow so that this passes through in about 15 min. Suck the column of adsorbent dry on a filter-pump, add the second 10-ml portion of the yeast washings, and again suck dry. Finally, wash the column with 15 successive 5-ml portions of water. The only substances present in the filtrate which react are glycerol and inositol, and at low temperatures these react with periodic acid at different rates. Thus, at 8° C. glycerol reacts quantitatively in 90 min. and yields 90% of the theoretical amount of formic acid, whereas inositol reacts to the extent of only 2% under these conditions. Dilute the filtrate from the adsorbent to 200 ml and divide the soln. in half. Cool both portions to 8° C., add 10 ml of 0·1 *M* periodic acid to one portion and allow this to react for 48 hr. Add 2 ml of periodic acid to the other portion and allow to react for 90 min. Stop both reactions by adjusting the *p*H to 6·5 by adding 5 ml of phosphate buffer soln. (12 g of Na_2HPO_4. $12H_2O$ and 20 ml of *N* sulphuric acid per 100 ml) followed by 5 ml of 5% potassium iodide soln. After 5 min., titrate the liberated iodine in both solns. with 0·004 *N* sodium thiosulphate soln., using 1% soluble starch in saturated sodium chloride soln. as indicator, and stabilise the end-point by addition of a few drops of saturated sodium bicarbonate soln. Allow a soln. of 1 mg of inositol in 100 ml of water to react with 10 ml of 0·01 *M* periodic acid for 48 hr. at 8° C., and titrate the liberated iodine as above. Calculate the inositol content in mg by subtracting the value obtained after 90 min. from that obtained after 48 hr. and dividing the result by the titre given by the standard soln. Finally, multiply by 1·02 to correct for the slight oxidation of inositol after 90 min. Combined inositol is estimated by hydrolysing the portion of the tissue extract reserved for this purpose with acid according to the method of Woolley (*J. Biol. Chem.*, 1941, **140**, 453). Dilute the extract to 50 ml and reflux for 6 hr. with hydrochloric acid of such a strength that the final concn. is 18%. Remove the hydrochloric acid by distillation under reduced pressure and rinse out the flask with water to give a final vol. of 25 ml. Neutralise by the dropwise addition of sodium hydroxide soln. and then subject to the fermentation, adsorption and oxidation described above. F. A. R.

* Obtainable from Permutit Co., Ltd.

Measurement of Thiourea in Ultra-filtrate of Serum. T. S. Danowski (*J. Biol. Chem.*, 1944, **152**, 201–206)—Withdraw a vol. of blood under oil sufficient to yield 7–10 ml of serum. Transfer the serum by means of capillary pipettes into cellophane cylinders, and connect these to a source of nitrogen at a pressure of 15 cm of mercury. Maintain for 24–48 hr. at 6–10° C. and collect the ultra-filtrate (4–5 ml), which contains any thiourea injected into the animal or added to the serum prior to ultra-filtration. Prepare a quantity of Grote's reagent by dissolving 0·5 g of sodium nitroferricyanide in 10 ml of water at room temp., adding 0·5 g of hydroxylamine hydrochloride and 1·0 g of sodium bicarbonate. When the evolution of carbon dioxide stops add 10 small drops of bromine; this causes a second evolution of gas; when this stops filter and dilute to 25 ml with water; dilute five-fold immediately before use. Since the reagent has a slight and variable amount of colour, fresh standards must be prepared for each group of determinations. Prepare a calibration curve from a mixture of 9 ml of water, 1·0 ml of serum ultra-filtrate from an animal before treatment with thiourea, 1·0 ml of a standard soln. of thiourea and 1·0 ml of diluted Grote's reagent, added in that order. Four thiourea standards, containing 3, 5, 6 and 8 mg per 100 ml of thiourea, are used. Mix the contents of the tubes and leave at room temp. Measure the green, greenish-blue or blue colour after 60 min. in an Evelyn colorimeter, using filter 565. Follow the identical procedure with the unknown soln. using 10 ml and not 9 ml of water. Calculate the concn. of thiourea from the calibration curve. The method can also be used for the analysis of urine: Dilute 2 or 4 ml of urine to 100 ml and mix 1·0 ml with 10·0 ml of water and 1·0 ml of Grote's reagent. Compare the colour with that of a series of standards; the reading may be taken at any time up to 21 hr. after mixing. No blank is necessary with urine. The recovery of added thiourea was within 0·1 mg per 100 ml of the expected value. F. A. R.

Estimation of "True Ascorbic Acid" in Blood. D. Richter and P. G. Croft (*Biochem. J.*, 1943, **37**, 706–708)—Put 4 ml of oxalated blood into a 15-ml centrifuge tube containing 0·05 ml of *sec.*-octyl alcohol and bubble coal gas through the soln. Add 4 ml of water and continue the gassing for 10 min. Add 2 ml of freshly-prepared 32% hypophosphorous acid, 1 ml of 25% lead acetate soln. and 2 ml of saturated sodium acetate soln. Centrifuge the suspension and take 2-ml portions of the supernatant liquid for titration. Standardise the 2 : 6-dichlorophenolindophenol soln. (2 mg/100 ml of warm water) by titration against a freshly-prepared soln. containing 2 mg of ascorbic acid in 400 ml of 2% hypophosphorous acid. Add the dye soln. from a 2-ml micro-burette to 2·0 ml of ascorbic acid soln. in a small conical vessel, stirring the soln. after each addition until a faint pink colour persists for 10 sec. and matches that of a

soln. made by adding 0·05 ml of dye to 1 ml of 1% hypophosphorous acid in a similar conical vessel. Subtract the amount of dye soln. added in the blank from the final burette reading. A correction must then be applied for the ascorbic acid retained by the protein ppt. This contained 65% of water, and, with the vols. given above, occupied a vol. of about 2·2 ml. Thus the effective vol. of the aqueous soln. is 13·0 − (2·2 × 0·35) = 12·2 ml, and the amount of ascorbic acid found in whole blood (mg per 100 ml) is equal to the amount found in 2 ml of supernatant liquid multiplied by the factor 12·2/2 × 100/4 = 152·5. By this method appreciably lower values were obtained than by direct titration, between 10 and 30% of non-ascorbic acid-reducing substance being present. The method gave results in close agreement with the ascorbic acid oxidase method of Richter and Croft (*Lancet*, 1943, **1**, 802), but was more convenient to carry out.

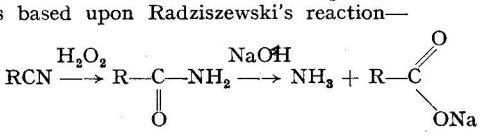
F. A. R.

Apparent Vitamin C in Foods. F. Wokes, J. G. Organ, J. Duncan and F. C. Jacoby (*Biochem. J.*, 1943, **37**, 695–702)—It has recently been established that the apparent vitamin C content of foods may become considerably higher than the true vitamin C content under certain conditions of manufacture and storage. Substances appear to be formed which, although antiscorbutically inactive, are titratable with 2 : 6-dichlorophenolindophenol. Vitamin C itself is destroyed by treatment with formaldehyde at *p*H 4·5 for 6 min., whereas little of the apparent vitamin C activity is lost under these conditions. By titrating the soln. before and after treatment with formaldehyde, therefore, using the same time intervals and rate of dye addition in each titration, the true vitamin C content can be estimated, the result of the second titration being subtracted from that of the first. The action of the formaldehyde is stopped at the end of 6 min. by adding excess hypophosphorous acid to bring the *p*H to 1·0. Apparent vitamin C is produced when potatoes, cabbages, carrots and germinated grains are treated with various concns. of acids, whilst with certain foods, apparent vitamin C is produced by heating to 100° C. without addition of acid; with malt extracts this effect is particularly marked. Apparent vitamin C may also be formed gradually during storage under normal conditions.

F. A. R.

Toxicological and Forensic

Determination of Small Amounts of Acrylonitrile in Air. G. W. Petersen and H. H. Radke (*Ind. Eng. Chem., Anal. Ed.*, 1944, **16**, 63–64)—The recent widespread use of acrylonitrile in the manufacture of one type of synthetic rubber necessitates a method for its determination in air. Toxicity determinations (Dudley and Neal, *J. Ind. Hyg. Toxicol.*, 1942, **24**, 27) indicate that the max. permissible limit is *ca.* 20 p.p.m. and an analytical method must be accurate to at least that concn. In the method an absorbing soln. containing the acrylonitrile is made strongly alkaline and then oxidised with hydrogen peroxide. Upon refluxing, acrylonitrile is converted quantitatively into ammonia which is determined by distillation into standard acid. The method is limited by the fact that the air must not contain other nitrogeneous compounds convertible to ammonia. The absorption trap consists of a 0·25-in. test tube forming one limb of a U-tube, the other limb (sealed into the bottom of the test tube) being a narrow tube bent at right angles so that its vertical portion is *ca.* half the length of the test tube. This serves as the inlet tube. The test tube contains glass beads to a depth of *ca.* 1 in and 2 ml of conc. sulphuric acid and is closed by a rubber stopper carrying a short outlet tube. Two such absorption traps are used in series and are placed in a water and ice bath. The air sample is drawn through at the max. rate of 0·4 litre per min., the rate being accurately measured by a rotameter, and the vol. of sample should represent *ca.* 6 mg of acrylonitrile. The sample is washed into a refluxing flask and 0·2 g of copper acetate is added to prevent polymerisation. Connect the refluxing flask with a vertical reflux water condenser which is connected with the top of a non-reflux vertical water condenser. A sintered glass bubbler attached to the lower end of the second condenser dips into 25 ml of 0·025 *N* sulphuric acid. The first condenser is closed by a stopper carrying the tube connecting the two condensers and a tap funnel. With the system closed, place 50 ml of 50% sodium hydroxide soln. in the tap funnel. Run the alkali into the refluxing flask through the first condenser, add 10 ml of water in the same manner and finally add 20 ml of 30% hydrogen peroxide from the funnel. After refluxing gently for 30 min. drain the water from the jacket of the first condenser and allow *ca.* half of the sample to distil into the standard acid. Rinse down the second condenser with water and titrate the excess of acid with 0·01 *N* sodium hydroxide, using methyl red as indicator. If N_A is the normality of the acid, V_B ml the vol. of sodium hydroxide used in titration, V_S litres the vol. of air sampled, N_B the normality of the sodium hydroxide and *C* the gas factor 22,400 corrected to sampling temp. and pressure, then the concn. of acrylonitrile in p.p.m. by vol. is $(25 N_A - N_B V_B) \times \frac{C}{V_S}$. The analysis is based upon Radziszewski's reaction—



A. O. J.

Spectrographic Determination of Lead in Blood. A. Tracy and J. McPheat (*Biochem. J.*, 1943, **37**, 683–685)—An improved method of estimating lead spectrographically was developed for investigating suspected industrial lead-poisoning. All the vessels and reagents used were, as far as possible, lead-free. A large Hilger quartz spectrograph with a range of 1910–8000A was used, with the following standard conditions: wave band, 2700–4300A; slit width, 0·015 mm (fixed slit); arc gap, 2 mm; voltage, 120 d.c.; amperage, 5; upper electrode, H.S. graphite pointed to 80° angle; lower electrode, H.S. graphite 10 mm diam. cut to form a crater to receive sample; exposure, 1 min.; plate, Ilford H. and D., 70; developed (*a*) 90 sec. at 19·5° C.; (*b*) fixed, acid "hypo." 15 min.; (*c*) washed in running water 15 min.; (*d*) dried in current of warm air. For the lead investigation the region 2700–4300A was used and the lines were identified by the Judd Lewis Comparator. The amount of lead present was calculated from a graph, prepared by measuring with a Hilger non-recording microphotometer the ratio of the line densities of different known amounts of lead and a constant amount of an internal reference element. From

this graph the amount of lead in the sample under examination was estimated. Originally, bismuth, with a line at 2898A, was chosen as internal reference element, but it was found that bismuth may be encountered normally in blood. Platinum, which has a line at 2830A, was subsequently found to be more satisfactory; it is added to a soln. of the ash in *aqua regia* in the form of a soln. containing 10 mg per 100 ml of sulphuric acid. By varying the amount of platinum and the exposure time and type of plate in accordance with the sensitivity of the element under investigation, platinum can also be used for the analysis of other elements.

As 2860·5A	Ni 3002A	Cr 2986·4A, 3015A, Pt 2998A	Au 2767A
Pt 2830A	Pt 2998A		Pt 2702A
Th 2837A	V 3063·7A	Mn 2798·3A	Cd 2980A
Pt 2830A	Pt 3064·7A	Pt 2830A	Pt 2998A

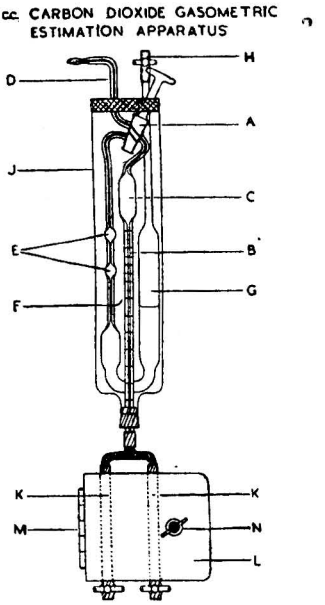
Platinum has the advantage that the lines are of high spectrographic intensity and are not overshadowed. The material used for preparing the standard curve was a synthetic ash which gave a main base spectrum similar to that of blood; it contained 1–2μg of lead per 100 g. Different known amounts of lead and a constant amount of platinum were added to 50-mg samples of this synthetic ash and these were spectrographed in duplicate; the standard curve was constructed from the mean of two sets of results. Five-ml samples of blood, urine, etc., were transferred to a lead-free silica crucible and treated with 3 ml of lead-free sulphuric acid. The samples were then ashed in a dust-proof electric furnace at 110° C., for 12 hrs. and then at 500° C. for 8 hrs. The low iron content of serum, urine and cerebrospinal fluid gave a weak base spectrum, and 20 mg of synthetic ash were therefore added to such samples. The results obtained with serum and urine were accurate to ±10μg/100 ml. The results indicate that in normal individuals the blood lead concentration may vary from 5 to 120μg/100 ml with a mean of 40μg/100 ml; the most common values lie between 40 and 60μg/100 ml. A value of 130μg/100 ml is regarded as the critical concentration for lead-poisoning. F. A. R.

Agricultural

Colorimetric Analysis of Xanthone Spray Residues. C. C. Cassil and J. W. Hansen (*Ind. Eng. Chem., Anal. Ed.*, 1944, **16**, 35–37)—Xanthone is reduced by sodium amalgam to xanthydrol. When this is treated with hydrochloric acid, the hydroxyl group is replaced by chlorine and the resulting compound gives a bright yellow soln. in conc. hydrochloric acid. The following procedure serves to estimate xanthone residues on apples which have been sprayed with xanthone as an insecticide. To prepare standards, dissolve 50 mg of pure xanthone in pure toluene and make up to 250 ml. Dilute 2 ml of the standard soln. to 20 ml with toluene in a 125-ml flask fitted with a ground-glass joint, add 10 ml of abs. methanol and 0·5–1 ml of sodium amalgam and heat the mixture beneath a reflux water condenser for 30 min. Cool the flask before removing it from the condenser, add 20 ml of water and shake vigorously to remove methanol from the toluene soln. and pour the soln. into a 50-ml Nessler glass, retaining the amalgam in the flask. Pipette 5 ml of the toluene layer containing the xanthydrol into a 100-ml flask containing exactly 10 ml of conc. hydrochloric acid and develop the colour by swirling the mixture gently for *ca.* 1 min. Pour into a cell and measure the colour of the acid layer in a photometer. Repeat the procedure with other aliquot portions of the standard soln. A glass colour filter having max. transmission at 424 mμ gives satisfactory results in a Type F Aminco photometer. Prepare a standard graph by plotting the quantities of xanthone against the log. of the corresponding photometer readings. The light transmission of 148μg of xanthone, read under the stated conditions in a 2·5 cm cell, is 50%. Statistical analysis has shown that 20 to 25 apples taken from different parts of a tree constitute a satisfactory sample for residue determination. Weigh the apples in a tared glass jar, calculate the surface area by means of a previously determined relationship, add 100–200 ml of toluene, according to the amount of xanthone and the size of the apples, and shake for 5 min. in a machine by the process described by Fahey (*J. Assoc. Off. Agr. Chem.*, 1943, **26**, 150). Filter a portion of the extract and use an aliquot portion (not exceeding 10 ml) of the filtrate for analysis, as described for the preparation of standards. Read the amount of xanthone from the standard graph. Toluene may contain an impurity, probably a thiophene derivative, that gives a yellow colour with hydrochloric acid. The following procedure serves both to purify the original toluene and to render toluene residues from the analyses available for subsequent use. Allow each litre of toluene to stand for 24 hr. with 50 ml of conc. sulphuric acid, separate the two layers and distil the toluene, discarding the first milky portion of the distillate. To prepare sodium amalgam, cautiously melt 9 g of sodium in 20 ml of toluene and add 750 g of mercury, drop by drop at first, and then more rapidly. Most of the toluene will evaporate but some should be kept over the amalgam stored in an air-tight container. The solubility of xanthone in toluene is 1·43 g per 100 ml. Since apple wax appears to dissolve, any xanthone covered by wax is included. Interference with the analysis by lead arsenate is negligible, and expts. with weathered apples showed that decomposition products of xanthone either do not remain on the apple or do not interfere with the analysis. A. O. J.

Simple Gasometric Apparatus for Estimation of Carbon Dioxide [in Grain]. T. A. Oxley (*Chem. and Ind.*, 1944, 24–25)—Howe and Oxley (*Bull. Ent. Res.*, in the press) have estimated insect infestation of grain by determining the concentration of carbon dioxide developed in inter-granular spaces of the sample sealed and incubated under standard conditions. The apparatus consists of: a graduated tube BC connected with tube D through three-way tap A; B is graduated in 0·01 ml from 0 (top) to 1 ml (bottom). The vol. (C) from the top graduation to the tap is 4 ml. A also connects with a U-tube containing potassium hydroxide soln., with level near the graduation F. The right-arm top of the U-tube (3–4 mm bore) is closed by rubber tubing and clip H. A glass water-jacket J, with divided cork closure at top, encloses the whole; B passes through a rubber stopper in the bottom and is connected, by rubber and wide glass T-piece, to short wide rubber tubing K closed at the bottom ends. These may be compressed by flap L, hinged at M; B and K contain mercury; adjustment of flap L varies the mercury level in B. *Method*—Close L by screw N until the mercury completely fills B, C and D. Connect the syringe containing the air sample to D, draw in

4·7–4·9 ml and, in order that air in the connections may not seriously contaminate the portion drawn in, re-expel into the syringe and re-draw several times, by repeatedly raising and lowering the mercury level. Connect C with the potash tube and adjust the mercury level until the potash level coincides with F, making the final adjustment after stirring J and closing H. After reading the level in B, raise and lower the mercury about 10 to 20 times to mix the sample in the potash tube. Bulbs E prevent the drawing of potash into B and C. Adjust the mercury level until the potash level again coincides with F, after stirring the water-jacket J by means of air blown in through a tube (not shown) passing through the divided cork.

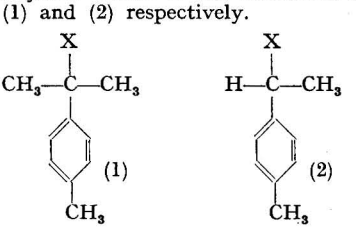


From the readings of B the vol. of carbon dioxide may be calculated. Possible sources of error are discussed. If air is drawn directly from grain into the apparatus the potash solution must be in equilibrium with 65% relative humidity (sp.gr. 1·255 at 15° C.); for grain with moisture content 12 to 15% the error is within ±0·2%. A table shows the vol. % of water vapour in air for various relative humidities at 10 to 30° C. From this, the vol. of air examined may be corrected for water vapour present. The apparatus is easy to construct, but improved portability, greater simplicity and greater operational speed are desirable. E. B. D.

Organic

Colour Reactions for Certain Amino Acids. H. Tauber (*J. Amer. Chem. Soc.*, 1944, **66**, 310)—Certain amino acids are converted by heating into chromogens, alcoholic extracts of which become more deeply coloured on addition of alkali, and colourless or much lighter on subsequent acidification. The procedure is: Heat 10 mg of amino acid in a dry Pyrex test-tube until the first colour change occurs, avoiding over-heating. Cool, add 3 ml of ethanol, boil for 30 sec. and divide the soln. into 3 portions. To the first add 0·2 ml of 0·1 *N* sodium hydroxide, to the second 0·2 ml of 0·1 *N* sulphuric acid, and to the third 0·2 ml of water. The behaviour in this test of 21 biologically important amino acids is discussed. Three (*dl*-alanine, *dl*-valine and *dl-iso*leucine) leave no pigment or residue. Twelve (*l*-cystine, *l*-cysteine, glycine, *l*-hydroxyproline, *dl*-methionine, glutamic acid, *dl*-aspartic acid, *dl*-serine, *l*-proline, *d*-arginine monohydrochloride, *dl*-lysine and *d*-lysine) change to yellow, brown or black decomposition products without chromogenic properties. Six form chromogens, which react as follows in the above tests: Red-brown on heating, light brown or reddish in alcoholic soln., darkening on addition of alkali: *l*-tyrosin, *l*-tryptophan and *dl*-threonine; on addition of acid to alkaline alcoholic soln., light yellow: *l*-tyrosin, or light brown: *l*-tryptophan and *dl*-threonine. Light brown on heating, light yellow in alcoholic soln., deep yellow on addition of alkali, and almost colourless on acidification: Histidine monohydrochloride. Partly subliming on heating, changing to yellow, and giving a yellow or light yellow colour in alcoholic soln., becoming deep yellow on addition of alkali and almost colourless on subsequent acidification: *dl*-β- or *l*-β-phenylalanine and *l*-leucine. The alkaline alcoholic soln. of the β-phenylalanines shows a greenish-yellow fluorescence, which is particularly strong in ultra-violet light, without addition of alkali. The isolation of this fluorescent pigment is described. E. M. P.

Determination of α-*p*-Dimethylstyrene in presence of *p*-Methylstyrene, Styrene and *p*-Cymene. J. H. Elliott and E. V. Cook (*Ind. Eng. Chem., Anal. Ed.*, 1944, **16**, 20–23)—According to Markownikoff's rule, addition of HX (where X is chlorine or bromine) to α-*p*-dimethylstyrene or *p*-methylstyrene results in the formation of compounds (1) and (2) respectively.



The halogen in (1) is attached to a tertiary carbon atom, that in (2) to a secondary carbon atom. The halogen in (1) is much more easily hydrolysed than the halogen in (2). The secondary bromide formed by action of hydrogen bromide on styrene is hydrolysed under relatively mild conditions, and the conditions of hydrolysis of tertiary halide must be so chosen as to avoid interference from the secondary halides present. *p*-Cymene does not react with HX. Treat the sample (0·5–1 g), dissolved in 25 ml of carbon tetrachloride, with gaseous hydrobromic acid (generated by dropping bromine on to naphthalene and purified by means of several towers filled with naphthalene and one filled with Drierite) for 30 min. at the rate of 3 or 4 bubbles per sec. Rinse the addition tube with 10 ml of carbon tetrachloride and remove unreacted hydrogen bromide by means of a brisk stream of nitrogen. Cool the product in ice and salt for 10 min., add 30 ml of cooled 90% alcohol, swirl the mixture for a few sec. and titrate the cold liquid rapidly with 0·1 *N* alcoholic potassium hydroxide (methyl red as indicator) until the bright yellow end-point persists for 10 sec. Hydrogen chloride (prepared by dropping conc. hydrochloric acid into conc. sulphuric acid and drying the gas with conc.

sulphuric acid) may be used instead of hydrogen bromide, with the following changes. Use benzene as solvent, hydrolyse the tertiary halide with 40 ml of 80% alcohol and titrate at room temp. to the bright yellow end-point of methyl red persisting for 30 sec. With hydrogen chloride the process is somewhat easier and the end-point sharper. Analyses of known mixtures showed that the recovery of α-*p*-dimethylstyrene is consistently *ca.* 95% and a correction factor of 1·05 is therefore used. The low recovery is probably due to the fact that a small fraction of the addition takes place contrary to Markownikoff's rule. The precision of the method is *ca.* ±3% of the amount of α-*p*-dimethylstyrene present. The spectrophotometric method has been successfully used to determine the amount of each styrene present in the mixture within 2% of the known amount. Absorption data were obtained with a Beckman quartz spectrophotometer, the solvent being ethanol. The formula used to obtain a specific absorption coefficient α was

$$\alpha = \frac{\log_{10} \frac{I}{I}}{cl},$$

where I_0 and I are the intensities of transmission of solvent and soln. respectively, c is the concn. of solute in g/100 ml and l is the length (cm) of the soln. The styrenes have strong absorption bands in the region 248–252*mμ* with distinct but less pronounced bands in the region towards 300*mμ*. For the three styrenes the bands in the first mentioned region are so similar as to be unsuitable for quantitative measurements. A larger scale graph of the absorption in the region 280–300*mμ* shows that at certain wavelengths there are sufficient differences in α to permit quantitative measurements. Cymene has absorption values below 280*mμ* and therefore does not appreciably affect absorption values above 280*mμ* even when it is present up to 70 or 80%. At a given wavelength, components *a*, *b*, *c*, present in the soln. to the extent of *x*, *y*, *z*% respectively will give a total absorption value represented by $\alpha_{soln} = \frac{(\alpha_a)x + (\alpha_b)y + (\alpha_c)z}{100}$ and with three equations at different wavelengths the values of *x*, *y* and *z* can be determined. With mixtures of the pure compounds the absorption data at wavelengths 285, 291 and 295*mμ* gave values differing from the known composition by less than 2%. Data at wavelengths 283, 287 and 291 gave equally accurate results, and it is therefore possible to use two sets of wavelengths to serve as checks on each other. A. O. J.

Determination of *d*-Galactose in Plant Mucilages by Selective Fermentation. L. E. Wise and J. W. Appling (*Ind. Eng. Chem., Anal. Ed.*, 1944, **16**, 28–32)—The method depends upon the use of two yeasts: *Saccharomyces carlsbergensis* var. *manchuricus*, N.R.R.L. No. 379 and *S. bayanus*, N.R.R.L. No. 966. Neither organism has more than a slight effect upon arabinose, xylose and glycuronic acid. No. 379 ferments *d*-glucose, mannose, fructose and galactose almost quantitatively within 48 hr., whilst No. 966 ferments the first three readily within the same period but has no action on galactose. These yeasts can be kept in good condition by monthly transfers on glucose agar (Bacto-Dextrose Agar, dehydrated) and show no decrease in potency in 8 months. Agar slant cultures 2 to 7 days old are used for preparation of the suspensions required for inoculation of the sugar solns. The following procedure is for either yeast. Pipette *ca.* 2 ml of sterile water into the tube containing the agar slant culture and remove the surface growth by means of the pipette, which serves also to stir the suspension. For each bottle slant of glucose agar required, remove 0·5 ml of the suspension and spread it over the surface by tilting the bottle. Incubate the slants for *ca.* 48 hr. at 30° C. One bottle slant furnishes enough inoculum for four sugar analyses. Add 10 ml of sterile water to the slant, and gently tilt the bottle to loosen the growth. Pipette 5 ml of the dense yeast suspension into a sterile dilution bottle and add 20–30 ml of sterile water according to the turbidity shown by a Cenco-Sheard-Sanford photelometer. Suspensions giving readings of 10–16 (water ≡ 90) ferment sugar mixtures satisfactorily. Adjust suspensions falling outside this range by addition of yeast or water. Plate counts showed this density range to correspond with 35 million cells per ml. It is expedient to use 25-ml portions of sugar soln. in the fermentations and to make all expts. in 125-ml Erlenmeyer flasks. The total reducing sugar in such solns. should not exceed 2% and the galactose concn. should be within the range 40–250 mg per 25 ml. To the sugar soln. add 15 ml of filtered yeast extract (*infra*), sterilise the mixture at 15 lb. pressure for 15 min., cool to 30° C. and inoculate under aseptic conditions with 10 ml of the appropriate yeast suspension. Incubate at 30° C. for at least 48 hr. Run the fermentations in pairs under identical conditions, inoculating one with No. 966 and the other with No. 379. Rotate the flasks 3 or 4 times during incubation to bring the bottom yeasts into contact with the sugars. At the end of fermentation dilute the solns. to 100 ml, mix thoroughly and filter through two Whatman No. 50 papers. Analyse aliquot portions of the filtrates by the usual Munson-Walker method. Asbestos used in the Gooch crucibles should be digested thoroughly with hot Fehling's soln. and with conc. nitric acid until it permits the filtration of hot Fehling's soln. containing yeast extract. Subtract the wt. of cuprous oxide obtained with organism 379 from that obtained with organism 966 and determine the corresponding amount of galactose from a curve prepared by the analysis of pure galactose solns. Errors due to presence of pentoses, glycuronic acid and galacturonic acid are negligible in this differential process. In higher concns. galactose shows a slight but persistent residual reduction after fermentation with organism 379, and this accounts for a slightly low recovery of galactose from solns. of known composition. This reduction is due either to very small amounts of unfermented galactose, or more probably, to the slight reducing power of the products of fermentation. The method was applied successfully to a series of mannogalactan mucilages isolated from seed endosperms by hot water extraction and pptn. with alcohol. The dried mucilage was hydrolysed by boiling for *ca.* 6 hr. with 2% sulphuric acid and the cooled soln. was brought to *p*H 5–6 by addition of solid sodium carbonate before being subjected to differential fermentation. Yeast extract for use in the method described may be prepared as follows. Heat a 10% suspension of starch-free yeast for 1 hr. in an Arnold steriliser at *ca.* 100° C. and subsequently for 20 min. at 15 lb. pressure (121° C.). Filter the cooled suspension several times with Cellite through fluted paper. Sterilise the slightly turbid filtrate in 80-ml portions in plugged flasks for 20 min. at 15 lb. pressure and store in a refrigerator. A. O. J.

Analysis of Petroleum Oil-soluble Sodium Sulphonates by Adsorption. J. M. Koch (*Ind. Eng. Chem., Anal. Ed.*, 1944, **16**, 25–28)—In the manufacture of medicinal white oil by treatment of petroleum stocks with fuming sulphuric acid sulphonic acids are formed, some of which remain dissolved in the oil layer after its separation from the acid sludge. Neutralisation with alkali results in the formation of sodium sulphonates (mahogany soaps) which are extracted with aq. alcohol, refined and marketed as emulsifiers consisting of approx. equal proportions of oil and soap contaminated with a little organic matter. To control the ratio of oil to soap it is necessary to estimate at least one of these constituents. Distillation and crystallisation are not suitable methods for separation, and, although separation can be effected by systematic multiple extractions with light petroleum and aq. alcohol, the method is tedious and subject to difficulties caused by emulsions. Sodium sulphonates can be selectively adsorbed from naphtha soln. and can be completely displaced from the adsorbent by methanol. The apparatus consists of a 250-ml separating funnel connected with a 250-ml extraction flask by a percolation tube, 40 cm long and of internal diam. 1·5 cm, with a delivery tube 5 cm long and of internal diam. 3 mm. To prepare the column, which is supported on a cotton-wool plug, tamp successive 5-cm portions of fresh Attapulgus clay (Attapulgus Clay Co., 260, South Broad St., Philadelphia) tightly with a glass rod until *ca.* 35 g have been introduced into the percolator. The clay should be 30-60 mesh calcined at 482° C. Dissolve *ca.* 2 g of the sample in 25 ml of petroleum naphtha in the extraction flask. Naphtha of boiling range 30°–80° C., distilled from a paraffin base crude oil and A.S.T.M. precipitation naphtha of 50°–130° C. b.r. are satisfactory. Transfer the soln. to the separating funnel and stopper the funnel, and place the stem in the percolation tube so that it just touches the surface of the clay. Open the tap of the funnel and allow the soln. to percolate. The stoppered funnel will then act as an automatic feeding device and the percolate will be received in the tared extraction flask. As soon as the last drop of soln. has entered the percolator, rinse the stem of the funnel and the top of the percolator with naphtha and run the washings through the percolator. Finally allow 100 ml of naphtha to percolate from the funnel. If the total percolate is not clear, run it through a second column with the same procedure. Evaporate the combined percolates and dry the residual oil for 15-min. periods at 100–105° C. until its wt. is constant. Allow 100 ml of abs. methanol to percolate through each of the columns used, evaporate the solvent and dry the residual soap for 15 min. periods at 120°–130° C. until its wt. is constant. With soap and oil mixtures containing resins the method needs modification, since resin is adsorbed on the clay. Expts. showed that ethyl acetate and diethyl ether are good resin eluants. The procedure is as already described, but, after washing the column with 100 ml of naphtha, extract the resin from the column with either eluant. Percolation with methanol then follows. A. O. J.

[Estimation of] Amorphous Swollen Cellulose by an Improved Thallous Ethylate Method. A. G. Assap, R. H. Haas and C. B. Purves (*J. Amer. Chem. Soc.*, 1944, **66**, 59–65)—One method of estimating amorphous cellulose is to measure the density of the diffuse "background" radiation in the X-ray diffraction pattern of the fibre (Mark, *Ind. Eng. Chem.*, 1942, **34**, 449). In other methods the solid cellulose is submitted to a chemical reaction and the proportion of the more accessible, amorphous portion is calculated from the initial, more rapid change. There is evidence that the amount of amorphous cellulose has an important bearing on the elasticity, the absorptive capacity and other valuable industrial properties of the fibre (*J. Phys. Chem.*, 1940, **44**, 764). Some of the chemical methods of estimation suffer from the drawbacks that the reagents may penetrate the cellulose crystallites and also cause swelling during the estimation. To prevent this, Harris and Purves (*Paper Trade J.*, 1940, Feb. 8, **110**, 29) used thallous ethylate dissolved in ether or benzene (non-hydroxylic, non-swelling liquids) to measure the amorphous cellulose in ramie. The reaction depends upon the conversion of cellulose hydroxyl groups into thallium cellulosates—

Cellulose OH + TlOEt ⟶ Cellulose OTl + EtOH

The thallium cellulosate is heated with excess of methyl iodide or dimethyl sulphate in benzene and, after purification, the methoxyl content of the partly methylated cellulose is determined, and the methoxyl substitution is regarded as proportional to the number of hydroxyl groups that came in contact with the thallous ethylate reagent. The present paper describes an improved technique for the procedure, with exact details for thallation and methylation and a description of the Pyrex apparatus used. The tabulated results show that the absolute amount of amorphous cellulose in cotton linters, ramie and viscose rayon shows great variations. *Estimation of Cellulose Hydroxyl Groups by means of Methyl magnesium Iodide*—Dried, highly swollen linters (0·117 g) were brought into contact with an approx. 0·4 *M* soln. of methylmagnesium iodide in isoamyl ether (*J. Amer. Chem. Soc.*, 1930, **52**, 3736) in a modified Zerewitinoff apparatus (*Ind. Eng. Chem., Anal. Ed.*, 1938, **10**, 195; 1939, **11**, 164), and 4·96 ml of methane (corr. to N.T.P.) were collected. This amount corresponded to the active hydrogen in 10·2% of the hydroxyl groups in the sample. Parallel estimations by the thallation-methylation method gave 6·5–6·8% of methoxyl with thallous ethylate in the same solvent, indicating that 11·5% of the hydroxyl groups had been methylated. Although the Zerewitinoff method can be used to check the thallous ethylate technique, its utility is limited by its sensitivity to traces of moisture in the sample and by the fact that it is necessary to use a solvent with low vapour pressure.

Inorganic

Spot-Test for Small Amounts of Chromium in Steel. R. H. Williams (*Metal Progress*, 1943, **44**, 1125; *Nickel Bull.*, 1944, **17**, 38)—A sensitivity down to 0·02% of chromium is claimed for the following test. Clean the area to be tested by filing or grinding. Place one drop of acid mixture (water, 250 ml; conc. nitric acid, 200 ml; 85% phosphoric acid, 50 ml) on the surface. After exactly 30 sec., transfer the drop by means of a glass rod to a spot on filter-paper which has received 1 drop of oxidising soln. (water, 400 ml; sodium hydroxide, 20 g; bromine, 2·5 ml). Add 1 drop of dilute (1 : 5) sulphuric acid, followed by 1 drop of phenol solution (phenol, 5 g; acetic acid, 50 ml). Add finally 1 drop of freshly prepared diphenylcarbazide solution (diphenylcarbazide, 1 g; acetic

acid, 5 ml; ethyl alcohol, 50 ml). A faint pink colour indicates "low chromium content."
S. G. C.

Spectrographic Determination of Thorium in Tungsten Filament Wire. S. L. Parsons (*J. Opt. Soc. Amer.*, 1943, **33**, 659–662)—For the detn. of thorium in tungsten wires over the range from 0·9% to 2% the following method is used. Square the ends of two ¼ in. graphite electrodes and drill each with a small hole (0·04 in. diam.) to a depth of about ¼ in. Pack several pieces of the tungsten wire into each hole and cut off so that they project about 3/32 in. above the graphite. Run on a 2200 volt, 2·4 amp. A.C. arc, giving a 50-sec. preburn and a 60-sec. exposure on an Eastman 33 plate. Calibrate each plate (blackening/intensity relation), using an iron arc and a rotating stepped sector; then use this calibration to determine the log. intensity ratio of the thorium line 2899·3A to the tungsten line 2904·0A. Deduce the corresponding thorium concn. from a previously constructed analytical curve obtained by the use of chemically analysed standards. This method has proved suitable for thoriated tungsten wires of diameters ranging from 0·004 in. to 0·013 in. Trials made on 84 samples from a batch of known composition gave a standard deviation of 3·54% from the mean. The method is being used for routine examination.
B. S. C.

Separation of Erbium from Yttrium. T. Moeller and H. E. Kremers (*J. Amer. Chem. Soc.*, 1944, **66**, 307)—Fractional pptn. with ferrocyanide is more rapid than basicity methods; the sliminess of the ppt. can be reduced by a high concn. of nitric acid (Prandtl, *Z. anorg. allgem. Chem.*, 1931, **198**, 157). The authors' alternative process avoids undesirable oxidation of the ferrocyanide. To the cold, 3 *N* hydrochloric acid soln. containing 2% of rare earths and 10% of ammonium chloride, slowly add during constant stirring enough 20% potassium ferrocyanide soln. to ppt. about one-half of the rare earths. The pptn. is gradual and begins only after all of the ferrocyanide has been added. Set aside for some hours, collect ppt. by suction filtration, and boil it with 50% sodium hydroxide soln. Dilute, filter, and ignite the ppt., dissolve it in hydrochloric acid, and eliminate the last of the iron by double oxalate pptn. Even traces of iron interfere by producing gelatinous ferrocyanide ppts. The products were examined by spectrophotometry (489 and 653mμ erbium absorption bands). 6·9 g of a binary mixture containing 87% Er_2O_3 yielded 3·14 g of 98·5% erbia after two fractionations. Three fractionations of 45 g of 17% Er_2O_3 gave 5·6 g of 48% erbia.
W. R. S.

Reaction of Indium Sulphate with Potassium Chromate. M. F. Stubbs (*J. Amer. Chem. Soc.*, 1944, **66**, 498)—The salts react in dil. aqueous solution to yield a yellow ppt. which begins to form at *p*H 3·30 and flocculates out at *p*H 3·36–3·42. Analysis of the ppt. after filtering off, washing with water and drying, showed that it consisted mainly of indium hydroxide with a small amount of indium chromate. Indium hydroxide is pptd. in absence of chromate at *p*H 3·41–3·43.
S. G. C.

Photometric Determination of Sulphate in Nickel Nitrate. P. von Stein. (*Chemist-Analyst*, 1943, **42**, 62–63; *Nickel Bull.*, 1944, **17**, 30)—The solution of nickel nitrate hexahydrate (100 ml, containing 14·6 g of the salt) is mixed with 5 ml of glycerin and 20 ml of barium chloride reagent (200 g of barium chloride and 50 ml of conc. hydrochloric acid per litre) at room temperature. The liquid is transferred to a rectangular glass cell illuminated on one face by a 15-watt electric bulb (this face has an etched surface for diffusion of light), and after 3 minutes the light transmitted to the opposite face is measured by a photoelectric light meter (a "General Electric Exposure Meter Type DW–48"). The sulphate content is determined by reference to a calibration curve which has been plotted from measurements made on a series of solutions of known sulphate content with the same illumination. The 3-min. pptn. period was selected as the min. time required for comparative results; no great variation occurred within the next 15 min. Amounts of sulphate from 0·005 to 0·11% were determinable. A suitable apparatus can be extemporised from a wooden block bored to accommodate the light source, cell and light meter.
S. G. C.

Physical Methods, Apparatus, etc.

A Compendium of Line Pairs and Operating Conditions used in Spectrographic Analysis. E. S. Hodge (*J. Opt. Soc. Amer.*, 1943, **33**, 656–659)—In making a compilation of information on spectrographic analysis from most of the available literature it was noted that many good papers, as judged by the quantitative results, were of little value because important operating conditions had been omitted. It is considered essential that data relating to the following three items be included in published methods: The spectrum line pairs used, the concentration range covered and the type of source used for excitation of the spectra. It is also suggested that the fullest data on the operating conditions would include: Instrument used; source voltage; source current or power; inductance; capacity; source gap; upper and lower electrode materials; slit width; exposure (expressed as preburning time, exposure time, radial sector opening or screen used); photographic emulsion; developer; development time and temperature; calibration method used; buffer used; lens used and method; other remarks (*e.g.*, sample preparation).
B. S. C.

Sampling, Mixing and Grinding Techniques in the Preparation of Samples for X-ray and Spectrographic Analysis. J. W. Ballard, H. I. Oshry and H. H. Schrenk (*J. Opt. Soc. Amer.*, 1943, **33**, 667–675)—During the investigation of the composition of atmospheric particulate matter a study was made of the most suitable techniques for preparing very small samples for quantitative analysis by X-ray diffraction and by spectrographic methods. The factors common to both techniques are that a very small sample must often be prepared as representative of a large gross sample, and that the small sample must usually be intimately mixed with another material to serve as an internal standard. For gross materials a "Diamond" mortar, ore grinder, mixing cloths and riffle are used for obtaining a small representative sample. One interesting feature in the preparation of the spectrographic samples is the use of specially designed miniature ball mills made of nickel. Since this element is also used as the internal standard, contamination is thus reduced to a minimum.
B. S. C.

Base Electrolytes for Use in Polarographic Determinations. H. Wolfson (*Nature*, 1944, **153**, 375–376)—The method was devised originally for the determination of iron (12–18%) and molybdenum (2–5%) in presence of more than 70% of nickel, with other metals (*e.g.*, copper and manganese). Dissolve the sample in a mixture of sulphuric and nitric acids, dilute to a known vol. and transfer aliquot portions to a graduated flask. For the determination of molybdenum all the nitric acid should first be removed by heating the soln. until the sulphur trioxide fumes appear. Then add the base electrolyte, a mixture of citric and sulphuric acids in proportions such as to produce final concns. in the flask, of 0·5 *M* and 0·75 *N*, respectively. Under these conditions the polarogram for molybdenum shows 2 sharply-defined waves, having half-wave potentials (against the sulphate electrode) of −0·35 and −0·85 volt; iron and copper, however, tend to interfere with the former, and poorer results are obtained when the drop time of the capillary in this soln. is less than 2·5 sec. at zero applied potential. With alloys of the above type (*e.g.*, "Permalloy") normal polarographic methods failed to give accurate iron contents, but improved results were obtained when the base electrolyte contained 0·3 *M* triethanolamine, with 1·0 *M* ammonia and 0·85 *M* ammonium chloride. The iron is then reduced in stages (−0·5 and 1·6 volt; standard calomel electrode, S.C.E.); the first gives the better defined wave. Copper interferes to some extent, and most of it should be first removed. This work has led to a general study of the effects of organic amines mixed with certain inorganic salts, on the suppression, shifting and separation of the waves corresponding with the various metal ions. Base electrolytes of this type are likely to be of great use in the determination, *inter alia*, of iron, copper, nickel, lead, cadmium, barium, cobalt and zinc.

J. G.

Modification to the Cryoscopic Equation. A. V. Brancker, S. J. Leach and V. A. Daniels (*Nature*, 1944, **153**, 407)—The relationship between the depression of the freezing pt. (Δt) of the solvent in a soln. of a pure solute (mol. wt. M) of molecular concentration m is usually expressed by the equation $\Delta t = K_f m$, where K_f is the cryoscopic constant of the solvent. K_f is, however, not strictly constant, but decreases as the concentration of solute rises (*cf.* Meldrum, Saxer and Jones, *J. Amer. Chem. Soc.*, 1943, **65**, 2023). The authors suggest the use of a modified equation $\Delta t = K_f m^b$. They have examined a number of solutes of known mol. wt., dissolved in benzene and camphor respectively, and have found that a straight line was obtained when log Δt was plotted against log m, indicating that b was a constant, which could be determined. With solns. containing from 0·68 to 11·13% by wt. of tetralin in benzene, the simple equation ($\Delta t = 5{\cdot}229\, m$) gave values of M from 111·6 to 134·9, and the modified equation ($\Delta t = 5{\cdot}229\, m^{0{\cdot}9394}$) values from 132·4 to 137.0.

Projection Arrangement for Determination of Fibre Dimensions. J. H. Graff and J. R. Feavel (*Paper Trade J.*, 1944, **118**, *T.A.P.P.I. Sect.*, 53–57)—Existing methods are inaccurate and lengthy and give little consideration to the relative widths of the fibres, and to the relative % of whole and broken fibres, ray cells and fibre fragments. Projection type methods are rendered more accurate if fibres are straightened out in parallel lines under a dissecting microscope, and when the angular aperture of the projection lens is large enough to cover the whole field of the parallel fibres; also, the focal length of the lens must be short enough to ensure that the projection distance corresponding with the magnification necessary to attain a desired accuracy is not unduly great. An apparatus which fulfils these requirements is described. It consists of a projection microscope from which the microscope tube has been removed; a f. 2·8 projection lens (focal length, 12·5 mm) replaces the objective. The microscope and illuminant are placed in a blackened box on top of which is a finely-ground glass plate, on which the image is projected *via* a reflecting mirror. A series of parallel lines are drawn at 7·5–mm intervals on the ground glass plate, and the plate is protected from extraneous light by a blackened hood. Thoroughly disintegrate 1·5 g of sample (*e.g.*, paper or pulp), squeeze it between the fingers, and place it in a mixture of Bright's stains *A* and *B* at room temp. for *ca.* 1 min.; this stain is preferred because the stained fibres do not fade readily, and both bleached and unbleached fibres are stained. Strain the pulp through nainsook cloth, wash it with 500 ml of water, strain it again, squeeze it, and immerse it in 50 ml of Bright's stain *C* for 2 min.; strain, squeeze and wash in 500 ml of water. Finally, strain the pulp suspension and dilute it so that 1 drop (delivered from a dropper having a jet of 5 mm internal diam.) contains *ca.* 25 fibres. Divide this suspension between 4 test tubes, shake each in turn and at once transfer 2 drops from each to a pair of microscope slides (*i.e.*, so as to obtain 4 pairs of 1-drop preparations). Dry the preparations at 50° C., add 1 drop of a soln. of calcium chloride (sp.gr., 1·36 at 28° C.) and after 2 min. remove the surplus liquid partly by tilting the slide and partly with the aid of filter paper; take care that no fibres are lost. Tease out all the fibres, so that they are straight and parallel, under a dissecting microscope, and cover them with a square cover slip, so that one edge is close to the ends of the fibres; irrigate the calcium chloride soln. under this edge of the cover slip. Project the preparation on to the ground glass, and measure the length and width of each fibre to within 0·1 and 0·01 mm., respectively, with the aid of a transparent micrometer scale. With a magnification of ×75 it is possible to distinguish whole fibres, fibre débris and ray cells. At least 200 measurements should be made; the 8 slides are usually sufficient. Data are tabulated for the arithmetic and weighted average fibre lengths, the average width, the ratio (length : width), the average area of the fibres, and the % frequencies of the length and area. For all practical purposes it is safe to calculate the wts. from the areas, since it may be assumed that all fibres of the same origin have the same sp.gr. By plotting the % distribution by wt. of short (0·5 mm, and less) medium (0·5–1·5 mm) and long (over 1·5 mm) fibres on a triangular chart, it is possible to see at a glance the relative proportions of the 3 types of fibres present. The % probable errors for the weighted average fibre length and for the average fibre width are considerably less than for the arithmetic average fibre length. This is important, because the weighted average fibre length is inversely proportional to the no. fibres in 1 g of pulp.

J. G.

Testing Wood Preservatives. J. Leutritz (*Nature*, 1944, **153**, 441)—In the Bell Laboratory

method outlined (*cf. Bell Lab. Rec.*, 1943, **22**, No. 4) sticks (0·75 × 0·75 × *ca.* 36 in.) are cut from boards of Southern pine sapwood, and 20 of them are placed in a cylinder, which is then evacuated and, after a specified interval, filled with the preservative (*ca.* 30 lb./ft.^{3} of wood), under an appropriate pressure for the "full cell" method. With the "empty cell" method the pressure of the air in the cylinder is first raised to 25–50 lb./in.^{2}, the preservative is pumped in, and the pressure raised still further so as to force it into the wood; on releasing the pressure the expansion of the air trapped by the preservative forces the excess of the latter out of the wood so that, assumedly, only the cell walls are coated. The cells are then almost completely emptied by applying a vacuum after the pressure is released. The initial air pressure thus largely determines the amount of preservative which will be forced out of the cells, while the difference between the initial and final pressures determines its distribution and degree of penetration. The amount of preservative retained is given by the increase in wt. resulting from the treatment. The specimens are then cut into halves, one of which is subjected to a laboratory rot test, whilst the other halves are buried 7 in., in a uniform distribution, throughout the test plot. They are examined at suitable intervals (*e.g.*, annually), and the amounts of decay at or below the groundline are noted. If a material gives promising results, then the field test is extended first to samples of fence posts size and then to 10-ft. posts. J. G.

Reviews

THE EXTRA PHARMACOPOEIA (MARTINDALE). Volume II. Editor: C. E. Corfield, B.Sc., F.R.I.C., Ph.C. 22nd Edition. Pp. xxxiv + 1217. London: The Pharmaceutical Press. 1943. Price 27s. 6d.

There can be no doubt that Vol. II of the twenty-second edition of "Martindale" abundantly supports the high tradition now so long associated with this classic work. It is divided into forty-one main sections containing detailed descriptions of practical procedures together with numerous notes and references on analytical chemistry, biochemistry, bacteriology, pathology, nutrition and clinical methods. The volume opens with 372 pages of Analytical Addenda to Materia Medica which, besides embracing all significant data embodied in the British Pharmacopoeia and the British Pharmaceutical Codex together with their respective Addenda and Supplements, also includes assays and standards of the United States Pharmacopoeia XII and the American National Formulary VII. The methods for the alkaloidal assay of aconite, gelsemium and lobelia developed by the Poisons Sub-Committee appointed by our Society are given in full. This first section, which by itself constitutes a substantial book of reference, has been brought up to date in a truly commendable manner.

The next chapter, dealing with Proprietary Medicines, gives a detailed history of the law controlling these commodities and the formulae as disclosed on the labels of approximately 750 so-called "patent" medicines. This list does not include the "ethical" proprietaries customarily developed under direct medical guidance and now described in Vol. I of the Extra Pharmacopoeia. There follows a helpful section on the Nomenclature of Organic Compounds, a discourse on the Relation between Chemical Constitution and Therapeutic Effect, then more analytical data, including a Scheme for the Recognition of Organic Chemical Substances used in Therapeutics together with tables of corroborative tests for 378 compounds. Notes on Hydrogen Ion Concentration, Oxidation-Reduction Potentials, Indicators, Chromatographic, Microchemical and Spectrophotometric Analysis constitute useful introductions to the study of these specialised subjects, and in this connection an interesting and newly introduced section on Polarographic Analysis seems to be indicative of "the shape of things to come."

Analytical and legal information concerning Margarine, Jam, Vinegar, Food Preservatives, Food Colouring Materials, a re-written section on Flour and Bread, a general account of the composition and control of war-time Food Substitutes and a useful summary of the more important Statutory Rules and Orders affecting the composition of Food are features of vital interest to Public Analysts. Much work has gone into the preparation of the monograph on Vitamins and the chapter on the all important subject of Nutrition has been revised in the light of modern experience.

Copious notes on the Chemical and Microscopical Examination of Urine, Blood, Cerebrospinal Fluid and Stomach Contents fill 78 closely printed pages, while the extensive section on Bacteriological and Clinical Notes with Reference to Special Diseases is largely complementary to the information concerning anti-toxins, sera and vaccines given in Vol. I of Martindale. Other monographs of allied interest are headed Culture Media, Sterilisation of Materials for Injection and Surgical Dressings, Disinfectants and Antiseptics, and the book

winds up with chapters devoted to accounts concerning special principles of medical treatment such as X-Ray Diagnosis, Radium Therapy, Electrotherapy, Diathermy and Actinotherapy. The Index covers both volumes of the work and occupies 111 pages printed in double columns.

One hesitates to offer even constructive criticism on such a monumental work: what is the limited purview of a reviewer compared with this encyclopaedean survey? However, a few minor points concerning analytical practice may be mentioned in the hope that they will be of assistance for the next revision. Thus, under Blood Analysis it might have been possible to include an account of the highly successful alkaline haematin method of Clegg and King (*Brit. Med. J.*, 1942, ii, 329) for the determination of haemoglobin. Again, one notices that for the determination of sulphanilamide the obsolete method of Marshall *et al.* (*J. Amer. Med. Assoc.*, **1937**, **108**, 953, and *J. Biol. Chem.*, **1937**, **122**, 263), using dimethyl-α naphthylamine as coupling reagent, is given, whereas the modified method of Bratton and Marshall (*J. Biol. Chem.*, 1939, **128**, 537) using N-(1-naphthyl)-ethylenediamine dihydrochloride is unquestionably the method of choice; fortunately, King's micro modification (*Lancet*, 1942, **242**, 207) of the latter is included as an alternative. Under Milk and Milk Products one looks in vain for an account of the resazurin test, fully developed by 1940, and so valuable for detecting the incidence of mastitis in herds. The Notes on Water Analysis include a method for the direct colorimetric determination of fluorine, using the zirconium-alizarin S reaction, but there can be little doubt that preliminary distillation from perchloric acid is essential (Lockwood, ANALYST, 1937, **62**, 774), while the volumetric method involving the use of thorium nitrate has superseded the unsatisfactory colour test. Turning to the examination of potable waters for nitrates, it may not be inappropriate to observe that if one applies the strangely forgotten technique due to Frederick (ANALYST, 1919, **44**, 281) it is possible to conduct accurate determinations with the use of phenoldisulphonic acid without removing chlorides, even when the concentration of the latter amounts to 100 parts of chlorine per 100,000, and a description of this procedure might well replace portions of the existing account.

Against the weight of so great a book these trifling details are imponderable. The editor, his assistants, the Revision Committee and that little band of people with familiar names acknowledged at the end of the Preface may justifiably congratulate themselves upon the successful consummation of a task so well completed despite more than four years of direful war.

N. L. ALLPORT

MODERN SYNTHETIC RUBBERS. By H. BARRON, Ph.D., F.R.I.C. Second Edition. Pp. xii + 355. London: Chapman & Hall, Ltd. 1943. Price 28s.

The present edition of this topical work follows closely the arrangement of the first (Review, ANALYST, 1943, **68**, 97), but there is an increase of 81 pages, containing 76 additional tables. Much information is therefore available for the extra 3s. demanded for the second edition. The new material is dispersed throughout the book, but the most immediately obvious is a chapter devoted to ethyl cellulose. The section on the history and future prospects of the synthetic rubber industry, with special reference to competition with the natural product, is brought up to date and the new data support the author's optimism concerning the former (an enthusiasm maintained in spite of some adverse criticism), but peace will be accompanied by conditions at present difficult to forecast.

One may be certain, however, that elastomers have already become established and analysts will be well advised to study the later parts of this book, describing the chemistry and processing of the various synthetic materials. An account is given of the polymerisation of the raw materials, also their compounding and "vulcanisation" where these processes are appropriate. Here the non-specialist may be somewhat mystified through the use of trade names, without explanation, for grades of carbon and other ingredients of compounded rubbers. Although the author is concerned primarily with the technological aspects of his subject, many characteristics of the elastomers are noted in these pages and the book ends with a chapter on their properties. This chapter has been extended by 6 pages but, from the analytical standpoint, the position has not changed since the earlier edition was published, and the additional information now given presents the practising analyst with little foundation for his determinations. Consideration of the complexity of the chemistry of the elastomers immediately reveals the difficulty of devising a general scheme of analysis, but one must agree with Dr. Barron that "it is not rubber that is wanted, but the properties of rubber." This attitude of mind leads to the establishment of performance tests, many of which are

already familiar in the rubber industry, yet one can visualise circumstances (*e.g.*, in forensic chemistry) in which the chemical nature of the material might well prove important.

In spite of corrections to the first edition, the book retains numerous typographical errors, the most important being the omission of a line from page 279 and the transposed titles of Figs. 55 and 56, but the findings of the previous review deserve repetition: "The book can be thoroughly recommended to all interested in the subject." G. H. WYATT

ELECTRONIC THEORY AND CHEMICAL REACTIONS. By R. W. STOTT, M.A. Pp. viii + 112. London: Longmans, Green & Co., Ltd. Price 6s.

Probably the major advance in chemistry during the last decade is in the application of electronics to chemical reactions. Since very little information on this matter is to be found outside original papers and highly specialised books, the appearance of an elementary treatment of the subject should be very welcome.

The author, assuming a general acquaintance with the electronic structure of the elements and the various types of valency arising therefrom, plunges, after a brief introduction, into a lucid and convincing account of the causes of the uniqueness of water and of acidity in certain inorganic compounds. Of necessity, most of the book deals with organic compounds. Among the subjects treated are: the activity of hydrogen atoms adjacent to carbonyl groups, the hydrolysis of esters and esterification. Included are two excellent chapters on the structure of benzene and substitution in the benzene nucleus. The book closes with an account of some uses of radioactive and other isotopes in elucidating reaction mechanisms and in biological investigation. In passing, the increasingly important subject of the hydrogen bond receives considerable attention.

Throughout, stress is laid on the tentative nature of the theories discussed. Nevertheless the author has succeeded in producing a very stimulating and interesting volume which should be useful to scholarship candidates and first-year university students. Perhaps, too, graduates of long standing may find in it the key to the understanding of much modern organic chemical theory. HAROLD TOMS

INAUGURAL MEETING OF THE MICROCHEMICAL GROUP

IN view of the present uncertain conditions and of the extreme difficulties of travelling long distances, the Council has decided to postpone the inaugural meeting of the Microchemical Group until the day of the Ordinary Meeting of the Society in October. Members of the Group will receive notification of the meeting in due course.

PROPOSED FORMATION OF A GROUP DEALING WITH PHYSICAL METHODS OF ANALYSIS

IN pursuance of the policy for the formation of Groups for special branches of analytical chemistry proposed by the Council and approved by a meeting of the Society on November 3rd, 1943, the Council has had under consideration a proposal to form a Group dealing with physical methods of analysis. The Group would deal with such methods as for example:

1. Spectrographic methods
   - (*a*) Emission spectrograph;
   - (*b*) U.V. and visible absorption spectrograph;
   - (*c*) Infra-red absorption spectrograph;
   - (*d*) Mass spectrograph.
2. Quantitative photometric methods by means other than spectrophotometry.
3. Polarographic methods.
4. X-Ray diffraction.

The Council's decision as to the formation of the Group will depend on the number of members of the Society desirous of joining it. Members of the Society who wish to become members of the Group are asked to notify the Hon. Secretary of the Society, 7/8, Idol Lane, London, E.C.3.

Volume 69 **June, 1944**
No. 819, Pages 169—200

Contents

PROCEEDINGS OF THE SOCIETY OF PUBLIC ANALYSTS AND OTHER ANALYTICAL CHEMISTS

ABSTRACTS OF PAPERS PUBLISHED IN OTHER JOURNALS

Printed for the Society of Public Analysts and other Analytical Chemists by W. Heffer & Sons Ltd., Hills Road, Cambridge. **Communications to be addressed to the Editor, C. Ainsworth Mitchell, The Close, Weedon, Aylesbury, Bucks.**