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Comparison of Particle-size Analysis Results Obtained by Using a Centrifugal Photosedimentometer with those Obtained with Centrifugal Pipette Equipment

By M. W. G. BURT
(Atomic Weapons Research Establishment, Aldermaston, England)

AND B. H. KAYE
(I.I.T. Research Institute, 10 W. 35th Street, Chicago, Illinois, U.S.A.)

Size analysis results obtained with a centrifugal disc photosedimentometer are compared with those obtained with the Slater - Cohen disc centrifuge. It is shown that for three of the powders tested, agreement between the powders was good. The discrepancies noted with the fourth powder are probably due to dispersion difficulties.

SEVERAL new techniques for measuring the size distribution of powders by centrifugal sedimentation have been reported recently.[1,2,3,4] From theoretical reasoning alone it is difficult to predict the relationship between results obtained with the various instruments because of the different physical phenomena used to measure concentration changes within the settling suspension. The experiments reported in this paper form the first part of an investigation of the performance of centrifugal particle-size analysers. The instruments compared are the Slater - Cohen disc centrifuge[1] and the centrifugal disc photosedimentometer developed by Kaye.[2]

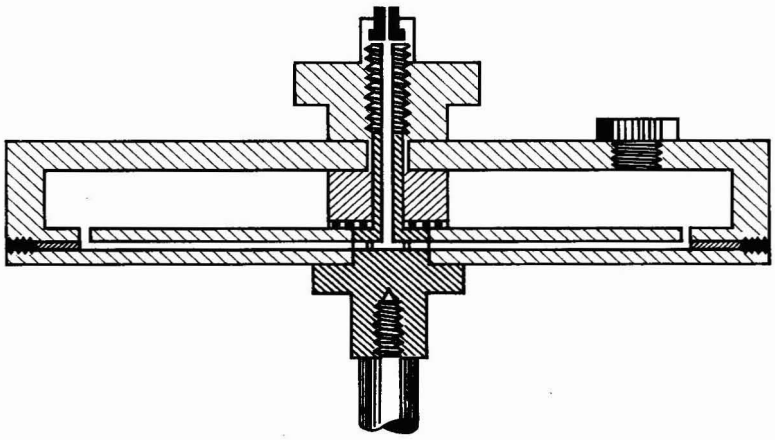

Fig. 1. Slater - Cohen disc centrifuge

The basic system of the Slater - Cohen centrifuge is shown in Fig. 1. It is essentially a centrifugal analogue of the Andreasen pipette method. The centrifuging chamber is a hollow disc with a central pillar. Several holes are drilled through the base of the disc from the central pillar and terminate in a series of apertures equally spaced around the perimeter of a circle concentric with the axis of rotation. The disc is partly filled with suspension and rapidly accelerated to the speed at which the analysis is to be carried out. Under centrifugal forces the free surface of the suspension is a cylinder concentric with the axis of rotation. When the suspension has been spun in the centrifuge for a given time, a sample of suspension is withdrawn through the base by applying suction to the drainage pillar. The solids concentration of the sample is then determined directly. The process is repeated at a series

of times. A discussion of the method and the treatment of the results has been presented fully.[1] In the centrifugal disc centrifuge developed by Kaye the disc has transparent portions through which a beam of light is passed to measure the concentration of the particles as they are centrifuged outwards. On the top plate of the disc there is a cylindrical entry port at the axis of rotation through which the suspension is injected. In the experiments reported in this communication the two-layer analytical procedure was used.[5] When this technique is used the centrifuge is run up to speed partially filled with clear liquid. When steady-state conditions are attained, the free surface of the fluid approximates to a cylinder, its axis being coincident with the axis of rotation. The powder to be examined is injected as a low concentration suspension through the entry port. Initially the incoming suspension does not have the energy to penetrate the free surface of the clear fluid, which has high angular momentum.

During this initial period, which appears to last for about 2 or 3 seconds, the suspension forms a thin uniform layer over the free surface of the clear fluid. Therefore, all the particles start off at essentially the same distance from the centre of rotation, and it follows that each size of particle reaches the light beam at the measuring zone at a definite time. By measuring the changes in light intensity as the particles cross the beam, the concentration of each size group is measured continuously. Claims of absolute accuracy have not previously been made for the centrifugal disc photosedimentometer because of the difficulty of interpreting light scattering measurements of concentration changes.

The light obscured by particles suspended in the path of a parallel beam of light is related to the concentration by the equation—

$$\log_e \frac{I_o}{I_t} = B\Sigma_{i=s}^{i=m}\, n_1 d_1^2 K_1 \qquad (1)$$

where I_o is the intensity of light beam entering the suspension;
I_t is the intensity of light beam leaving the suspension;
B is a constant, dependent on the dimensions of the beam of light and on the shape of the particles;
n_1 is the number of particles of diameter d_1;
K_1 is the extinction coefficient of a particle of diameter d_1;
s is the smallest particle present; and
m is the largest particle present.

By definition, K_1 is the ratio of light obscured by a particle to the light which it would have obscured if the laws of geometric optics were valid for the system under consideration. If, for a given system, the ratio of the particle diameter to the incident wavelength (λ) is greater than 100, the value of K_1 is constant, and can be taken as unity. For smaller ratios, K_1 becomes a complex function of d_1/λ, the relative refractive index of the particle with respect to that of the supporting medium and the shape of the particle.

The quantity, $\log_e I_o/I_t$, is usually termed the optical density of the suspension, and changes in the optical density with time can be related to the concentration of particles in specific size groups. The need to know B is usually eliminated by carrying out the analysis until all the particles are removed from the suspension, and then calculating the percentage concentrations. The difficulty in absolute interpretation of the results arises from the need to know K_1. In theory its value can be either calculated or measured experimentally; however, insufficient results are available for the analyst to do this often.

The calculations are so complex that in the past the writers[6,7,8] have recommended that a white-light source be used in conjunction with a relatively wide-angle acceptance receiver for the transmitted light to minimise fluctuations in K_1, and then assume that $K_1 = 1$. By making this assumption, the equation can be manipulated readily, although the results can only be regarded as having a high precision, but an unknown accuracy.

Recent work[9,10] has demonstrated the high precision that can be obtained with the centrifugal photosedimentometer, however, as it is well known that particles of the same order of magnitude as the wavelength of light scatter light more effectively per unit-weight than larger particles in comparable systems. There is a possibility that the percentage weight of fines within a powder could be over-estimated when using the centrifugal disc photosedimentometer.[6] It was to explore the influence of the light-scattering properties of the small particles that the performance of the centrifugal disc photosedimentometer was compared with the Slater - Cohen centrifuge, as in this centrifugal equipment the concentration changes in the suspension are measured gravimetrically.

Experimental

Four powders that had been analysed on the Slater - Cohen disc centrifuge and the appropriate analytical results were made available by Dr. Cohen. These four powders were analysed on the centrifugal disc photosedimentometer.

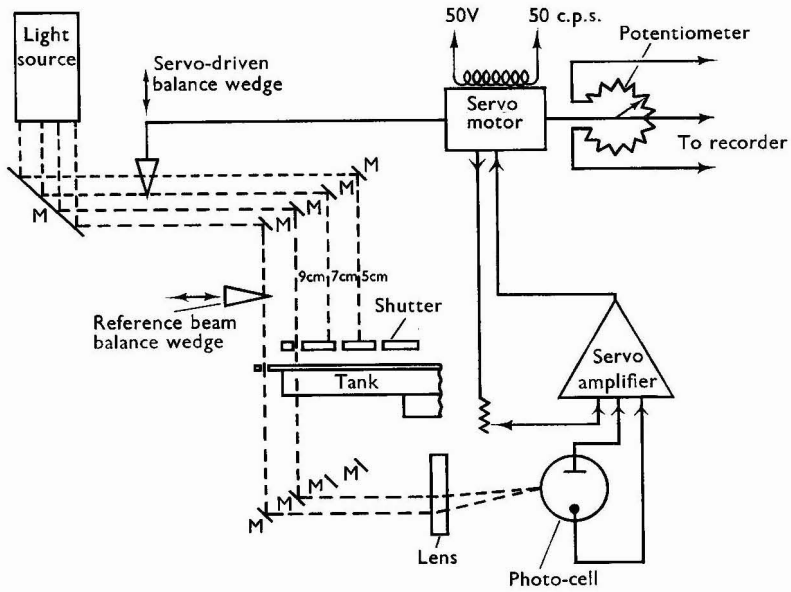

Fig. 2. Optical circuit of the Atomic Weapons Research Establishment centrifugal photosedimentometer

The equipment used in this investigation was based upon the original specification by Kaye,[2] and was designed and built at the Atomic Weapons Research Establishment, Aldermaston, by the Electronics Engineering Department. It consists of a shallow cylindrical stainless-steel tank with a system of slotted windows set at 3 fixed radii (5, 7 and 9 cm) such that light can pass through the tank and its contents at these radii. The tank can rotate synchronously at either 750 or 1500 r.p.m.

An incandescent lamp and lens unit directs a parallel beam of white light into four separate 45° mirrors. Three of these mirrors are able, one at a time (as selected by the movement of a shutter), to pass this measuring beam through the tank and its contents at the required radius on to a similar mirror beneath the tank, and thence on to a photoelectric cell. The fourth mirror reflects a reference beam, through a slot in the rim of the tank, avoiding the contents of the tank, on to another mirror below the tank that directs the beam on to the same photoelectric cell.

The system of slots is arranged to allow the cell to pick up the reference beam for one half cycle while the measuring beam is blocked, and the measuring beam on the next half cycle while the reference beam is blocked. The cell is thus continuously comparing the two beams.

In the path of the reference beam is a manually controlled optical wedge (the reference wedge). A similar wedge is placed in the measuring path, and is calibrated in optical density over the range 0 to 0·4, and automatically controlled by a continuously self-balancing servo-system. The movement of the latter optical wedge is recorded on a 10-inch chart recorder. With the rotating tank partly filled with clean liquid, the position of the pen recorder is adjusted to zero by operating the reference wedge manual control. The sample of powder suspension is then injected into the tank. Provided the solids concentration is low (see later discussion), the suspension forms a thin layer on the free surface of the fluid and the particles move outwards under centrifugal force. When the particles are in the path of the measuring beam, previously focused at the selected sampling radius, the intensity of the beam decreases. The strength of the beam passing through the disc is continuously compared, by means of the

photocell, with that of the constant reference beam. Any difference between the two beams constitutes an error signal that is amplified and fed to the servo-system. This servo-system causes the measuring wedge to take up a new position to restore the balance between the two beams. The movement of the wedge is recorded on the pen recorder, thus providing a record of optical density at the sampling zone with time. A diagram of the optical circuit is shown in Fig. 2; further technical data concerning the instrument have been published.[10]

PROCEDURE—

All four powders were analysed in the Atomic Weapons Research Establishment disc centrifuge by using water containing 0·01 per cent. of Cetavlon as the dispersed phase and spinning in a centrifuge at 750 r.p.m. It has been found experimentally that for the injected layer to be stable, very low solids concentrations have to be used in the injected suspension.[5,9,10] The value of the solids concentration used was determined by carrying out successive analyses at increasing solids dilution until the measured distribution was independent of the concentration used. The concentrations used in these experiments were—

	Percentage by volume
Barium sulphate	0·002
Silica	0·008
Titanium dioxide	0·0015
Zinc oxide	0·003

Suspensions were made up as follows:

The dry powder was coned and quartered, and the amount needed to make approximately 100 ml of suspension was taken and weighed.

The sample was gently mixed with water into a paste by using a rubber-tipped rod; it was then washed into a beaker. The suspension was made up to the required volume and viewed under a microscope to determine whether adequate dispersion had occurred.

With moderate stirring, 20-ml samples of suspension were withdrawn from the bulk by using a syringe. (The syringe was moved about in the suspension during this process.)

All the materials, with the exception of the zinc oxide, were readily dispersed in 0·01 per cent. Cetavlon solution, although some gentle working in a paste was required to break up agglomerates each time.

It was necessary, however, first to disperse the zinc oxide powder in a few drops of an 0·01 per cent. solution of sodium hexametaphosphate, and rather vigorous paste-mixing was required to break up some agglomerates that were quite large. Only the zinc oxide had both compounds added to it, the other three materials were dispersed with the Cetavlon alone.

CALCULATION OF SIZE DISTRIBUTIONS

Treasure[11] has shown that the range of sizes present in the sampling zone is proportional to d_t and that, therefore, the optical density of the zone at time t is directly proportional to d_t^3.

TABLE I

CALCULATION OF THE PROPORTION UNDERSIZE, PER CENT. W/W

Stokes' diameter limits, μ	Area under optical density *versus* d_t curve, cm² or inch²	Percentage by weight within d_t limits	Proportion undersize, per cent. w/w
d_1 to d_2	a_1	$w_1 = \frac{a_1}{A} \times 100$	100
d_2 to d_3	a_2	$w_2 = \frac{a_2}{A} \times 100$	$100 - w_1$
d_3 to d_4	a_3	$w_3 = \frac{a_3}{A} \times 100$	$100 - w_1 - w_2$
.	.	.	.
.	.	.	.
etc.	etc.	etc.	etc.
.	.	.	.
.	.	.	.
d_n to d_{n+1}	a_n	$w_n = \frac{a_n}{A} \times 100$	$100 - w_1 - w_2 \ldots - w_n$

where A = total area under curve
$d_1 > d_2 \ldots . > d_n$

By using the optical density *versus* time results given by the recorder, and applying equation (1) to determine the values of d_t at various times, a graph of optical density against d_t was constructed. This is the weight - frequency distribution curve for the powder. This curve is integrated by areas to give the percentage by weight within chosen particle diameter limits, and hence, cumulative proportions undersize, per cent. w/w, can be calculated. This is illustrated in Table I. The results of the two separate analyses on the centrifugal disc photosedimentometer are given in Table II. The data from the Slater - Cohen analysis are given in Table II.

TABLE II

DATA FOR THE SLATER - COHEN DISC CENTRIFUGE

Zinc oxide				Barium sulphate				Titanium dioxide (rutile)				Silica			
	Cumulative proportion undersize, per cent. w/w				Cumulative proportion undersize, per cent. w/w				Cumulative proportion undersize, per cent. w/w				Cumulative proportion undersize, per cent. w/w		
Stokes' diameter, μ	A	B	B′	Stokes' diameter, μ	A	B	B′	Stokes' diameter, μ	A	B	B′	Stokes' diameter, μ	A	B	B′
4·0	90	94	95	2·0	96	98	98	2·0	95	98	98	8·0	92	96	95
3·0	87	91	92	1·5	92	95	95	1·5	92	96	95	5·6	80	86	85
2·0	80	85	86	1·0	82	80	79	1·0	81	86	85	4·0	68	71	70
1·5	74	74	75	0·9	77	74	73	0·9	77	79	78	2·8	53	53	52
1·0	61	48	46	0·8	72	67	66	0·8	70	71	60	2·0	36	33	33
0·9	56	40	37	0·7	64	57	56	0·7	61	62	60	1·4	23	16	16
0·8	50	32	28	0·6	53	46	44	0·6	48	48	47	1·0	13	9	10
0·7	43	25	21	0·5	41	34	32	0·5	37	30	31	0·7	6	4	5
0·6	34	18	15	0·4	27	22	20	0·4	17	18	19	0·5	3	2	2
0·5	25	13	10	0·3	13	11	12	0·3	5	10	10	—		—	
0·4	16	8	6	0·2	3	5	4	0·2		4	5	—		—	
0·3	7	5	3	—		—		—		—		—		—	
0·2	2	2	1	—		—		—		—		—		—	

A = Analysis as determined on the Slater - Cohen disc centrifuge.
B and B′ = Analysis on the centrifugal disc photosedimentometer.

The data for the Slater - Cohen disc centrifuge are published by permission of the management of Simon Carves Ltd.

DISCUSSION

The agreement between the two separate analyses on the centrifugal disc photosedimentometer further confirms the precision attainable with this instrument. The agreement between the results from the two instruments for the barium sulphate, titanium dioxide and silica is of the order to be expected from differences in the dispersion techniques and sampling variations. An important factor governing the scattering power of a small particle is the difference in refractive index between the particle and the surrounding medium.[12] Taking the appropriate values of the refractive indices to be 1·64 for barium sulphate, 1·55 for silica, 2·76 for titanium dioxide (rutile) and 1·33 for water, the values[11] for the difference in refractive indices are 0·31, 0·22 and 1·43. Over the wide range of an important variable, and although the barium sulphate and titanium dioxide both contained a high percentage of particles, below 1-μ Stokes' diameter there is no indication that analysis by centrifugal disc photo sedimentometer over-estimates the fines content as compared to techniques in which the particle concentrations are measured gravimetrically.

The analysis of the zinc oxide by the centrifugal disc photosedimentometer gives a coarser estimate of the particle-size distribution than by the Slater - Cohen disc centrifuge. As reported earlier we had difficulties in dispersing this powder. It has been brought to our attention since we completed this work that the mixture of sodium hexametaphosphate with Cetavlon solution eventually used to disperse the zinc oxide was an unfortunate choice because Cetavlon contains quaternary ammonium compounds that are incompatible with soluble phosphates. The resultant precipitations of the insoluble quaternary phosphate probably interfered with the efficient dispersion of the zinc oxide. In the absence of information on the dispersion technique used by Dr. Cohen, no further comment on this discrepancy is possible.

In conclusion, it can be said that the results reported here indicate that the accuracy of the centrifugal disc photosedimentometer is probably comparable with that of the Slater - Cohen disc centrifuge, and the the fines content is not over-estimated for the powders considered.

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A Colorimetric Method for the Determination of Oxides of Nitrogen

BY GEORGE NORWITZ

(*Pitman–Dunn Laboratories, Frankford Arsenal, Philadelphia, Pennsylvania, U.S.A.*)

A method is described for determining oxides of nitrogen, and it has been applied to the gaseous products derived from initiating compositions. The oxides of nitrogen are absorbed from the sample into sulphuric acid, iron(II) sulphate is added, and the pink colour is measured. The interference effects of a number of gases such as hydrogen sulphide and sulphur dioxide have been investigated. The range of the method is 0·005 to 5 per cent. of oxides of nitrogen (calculated as nitrogen dioxide).

A METHOD was required for determining oxides of nitrogen in gases from initiating compositions. The oxides of nitrogen include nitric oxide, nitrogen dioxide, nitrogen trioxide and nitrogen tetroxide (but not nitrous oxide). The determination of oxides of nitrogen is usually made as the nitrate by the phenoldisulphonic acid method,[1,2] which is time-consuming. Another method for determining oxides of nitrogen involves the formation of nitrite by their absorption in alkali followed by diazotisation in which acid and α-naphthylamine or similar compounds are used.[1,3,4,5,6] This method has the disadvantage that the reaction is not stoicheiometric. Methods have also been proposed for determining the individual oxides of nitrogen in the presence of each other by titration[7,8] and by physical means[2,9,10,11,12] but, in general, these methods lack sensitivity.

It seemed that a relatively simple colorimetric method could be developed for determining oxides of nitrogen based upon absorption of the gases into sulphuric acid to form nitrate and nitrite, followed by the addition of iron(II) sulphate to form a pink colour. The formation of a pink colour with iron(II) sulphate has previously been used as the basis of a method for determining nitrate in many materials.[13 to 19]

METHOD

APPARATUS—

The apparatus (Fig. 1) consists of a gas bulb (A), an adaptor (B) and a solid cap (C). The cap with the opening (D) is needed only if standard samples are to be prepared to check the method. The gas bulb has a capacity of about 800 ml (the volume to the top stopcock should be determined by filling with water). The ground-glass joints of the gas bulb, adaptor and caps have hooks for the purpose of attaching springs. The stopcocks are greased with Chorofluorolube grease (obtainable from Hooker Chemical Corporation, Niagara Falls, New York) as other greases are attacked by oxides of nitrogen.

REAGENTS—

Sulphuric acid (10 + 3, *v/v*)—Place 600 ml of water in a Pyrex bottle, add 2 litres of concentrated sulphuric acid (sp.gr. 1·84) and cool the resulting mixture to room temperature.

Iron(II) *sulphate solution*—Add 4·0 g of iron(II) sulphate ($FeSO_4.7H_2O$) to a mixture of 55 ml of water and 5 ml of concentrated sulphuric acid, and stir to dissolve. Add 200 ml of concentrated sulphuric acid and cool to room temperature. Prepare daily.

Standard potassium nitrate solution—Dry analytical-reagent grade potassium nitrate at 100° C for 2 hours. After cooling it, dissolve 2·1973 g of it in water and dilute the solution to 1 litre in a calibrated flask.

1 ml ≡ 1·0 mg of nitrogen dioxide.

Sodium hydroxide solution (0·4 *per cent. w/v*).

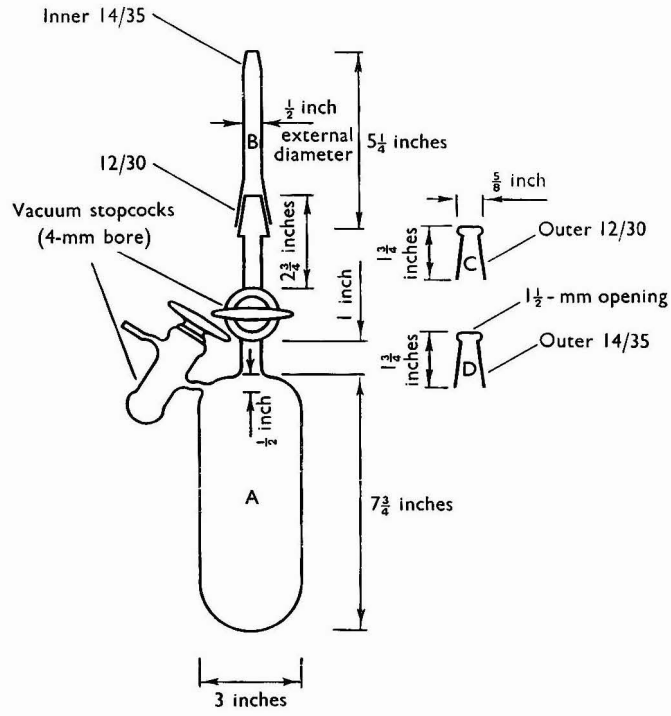

Fig. 1. Diagram of the apparatus used for the determination of oxides of nitrogen and preparation of standards

PREPARATION OF CALIBRATION GRAPH—

Transfer 2·0, 3·0, 4·0, 5·0, 6·0 and 7·0-ml portions of standard potassium nitrate solution, measured with a semi-microburette, to 250-ml beakers and add 5 drops of sodium hydroxide solution (0·4 per cent.). Carry out a determination on a reagent blank. Evaporate the solutions to dryness at low heat on the hot-plate. Cool the beakers to room temperature and add to each 25·0 ml of sulphuric acid (10 + 3, v/v) measured with a 25-ml graduated cylinder with 0·2-ml divisions. Allow the solutions to stand for several minutes to ensure dissolution of the salts. Add 25·0 ml of iron(II) sulphate solution to each beaker, swirl the solutions, and allow them to stand for 10 minutes. Transfer a portion of each solution in turn to a 1-cm cell and read the transmissions within 2 hours at 520 mμ with a spectrophotometer that has been set to 100 per cent. transmission with the reagent blank. Plot milligrams of nitrogen dioxide against percentage transmission.

PROCEDURE—

Collect the gas to be examined in the recommended gas bulb (A) (see Fig. 1) so that the pressure is less than 500 mm and the content of nitrogen oxides (as nitrogen dioxide) is less than 0·7 per cent., calculated at atmospheric pressure. For higher percentages of oxides of nitrogen use a correspondingly lower pressure. If desired, the gas can be collected in any suitable device and transferred to the gas bulb. Measure the pressure in the gas bulb by connecting it to an open-end manometer, observing the difference in the mercury levels and subtracting this difference from the atmospheric pressure. Attach the adapter (B) and add 25 ml of sulphuric acid, while manipulating the stopcock so that a little sulphuric acid remains in the adaptor. After all of the sulphuric acid has been added, turn the stopcock to admit air to bring to atmospheric pressure. Detach the adaptor and place it in a clean beaker. Attach the solid cap (C) to the stopcock and shake the bulb vigorously for 10

minutes. Re-attach the adaptor, open the second stopcock and add 25·0 ml of iron(II) sulphate solution. Shake the bulb gently once and allow it to stand for 5 minutes. Transfer a portion of the iron(II) sulphate to a 1-cm cell and read the transmission within 2 hours at 520 mμ with a spectrophotometer that has been set to 100 per cent. transmission with the reagent blank (equal volumes of sulphuric acid and iron(II) sulphate solution). Convert the reading obtained to milligrams of nitrogen dioxide by reference to the calibration graph. Calculate the corrected volume of the gas as follows—

$$V_c = \frac{V_b \times P \times 293}{760 \times T}$$

where V_c is the corrected volume of gas, ml;
V_b is the volume of gas bulb, ml;
P is the pressure of gas in bulb, mm; and
T is the room temperature, ° absolute.

Calculate the percentage of nitrogen dioxide as follows—

$$\text{Percentage of nitrogen dioxide} = \frac{W \times 24{\cdot}1 \times 100}{46\ V_c}$$

where W is the nitrogen dioxide found, mg; and
V_c is the corrected volume of gas, ml.

(The formula weight of nitrogen dioxide is 46 and the gram molecular volume at 20° C and 760 mm pressure is 24·1.)

Combining the above equations—

$$\text{Percentage of nitrogen dioxide} = \frac{136 \times W \times T}{V_b \times P}$$

To prepare for the next run, wash out the bulb with water and heat it over a Meker burner to drive off most of the moisture; while still hot connect the bulb to a vacuum pump.

Results

Application of the method to the determination of nitrate and nitrite ions—

For the direct application of the iron(II) sulphate method to the sulphuric acid solution of the oxides of nitrogen, the complexes of nitrate and nitrite with iron(II) sulphate must be the same. The relationship between the two complexes was therefore investigated by using potassium nitrate solution (2·1973 g per litre) and sodium nitrite solution (1·500 g per litre), both solutions containing the equivalent of 1·0 mg of nitrogen dioxide per ml. After treating 1 to 5 ml of the solutions with 5 drops of sodium hydroxide, the solutions were evaporated to dryness in beakers on the hot-plate. The beakers were cooled to room temperature, 25·0 ml of sulphuric acid were added to each beaker and the solutions were then allowed to stand for a few minutes to dissolve the salts. Iron(II) sulphate solution was then added to each beaker in 25-ml portions and after a few minutes the transmissions were measured at 520 mμ.

Nitrite solutions are not oxidised during the evaporation on the hot-plate.[20]

The potassium nitrate dissolved in the sulphuric acid without loss as was expected, considering that up to 1 g of potassium nitrate may be dissolved in sulphuric acid (94·5 per cent.) in the nitrometer method for determining nitrate.[21] The sodium nitrite dissolved in the sulphuric acid in the range indicated (up to 5 mg of nitrogen dioxide) without loss, but with amounts larger than 5 mg of nitrogen dioxide, some loss seemed to occur.

During the course of the work an improvement was made to the method for preparing the iron(II) sulphate solution. It is recommended that it should be prepared by dissolving the iron(II) sulphate in dilute sulphuric acid (5 parts of sulphuric acid + 55 parts of water, v/v) and then adding concentrated sulphuric acid. The iron(II) sulphate dissolved immediately without oxidation and clear water-white solutions were always obtained.

The sulphuric acid and iron(II) sulphate solution were added by means of a 25-ml graduated cylinder with 0·2-ml divisions. By using this technique the necessity of transferring to, and diluting in, calibrated flasks was avoided; this was convenient when working with gas bulbs.

The results for the potassium nitrate and sodium nitrite solutions showed that the transmissions obtained with the two solutions were the same, indicating that the complexes with the nitrate and nitrite were similar. Further confirmation that the complexes were identical was obtained by making a spectrophotometric curve for the nitrite complex; this curve was the same as that obtained for the nitrate complex.[16] The apparent explanation for the colour complexes being the same is presumably that the iron(II) ion reduces the nitrogen to the tervalent state (nitrous acid) in the concentrated sulphuric acid, and that the tervalent nitrogen then reacts with the iron(II) ion to form the pink complex. If the nitrogen is already in the tervalent state no reduction is involved.

These findings concerning the similarity of the two complexes are at variance with the conclusions of English[14] who stated that the complexes were not the same. English, however, used conditions that were different from those used by the present investigator.

APPLICATION OF THE COLORIMETRIC PROCEDURE TO OXIDES OF NITROGEN—

In order to check the application of the method to the analysis of oxides of nitrogen, it was necessary to prepare standard gas mixtures. In this investigation standard samples were prepared by using a Hamilton gas syringe (No. 1001), small cylinders of nitric oxide and nitrogen dioxide, and a special gas bulb (Fig. 1).

The cylinders of nitric oxide and nitrogen dioxide were of the standard type, 2 inches in diameter and 15 inches in height, (Matheson size No. 9). They were equipped with a Matheson No. 59 valve, which is a combination of a stainless-steel valve with a vertical stainless-steel tube (12 inches in height and $\frac{1}{4}$ inch in diameter).[22]

The procedure in preparing the gas mixture was as follows: the gas bulb (A) was evacuated and the adaptor (B) and the cap with the 1·5-mm opening (D) were attached. The end of the stainless-steel tube from the valve of the cylinder of nitric oxide or nitrogen dioxide was connected by a short piece of Tygon tubing (obtainable from U.S. Stoneware Co., Tallmadge, Ohio) to a piece of glass tubing that was several inches in length, and dipped below the surface of water contained in a small Erlenmeyer flask. The stainless-steel tubing was flushed at a moderate rate (under a hood) with nitric oxide or nitrogen dioxide, the gas-flow reduced to a very slow rate, and the Tygon tubing finally disconnected. The Hamilton syringe was inserted into the stainless-steel tube, filled and emptied, and then filled and emptied again. The syringe was filled with gas to the appropriate mark, inserted through the 1·5-mm opening of the cap of the adaptor, and depressed. The syringe was withdrawn and a finger immediately placed over the 1·5-mm opening. The top stopcock was opened a little, the finger withdrawn, the nitrogen oxide flushed into the system by the air, and the stopcock closed. Some vacuum should remain in the gas bulb after the flushing. Up to five 1-ml portions of the gas could be added by using this technique.

After adding the nitric oxide or nitrogen dioxide to the gas bulb, the cap was detached, and 25·0 ml of sulphuric acid was added while manipulating the stopper so that a little sulphuric acid remained in the adaptor. After all of the sulphuric acid had been added, air was admitted to bring the bulb to atmospheric pressure and the analysis was carried out as described in the section headed Method.

Experiments showed that vigorous shaking of the bulb for 10 minutes was sufficient.

It is necessary that at least half of the nitric oxide should be oxidised (an equimolar mixture of nitric oxide and nitrogen dioxide behaves like nitrogen trioxide and dissolves in sulphuric acid to form nitrous acid, while nitrogen dioxide alone dissolves in sulphuric acid to form nitrous acid and nitric acid). It would be expected that much more than enough oxygen to oxidise half of the nitric oxide to nitrogen dioxide would be present if the recommended method is followed, which requires that the initial pressure of 500 mm or less in the 800-ml gas bulb should be raised to atmospheric pressure by the admission of air. This conclusion was checked by filling two evacuated gas bulbs to 500-mm pressure with nitrogen, carefully admitting 0·1 and 1·0 ml of nitric oxide and proceeding as in the described method. Transmissions of 95 and 66 per cent. were obtained for the 0·1 and 1·0 ml of nitric oxide, respectively. When the same experiment was repeated without the initial nitrogen being present, transmissions of 96 and 66 per cent. were obtained.

Experimental calibration graphs were prepared with 0·1 to 4 ml of nitric oxide and 0·1 to 2·5 ml of nitrogen dioxide. The upper limits cited gave a transmission of approximately 20 per cent. These experimental calibration graphs followed Beer's law.

The volumes used for the preparation of the experimental calibration graph for nitric oxide were corrected to S.T.P., and the theoretical amount of nitrogen dioxide that would be produced was calculated by using the gram molecular volume. Actual recovery averaged 97·4 per cent. of theoretical (Table I). This is an excellent result considering the uncertain

TABLE I

RECOVERY OF NITRIC OXIDE

Nitric oxide		Nitrogen dioxide		
added, ml	corrected to S.T.P., ml	found, mg	theoretical, mg	Percentage recovery
0·10	0·09	0·18	0·19	94·7
0·20	0·18	0·36	0·37	97·3
0·50	0·46	0·90	0·94	95·7
1·00	0·91	1·85	1·87	98·9
1·00	0·91	1·85	1·87	98·9
2·00	1·82	3·68	3·74	98·4
3·00	2·73	5·50	5·61	98·0
4·00	3·64	7·30	7·48	97·6
			Average ..	97·4

state of purity of nitric oxide.[9,10] The reproducibility and the ratios of the nitrogen dioxide recovered in the experiments with nitrogen dioxide were good (Table II). Application of the gram molecular volume calculations to nitrogen dioxide is uncertain because of the equilibrium between nitrogen dioxide and nitrogen tetroxide, the impurities that are present and the fact that nitrogen dioxide does not behave as a perfect gas. Accepting that nitrogen tetroxide is 20 per cent. dissociated into nitrogen dioxide at 27° C at atmospheric pressure,[23] the recoveries in Table II are approximately 90 per cent. of theoretical values.

TABLE II

RECOVERY OF NITROGEN DIOXIDE

Nitrogen dioxide		
added, ml	corrected to S.T.P., ml	found, mg
0·10	0·09	0·30
0·20	0·18	0·60
0·50	0·46	1·50
0·50	0·46	1·58
1·00	0·92	3·00
1·00	0·92	2·96
2·00	1·84	5·91
2·00	1·84	5·91
2·50	2·30	7·48

The calibration curve obtained with nitric oxide could be readily used for calculating the percentage of nitric oxide in a gas sample, especially if all volumes were referred to fixed conditions, *e.g.*, 760 mm pressure and 20° C. The graph obtained with nitrogen dioxide cannot be used advantageously for calculating the percentage of nitrogen dioxide, as a mixture of nitrogen tetroxide and nitrogen dioxide rather than nitrogen dioxide is used for plotting the graph. The graphs for nitric oxide and nitrogen dioxide prepared as above were found to be useful for testing interferences.

The method in which the 800-ml bulb and 500-mm pressure are used will give good results for up to 0·7 per cent. of oxides of nitrogen, calculated as nitrogen dioxide For the analysis of larger amounts of nitrogen dioxide it is only necessary to decrease the pressure.

The minimum amount of oxides of nitrogen that can be detected (by using as a criterion the amount that will give 98 per cent. transmission) is approximately 0·005 per cent. of nitrogen dioxide.

If it is known that nitric oxide is the only oxide present, the calculation can, of course, be made as nitric oxide.

Low results were obtained each time on shaking with the iron(II) sulphate solution without first dissolving the nitrogen oxides in sulphuric acid.

INVESTIGATION OF THE EFFECT OF OTHER GASES—

A study of possible interferences (Table III) showed that carbon monoxide, carbon dioxide, hydrogen, oxygen, nitrogen, methane and nitrous oxide did not interfere. The presence of as much as 0·6 per cent. of sulphur dioxide (calculated at atmospheric pressure) did not cause interference, but the presence of more than 0·04 per cent. of hydrogen sulphide (calculated at atmospheric pressure) gave low results.

TABLE III

TESTS FOR INTERFERENCES

Present (in 805 ml volume)	Volume found, ml: Nitrogen dioxide	Volume found, ml: Nitric oxide
Nitrous oxide (500 mm pressure)	0·00	—
Nitrous oxide (100 mm pressure) + 1·00 ml of nitric oxide	—	1·02
Nitrous oxide (100 mm pressure) + 1·00 ml of nitrogen dioxide	1·00	—
Gas mixture* (300 mm pressure) + 1·00 ml of nitric oxide	—	0·99
Gas mixture* (300 mm pressure) + 1·00 ml of nitrogen dioxide	0·98	—
Sulphur dioxide (1 ml) + 0·50 ml of nitrogen dioxide	0·52	—
Sulphur dioxide (200 mm pressure) + 0·50 ml of nitrogen dioxide	0·41	—
Hydrogen sulphide (0·1 ml) + 1·00 ml of nitrogen dioxide	1·00	—
Hydrogen sulphide (0·3 ml) + 1·00 ml of nitrogen dioxide	0·98	—
Hydrogen sulphide (0·5 ml) + 1·00 ml of nitrogen dioxide	0·68	—
Hydrogen sulphide (1 ml) + 1·00 ml of nitrogen dioxide	0·36	—

* Contains: carbon dioxide, 40·0 per cent.; nitrogen, 44·0 per cent.; oxygen, 2·9 per cent.; hydrogen, 4·0 per cent.; carbon monoxide, 8·7 per cent.; methane, 0·6 per cent.

GASES DERIVED FROM INITIATING COMPOSITIONS—

The gases derived from 5 different initiating compositions were analysed. The results obtained were 0·56, 0·021, 1·21, 1·07 and 0·53 per cent. of nitrogen dioxide.

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Flame-spectrophotometric Determination of Calcium in Human Saliva

By J. G. JONES and J. D. R. THOMAS
(*Department of Chemistry, Welsh College of Advanced Technology, Cardiff, Wales*)

A method is described for the determination of calcium in saliva by using the Unicam SP900 flame spectrophotometer with an acetylene - air flame. Interference by added orthophosphate is negligible. This, together with the good recoveries of calcium added to saliva, indicates that the method is applicable to the direct determination of calcium in saliva and that no special precautions need be taken to insure against interferences.

The determination of calcium in biological fluids continues to be a subject of intense interest. The methods that have been developed range from the classical oxalate - permanganate titration and oxalate - carbonate gravimetric method, through the more recent complexometric and colorimetric techniques, to the physico-chemical methods of emission flame photometry and atomic-absorption spectroscopy.

Saliva has been examined for calcium by several of the above methods, but, unfortunately, most are subject to certain disadvantages. For example, the oxalate methods require large amounts of saliva, while the flame-spectroscopic methods are frequently subject to interferences, especially by refractory-forming materials. In this latter connection, the presence of phosphorus is a major source of interference in both the flame-photometric[1] and atomic-absorption spectroscopic[2] methods.

Acetylene - air mixtures give hotter flames than either coal gas - air or propane - air mixtures, a feature which facilitates the dissociation of the refractory compounds that tend to arise in the presence of phosphates and other materials present in biological fluids. Acetylene - air mixtures are, therefore, used on a wide scale in flame spectroscopic methods. However, there does not appear to be any reference in the literature to the determination of calcium in saliva by using these mixtures in conjunction with the flame-spectrophotometric method.

In view of the rather complex measures that frequently have to be taken to overcome interferences in the determination of calcium in saliva, particularly with regard to phosphorus, the present investigation was undertaken to explore the possible application of the use of the acetylene - air flame in conjunction with the flame-spectrophotometric method for the direct determination of calcium in whole saliva.

Experimental

Apparatus—

A Unicam SP900 flame spectrophotometer, fitted with an acetylene - air burner unit is used. To ensure sound reproducibility of results, the instrument should be allowed a warm-up period of several hours in accordance with the manufacturer's instructions.

Preparation of solutions—

Standard calcium solution—Dissolve 0·2518 g of calcium carbonate in 60 ml of 0·1 M hydrochloric acid and dilute to 1 litre with de-ionised water. This stock solution contains 100 μg of calcium per ml and is diluted to the required concentration with de-ionised water, or as otherwise described for the phosphorus interference studies.

Standard orthophosphate solution—Dissolve 1·8700 g of disodium hydrogen orthophosphate, $Na_2HPO_4.12H_2O$, in 1 litre of de-ionised water. This stock solution contains 500 μg of orthophosphate per ml (PO_4^{3-}), and for the phosphorus interference studies it is added in appropriate aliquots when preparing the standard calcium solutions. However, for the study of the effect of orthophosphate concentration at 1500 μg per ml (as PO_4^{3-}), prepare the solution directly by dissolving 1·4020 g of the disodium hydrogen orthophosphate in de-ionised water. Add 5 ml of the stock calcium-containing solution, and dilute to 250 ml with de-ionised water.

SETTING-UP AND CALIBRATION PROCEDURE—

Set up the flame spectrophotometer. Calibrate the instrument at 4227 Å and a slit width of 0·065 mm, with the acetylene - air flame controlled at 28·5 p.s.i. for the air and at 13·2 inches pressure for the acetylene.

With the voltage selector at 1, the filter selector at 3 and the electrical band-width at 1, adjust the galvanometer controls to give a zero galvanometer deflection while de-ionised water is sprayed into the flame. With a calcium solution containing 10 μg per ml being sprayed into the flame, adjust the instrument gain selector to give a galvanometer deflection of 80.

Continue adjusting the galvanometer controls and the instrument gain selector, whilst water and the calcium solution are alternately sprayed into the flame, to a galvanometer deflection of zero and 80, respectively, until no further adjustment is necessary.

Switch the electrical band-width control to 4, and by using standards containing 0 to 10 μg of calcium per ml, check the linearity of the calibration.

DETERMINATION OF CALCIUM IN WHOLE SALIVA—

After following the setting-up and calibration procedure described above, dilute 1 ml of saliva to 25 ml (or alternatively, to 10 ml) with de-ionised water and spray into the acetylene - air flame. When the dilution factor has been allowed for, the calcium content can then be determined directly from the galvanometer deflection.

INTERFERENCE CHECKS—

Phosphorus as orthophosphate—Check the calibration for absence of interference by phosphorus, in the form of orthophosphate, on solutions containing 2 μg of calcium per ml together with varying amounts of orthophosphate ranging from 0 to 100 μg of orthophosphate per ml. The solution containing 1500 μg of orthophosphate per ml is prepared by the method described above.

As a further check on the absence of interference by added orthophosphate, take readings on various calcium-containing solutions, each containing 100 μg per ml of added orthophosphate.

De-proteinisation—Check whether de-proteinisation has any effect on the apparent calcium content of the saliva by adding 5 ml of an aqueous 4 per cent. solution of trichloroacetic acid to 1 ml of saliva and spinning the solution in a centrifuge. Dilute the supernatant liquid to 25 ml with de-ionised water and determine the calcium content in the manner described above.

COMPARISON OF PROPOSED FLAME-SPECTROPHOTOMETRIC METHOD WITH THE CLASSICAL OXALATE - PERMANGANATE TITRATION METHOD—

De-proteinise[3] a 10-ml aliquot of saliva with 1·0 ml of 30 per cent. trichloroacetic acid. Spin the solution in a centrifuge. Use the supernatant liquid to determine the calcium content by the oxalate - permanganate titration method.[4] Compare the result with that obtained for a 1-ml aliquot of the same sample of saliva by the proposed flame-spectrophotometric method.

RESULTS

CALIBRATION—

Setting the galvanometer deflection to zero with de-ionised water required the galvanometer controls to be set at 8·6, while the instrument gain selector was set at 3·0 at the deflection of 80 for the solution containing 10 μg of calcium per ml.

The check on the setting-up calibration for calcium gave the following linearly related figures—

Calcium concentration, μg per ml ..	0	2	4	6	8	10
Galvanometer deflection	0	16	32	48	64	80

Calibration checks on a solution containing 2 μg of calcium per ml and varying amounts of orthophosphate gave the following galvanometer deflections, which indicate negligible interference by phosphorus under these conditions—

Phosphorus concentration, μg of PO_4^{3-} per ml ..	0	10	30	50	70	100	1500
Galvanometer deflection	16	17	17	17	17	17	17

The negligible interference by phosphorus in the form of orthophosphate was confirmed by the following galvanometer deflections obtained on solutions, each containing 100 μg of orthophosphate per ml in addition to calcium—

Calcium concentration, μg per ml ..	0	2	4	6	8	10
Galvanometer deflection	0	17	30	45	63	78

CALCIUM IN WHOLE SALIVA—

The procedure for the determination of calcium in whole saliva was tested on spat saliva collected from a number of students. This was carried out 2 hours after the previous meal, no brushing of teeth, rinsing of mouth, smoking or any other treatment having occurred in the intervening period. Results were obtained on saliva collected on each of 4 different days, and the calibration curve obtained as above for pure calcium-containing solutions was used.

The results quoted in Table I were obtained on 1 in 25 dilutions of saliva in de-ionised water. It was ascertained several times that 1 in 10 dilutions gave identical figures for the calcium content of the whole saliva. On the other hand, direct determinations on undiluted saliva are not feasible, as the concentrations of calcium are then beyond the range of the flame spectrophotometer when used with the acetylene - air flame.

TABLE I

CALCIUM CONTENT OF WHOLE SALIVA BY THE METHOD DESCRIBED

	Calcium content, μg per ml of whole saliva			
Student	Day 1	Day 2	Day 3	Day 4
1	50	50	49·5	50
2	50	50	50	50
3	54	53	54	54
4	50	50	50	50
5	49	49	50	49
6	50	50	51	50
7	57	57	58	57
8	56	56	55	57
9	50	49	49	51
10	47	46	46	48

The higher dilution for the saliva (1 in 25) was selected for the investigation as it gave saliva solutions with calcium content nearer the recommended working range of the SP900 flame spectrophotometer.

Readings taken on each side of the region of calcium emission, *i.e.*, at 4180 Å and 4280 Å, gave zero galvanometer deflections for both 1 in 25 and 1 in 10 diluted samples. This indicates the absence of direct sodium interference.

Apart from the determination of calcium on ordinary diluted samples, the saliva taken on Day 1 was also de-proteinised and treated in the manner described above. The apparent calcium content obtained in this way corresponded exactly with the values quoted in Table I for this day. A blank determination on the trichloroacetic acid treated in an analogous manner gave a zero content for calcium. However, a solution containing 50 μg of calcium per ml prepared from the stock solution and treated in the same way as the saliva gave a reading for the calcium content corresponding to 50 μg per ml.

In a selected number of instances, 1 in 25 diluted solutions of saliva were prepared to contain 150 μg of added orthophosphate per ml. On no occasion was there any significant difference in the apparent calcium content over that observed for the normal uncontaminated diluted solutions.

Determination of the calcium content of a series of samples of saliva by the proposed method and by the classical oxalate - permanganate titration method indicated a favourable measure of agreement between the two methods—

Proposed method (calcium in μg per ml)	47	52·5	55	58	42	42
Oxalate - permanganate method (calcium in μg per ml) ..	45	53	53·5	57·5	42	40·5

RECOVERY OF CALCIUM ADDED TO WHOLE SALIVA—

Statistical experiments were designed to determine the standard deviation of percentage recoveries of added calcium to whole saliva. For a series of 20 samples, the standard deviation was found to be $\pm 1{\cdot}62$ per cent., and for a further series of 100 samples, $\pm 1{\cdot}50$ per cent., the added calcium ranging from 25 to 100 p.p.m. for each series.

DISCUSSION

Flame spectrophotometry used in conjunction with the acetylene - air flame has revealed a quick method for the determination of calcium in saliva. Reproducibility is excellent, provided an adequate warm-up period has been allowed for the instrument.

The negligible effect of added orthophosphate, even in considerable excess, on the flame-spectrophotometric response for standard calcium solutions suggests that the acetylene - air flame is effective in dissociating refractory compounds of calcium, and it indicates that no special precautions need to be taken to overcome possible interference by phosphorus. This is confirmed by the reproducibility of the calcium readings on saliva, even in the presence of orthophosphate.

The consistency of the results of untreated whole saliva and de-proteinised whole saliva, the good recoveries obtained for calcium added to whole saliva, and the zero galvanometer deflections each side of the calcium emission, all provide further confirmation that the method is apparently free from interference effects.

A propane - air flame can cope with larger concentrations of calcium, but, as indicated in the opening remarks, interference by phosphorus creates a problem so that the advantages of being able to use undiluted saliva are lost. The disadvantages of the time consumed in preparing diluted samples is greatly outweighed by the advantage of overcoming interference effects.

A final point calling for comment is that dilution of saliva at the different levels of 1 in 10 and 1 in 25 produces no difference in the apparent calcium content. This is contrary to what might have been expected in the light of Newburn's remarks[2] to the effect that dilution with water reduces the apparent calcium content in the determination by atomic-absorption spectroscopy. However, there is no indication of Newburn having used an acetylene - air flame.

We thank the authorities of the Unilever Research Laboratory, Isleworth, for suggesting the project.

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Colorimetric Determination of Sodium Isethionate by Means of Ammonium Ceric Nitrate

By D. W. G. DICKER* and T. H. NEWLOVE
(*Unilever Research Laboratory, Port Sunlight, Cheshire*)

A direct colorimetric method is described for determining sodium isethionate (2-hydroxyethane sulphonate) in aqueous solution by means of its red complex with ammonium ceric nitrate. Ethylene glycol, if present, is removed from the sample by extraction with ethyl acetate before colorimetric determination.

The over-all precision of the method is within ±1 per cent.

Sodium 2-hydroxyethane sulphonate (isethionate) is manufactured commercially by the addition of sodium bisulphite to ethylene oxide[1] and is used for the synthesis of surface-active agents by esterification with fatty acids. No method of analysis appears to have been published for this compound, and it is our object to describe a suitable direct colorimetric method. This method is based on the use of ammonium ceric nitrate (ammonium hexanitratocerate $(NH_4)_2Ce(NO_3)_6$) for the colorimetric determination of alcohols.

The formation of coloured complexes of alcohols and ammonium ceric nitrate was first reported in 1940 by Duke and Smith[2] as a qualitative test for the alcoholic hydroxyl group. Later, Duke[3] described how the reaction could be used for determining relatively small amounts of alcohol in mixtures with approximately 5 per cent. accuracy. In 1952, Reid and Truelove[4] reported the use of ammonium ceric nitrate for the precise determination of alcohols in dilute aqueous solution. A further paper by Reid and Salmon[5] described a modified procedure for trace amounts (up to 0·1 per cent. w/w) of alcohols, and this is the basis of the method described in the present report.

It may be noted that the ammonium ceric nitrate reagent has also been shown to give colour reactions with certain thiophen derivatives and other heterocyclic compounds[6] and has been used for the determination of 1-chloropropan-2-ol.[7]

Mechanism of reaction—

A study of the mechanism of the reaction was made by Duke and Smith.[2] These workers carried out potentiometric titrations of aqueous potassium ceric nitrate and of ammonium ceric nitrate in absolute ethanol with aqueous and absolute ethanolic standard solutions of sodium hydroxide, respectively. They deduced that the basis of the test was the formation of the red complex anion according to the equation—

$$[Ce(NO_3)_6]^- + ROH = [Ce(OR)(NO_3)_5]^- + H^+ + NO_3^-$$

They also found that alcohols with up to 10 carbon atoms were responsive to the test.

Method

Reagents—

Ammonium ceric nitrate reagent[4]—Dissolve 20 g of pure ammonium ceric nitrate in 100 ml of standardised 4 N nitric acid. Allow to stand overnight and then filter through a No. 4 sintered-glass funnel. Standardise by titration with 0·1 N ammonium ferrous sulphate solution, and adjust the strength to 0·36 N, if necessary.

Acetone—Re-distil from anhydrous calcium chloride.

Ethyl acetate.

All the reagents should be of the highest purity obtainable.

Apparatus—

Spectrophotometer—A suitable instrument is the Unicam SP600.

* Present address: Unilever House, Blackfriars, London, E.C.4.

PROCEDURE—

Accurately weigh about 4 g of the solution containing approximately 50 per cent. w/w of sodium isethionate into an evaporating dish and add 5 ml of dry acetone. Place the dish on the steam-bath and gently evaporate the acetone. Add a further 5 ml of acetone and evaporate again while stirring with a glass rod, taking care to avoid loss of any solid material by spitting of the solution.

Remove the dish from the steam-bath and allow the solid residue to cool. Extract the residue by stirring with 10 ml of ethyl acetate and filter the suspension through a sintered-glass funnel (No. 4 porosity) with suction. Transfer quantitatively any residue from the dish to the filter with three 5-ml portions of ethyl acetate and then wash the sinter with two 5-ml portions of this solvent. Continue air suction for about 5 minutes to ensure that the solvent has been removed. Discard the filtrate. Wash the glass dish with warm distilled water (at a temperature of about 40° C) and pass the solution through the funnel into a clean Büchner flask. Use about 200 ml of warm water in this way and finally transfer the solution to a 500-ml calibrated flask. Wash the Büchner flask four times with 25-ml portions of distilled water, and transfer these to the calibrated flask. Allow to cool, then make up to volume with distilled water.

Transfer by pipette 5 ml of this solution (containing about 0·02 g of sodium isethionate) into a small flask and add by pipette (with a safety bulb) 2 ml of the ammonium ceric nitrate reagent. Mix the solutions well and, after exactly 5 minutes, read the optical density of the resultant solution at 486 mμ in a 1-cm cell, with 5 ml of distilled water and 2 ml of the ammonium ceric nitrate reagent in the reference cell.

Read off the amount of sodium isethionate in the sample from a previously prepared calibration graph (which should be linear and should pass through the origin) relating optical density to concentration. Alternatively, calculate the amount of isethionate by multiplying the optical density by a factor given by the slope of the line.

DISCUSSION OF RESULTS

SPECTROPHOTOMETRIC EXAMINATION—

Spectrophotometric studies of the red anionic complex formed by sodium isethionate and the reagent were made with a pure sample of sodium isethionate, that had been re-crystallised 4 times from acetic acid to a constant melting-point of 196° to 198° C, and a Cary model 14 (ultraviolet and visible) recording spectrophotometer. The absorption spectrum of the red complex was measured over the range 400 to 600 mμ, during the period of 4·5 to 6 minutes after mixing the isethionate solution and the reagent, the reagent mixed with water (in place of the sodium isethionate) being used in the reference cell. The spectrum showed a fairly sharp peak at 448 mμ. Reid and Salmon,[5] however, specified 486 mμ as the wavelength for measurement of simple aliphatic alcohol complexes with the reagent.

Further tests with the reagent alone against water in the reference cell showed rapidly increasing optical absorption with decreasing wavelength in the region of 486 to 448 mμ. By using the SP600 spectrophotometer recommended for the procedure, results could not be obtained at wavelengths below 460 mμ because of the high absorption of the blank. The wavelength 486 mμ was therefore chosen for our measurements; subsequent absorption measurements were made with a Unicam SP600 spectrophotometer.

THE EFFECT OF TIME ON THE OPTICAL DENSITY OF THE COMPLEX—

The colour of the complex formed by sodium isethionate solution (0·25 per cent. w/v) and the reagent fades linearly with time at a rate corresponding to a reduction of 0·002 in optical density per minute. Provided that the readings are taken within ±1 minute of a given time interval after mixing the solution of the sample and the reagent (5 minutes is the recommended time), the variation in optical density may be neglected.

THE EFFECT OF TEMPERATURE ON THE OPTICAL DENSITY OF THE COMPLEX—

A rise in temperature from 10° to 30° C had the effect of raising the optical density of the complex (with 0·30 per cent. w/v sodium isethionate solution) from 0·423 to 0·440, but with a further rise from 30° to 40° C, the optical density fell from 0·440 to 0·407, showing instability of the complex.

REAGENT STABILITY—

Tests were made with 5-ml portions of 0·25 per cent. w/v solution of sodium isethionate and 2 ml of ammonium ceric nitrate reagent after the reagent had been allowed to age. For up to 67 days there was no significant change in the optical density observed.

INTERFERING SUBSTANCES—

Tests were made to determine whether certain substances that are liable to be present in commercial sodium isethionate would interfere with the determination. The comparisons which follow refer to the standard 0·4 per cent. w/v strength recommended for sodium isethionate solution, although isethionate was absent from the test solutions.

Sodium sulphate—The effect was negligible even at the same weight concentration as the sodium isethionate. At higher concentrations there is an effect, but this is of no practical significance as, in an isethionate sample of the type envisaged, sulphate would approximate to 1 per cent. of the isethionate content.

Sodium chloride—This substance interfered, but the effect was approximately 1/20th of that of sodium isethionate at the same weight concentration. As the level of chloride present in commercial sodium isethionate would be about 0·05 per cent., the effect is negligible.

Sodium sulphite—This substance caused a reduction in optical density by bleaching. It was found that 0·0002 g of sodium sulphite in 5 ml of solution reduced the optical density by 0·012, which is negligible. This amount of sodium sulphite would correspond to 0·5 per cent. in the sodium isethionate sample when using the specified amount of the latter for test. Hence sodium sulphite can be tolerated up to at least 0·5 per cent.

Ethylene glycol—This substance produced optical densities approximately twice those given by sodium isethionate solutions at the same level of concentration.

As approximately 1 per cent. of glycol is generally found in commercial sodium isethionate, a procedure for its removal before determining isethionate was sought. It was found that extraction of the total solid material in the isethionate sample with ethyl acetate was effective in removing the glycol, and was accordingly adopted in the recommended procedure.

CALIBRATION GRAPH—

A calibration graph was prepared by treating 5-ml samples of standard solutions of pure re-crystallised sodium isethionate with 2-ml portions of the reagent, according to the standard test procedure. The resulting optical densities are given in Table I.

TABLE I

OPTICAL DENSITIES OF STANDARD SOLUTIONS OF SODIUM ISETHIONATE

Weight of sodium isethionate in 5-ml sample, mg	Optical density (1-cm cell), 486 mμ*
5	0·158
10	0·296
15	0·438
20	0·580
25	0·721
30	0·858

* Measured with the SP600 spectrophotometer.

The above results fall on a straight line and, within the limits of accuracy of the instrument, this line passes through the origin.

ACCURACY AND PRECISION—

The accuracy and precision of the method were measured by subjecting to the recommended procedure mixtures (all of which were made up to 4·0 g with water) containing a known amount of pure sodium isethionate, together with various amounts of sodium sulphate, sodium chloride, sodium sulphite and ethylene glycol. The results are given below.

	Sodium isethionate		Standard deviation	
No. of determinations	taken, g	found, g	absolute, g	relative, per cent
11	2.00	1.99 (mean)	0.015	0.75

The relative precision, as defined by standard deviation, is within ± 1 per cent.

With amounts normally of about 50 per cent. w/w of sodium isethionate on 4 g of test solution, the standard deviation would be 0·38 per cent. and the 95 per cent. confidence limits $\pm 0{\cdot}84$ per cent.

It may be noted that in the solutions tested, ethylene glycol was present up to 3·75 per cent. and the recoveries of sodium isethionate were still satisfactory.

Comparative tests—

The ammonium ceric nitrate colorimetric method described in this paper was applied to three manufactured samples of sodium isethionate (of unknown composition) and the values thereby obtained for sodium isethionate content were compared with the respective values calculated by subtraction of the separately determined sodium sulphate, sodium chloride, sodium sulphite and ethylene glycol from the total solid content in each sample. The results are given in Table II and show satisfactory agreement.

Table II

Comparison of results by the colorimetric and weight difference methods

Sample	Sodium isethionate, per cent., found by—	
	Colorimetric method	Weight difference method
1	51·7	52·1
2	57·6	58·0
3	59·5	59·2

Conclusions

A rapid colorimetric method for the determination of sodium isethionate in commercial samples of this substance has been described, and has been shown to give results that are both accurate and of good precision.

We thank the Directors of Unilever Ltd. for permission to publish this work, and acknowledge the assistance of Mr. C. J. Goodwin in performing the analyses.

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The Spectrophotometric Determination of Vitamin D in Fresh-water Fish Liver Oils

By R. K. BARUA and M. V. K. RAO
(*Department of Chemistry, Gauhati University, Jalukbari, Assam, India*)

There are many difficulties associated with the determination of vitamin D, especially in natural products such as fish liver oils. Vitamin A is the chief interfering material; it masks the absorption of vitamin D both in the ultraviolet region and in the antimony trichloride colour test, making the determination of vitamin D almost impossible. In addition to vitamin A_1, fresh-water fish liver oils contain vitamin A_2, which also interferes in direct spectrophotometry. A method for determining vitamin D is described, in which vitamins A_1 and A_2 are eliminated by converting them to anhydrovitamins A_1 and A_2 and separating them from vitamin D by chromatography on an alumina column.

A method for determining vitamin D in fresh-water fish liver oils is described, in which both vitamins A_1 and A_2 are eliminated by converting them to the anhydrovitamins A_1 and A_2, by treatment of the unsaponifiable material from the liver oils with 0·033 N ethanolic hydrogen chloride. The light petroleum extract of the resulting product is chromatographed by placing it on a column of weakened alumina containing 8 per cent. w/v of water. On developing the chromatogram with light petroleum, both anhydrovitamins A_1 and A_2 flow through the column. Three distinct yellow bands develop on the column; the colourless portion between the second and third yellow zones contains all the vitamin D. The colourless portion is extruded and eluted with diethyl ether, the eluate is evaporated to dryness under reduced pressure, and the colour developed with antimony trichloride - acetyl chloride reagent is measured within 30 seconds of adding the reagent. Over 90 per cent. of the vitamin D is recovered.

The determination of vitamin D in natural products like fish liver oils is beset with many difficulties. The presence of various interfering materials such as vitamins A_1, A_2 and their congeners, sterols and provitamins D, free fatty acids, phospholipids and wax alcohols make the determination of vitamin D by the spectroscopic method almost impossible. Of these, vitamin A seems to be the chief interfering material as it masks vitamin D in both the ultraviolet absorption and colour tests.

Vitamin A_2, in the liver oils of fresh-water fish predominates over vitamin A_1, from both European[1] and Indian[2] waters. Vitamin A_2 has an absorption maximum at 351 mμ and a subsidiary maximum at 286 mμ with an inflection at 276 mμ. In fresh-water fish liver oils the presence of vitamin A_2 will therefore cause an additional interference in the spectrophotometric determination of vitamin D, which absorbs at 265 mμ. Although fresh-water fish liver oils have a preponderance of vitamin A_2 over vitamin A_1, it has been found that vitamin A_1 is invariably present in most of these oils in varying proportions[3] and, as such, these oils present a problem for the determination of vitamin D after elimination of both vitamin A_1 and A_2. Vitamin A_2 is a highly labile compound. Gillam *et al.*[4] reported that vitamin A_2 decomposed when absorbed on alumina giving rise to a considerable amount of material that exhibited a strong 650-mμ band in the antimony trichloride test. On treatment with ethanolic hydrogen chloride, vitamin A_2 yields ethoxy anhydrovitamin A_2,[5,6] which is more strongly adsorbed than anhydrovitamin A_1 on columns of alumina.

In an earlier paper,[7] we described a method for determining vitamin D in the presence of vitamin A_1 by converting vitamin A_1 to anhydrovitamin A_1 and separating it from vitamin D by chromatography on a column of alumina (weakened by addition of 8 per cent. w/v

of water). In the present paper, a method is described for determining vitamin D in the presence of both vitamins A_1 and A_2, by converting these two vitamins into their anhydro products, and separating them from vitamin D by chromatography. The anhydrovitamins A_1 and A_2 will flow down the column, and all of the vitamin D will be located during development of the chromatograms in a colourless zone between two yellow zones at the top of the column. The colourless zone is extruded and vitamin D is eluted and determined by the Zimmerli - Nield - Russel antimony trichloride reagent.

Method

Apparatus—

The spectrophotometric determinations were performed on a Beckman DK–2 spectrophotometer.

Reagents—

Calciferol—B.P. grade.

Other reagents (alcohol, light petroleum, diethyl ether, anhydrous sodium sulphate, antimony trichloride reagent, alcoholic potassium hydroxide, weakened alumina and ethanolic hydrogen chloride) should be purified or prepared by the procedures described previously.[7]

Extraction of liver oil—

The liver oils were obtained from the livers of four fresh-water fishes, *viz.*, *Wallagu attu*, *Bagarius bagarius*, *Mystus seenghala* and *Silonia silondia*. Mix the livers with acid-washed silver sand and anhydrous sodium sulphate in a glass mortar. Extract the thoroughly ground material repeatedly with light petroleum until the extract gives no blue or green colour with antimony trichloride reagent. Then combine the extracts and dry over anhydrous sodium sulphate. Remove the solvent under reduced pressure and store the oils obtained in amber coloured bottles in a refrigerator. Determine the vitamin A_1 and A_2 by the method described by Cama and Morton.[8] The results for these oils are given in Table I.

Table I

Vitamin A_1 and A_2 content of fresh-water fish liver oils

	Vitamin A_1, i.u. per g	Vitamin A_2, i.u. per g
Bagarius bagarius	4000	143,200
Wallagu attu	19,550	113,000
Mystus seenghala	11,420	41,730
Silonia silondia	2436	213,200

Determination of vitamin D—

Saponification—Take a convenient weight of the oil which contains not less than 300 i.u. of vitamin D and saponify with freshly prepared potassium hydroxide solution under a slow stream of nitrogen for 10 to 15 minutes. Maintain a ratio of 2·5 g of potassium hydroxide to 1 g of oil. Dilute the ethanolic soap solution with an equal volume of water and extract 4 times with suitable volumes of light petroleum. Combine the extracts and wash them with water until the washings are neutral to phenolphthalein. Dry the solution over anhydrous sodium sulphate. Evaporate the solution (a known volume) to dryness and treat the residue with a convenient volume, 10 to 40 ml, of ethanolic hydrogen chloride for 40 minutes under an inert atmosphere. Neutralise the excess of acid with the least amount of solid sodium hydrogen carbonate. Extract the solution with petroleum as described above. Wash the extract free from alkali and dry it over anhydrous sodium sulphate.

Chromatography—Evaporate the light petroleum solution to a small volume (1 to 2 ml) and transfer to a chromatographic column (1 × 9 cm) packed with alumina weakened by the addition of 8 per cent. of water. Develop the chromatogram with light petroleum. Maintain the rate of flow at approximately 2 ml per minute. Anhydrovitamin A_1 will flow out in the first 25 ml of the eluate. Anhydrovitamin A_2 will flow out in the next 40 to 50 ml

of the eluate. At this stage several yellow bands can be observed on the column. Further development of the column with about 100 ml of the solvent elutes all the yellow bands except 3, and a colourless portion between the second and third yellow bands. Described from the top of the column they will be—

(*i*) A thin yellow band. This substance exhibits a broad absorption band at 318 to 330 mμ and a subsidiary band at 271 mμ. The antimony trichloride reaction product exhibits absorption maximum at 650 mμ and at 580 mμ.

(*ii*) A relatively wide yellow band about 3 to 4 mm below the first. This material has values for $\lambda_{max.}$ of 335, 349, 368 and 390 mμ. The antimony trichloride reaction product has an absorption band at 650 mμ.

(*iii*) A colourless portion, 4 to 5 cm long, containing vitamin D.

(*iv*) A wide yellow band about 4 to 5 cm below the second yellow band. This material shows a broad absorption band at 325 mμ, with inflections at 300, 310, 347 and 367 mμ. The antimony trichloride reaction product of this material has an absorption band at 650 mμ.

From a set of preliminary experiments it has been found that all the vitamin D was contained in the colourless portion (*iii*) of the column.

Extraction and determination of vitamin D—

Extrude the column, remove the upper half of the colourless portion between the second and third yellow bands and elute it with two 15-ml portions of diethyl ether. Evaporate the eluate to dryness under nitrogen, and dissolve the residue in a convenient volume of light petroleum (10 ml). Evaporate a 5-ml portion of this to dryness under nitrogen in a small flask. Add 4 ml of antimony trichloride reagent to the residue and gently swirl the flask for 30 seconds and transfer the solution quickly to an optical cell. Measure the extinction at 500 mμ. Calculate the amount of vitamin D present, assuming $E^{1\%}_{1cm}$ value at 500 mμ for pure vitamin D to be 1800.

Results

Table II shows the results of the determination of vitamin D in fresh-water fish liver oils by the above method.

Table II

Vitamin D in fresh-water fish liver oils

Name of fish	Weight of oil, g	$\epsilon_{500m\mu}$	$E^{1\%}_{1cm}$, at 500mμ	Vitamin D present, i.u. per g
Bagarius bagarius ..	2·0670	0·34	0·01317	292·7
	2·1210	0·34	0·01313	291·8
	2·0240	0·32	0·01265	281·1
Wallagu attu	0·3421	0·65	0·1521	3379
	0·3562	0·67	0·1505	3345
	0·3611	0·69	0·1529	3398
Mystus seenghala ..	0·5426	0·58	0·08551	1901
	0·5212	0·55	0·08443	1876
	0·5365	0·57	0·08504	1900
Silonia silondia ..	0·3741	0·68	0·1455	3232
	0·3561	0·64	0·1438	3197
	0·3420	0·61	0·1427	3171

Recovery experiments—

To assess the accuracy of the method described for determining vitamin D in the fish liver oils, recovery experiments were carried out after adding vitamin D to these oils, as recovery experiments are considered the most critical proof that vitamin D is, in fact, being measured. Measured volumes of an ethanolic solution of calciferol of known strength were added to different weights of these oils and the method outlined above was followed to determine vitamin D. The recoveries of total vitamin D are shown in Table III.

TABLE III

RECOVERIES WITH ADDED CALCIFEROL TO THE LIVER OILS

Name of fish	Weight of oil, g	Vitamin D, i.u. per g present	added	total	found	Recovery, per cent.
Bagarius bagarius ..	0·5112	288	982	1270	1201	94·5
	0·5235	288	959	1247	1207	96·8
	0·6702	288	700	988	995	100·7
	0·7104	288	985	1273	1221	95·9
	0·1477	288	2993	3281	2943	89·7
	0·1548	288	2842	3130	2872	91·7
Wallagu attu	0·4121	3374	1535	4909	4680	95·3
	0·3802	3374	1660	5034	4325	86·9
	0·1244	3374	13,460	16,834	15,940	94·7
	0·1652	3374	5501	8875	8878	100·0
Mystus seenghala ..	0·3132	1892	2618	4510	4302	95·4
	0·3201	1892	2624	4516	4228	96·6
	0·4649	1892	1161	3053	2885	94·5
	0·4456	1892	1212	3104	2981	96·0
Silonia silondia ..	0·2124	3200	4341	7541	7001	92·8
	0·2090	3200	4412	7612	7516	96·5
	0·3462	3200	1863	5063	4790	94·6
	0·3451	3200	1869	5069	4837	95·4

The recoveries of total vitamin D were usually over 90 per cent. and show the reliability of this method for the determination of vitamin D in fish liver oils.

DISCUSSION

In all of these experiments in the determination of vitamin D in different fish liver oils, the chromatograms were found to be similar, with the three yellow zones and a wide colourless portion between the second and the third yellow cones.

In all instances, only the eluates from the upper half of the colourless portion showed an absorption band at 500 mμ when treated with antimony trichloride reagent. The intensity of this band remained constant even after 5 minutes, but a weak second absorption band developed at 420 mμ (406 mμ with *bagarius bagarius* liver oil) as shown by the spectra recorded 5 minutes after adding the reagent. With antimony trichloride reagent, a weak band at 650 mμ also appeared, the intensity of which increased with time. Both of these bands made little contribution at 500 mμ at which wavelength the extinctions for vitamin D were measured. The 420 mμ band was of very low intensity and initially absent 30 seconds after adding the antimony trichloride reagent. The 420 mμ band may be due to sterols and, as these have little effect,[9,10] the extinction at 500 mμ gives a measure of the vitamin D present if all extinctions are measured within 30 seconds after adding the antimony trichloride reagent.

In these investigations no attempt has been made to determine the bio-potency of the oils, but from the results of the recovery experiments, and also from the analysis of the fresh-water fish liver oils that gave reproducible results, it may be suggested that this method can be used for the determination of vitamin D in fish liver oils.

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The Gas-chromatographic Analysis of Gases Extracted from Metals by Vacuum Fusion

By M. T. LILBURNE

(*Metallurgy Division, National Physical Laboratory, Teddington, Middlesex*)

The equipment necessary to use a micro-ionisation detector of the Lovelock type with helium carrier gas is described. Although not used at its highest sensitivity, the apparatus can measure quantitatively 1 per cent. v/v of components in a mixture of carbon monoxide, nitrogen, hydrogen and methane of total volume 5×10^{-4} cm³ at S.T.P.

The purpose of this paper is to describe the application of gas chromatography and Lovelock ionisation detectors[1] to the analysis of gas mixtures obtained from vacuum fusion experiments on metals. A typical gas mixture extracted by the vacuum fusion procedure contains carbon monoxide (from oxygen in the metal), nitrogen, hydrogen and, occasionally, methane. With present-day trends towards high purity metals and smaller samples being made available to the analyst, existing methods of gas analysis have been found to be inadequate. These methods remove the reactive gases from the mixture in succession, either chemically, (*e.g.*, by absorption of carbon monoxide in ammoniacal cuprous chloride solution) or physically, (*e.g.*, by diffusion of hydrogen out of the system through a palladium thimble), and the gas remaining is considered to be nitrogen. As a result, the determination of nitrogen is not as precise as the determination of oxygen and hydrogen. Moreover, in order to determine oxygen, a commonly used physical method involves oxidation of the carbon monoxide in the mixture to carbon dioxide by exposure to hopcalite, the resulting carbon dioxide then being frozen out in a cold trap. The hopcalite, however, absorbs small amounts of hydrogen, and, as a result, the oxygen determination can be in error. This error is not readily apparent until the total gas content in the sample falls below 10 p.p.m. w/w. An obvious way of solving this problem is to remove the hydrogen first, but it is not always convenient to do so. The sensitivity of the chemical methods of analysis is such that 3×10^{-1} cm³ at S.T.P. of the mixture is required in order that a 1 per cent. v/v component may be accurately determined; for physical methods 3×10^{-2} cm³ at S.T.P. is required.

Several workers[2,3,4] have used gas chromatography to separate the gases in the mixture and calibrated thermal-conductivity detectors for their determination. As the gases have been separated before reaching the detector, the precision of determination depends only on the response of the detector for the particular gas and the subsequent recording and integrating systems. Because it is no longer obtained by difference the nitrogen figure is more reliable. However, when using a thermal-conductivity detector, a choice has to be made between high sensitivity for carbon monoxide and nitrogen, and low sensitivity for hydrogen, or *vice versa*. The former is obtained when helium is used as carrier gas, and the latter when argon is used. The thermal-conductivity detector acts as a balanced device comparing heat transfer in the pure carrier gas with that in the carrier gas *plus* sample, and hence for high sensitivity, a requirement is for the thermal conductivities of the carrier and sample gases to be quite different. As can be seen from Table I, if the gas mixture contains carbon monoxide, nitrogen and hydrogen, it is impossible to obtain simultaneously a high sensitivity for these three gases. Of the two choices of carrier gas available, helium

TABLE I

THERMAL CONDUCTIVITIES OF SOME GASES

Gas	Thermal conductivity × 10^{-5} cal. per second per cm per °C
Hydrogen	41·3
Helium	34·3
Argon	3·89
Carbon monoxide	5·58
Methane	6·47
Nitrogen	5·81

is the better because approximately 10 times the volume of hydrogen per p.p.m. by weight in the sample is released compared with the other gases, thus diminishing the effect of low sensitivity of the detector for this gas. The minimum amount of gas required is the same as that for the physical methods, *viz.*, 1 to 3 $\times$ 10^{-2} cm^3 at S.T.P. of gas mixture.

In principle, the Lovelock type of micro-ionisation detector, which has an approximately equal sensitivity to all permanent gases, can be used with helium as carrier gas. The sensitivity of this detector can be as much as 1000 times that of the thermal-conductivity type. The linear range of the ionisation detector is three orders of magnitude, and the maximum concentration of permanent gases in the helium carrier gas that can be handled is 1 in 10^3. Thus in order to use this detector over its full range, the carrier gas must have a total impurity content of no more than 1 p.p.m. by volume. Water vapour, probably the most difficult impurity to eliminate from a gas system, alters the mode of operation of these detectors thereby producing a disastrous effect on sensitivity and reproducibility. The purification of the carrier gas and the design of the train to introduce the sample mixture are the critical points in the application of ionisation detectors to permanent gases. A description is now given of the apparatus developed at the National Physical Laboratory.

Experimental

Helium purification—

Berry[5] was the first to apply ionisation detectors successfully to the detection of the permanent gases in helium. His success was due to the use of a complex chemical purification train with chilled molecular-sieve traps (−196° C) and heated titanium and manganese dioxide traps (900° and 350° C, respectively). Initial experiments at the National Physical Laboratory were conducted with a system following that of Berry. Although excellent results at high sensitivity were obtained for limited periods, the performance of the system was erratic, and it had a lifetime of only 2 months, presumably because the traps were not large enough. A helium flow-rate of 100 cm^3 per minute was used.

A helium diffusion cell to the design of McAfee (from a private communication) consisting of a large bundle of capillary silica tubes, housed in a steel tube that was capable of withstanding high pressures and temperatures, was then obtained from Electron Technology Inc., Kearny, U.S.A. When the cell operates at a pressure of 700 p.s.i.g. and a temperature of 350° C, a flow-rate of 100 cm^3 per minute at 10 p.s.i.g. is obtained on the low pressure side. The only helium flow involved is that which diffuses through the silica barrier. This type of cell allows diffusion through the silica of small amounts of hydrogen and neon in addition to helium, but with the helium supplies commonly available, the concentration of these impurities will never reach the level of 1 p.p.m. by volume. This device has been operating successfully for more than 9 months.

Sampling valve and apparatus tubing—

Stainless-steel connecting tubing was used throughout, and joints were made with neoprene O-rings that were later replaced by silicone O-rings. The carrier gas exhibited evidence of being impure when both of these types of O-ring were used, probably owing to gas diffusion through the elastomer itself. Finally, all joints were made with compression fittings of the "Swagelok" type with indium ferrules. The ferrules were made from a strip of indium that was wrapped around the tube and placed into the fitting immediately before screwing down the backing nut. Joints made in this manner were found to be more reliable than those made with the stainless-steel ferrule provided. It was found necessary to outgas the tubing progressively (from the diffusion cell to the detector) after the apparatus had been assembled and with the full helium flow passing through, taking care not to overheat any fitting containing indium.

The sampling valve was designed so that the primary gas-tight seal was between stainless-steel and PTFE plates. Any O-rings present were of secondary importance.

This valve consisted of two circular plates, one of stainless steel and one of PTFE, whose mating surfaces were polished flat and clamped together via a thrust bearing so that the PTFE plate was free to rotate. The polished surfaces provided an excellent gas-tight joint after a very small application of vacuum grease. The stainless-steel plate contained 4 ports: two, close together, to allow the entry and exit of carrier gas; the third, to allow

the sample gas to enter; and the fourth, to enable the valve to be evacuated. The vacuum port is drawn in Fig. 1 (*a*) and it shows the internal pumping lines necessary to evacuate the annular rings at the centre and edge of the flat plates. Thus when a collar containing suitably placed O-rings is fixed round the plates, the mating surfaces of the plates can be isolated from the atmosphere. The PTFE plate has three equally-spaced slots each of 2×10^{-2} cm^3 volume. The slots are long enough to connect the gas inlet and exit ports, Fig. 1 (*b*). In operation, slot 1 is connected to the sample gas contained in a small reservoir at a pressure of about 20 torr; slot 2, to the carrier-gas ports; and slot 3, to a vacuum line. On being rotated in the correct direction through 120°, a volume of 5×10^{-4} cm^3 at S.T.P. of sample gas is injected into the carrier stream; an evacuated slot is carried to the reservoir; and the third slot is evacuated.

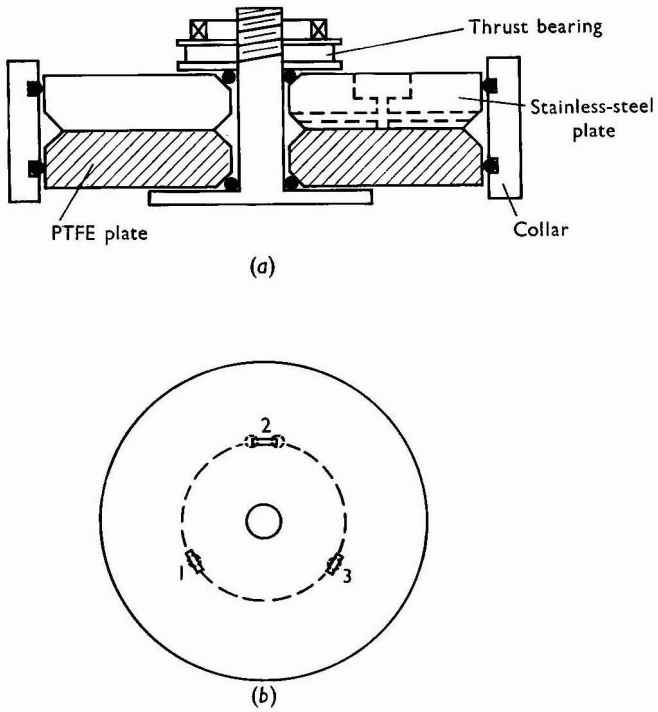

Fig. 1. The sampling valve: (*a*), the vacuum port; (*b*), the PTFE plate with the positions of the ports in the stainless-steel plate shown

In a vacuum-fusion experiment the extracted gases are trapped at low pressure in the fore-line of a high-speed mercury diffusion pump, and then removed to calibrated volumes by a Toepler pump. After measuring the volume of the gas, it is collected over mercury at atmospheric pressure in a small transfer vessel fitted with a greased vacuum tap. This enables separate analyses by chemical and gas-chromatographic methods to be undertaken. 5×10^{-2} cm^3 at S.T.P. of this gas mixture is transferred to the small sample reservoir of 1·5 cm^3 volume attached to the chromatograph through a modified three-way glass stopcock. The pressure in the sample reservoir is then 20 to 25 torr. The usual "T" bore of the three-way stopcock was replaced by a straight-through bore of 2 mm diameter and 5×10^{-2} cm^3 volume so that the gas could be trapped across the tap between mercury columns. Thus, when the tap is rotated through 90°, the trapped gas is allowed to enter the reservoir.

CHROMATOGRAPH—

Analytical column—A column 120 cm long and 0·5 cm in diameter, coiled and used at 120° C.

Stationary phase—Molecular-sieve (Linde 5A) powder of 36 to 60 mesh. After crushing, the "fines" were floated off by stirring the powder supported in a tall beaker vigorously with a water jet. The sieve was activated by heating at 400° C overnight in a stream of carrier gas.

Detector and power supply—Type I.E.103B, available from Gas Chromatography Ltd., Maidenhead, England.

Integrator—Type 92314 available from Electro Methods Ltd., Stevenage, England. This was driven by a voltage delivered from a transmitting slide-wire on the recorder. It was found necessary to integrate the area under the peaks because the carbon monoxide peak showed some "tailing." All other peaks were gaussian in shape.

Results

The apparatus was calibrated by injecting, in turn, known amounts of the pure gases required, and noting the response. The sensitivity was found to be highly dependent on the voltage applied to the detector and was more than 60 coulombs per mole at 500 volts. In the present application it was only necessary to operate the detector at 220 volts when the sensitivity was about 1 coulomb per mole (Fig. 2). So long as the peak height was greater

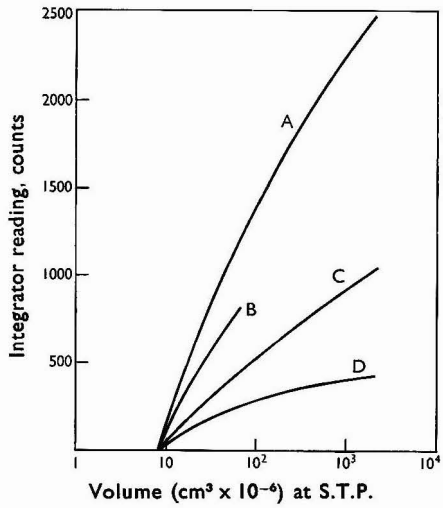

Fig. 2. Calibration curves (1 count = 3·6 × 10^{-11} coulombs); A, carbon monoxide; B, methane; C, nitrogen; D, hydrogen

than 50 per cent. of the scale reading, the integrated area of the peaks obtained was reproducible to within 4 per cent. Frequent calibration checks over a period of several months established that there was no variation in sensitivity. Table II gives the results of several analyses, chosen at random from a large number, compared with the analyses obtained by chemical means. The time taken for analysis by gas chromatography was 2 minutes and compared favourably with the 20 minutes taken by the chemical method.

Table II

Comparison of gas-chromatographic and chemical methods

Sample	Gas chromatography, volume used ≏ 5 × 10^{-4} cm³ at S.T.P., p.p.m. by weight in sample			Chemical method, volume used ≏ 2 × 10^{-1} cm³ at S.T.P., p.p.m. by weight in sample		
	Oxygen	Hydrogen	Nitrogen	Oxygen	Hydrogen	Nitrogen
Nimonic	194	3·8	17·7	185	4·9	18·5
Chromium	18·0	1·0	6·2	18·6	0·8	7·3
Pure iron	16·1	0·2	7·6	15·9	0·2	7·6
Chromium flake ..	56·0	1·3	14·7	57·3	1·3	13·3
Chromium flake ..	16·2	0·8	18·4	15·2	0·8	19·5
Iron-manganese ..	7·2	0·4	3·9	6·5	0·5	4·0

Discussion and conclusions

The speed and accuracy of gas analysis by gas chromatography have been demonstrated particularly for samples having low gas contents. The result obtained by this method of analysis is superior to those obtained by chemical and other physical methods, in that a direct determination of nitrogen is made, and it is therefore of higher accuracy. The sensitivity of the method is not as great as that obtainable by using mass-spectrometric methods but can, if necessary, be increased by a factor of more than 10. On the other hand, these latter methods have to use an indirect approach as both carbon monoxide and nitrogen have an atomic mass of 28.

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The Determination of Boron in Mild Steel

By T. S. HARRISON and W. D. COBB

(*Appleby-Frodingham Steel Company, Central Laboratory, Scunthorpe, Lincolnshire*)

A method of controlling the reaction between acetic anhydride and water to permit the development of the boron - curcumin complex without the previous separation of iron is described. The effect of other elements in amounts normally found in mild steel has been examined. The method has been applied to a series of British Chemical Standards and also to commercial samples.

The purpose of this work was to try and adopt Hayes and Metcalfe's[1] curcumin colour procedure so as to be able to make a direct determination of boron in the presence of iron, thus giving a method for the determination of boron in mild steel that would be suitable for routine batch work.

To test the colour procedure in the presence of iron, additions of 10 N sodium hydroxide were made to 5-ml aliquots of sulphuric acid (20 per cent. v/v) containing iron and boron so as to nearly neutralise all of the acid. These solutions were evaporated to dryness on a steam-bath and the Hayes and Metcalfe colour procedure was then applied. Not all the iron salts went into solution when the curcumin - acetic acid reagent was added, and the final colours produced had approximately one third of the intensities of those found with the Hayes and Metcalfe procedure. These tests were useful in that they suggested that iron did not interfere positively with the curcumin - boron reaction.

Whereas Hayes and Metcalfe[1] found that their colour procedure could tolerate the introduction of up to 0·25 ml of water, their direct procedure based on a 0·25-ml aliquot was not sufficiently sensitive or practical for our purpose. In order, therefore, to increase the sensitivity without resorting to the preliminary separation of boron by distillation, a means had to be found of permitting an adequate sample fraction to be taken and at the same time restricting the amount of water present in the colour procedure.

Acetic anhydride has been used to eliminate small volumes of water, and Crawley[2] used hydrochloric acid to catalyse the reaction between them. As it was anticipated that any volume taken would contain sulphuric acid (20 per cent. v/v), it was thought that the addition of hydrochloric acid might not be required. Tests proved this to be correct. It was also confirmed that an explosive reaction could occur, and that an addition of glacial acetic acid gave a more controlled reaction. Before the addition of acetic anhydride, it was found advisable to keep the temperature of the solutions below 20° C, because at higher temperatures the reaction was more violent.

Tests showed that an excess of acetic anhydride resulted in greatly reduced optical densities due to boron (*e.g.*, when applying colour procedure A with 6 ml of acetic anhydride), and so the amounts of acetic anhydride used in the experimental colour procedures A and B were calculated to allow for less than 0·1 ml of water to remain in solution.

It was found that by substituting a 50 per cent. acetone - water solvent for the ethanol used in Hayes and Metcalfe's procedure to extract the curcumin - boron complex, a filtration stage was avoided.

Colour procedures

Colour procedure A—

To a 1-ml aliquot in a polythene bottle add 2 ml of glacial acetic acid at room temperature. Add 4 ml of acetic anhydride. Mix the contents, stopper the bottle and immerse the lower half of it in water until the reaction has taken place (about 30 seconds). Cool to room temperature. Add 3 ml of the curcumin - acetic acid reagent, mix the liquids and add 4 ml of sulphuric acid - acetic acid reagent. Mix them again, and allow them to stand for the specified period of time. Extract the solutions with a 50 per cent. acetone - water solvent. Transfer the solutions to 100-ml calibrated flasks and measure the optical density as indicated in Table I.

COLOUR PROCEDURE B—

To a 2-ml aliquot in a polythene bottle add 10 ml of glacial acetic acid and cool to below 20° C. Add 8·5 ml of acetic anhydride and, after the reaction has taken place (almost immediately), stopper the bottle and cool it to room temperature. Continue as in colour procedure A from "Add 3 ml of curcumin - acetic acid reagent."

TABLE I

TIME REQUIRED FOR THE FORMATION OF THE BORON - CURCUMIN COMPLEX WITH VARYING BASE SOLUTIONS, WITH AND WITHOUT IRON

Standing time allowed for the formation of the boron - curcumin complex, minutes	Optical density corrected for the blank. Obtained by taking the indicated aliquot from the following solutions through the colour procedure A or B							
	No iron. 150 μg of boron + H_2SO_4, diluted to 100 ml	2 g of iron, 150 μg of boron + H_2SO_4, diluted to 100 ml	2 g of iron, 75 μg of boron + H_2SO_4, diluted to 50 ml	No iron. 75 μg of boron + 2ml HCl (conc.) + H_2SO_4, diluted to 50 ml	2 g of iron, 75 μg of boron + 2ml HCl (conc.) + H_2SO_4, diluted to 50 ml	No iron. 75 μg of boron + H_2SO_4, diluted to 100 ml	1 g of iron, 75 μg of boron + H_2SO_4, diluted to 100 ml	2 g of iron, 75 μg of boron + H_2SO_4, diluted to 100 ml
	1-ml aliquot through colour procedure A				2-ml aliquot through colour procedure B			
5	0·520	0·411	—	—	—	—	—	—
15	0·730	0·676	—	0·715	0·771†	0·579	0·548	0·672
30	0·807	0·778	—	0·799	0·834†	0·738	0·710	0·714
	0·791	—	0·817†	—	—	—	—	—
60	0·810	0·809	—	0·815	0·839	0·798	0·806	0·813
	0·804	—	0·825*	—	—	0·781	0·785	0·804
	0·810	—	0·826*	—	—	0·799	0·808	0·809
	0·819	—	0·829†	—	—	0·793	0·811	0·819
120	0·803	0·796	—	—	—	0·793	0·816	0·816
	—	—	—	—	—	0·802	0·804	0·809
	—	—	—	—	—	0·799	0·806	0·806
	—	—	—	—	—	0·804	0·809	0·819
180	0·780	—	0·790	—	—	0·786	0·804	0·810
	0·797	0·784	—	—	—	—	—	—

* Solutions filtered after extraction with 50 per cent. acetone - water solvent.
† Salts were slow to dissolve.

A series of tests was carried out (Table I) to find the time required for the formation of the boron complex consistent with a suitable form of base solution. As a result of these tests, colour procedure B was adopted for the final method given under Experimental.

The graph was found to be linear up to 1·6 μg per 100 ml for a 4-cm cell. This is equivalent to a range of up to 0·0040 per cent. of boron.

COLOUR STABILITY—

The final coloured solutions were found to be stable for 3 hours. Overnight standing (18 hours) showed a loss of up to 4 per cent. in optical density owing to boron.

EXPERIMENTAL

PRINCIPLE—

After the elimination of water from the sample solution, the boron - curcumin complex is developed in the presence of the iron, and is measured photometrically.

REAGENTS—

Acetone - water solvent—Mix equal volumes of distilled water and acetone, AnalaR.

Curcumin - acetic acid reagent—Dissolve 0·5 g of curcumin in 400 ml of glacial acetic acid, AnalaR, with warming and stirring.

Sulphuric acid - acetic acid reagent—Cautiously add, with cooling and stirring, 200 ml of sulphuric acid (sp.gr. 1·84) that has been tested for low boron content, to 200 ml of glacial acetic acid, AnalaR.

Dilute sulphuric acid (20 per cent. v/v).

Glacial acetic acid, AnalaR.

Acetic anhydride, AnalaR.
Sodium carbonate, anhydrous, AnalaR.
Hydrogen peroxide, 5 volume, AnalaR.
Iron(II) solution—Dissolve 10 g of boron-free iron in 200 ml of sulphuric acid (20 per cent. v/v). Cool, dilute to 250 ml with sulphuric acid (20 per cent. v/v) and filter.

(5 ml of solution = 0·2 g of iron.)

NOTE—All of the reagents are stable and stock solutions may be kept.

APPARATUS—

Boron-free glassware.
Polythene bottles—100-ml capacity fitted with polythene stoppers.
Photo-electric absorptiometer—The Spekker is a suitable instrument used in conjunction with a mercury lamp, Ilford No. 605 colour filters and Calorex H.503 heat filters.

PREPARATION OF THE SAMPLE SOLUTION—

Transfer 2 g (Note 1) of the sample to a 100-ml conical flask and add 40 ml of sulphuric acid (20 per cent. v/v). Insert an air condenser (Note 2) and digest on a water-bath at 80° C until the reaction ceases. Add 1 ml of hydrogen peroxide. Cool, rinse the condenser with sulphuric acid and filter through a small paper-pulp pad that has been previously washed with sulphuric acid of the same strength. Collect the filtrate in a 100-ml calibrated flask and wash the pad with a little sulphuric acid. Transfer the pad to a platinum crucible, sprinkle with 0·2 g of sodium carbonate, dry and ignite at a low temperature to remove carbon. Add 1 g of sodium carbonate and fuse at a temperature of 1100° C. Cool, and cautiously add sulphuric acid until the fused mass has dissolved, keeping the crucible covered with a polythene cover. Filter the solution through a small paper-pulp pad that has been previously washed with sulphuric acid into the same calibrated flask and wash with sulphuric acid. Dilute to the mark with sulphuric acid and mix (Note 3).

ADOPTED COLOUR PROCEDURE—

Transfer a 2-ml aliquot of the sample solution to a dry polythene bottle, add 10 ml (Note 4) of glacial acetic acid and cool to below 20° C. Hold the base of the bottle under cold water and add 8·5 ml of acetic anhydride. Stopper the bottle and allow the reaction to take place (this occurs almost immediately) and then cool to room temperature. Add 3 ml of curcumin - acetic acid reagent, mix and add 4 ml of sulphuric acid - acetic acid solution. Mix the contents and leave them for 1 hour (Note 5). Extract the solution with the 50 per cent. acetone - water solvent and transfer to a 100-ml calibrated flask, making up to the mark with 50 per cent. acetone - water.

Measure the absorption of the clear solution with an absorptiometer, with Ilford No. 605 colour filters and H.503 heat filters in the appropriate cell. Deduct the value for the blank obtained by taking boron-free iron through the procedure, and obtain the boron content by reference to the calibration graph.

CALIBRATION—

Standard boron solution (A)—Dissolve 0·2857 g of boric acid in water and dilute to 500 ml with water.

(1 ml of solution = 100 μg of boron.)

Standard boron solution (B)—Dilute 50 ml of solution (*A*) to 250 ml with sulphuric acid (25 per cent. v/v), giving a final acidity of 20 per cent. v/v.

(1 ml of solution = 20 μg of boron.)

To solutions of 2 g of boron-free iron in 20 per cent. v/v sulphuric acid contained in 100-ml calibrated flasks add measured amounts of boron solution (*B*) as indicated by Table II.

Dilute to the graduation mark with sulphuric acid and mix.

Transfer a 2-ml aliquot to a dry polythene bottle and continue according to the adopted colour procedure.

Prepare calibration graphs by plotting the optical density due to boron against the boron concentration for each cell size.

TABLE II

ADDITIONS OF BORON SOLUTION (*B*)

Boron solution (*B*), ml	Boron equivalent, μg	Boron equivalent, per cent.	Cell size, cm
0·5	10	0·0005	4
1·0	20	0·001	4
2·0	40	0·002	4, 2
3·0	60	0·003	4, 2
4·0	80	0·004	4, 2, 1
6·0	120	0·006	2, 1
8·0	160	0·008	2, 1
12·0	240	0·012	1
16·0	320	0·016	1
Nil	Nil	Nil	4, 2, 1

NOTES—

1. The range of the method may be increased by reducing the sample weight.
2. A suitable form of condenser is a 30-inch length of glass tubing, internal diameter 3 mm, carrying a one-hole rubber bung to fit the 100-ml conical flask.
3. If it is desired to determine the insoluble portion separately, treat the extract from the fusion as a separate determination and add 5 ml of iron(II) solution to both blank and samples before making up to 100 ml with sulphuric acid.
4. All additions for the colour procedure must be accurately measured by burette.
5. The standing time may be increased to 2 hours to suit individual requirements.

DISCUSSION

THE USE OF HYDROGEN PEROXIDE TO AID SOLUTION OF INSOLUBLE CARBIDES—

It was found that the addition of hydrogen peroxide to a solution of steel in sulphuric acid (20 per cent.) resulted in an increased blank value.

The volume of hydrogen peroxide added, however, had to be kept to a minimum so that the final acidity remained close to 20 per cent. v/v.

Tests were carried out on 2 g of steel that contained 0·5 per cent. of molybdenum, to ascertain the minimum amount of hydrogen peroxide required to effect a solution of the insoluble carbides. It was found that 1 ml of a 5-volume solution was sufficient, giving an increase in the blank, when measured in a 4-cm cell, of less than 0·02 optical density.

THE SEPARATE DETERMINATION OF INSOLUBLE BORON—

When insoluble portions containing vanadium were treated separately, a brown colouration resulted. An addition of iron(II) was found to overcome this interference.

CALIBRATIONS—

A series of calibrations was made, and the results are detailed in Table III.

TABLE III

CALIBRATIONS

		Optical density *minus* the blank								
		4-cm cell					2-cm cell		1-cm cell	
Boron, μg	Boron equivalent, per cent.	A_1	A_2	B	C_1	C_2	C_1	C_2	C_1	C_2
10	0·0005	0·115	0·103	0·109	0·111	0·113	—	—	—	—
20	0·001	0·226	0·212	0·219	0·219	0·223	—	—	—	—
40	0·002	0·438	0·415	0·434	0·421	0·438	0·214	0·222	—	—
60	0·003	0·648	0·616	0·657	0·628	0·654	0·316	0·332	—	—
70	0·0035	0·761	0·758	0·751	0·754	0·762	0·376	0·382	—	—
80	0·004	0·877	0·878	0·848	0·879	0·883	0·443	0·444	0·226	0·228
120	0·006	—	—	—	—	—	0·642	0·635	0·328	0·324
160	0·008	—	—	—	—	—	0·863	0·865	0·434	0·435
240	0·012	—	—	—	—	—	—	—	0·628	0·632
320	0·016	—	—	—	—	—	—	—	0·850	0·861

Calibrations A_1 and A_2 were obtained by taking 2-g portions of boron-free iron together with the boron fractions, dissolving and treating according to the full procedure given under Experimental.

Calibration B was carried out by taking 0·2-g portions of boron-free iron, in solution, together with the boron fractions and adding 1 g of sodium carbonate, making the solution up to 100 ml with sulphuric acid and taking 2-ml aliquots through the adopted colour procedure given under Experimental.

Calibration C_1 was carried out according to the calibration procedure given in the same section. Calibration C_2 resulted from taking further 2-ml aliquots from the C_1 solutions but allowing 2 hours' standing time for the formation of the boron - curcumin complex.

The results show that the simplified calibration procedure finally adopted would serve for both soluble and insoluble determinations.

A comparison of C_1 with C_2 shows that the standing time allowed for the formation of the boron - curcumin complex may be varied between 1 and 2 hours to suit individual requirements.

The effect of other elements—

Synthetic tests were carried out to study the effect of other elements on the determination of total boron by the method.

The element under investigation, together with 2 g of boron-free iron and the equivalent of 0·0020 per cent. of boron, was taken through the full procedure.*

Further synthetic tests were carried out on the basis of a separate insoluble determination. The element under investigation was fused with 1 g of sodium carbonate, extracted with sulphuric acid and added to 0·2 g of boron-free iron and the equivalent of 0·0020 per cent. of boron.† The method is free from interference from elements at the levels likely to be encountered in mild steels.

Samples—

Total boron was determined on a series of mild steel B.C.S. standards, by using the method described on two separate occasions. The results shown in Table IV are compared with the certificate value.

Table IV

Results obtained with B.C.S. mild steel standards, compared with certificate values

B.C.S. No.	Total boron, per cent.	
	Certificate value	Direct curcumin
273	0·002	0·0020
		0·0020
275	0·001	0·0010
		0·0009
277	< 0·001	0·0001
		0·0001
326	0·001	0·0010
		0·0010
327	0·003	0·0020
		0·0021
328	0·004	0·0037
		0·0037
329	0·008	0·0078
		0·0076
330	0·007	0·0064
		0·0064

* The results showed no interference from 2 per cent. of manganese, 1 per cent. of molybdenum, chromium, copper and silicon, or 0·5 per cent. of niobium, titanium, vanadium, nickel, tungsten, zirconium, tin, phosphorus, cobalt, zinc, magnesium, lead and tantalum, or 0·1 per cent. of beryllium, arsenic and antimony. All the recoveries were between 0·0018 and 0·0020 per cent. of boron.

† The results showed no interference from 1 per cent. of silicon, molybdenum and chromium, or from 0·5 per cent. of vanadium, titanium and tungsten. All the recoveries were between 0·0018 and 0·0020 per cent. of boron.

Soluble and insoluble boron determinations were carried out on a series of steel plate and pit samples (Table V) from Fortiweld quality steels.

The results for the distillation procedure were obtained on aliquots taken from the same sample solutions obtained for the direct procedure.

The quinalizarin values were those obtained on a routine basis.

TABLE V

RESULTS FOR A SERIES OF PLATE AND PIT SAMPLES OF FORTIWELD QUALITY STEELS

Sample No.	Boron, per cent.	Distillation curcumin		Direct curcumin		Routine quinalizarin
1	Soluble	0·0012	0·0011	0·0011	0·0011	0·0008
	Insoluble	0·0023	0·0023	0·0023	0·0023	0·0025
	Total	0·0035	0·0034	0·0034	0·0034	0·0033
2	Soluble	0·0032	0·0029	0·0030	0·0028	0·0024
	Insoluble	0·0010	0·0010	0·0009	0·0011	0·0008
	Total	0·0042	0·0039	0·0039	0·0039	0·0032
3	Soluble	0·0019	0·0017	0·0018	0·0016	0·0015
	Insoluble	0·0019	0·0019	0·0017	0·0019	0·0013
	Total	0·0038	0·0036	0·0035	0·0035	0·0028
4	Soluble	0·0022	0·0021	0·0021	0·0020	0·0018
	Insoluble	0·0018	0·0018	0·0016	0·0017	0·0017
	Total	0·0040	0·0039	0·0037	0·0037	0·0035
5	Soluble	0·0014	0·0013	0·0014	0·0012	0·0008
	Insoluble	0·0027	0·0026	0·0022	0·0024	0·0020
	Total	0·0041	0·0039	0·0036	0·0036	0·0028
6	Soluble	0·0025	0·0025	0·0024	0·0024	0·0017
	Insoluble	0·0010	0·0013	0·0010	0·0013	0·0011
	Total	0·0035	0·0038	0·0034	0·0037	0·0028
7	Soluble	0·0028	0·0028	0·0027	0·0026	0·0018
	Insoluble	0·0009	0·0011	0·0009	0·0011	0·0008
	Total	0·0037	0·0039	0·0036	0·0037	0·0026
8	Soluble	0·0021	0·0021	0·0020	0·0019	0·0014
	Insoluble	0·0015	0·0018	0·0014	0·0017	0·0014
	Total	0·0036	0·0039	0·0034	0·0036	0·0028
9	Soluble	0·0028	0·0029	0·0028	0·0027	0·0022
	Insoluble	0·0003	0·0005	0·0004	0·0005	0·0003
	Total	0·0031	0·0034	0·0032	0·0032	0·0025

CONCLUSIONS

A method for the determination of boron in mild steel has been described. The elements in amounts normally found in mild steel have been shown not to interfere. The method is suitable for routine batch work.

We thank Messrs. O. S. Bell and W. W. Foster for their co-operation, and the Directors of The Appleby-Frodingham Steel Company for permission to publish this paper.

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Distillation Method for Determining Total Carbon in Sodium

By V. M. SINCLAIR, J. L. DRUMMOND and A. W. SMITH

(Dounreay Experimental Reactor Establishment, United Kingdom Atomic Energy Authority, Thurso, Caithness, Scotland)

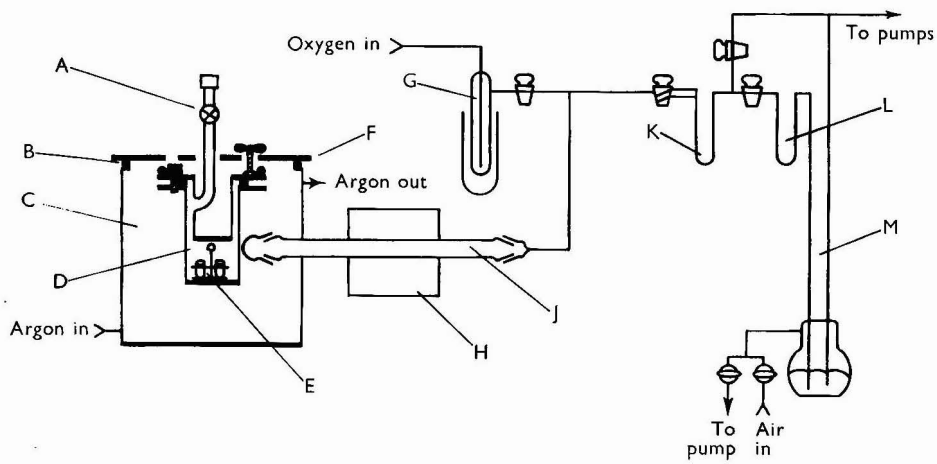
A procedure is described for measuring total carbon in sodium by the removal of the alkali metal by distillation, combustion of the residue in oxygen and manometric determination of the resultant carbon dioxide. It has been shown by radio-tracer techniques that there is no loss of carbon from the sample during the distillation stage, and that recovery of various forms of carbon is essentially complete. The coefficient of variation of a single determination at the 20 p.p.m. level is about 10 per cent. The bias is believed to be less than 10 per cent.; the average blank value is about 1·5 p.p.m. of carbon.

A distillation method by Walker and France[1] for the determination of "free" carbon in sodium has been published. A related procedure has been developed concurrently at Dounreay for determining total carbon in sodium and sodium - potassium alloys. The main differences from Walker and France's method are considered in this paper.

Experimental

Apparatus—

Silica crucibles (1·8 *cm i.d.* × 4 *cm high*)—The silica tubing must be washed with water before fabrication of the crucibles. It must not be touched with bare hands before glass-blowing, or the surface of the silica will be adversely affected. Immediately before use the crucibles should be re-washed with water, dried at 120° C, and ignited at 800° C in oxygen for 10 minutes, or until the carbon dioxide blank is negligible.



A = Valve
B = O-rings
C = Glove-box
D = Distillation pot
E = Sample crucibles and holder
F = Perspex lid
G = Molecular-sieve trap
H = Sliding furnace
J = Silica combustion tube
K = First trap
L = Second trap
M = Manometer

Fig. 1. Transfer glove-box and combustion apparatus

Distillation pot—The stainless-steel distillation pot is similar to that described by Walker and France,[1] but with 3 of the 6 bolts retaining the top replaced by fixed studs for attaching the pot to the roof of the transfer glove-box (see Fig. 1). The pot, cold-finger and a nickel support to hold 4 crucibles must be washed thoroughly with dilute nitric acid and dried at 120° C before use.

Sampling and distillation rigs—The sampling and distillation rigs are similar to those described by Walker and France.[1] The cold-finger of the distillation pot is cooled with water for sodium or with a solid carbon dioxide - methanol mixture for sodium - potassium alloy samples. It has been shown by direct measurement that the sodium temperature during a distillation was about 350° C with the furnace at a temperature between 600° and 650° C. The crucible temperature rose to a maximum of about 400° C after all the sodium had been distilled.

Combustion rig—One end of the combustion tube protrudes into the inert-gas filled transfer box shown in Fig. 1, which is used for unloading the crucibles from the distillation pot under carbon dioxide free conditions. The combustion furnace operates at 800° to 900° C, and the carbon dioxide produced is separated and determined in a conventional manometric apparatus.

Procedure—

Prepare the samples and carry out the distillation as described by Walker and France.[1] After cooling the pot at the end of the distillation, connect the top of the pot to the underside of the Perspex lid of the combustion rig glove-box, replace the lid on the box, and purge the box with argon. Disconnect the lower half of the distillation pot inside the box, and remove the crucibles to a storage vessel in the box.

Carry out a blank combustion on the empty apparatus to ensure that the apparatus blank is less than 2 μg of carbon. Load a sample crucible from the glove-box into the cold end of the combustion tube and evacuate the apparatus to less than 10^{-3} torr. Isolate the combustion tube from the vacuum pumps, admit oxygen to a pressure of about 70 cm of mercury, and move the pre-heated furnace into position surrounding the crucible. After the apparatus has been at 800° C for 10 minutes, slowly evacuate the apparatus through an efficient trap cooled in liquid nitrogen to less than 10^{-3} torr, and continue the combustion under vacuum for a further 10 minutes to complete the decomposition of any sodium carbonate, either from the original sample or that formed during the combustion. Isolate the cold-trap, and substitute a solid carbon dioxide - methanol trap for the liquid nitrogen. At the same time, cool a second small trap in the gas-measuring system with liquid nitrogen to transfer the carbon dioxide but not moisture. Warm to room temperature, and measure the carbon dioxide pressure in a convenient volume.

Repeat the combustion with the other sample and blank crucibles. Calculate the weight of carbon in each, deduct the blank value, and relate to the weight of sample obtained from the known volume of the sample crucibles.

Results and discussion

Consideration has been given to the possible sources of error by which carbon might be lost or gained.

Distillation stage—

In the distillation stage the temperature of the liquid metal is raised to between 350° and 400° C, and the sodium is distilled on to the cold-finger under a pressure of less than 10^{-5} torr. During this distillation carbon could distil as an unknown, volatile, sodium - carbon species, or be lost as gases such as carbon monoxide or carbon dioxide as a result of reactions such as—

$$Na_2O + C \rightleftharpoons 2Na + CO \quad .. \quad .. \quad .. \quad .. \quad (i)$$

$$Na_2CO_3 + 2Na \rightleftharpoons 2Na_2O + CO \quad .. \quad .. \quad .. \quad .. \quad (ii)$$

$$Na_2CO_3 \rightleftharpoons Na_2O + CO_2 \quad .. \quad .. \quad .. \quad .. \quad (iii)$$

$$(NaCO)_x \text{ species} \rightleftharpoons xNa + xCO \quad .. \quad .. \quad .. \quad .. \quad (iv)$$

The gas could either escape or be gettered by the distilling liquid metal. These reactions were studied under the time and temperature conditions prevailing during the distillation stage.

(*i*) *Carbon and sodium oxide reaction*—From thermodynamic considerations, the equilibrium partial pressure of carbon monoxide at 400° C for the reaction between sodium oxide and free carbon should be very low. However, towards the end of a sodium distillation, any carbon monoxide or sodium produced by this reaction could be removed from the system by pumping or by transfer to the cold-finger; the reaction could proceed irreversibly. Possible losses of carbon monoxide were therefore examined both in a static system with evacuated sealed tubes and under distillation conditions.

In the static system, samples of sodium oxide were mixed with carbon-14 labelled elemental carbon, both with and without free metallic sodium present. After heating to about 400° C for several hours, the carbon-14 activity of the gas was measured. The results in Table I confirm that the amount of carbon monoxide found was insignificant.

TABLE I

REACTIONS OF SODIUM OXIDE AND CARBON

Excess sodium present	Carbon added, μg	Oxygen added, μg	Carbon in gas, μg
Yes	600	—	<0·5
No	600	290	<0·5
Yes	600	1450	<0·5
Yes	600	2000	<0·5
No	600	3500	<0·5
Yes	600	4000	<0·5

In similar experiments under distillation conditions in a glass and silica rig, any gas evolved was collected continuously and counted for carbon-14, and also the distilled sodium from the cold-finger was dissolved and analysed for carbon-14. The amounts of carbon-14 found were always less than 0·1 per cent. and less than 3 per cent. of that added, respectively.

(*ii*) *Sodium carbonate and sodium reaction*—The change in free energy calculated for the reaction between sodium carbonate and sodium is +32 K cal., and therefore the equilibrium partial pressure of carbon monoxide should be negligible. To confirm by experiment that no significant amount of carbon monoxide was evolved even during continuous pumping, sodium metal was added to known amounts of sodium carbonate labelled with carbon-14 and distilled off. The amounts of carbon-14 in the gases evolved during distillation of the sodium and in the sodium distillate, shown in Table II, suggest that there is no appreciable loss of carbon from a distillation residue arising from a reaction of sodium and sodium carbonate.

TABLE II

SODIUM - SODIUM CARBONATE REACTION

Sodium added, g	Sodium carbonate added, μg	Carbon in gas, per cent.	Carbon in distillate, per cent.
2	800	<1	3
1	900	1	2
1	300	<1	3
1	26	<1	3
1	160	<1	1
1	73	<1	<1

(*iii*) *Decomposition of sodium carbonate*—Results given by Preston and Turner[2] for the thermal decomposition of sodium carbonate indicate that the amount of carbon dioxide lost from sodium carbonate on pumping at 400° C is small. The loss of carbon by this route is therefore negligible; this is again confirmed by the results in Table II.

(*iv*) *Decomposition of sodium carbonyl*—The nature of sodium carbonyl species in liquid sodium is uncertain. However, specimens of hexasodium hexacarbonyl were prepared as described by Miller,[3] and the dissociation pressure of the carbonyl was measured over the range 600° to 800° C. Extrapolation to distillation temperatures, *viz.*, 350° C, suggested that the pressure of carbon monoxide would be low and that loss would be negligible. To check this hypothesis some carbon-14 labelled sodium carbonyl was prepared in a large excess of sodium to give a carbon content of about 10 p.p.m. A sample of this sodium was distilled, and it was found that about 97 per cent. of the carbon-14 was present in the residue, with only 3 per cent. in the distillate and less than 1 per cent. in the evolved gas.

(*v*) *Loss of carbon transferred from steel*—Although the most probable forms of carbon in sodium have been covered, it is possible that unidentified forms, present in the liquid metal, could be lost during the distillation procedure.

To guard against this possibility, 2 specimens of iron containing iron carbide labelled with carbon-14 were prepared by dissolving carbon-14 in molten iron and cooling. These were suspended in low temperature filtered sodium contained in 2 stainless-steel cans (type 18.8.1) and heated at 600° C for 24 hours. Analysis of samples of the can showed that significant amounts of carbon-14 had transferred from the carburised specimens to the stainless-steel container. Samples of the sodium were distilled, and the gas produced during the distillation, the distilled sodium and the residue were all examined for carbon-14. The results in Table III show that a significant amount of carbon-14 was found only in the residue.

TABLE III

LOSS OF CARBON TRANSFERRED FROM STEEL

Carbon-14 in residue, per cent.	Carbon-14 in distillate, per cent.	Carbon-14 in gas, per cent.
99	<1	<1
99	2	<1

MEASUREMENT OF BLANKS—

A positive error could be caused in results by the distillation procedure if carbon was picked up by samples at any stage of the procedure. The "blank" for the method was therefore measured by regularly placing empty crucibles alongside sample crucibles throughout the whole procedure, and measuring the carbon in these. The reproducibility of this blank effectively sets the lower limit for determination.

Initial results were discouraging in that very high carbon blanks were occasionally obtained, corresponding to about 10 to 20 p.p.m. of carbon in the sample. Some effort was therefore devoted to tracing the source of the carbon and eliminating the trouble.

It was found that up to 0·5 ml of carbon monoxide *plus* carbon dioxide was evolved from the steel distillation pot during a distillation, and that these gases were largely "gettered" by the sodium distillate. Attempts to eliminate these gases only showed that they were probably not the main cause of the high blanks. The preparation of the sample crucibles from silica tubing was more critical, because the surface of the silica appeared to become reactive to sodium vapour and to adsorb carbon dioxide from the atmosphere if it was touched with bare hands and then ignited. Following the precautions outlined in the Experimental section, it was found possible to obtain blanks equivalent to 1 to 2 p.p.m. of carbon.

RECOVERY OF CARBON—

The full analytical procedure was checked by adding known amounts of carbon (as graphite and as carbonate) to samples of low-carbon sodium. The recoveries of carbon, given in Table IV, were satisfactory.

TABLE IV

CARBON RECOVERY FROM THE COMPLETE METHOD

Form of carbon	Oxygen content of sodium, p.p.m.	Carbon added, p.p.m.	Recoveries, per cent.
Graphite	10 to 20	2 to 10	90 to 120
Graphite	1000	30 to 3000	85 to 95
Carbonate	10 to 20	3 to 100	94 to 98

PERFORMANCE OF THE METHOD—

Although these studies have been concerned primarily with sodium, a limited amount of work has indicated that the method is applicable to samples of sodium - potassium alloys containing up to 30 per cent. of potassium.

The method has been in routine use for some hundreds of samples of sodium and sodium - potassium alloys over a period of 4 years, and has operated without serious trouble apart from occasional high blanks. Over the past year, the blank level has varied between 0·5 and 5 p.p.m. with an average of 1·5 p.p.m. These blanks are realistic, because the empty crucible follows all stages of the method alongside the sample crucibles. It has been shown

that the blanks near 5 p.p.m. were usually caused by contamination during sampling; it is essential that sampling stations are kept scrupulously clean and that strict attention is given to the careful handling of sample crucibles.

Duplicate determinations on routine samples have shown that near the 20 p.p.m. level of carbon, the coefficient of variation of the complete procedure is about 10 per cent. All the evidence available to date suggests that the method is probably accurate to within 10 per cent. The operator time for the analysis of 3 samples and 1 blank (or 2 samples and 2 blanks) is about 6 hours. The total elapsed time is about 12 hours.

Conclusions

It has been confirmed that distillation is a suitable technique for the separation of sodium from samples for carbon determination, and Walker and France's procedure for determining free carbon has been extended to determine total carbon, irrespective of its nature. The method has been used with samples as large as 10 g of alkali metal, and hence low levels of carbon down to 1 or 2 p.p.m. can be determined. It would seem feasible to increase the amount of sample taken and to extend the method to even lower limits should this be required.

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The Use of Diphenylcarbazone for the Determination of Microgram Amounts of Lead

By N. TRINDER

(*Ministry of Agriculture, Fisheries and Food, National Agricultural Advisory Service, Kenton Bar, Newcastle upon Tyne* 3)

The conditions necessary for extracting the crimson diphenylcarbazone - lead complex into xylene from an aqueous cyanide solution have been examined. A procedure is described for the spectrophotometric determination of lead isolated by means of diphenylthiocarbazone.

Few organic reagents have been proposed for the colorimetric determination of very small amounts of lead. Diphenylthiocarbazone (dithizone) has been used almost exclusively since it was introduced by Fischer.[1] Dithizone was first used as a means of isolating lead from organic material,[2] and its extension to colorimetric determination soon followed.[3]

The nature of dithizone is such that for the determination of lead in complex mixtures it is necessary to adopt a double extraction procedure, first isolating the lead and then determining it with a weaker solution of dithizone. This procedure has the obvious disadvantage that metals giving a coloured dithizonate at the first extraction may again give a dithizonate at the second extraction unless special precautions are taken. Bismuth is especially troublesome in this respect and, to obviate interference, a preliminary extraction with a diethyldithiocarbamate solution may be necessary.[4] Nevertheless, no satisfactory complementary reagent to dithizone has so far emerged.

Diphenylcarbazone is known to give a red colour with lead,[5,6,7] but attempts to use this reaction for the spectrophotometric determination of lead have so far been unsuccessful.[6,7] This lack of success is probably caused by the number of compounds (for example, iron salts and citrates) that inhibit the development of the lead complex, preventing the use of the reagent on complex mixtures. Diphenylcarbazone is useful, therefore, only for solutions from which interfering agents have been removed and can be considered solely as complementary to an isolating reagent.

Experimental

In the presence of lead, diphenylcarbazone imparts a bright crimson colour to the solvent layer when a weakly alkaline solution is shaken with an aromatic hydrocarbon, amyl acetate or carbon disulphide. Other metals produce similar colours, but in the presence of cyanide, diphenylcarbazone is virtually specific for lead. The optimum conditions for developing the colour were determined with a Unicam SP600 spectrophotometer and 1-cm light path cuvettes.

Choice of solvent—

A number of solvents, in particular benzene, toluene and xylene, were tried, and xylene was found to be the most satisfactory. The blank colour (pale orange) due to the diphenylcarbazone was minimal with xylene, and this solvent was less objectionable to use than either toluene or benzene.

Reagent strength—

The reagent blank increased arithmetically over the range 0·01 to 0·05 per cent. w/v of diphenylcarbazone in xylene. Under the conditions described in the method (given below), the optical density at 525 mμ was about 0·065 for each 0·01 per cent. of diphenylcarbazone. Maximum production of colour in the presence of lead was at 0·02 per cent. Above this concentration there was no increase in colour due to lead.

SENSITIVITY OF REAGENT—

The lead complex gave maximum optical density at 525 mμ, and the molar extinction coefficient for a 1-cm light path at this wavelength was about 72,000 if the colour was read 15 minutes after development. After 2 hours this had increased to about 74,000 and after 20 hours (overnight refrigeration) to about 82,000. After subtraction of the reagent blank, Beer's law was obeyed over the range of 0·4 to 2·0 μg of lead per ml. The optical density was 0·35 for each μg of lead per ml of solvent when the colour was read after 15 minutes.

STABILITY OF THE LEAD - DIPHENYLCARBAZONE COMPLEX—

The complex dissolved in xylene is stable for at least 20 hours at 20° C if kept over aqueous potassium cyanide containing an excess of diphenylcarbazone. Once transferred by pipette into a cuvette the complex begins to fade slowly after 5 minutes or more. Readings should, therefore, be taken expeditiously after pipetting or the colour should be read in the preparation tube (see below).

EFFECT OF POTASSIUM CYANIDE—

Maximum optical density of the lead complex was obtained over the pH range 9·5 to 10·4 in the aqueous phase. This represented the addition of 0·25 to 0·5 ml of 20 per cent. potassium cyanide solution to 3 ml of 1 per cent. nitric acid. The value of the reagent blank fell as more cyanide was added, and the addition of 0·35 ml of 20 per cent. potassium cyanide solution was considered to be the best compromise (pH 9·9). There was a progressive reduction in the intensity of the lead complex colour as the pH of the aqueous phase rose above 10·4.

EFFECT OF AMMONIA—

The procedure outlined below involves the addition of excess potassium cyanide to a weak solution of nitric acid. Some workers might consider this a potential hazard, so the effect of first neutralising the acid with ammonia was studied. It was found that ammonia, in excess, slightly diminished the intensity of the lead complex, but the intensity of the colour was not affected when the acid was just neutralised.

INTERFERENCE DUE TO BISMUTH—

In the presence of nitrate or acetate, and in the absence of chloride, up to 10 μg of bismuth per ml of solvent did not reduce the colour due to the lead - diphenylcarbazone complex. At a solvent concentration of 100 μg of bismuth per ml the bismuth was precipitated in the aqueous layer and some interference with the lead colour occurred.

In the presence of nitrate, bismuth from 1 to 100 μg per ml of solvent caused a slight but constant increase in the reagent blank. The optical density due to these amounts of bismuth was 0·025, fading to 0·005 after 2 hours. This increase is thought to arise from an impurity in the reagent, but different batches of diphenylcarbazone have not been tested to confirm this.

STABILITY OF REAGENT IN XYLENE—

A 0·02 per cent. w/v solution of diphenylcarbazone in xylene stored for 9 months at room temperature in an amber glass bottle gave a constant optical density with 2·0 μg of lead per ml of solvent during this period. The reagent blank, however, rose by 13 per cent. during the 9 months.

METHOD

APPARATUS—

Observations should be made with a Unicam SP600 spectrophotometer or similar instrument. Tubes for the development of the lead - diphenylcarbazone colour must be rinsed with diluted nitric acid (5 volumes of nitric acid, sp.gr. 1·42, to 95 volumes of water) immediately after use.

REAGENTS—

(*i*) FOR THE ISOLATION OF LEAD—

See the report of Analytical Methods Committee of the Society of Analytical Chemistry on the determination of lead.[4]

The following reagent is also required.

Dithizone solution, 0·05 per cent. w/v—Dissolve 0·05 g of diphenylthiocarbazone in 100 ml of benzene. Store in a refrigerator above the freezing point of benzene in an amber glass bottle.

(*ii*) FOR THE DETERMINATION OF LEAD—

All reagents, including water, should be free from lead.

Nitric acid, 1 per cent. v/v—Mix 1 volume of nitric acid, sp.gr. 1·42, with 99 volumes of water.

Bismuth nitrate solution—Dissolve 0·023 g of bismuth nitrate, $Bi(NO_3)_3.5H_2O$, in 100 ml of 1 per cent. nitric acid.

Nitric acid reagent—Mix 1 volume of nitric acid, sp.gr. 1·42, 1 volume of bismuth nitrate solution and 98 volumes of water. This reagent contains 1 μg of bismuth per ml.

Diphenylcarbazone solution, 0·02 per cent. w/v—Shake 0·1 g of diphenylcarbazone with 500 ml of xylene in a vibratory shaker for 1 hour. Filter through a No. 1 Whatman filter-paper into a dry amber glass bottle. If a shaker is not available, heat 0·1 g of diphenylcarbazone with 500 ml of xylene on a water-bath until all the solid material has dissolved.

Potassium cyanide solution—Prepare a 20 per cent. w/v solution in water. Use after 2 days or low results may be obtained.

Ammonia solution, 20 per cent. v/v—Mix 1 volume of ammonia (sp.gr. 0·88) with 4 volumes of water.

Standard lead solutions—

A. Dissolve 1·60 g of lead nitrate crystals in water, add 10 ml of nitric acid (sp.gr. 1·42) and dilute to 1 litre.

B. Dilute 1 volume of solution A to 100 volumes with 1 per cent. nitric acid (1 ml contains 10 μg of lead).

C. Mix 1 volume of bismuth nitrate solution with 20 volumes of standard lead solution B and dilute to 100 volumes with 1 per cent. nitric acid (1 ml contains 2μg of lead).

D. Dilute 20 volumes of standard lead solution B to 100 volumes with 1 per cent. nitric acid (1 ml contains 2 μg of lead).

Xylene—This should be suitable for histological purposes and free from sulphur.

PROCEDURE—

Isolation of lead—Extract the lead into 0·05 per cent. dithizone solution under conditions similar to those described under method A in the Report of the Analytical Methods Committee of the Society for Analytical Chemistry on the determination of lead.[4] Shake the solution vigorously, discard the aqueous layer, and spin the benzene layer containing the lead dithizonate in a centrifuge at 3000 r.p.m. Transfer the cleared benzene to a funnel containing a small cotton-wool plug. Transfer by pipette a suitable volume of the filtered benzene (containing up to 8 μg of lead) to a dry 15-ml centrifuge tube.

Determination of lead in the absence of bismuth—Add 4 ml of 1 per cent. nitric acid. Shake the tube vigorously, then spin it in a centrifuge for 1 minute at 3000 r.p.m. Draw off the benzene layer to waste. Transfer by pipette 3 ml of the aqueous layer into a centrifuge tube. Add 0·35 ml of 20 per cent. potassium cyanide solution to the tube. (Alternatively, add 0·15 ml of 20 per cent. ammonia solution, mix, and add 0·2 ml of 20 per cent. potassium cyanide solution.) Mix the solutions and add 3 ml of 0·02 per cent. diphenylcarbazone solution. Shake the tube vigorously 40 times. Spin the solution in a centrifuge for 1 minute at 3000 r.p.m.

After a standard interval of time, transfer the solvent layer by pipette into a 1-cm cuvette and read the optical density at 525 mμ within 5 minutes. Compute the lead concentration from a calibration graph of 1·0 to 6·0 μg of lead by using standard lead solution D.

Determination of lead in the presence of bismuth—Either read the optical density 2 hours after preparing the lead - diphenylcarbazone complex, or use 4 ml of nitric acid reagent (containing bismuth) to remove the lead from the benzene layer instead of 4 ml of 1 per cent. nitric acid. Read the optical density 15 to 30 minutes after development of the colour. Prepare a calibration graph with the standard lead solution C instead of solution D.

Discussion

It is necessary to spin the solution in a centrifuge and filter the benzene layer containing the lead dithizonate to prevent small amounts of citrate remaining in the solvent phase after extraction. Full recovery of lead was not obtained until this step was introduced.

The procedure adopted for the determination will depend on the time factor, the possibility of bismuth being present and the reactivity to bismuth of the batch of diphenylcarbazone used. At least 9 μg of bismuth per ml of solvent can be present if the method for the determination of lead in the presence of bismuth is used. Precise details of the extraction of the lead dithizonate are not given because the procedure will vary according to the nature of the material under test—indeed, in some circumstances the use of benzene as the solvent for dithizone may not be suitable, and it may be necessary to investigate the possibility of some other solvent, *e.g.*, carbon tetrachloride.

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SHORT PAPERS

Polarographic Determination of 0·01 to 0·10 per cent. of Bismuth in Lead

By J. BASSETT and J. C. H. JONES*
(*Chemistry Department, Woolwich Polytechnic, London, S.E.*18)

The accurate determination of low concentrations (0·01 to 0·10 per cent.) of bismuth in lead is of importance to manufacturers, because the presence of this element can affect various properties of lead, *e.g.*, corrosion resistance and creep characteristics. The need for a rapid, reliable method prompted the present study of the application of polarography to this determination.

Several polarographic procedures have been reported in which the bulk of the lead is removed by precipitation either as the sulphate[1,2] or as the chloride.[3,4] These methods are lengthy and, as our own studies of the removal of lead as lead sulphate have indicated, may entail the loss of bismuth due to its adsorption on the precipitate.

Přibil and Matyska[5] have described an amperometric method for the determination of bismuth in lead that does not require any prior chemical separation. A similar base electrolyte of lead nitrate and nitric acid is used in the rapid polarographic method described in the present paper. The lead sample is dissolved in nitric acid, sodium tartrate added and the pH adjusted to between 1·1 and 1·3 by the addition of ammonia. The polarographic wave obtained (E = −0·01 volts, against an S.C.E.) with an aliquot of the solution is due to copper and bismuth. The addition of EDTA to a second aliquot complexes bismuth so that the polarographic wave now obtained is due to copper only. The difference in wave heights is proportional to the concentration of bismuth.

Method

Apparatus—

A Tinsley single-unit pen recording polarograph (Mark 19) was used. The rate of flow of mercury and the drop time of the capillary were 4·131 mg per second and 3·2 seconds, respectively.

All pH measurements were made with a Pye universal pH meter.

Reagents—

All reagents should be of analytical-reagent grade.

Nitric acid, 50 per cent. v/v, aqueous—Prepare from concentrated nitric acid, sp.gr. 1·42.

Nitric acid, 20 per cent. v/v, aqueous—Prepare from 50 per cent. v/v aqueous nitric acid.

Sodium tartrate solution, 25 per cent. w/v, aqueous.

Gelatin solution, 0·25 per cent. w/v, aqueous.

Disodium ethylenediaminetetra-acetate (EDTA).

Pure lead—Obtainable from Associated Lead Manufacturers.

Pure bismuth—Obtainable from Johnson, Matthey & Co. Ltd.

Standard bismuth solution—Dissolve 0·5000 g of pure bismuth in 200 ml of 50 per cent. nitric acid solution, and make up to 500 ml with water.

1 ml ≡ 0·0010 g of bismuth.

Procedure—

To 5 g of sample in a 100-ml beaker add 40 ml of 20 per cent. nitric acid and heat until dissolution of the sample is complete. Cool the solution, add 2·0 ml of sodium tartrate solution and adjust the pH to between 1·1 and 1·3 by the dropwise addition of concentrated ammonia from a burette (approximately 3 ml is required). The solution should be stirred vigorously during this stage to dissolve the white precipitate that is initially formed. Transfer the solution to a 50-ml graduated flask, add 2·0 ml of 0·25 per cent. gelatin solution and dilute to the mark with water.

* Present address: Associated Lead Manufacturers, 7 Wadsworth Road, Greenford, Middlesex.

By pipette, transfer 20 ml of the solution to a polarographic cell and de-oxygenate by bubbling hydrogen through it for 10 minutes. Record the polarogram (A) from +0·20 to −0·30 volts against an S.C.E.; the reduction of lead ions begins at −0·30 volts against an S.C.E. Dissolve 0·05 g of EDTA in a second 20-ml aliquot, de-oxygenate the solution and again record the polarogram (B) from +0·20 to −0·30 volts against an S.C.E.

Measure the wave heights, h_1 and h_2, obtained at a half-wave potential of −0·01 volts against an S.C.E. in polarograms A and B, respectively, and find the bismuth concentration from the difference in wave heights (h_1–h_2).

CALIBRATION—

Add 0·50, 1·0, 2·0, 3·0, 4·0 and 5·0 ml of standard bismuth solution to separate 5-g samples of pure lead contained in 100-ml beakers (this corresponds to 0·01 to 0·10 per cent. of bismuth). Prepare the solutions and record the polarograms in the absence of EDTA as described under Procedure. Plot wave heights ($E_{\frac{1}{2}}$ = −0·01 volts against an S.C.E.) against concentration of bismuth.

RESULTS

The method was applied to a number of synthetic samples, each containing 5 g of pure lead and a small amount of added impurity. The results (Table I) show that no interference occurs in the presence of these other elements which are normally found in commercial grades of lead.

TABLE I

DETERMINATION OF BISMUTH IN THE PRESENCE OF OTHER ELEMENTS

Impurity added	Form of added impurity	Bismuth added, per cent.	Bismuth found, per cent.	Relative error, per cent.
0·060% Copper	Copper(II) chloride	0·060	0·059	−2
0·010% Copper		0·060	0·060	0
0·050% Silver	Silver nitrate	0·050	0·050	0
0·050% Iron	Iron(III) chloride	0·050	0·051	+2
0·050% Zinc	Zinc nitrate	0·050	0·050	0
0·050% Cadmium	Cadmium nitrate	0·050	0·050	0
0·010% Tellurium	Tellurium dissolved in nitric acid	0·050	0·048	−4
0·010% Selenium	Selenium dissolved in nitric acid	0·050	0·048	−4
0·050% Nickel	Nickel chloride	0·050	0·052	+4
0·050% Arsenic	Lead sample containing 0·05% arsenic	0·050	0·050	0
0·050% Tin	Lead sample containing 0·05% tin	0·050	0·053	+6
0·050% Antimony	Lead sample containing 0·05% antimony	0·050	0·051	+2

The reproducibility of the method was investigated with 5-g samples of pure lead to which the equivalent of 0·05 per cent. of bismuth had been added as a known volume of standard bismuth solution. The standard deviation calculated from the results of 11 determinations was 0·002 per cent.

DISCUSSION

The method described is rapid and can be completed in about 30 minutes. With a conventional polarograph, the lower limit of bismuth concentration that can be determined is about 0·005 per cent.

The use of a cathode-ray polarograph would probably enable the method to be applied at lower concentrations of bismuth, *e.g.*, 0·001 per cent., but the analysis of samples containing a high copper-to-bismuth ratio (*e.g.*, 0·10 per cent. of copper and 0·001 per cent. of bismuth) would not be satisfactory by this method as bismuth is found from the difference in wave heights.

We thank Dr. A. I. Vogel, Head of the Chemistry Department, Woolwich Polytechnic, for his interest, and one of us (J.C.H.J.) thanks the Directors of Associated Lead Manufacturers for enabling him to undertake the present research.

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The Photometric Determination of Excess of Cadmium in Cadmium Oxide

By V. J. NORMAN

(*Australian Defence Scientific Service, Defence Standards Laboratories, Department of Supply, Adelaide*)

Cadmium oxide is considered to be a degenerate n-type semi-conductor,[1] in which the donors arise either from impurities or excess of interstitial cadmium.[2] From the results of precision measurements of lattice parameters of samples of doped cadmium oxide, Cimino and Marezio[3] inferred an excess of cadmium of the order of 0·01 per cent.

A photometric method developed by the author[4] for determining the excess of zinc in zinc oxide has been applied to the determination of excess of cadmium in cadmium oxide. The method, which is based on the reduction of dichromate, gives good reproducibility, and is sensitive to 0·1 p.p.m. of excess of cadmium by weight.

Method

The apparatus and reagents required, and the calibration and analytical procedure used for determining excess of interstitial cadmium in cadmium oxide are identical with those specified in the author's paper[4] for determining excess of zinc in zinc oxide except for the following modifications—

(1) Use 10 ml of acid mixture in place of 20 ml.
(2) Add 4·0 g of cadmium oxide instead of 5·0 g of zinc oxide.
(3) In the preparation of the blank solution (iii), dilute the 10 ml of acid mixture with 25 ml of water before adding the 4·0 g of cadmium oxide.

Note—

For samples of cadmium oxide in which the excess of cadmium exceeds 30 p.p.m., a proportionately smaller sample weight may be taken, provided that the amount of acid mixture used is correspondingly reduced.

Results

The results of analysis of three samples of cadmium oxide by this method are shown in Table I.

Samples A and B were analytical-reagent grade cadmium oxide. Sample C was prepared by dissolving high purity spectrographic grade cadmium metal in analytical-reagent grade acid and precipitating it as the carbonate. The cadmium carbonate was ignited for 5 hours at 600° C.

Table I

Results by the proposed method

Optical density of blank solution .. = 0·980
Optical density of "ignited reference" .. = 0·096
Net optical density .. .. .. = 0·884 (equivalent to 25 ml of 0·0001 N dichromate)

Sample	Reference, net optical density	Equivalent dichromate, a ml	Colour, net optical density	Equivalent dichromate, b ml	(a - b), ml	Excess of cadmium,* p.p.m.
A (4·0 g)	0·740	21·0	0·064	1·9	19·1	26·9
	0·746	21·2	0·091	2·6	18·6	26·2
	0·742	21·1	0·079	2·3	18·8	26·5
B (4·0 g)	0·812	23·1	0·441	12·5	10·6	14·9
	0·799	22·7	0·432	12·3	10·4	14·7
C (2·0 g)	0·879	24·9	0·176	5·0	19·9	56·1
	0·885	25·0	0·205	5·9	19·1	53·9

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* For a sample weight of 4·0 g of cadmium oxide, the excess of interstitial cadmium, in p.p.m. by weight, is given by the expression 1·41 (a–b).

The Refractive Index of Aqueous Perchloric Acid

By J. R. McLEAN and G. S. PEARSON

(*Ministry of Aviation, Rocket Propulsion Establishment, Westcott, Aylesbury, Bucks*)

Perchloric acid (nominal 72 per cent.) is used in laboratory experiments at this Establishment in research into the combustion mechanism of solid propellents containing ammonium perchlorate.[1,2] The acid used is the commercially available analytical-reagent grade, which is specified to be 71·0 to 73·0 per cent. w/w. It is necessary to check each bottle of acid supplied to ensure that it does contain 71·0 to 73·0 per cent. acid, as it has been reported[3] that nominal 72 per cent. acid can be as low as 65 per cent. Conventional analysis by titration is inconvenient because it requires special care, as 72 per cent. perchloric acid is hygroscopic.

Refractive-index measurements offer a rapid and convenient check on the perchloric acid concentration. Values of the refractive index are available in the literature of acid concentrations from 0 to 20 per cent.,[4] and from 50 to 70 per cent.[5] However, a marked discrepancy was observed between the present measurements and the literature values over the 50 to 70 per cent. range, hence the refractive index was determined for the complete range from 0 to 72 per cent., and the reliability of the present results established by using perchloric acid from several sources.

Experimental

Perchloric acid was obtained from 5 different sources: Hopkin and Williams Ltd. (Essex, England), AnalaR grade 72 per cent.; Eastman Kodak Co. (Rochester, N.Y.), 72 per cent.; Merck AG. (Darmstadt, Germany), guaranteed reagent, 70 per cent.; Merck Suprapur, 70 per cent.; and G. F. Smith Chemical Co. (Columbus, Ohio), C.P. vacuum distilled, 72 per cent.

The Hopkin and Williams acid was diluted with de-ionised, distilled water to make up solutions of nominal concentration of 10, 20, 30, 40, 50, 60, 62·5, 65, 67·5 and 70 per cent. of acid. These solutions were analysed by titration against N sodium hydroxide, with phenolphthalein as indicator. Titration against 0·1N sodium hydroxide was not satisfactory and gave consistently low values. This was attributed to weighing errors and water absorption.

The refractive index was measured on an Abbé refractometer maintained at 20° and 30° C. The refractometer was washed and allowed to dry thoroughly between readings. The refractive indices of AnalaR grade chloroform and carbon tetrachloride, and also of de-ionised, distilled water were determined at the same time as the acid refractive indices.

Results and discussion

The refractive indices for a range of perchloric acid concentrations from 0 to 72 per cent. (Hopkin and Williams acid) are presented in Table I, together with the values obtained for the

Table I

Refractive indices of aqueous perchloric acid,

(Hopkin and Williams Ltd. AnalaR grade)

Per cent. w/w	Refractive index	
	20° C	30° C
0·00	1·3330	1·3320
9·73	1·3395	1·3381
20·05	1·3470	1·3452
30·75	1·3580	1·3559
40·31	1·3680	1·3665
50·28	1·3813	1·3800
60·08	1·3983	1.3960
62·81	1·4034	1·4010
64·00	1·4054	1·4028
68·52	1·4129	1·4103
70·06	1·4151	1·4130
72·62	1·4190	1·4159
Chloroform*	1·4445	1·4390
Carbon tetrachloride* ..	1·4601	1·4450

* AnalaR specifications of n_{20}^{D} for chloroform and carbon tetrachloride are 1·4440 to 1·4450, and 1·4600 to 1·4610, respectively.

refractive indices of water, chloroform and carbon tetrachloride. The refractive indices of the concentrated acid from the five sources are presented in Table II. These give a good straight line plot.

TABLE II

REFRACTIVE INDICES OF CONCENTRATED PERCHLORIC ACID

Per cent. w/w (by analysis)	Refractive index at 20° C	Source
72·59, 72·65	1·4193	Hopkin and Williams AnalaR grade
72·55, 72·59	1·4193	Eastman Kodak
69·52, 59·52	1·4153	Merck Suprapur
69·88, 69·88	1·4156	Merck guaranteed reagent
70·68, 70·72	1·4170	G. F. Smith Chemical Co.

The literature values[4] from 0 to 20 per cent. are in reasonable agreement with those reported here. However, those obtained by Smith and Lamplough[5] (also made with an Abbé refractometer) with 50 to 70 per cent. acid are higher than the values we obtained by about 0·5 per cent., corresponding to a change of acid concentration of 4·5 per cent. in the range 50 to 70 per cent.

It is concluded that the previous values reported[5] for the refractive index of 50 to 70 per cent. acid are in error. This error may have arisen from an impurity in the acid used, or an error in the titration of the acid. In the present work, perchloric acid from 4 widely separated sources has been used and consistent results have been obtained. Further, refractive indices measured for chloroform, carbon tetrachloride and water are in excellent agreement with literature values, thus showing that the refractometer was not displaying a systematic error. It is therefore believed that the present results are reliable and can be used as the basis of a rapid analytical method for checking the concentration of aqueous perchloric acid.

It is to be noted that the cement used in the refractometer must *not* be a litharge - glycerine cement, because this produces a dangerously explosive compound with perchloric acid.[6] An inert cement such as polythene must be used.

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Semi-quantitative Determination of Organophosphorus Insecticides by the Ring-oven Technique with Preliminary Thin-layer Chromatography

BY I. PEJKOVIĆ-TADIĆ, M. B. ĆELAP, T. J. JANJIĆ

(*Institute of Chemistry, Faculty of Science, University of Belgrade, 16 Studentski trg, Belgrade*)

AND S. LJ. VITOROVIĆ

(*Institute for Application of Nuclear Energy in Agriculture, Veterinary and Forestry, Zemun, 15 Baranjska, Yugoslavia*)

THIS paper describes a method for the semi-quantitative determination of four organophosphorus insecticides in mixtures: Baytex (fenthion), diazinon, Guthion (azinphosmethyl) and malathion.

The insecticides were first individually determined with the Weisz ring oven and the following mean deviations were obtained: Baytex, 1·8 per cent.; diazinon, 3·9 per cent.; Guthion, 3·2 per cent.; and malathion, 3·4 per cent. (Table I).

TABLE I

DETERMINATION OF INDIVIDUAL INSECTICIDES ON THE RING OVEN

Baytex		Diazinon		Guthion		Malathion	
Present, μg per μl	Found, μg per μl	Present, μg per μl	Found, μg per μl	Present, μg per μl	Found, μg per μl	Present, μg per μl	Found, μg per μl
2·50	2·50	25·0	25·0	2·50	2·66	5·00	5·00
2·50	2·50	25·0	26·6	2·50	2·66	5·00	5·16
2·50	2·66	25·0	26·6	2·50	2·50	5·00	5·32
3·00	2·95	30·0	28·3	3·00	3·00	6·00	5·66
3·00	3·00	30·0	30·0	3·00	3·10	6·00	6·00
3·00	3·08	30·0	31·5	3·00	3·10	6·00	5·66

Mixtures of the four insecticides were then analysed. After chromatography of their solution on silica-gel G layers, the separated insecticides were scraped into a sintered-glass funnel mounted above the oven, washed into the ring zone and determined as above. The deviations from the calculated values were as follows: Baytex, 5·5 per cent.; diazinon, 5·5 per cent.; Guthion, 5·6 per cent.; and malathion, 6·1 per cent. (Table II).

TABLE II

ANALYSIS OF INSECTICIDE MIXTURES

	Concentration of insecticide present, μg per μl				Concentration of insecticide found, μg per μl			
Mixture	Baytex	Diazinon	Guthion	Malathion	Baytex	Diazinon	Guthion	Malathion
	2·50	25·0	2·50	5·00	2·66	24·1	2·25	4·50
I	2·50	25·0	2·50	5·00	2·66	22·5	2·33	5·00
	2·50	25·0	2·50	5·00	2·66	24·1	2·50	4·50
	3·00	30·0	3·00	6·00	2·83	28·3	2·83	6·32
II	3·00	30·0	3·00	6·00	2·91	31·6	2·83	5·66
	3·00	30·0	3·00	6·00	3·16	31·6	2·83	5·66

EXPERIMENTAL

RING-OVEN METHOD—

Standard scales were prepared by spotting 1, 2, 4, 6, 8 and 10 μl of stock solutions of the appropriate insecticide in chloroform (Baytex, 0·1 per cent.; Guthion, 0·1 per cent.; malathion, 0·2 per cent.; and diazinon, 1 per cent. w/v) on to the 5·5-cm diameter filter-paper (Schleicher and Schüll No. 589[2]) by means of a 10-μl micro-pipette. The substance was then washed with 0·5 ml of chloroform. The paper was air-dried, sprayed with palladium(II) chloride reagent[1] (a 0·5 per cent. aqueous solution acidified to pH 3 with two to three drops of concentrated hydrochloric acid) and washed with two 0·5-ml portions of 0·05 N hydrochloric acid. After thorough washing with water, the paper was dipped into ammonium sulphide solution for 30 seconds and dried in an oven at 105° C for 10 minutes. Stable brown rings on a straw-coloured background were obtained.

In the same manner 3 coloured rings were prepared for each insecticide to be determined and their intensities compared with those of the standard scale; the amounts present (C_a) were then calculated by the expression—

$$C_a = C_s \frac{n_s}{n_a}$$

where C_s is the concentration of the standard solution, n_a the volume of sample solution, and n_s the volume of standard solution used in the matching ring.

COMBINATION OF THE RING OVEN WITH THIN-LAYER CHROMATOGRAPHY—

A micro-pipette was used to transfer 1, 2 and 3 μl of chloroform solution of the insecticide mixture I or II to an air-dried 0·2-mm thick silica-gel G chromatoplate (20 × 20 × 0·5 cm). The mobile phase, consisting of hexane and acetone (4 + 1), was allowed about 30 minutes for a frontal migration of 14 cm, and the plate was then taken out of the jar (32 × 20 × 10 cm). At the margin of the plate a guide mixture containing 2 μl of the mixture I or II was run in parallel and sprayed with palladium(II) chloride reagent. The R_F values were: diazinon, 0·50; Baytex, 0·41; malathion, 0·29; and Guthion, 0·14. The areas of the samples were lightly marked with a spatula and they were then scraped quantitatively from the plate into the funnel (porosity G4). Each lot of scrapings

was washed with about 1 ml of chloroform on to the round filter-paper and the insecticide determined as above. The standard rings were obtained by carrying aliquots of solutions of the individual insecticides through the same chromatographic treatment.

Results

A mixture of organophosphorus insecticides Baytex, diazinon, Guthion and malathion was analysed by using thin-layer chromatography in combination with the Weisz ring oven. The determinations performed with 10 to 100 μg of the insecticide had a deviation of $\pm 5{\cdot}6$ per cent. from the calculated value.

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Comments on "The Effect of Nitrilotriacetic Acid Impurity on the Standardisation of Solutions of Ethylenediaminetetra-acetic Acid"

By R. G. MONK

(*U.K.A.E.A., Atomic Weapons Research Establishment, Aldermaston, Berkshire*)

The observations reported in the paper concerned[1] are of great importance to those people working with EDTA titrations, and it is possible that much of the large amount of work published on the subject over the last 15 years has been affected, to some degree, by the presence of unknown amounts of NTA in the EDTA used. Unfortunately, the paper contains certain errors and it is the purpose of this note to draw attention to them in the hope of avoiding future confusion.

The log$K$'s and logK_H's at pH 10 (K_H is the conditional stability constant), which are of relevance to the present discussion, are listed below. They are all taken or calculated from results compiled by Wilson and Wilson.[2]

Complex	Log$K$	LogK_H at pH 10
Zinc - EDTA	16·5	16·0
Zinc - NTA	10·6	10·4
Zinc - Solochrome Black T	—	11·4
Cadmium - EDTA	16·5	16·0
Cadmium - NTA	9·8	9·6

It is clear that the logK_H's of the NTA and EDTA complexes are sufficiently different to produce the two inflections in a potentiometric titration curve of zinc or cadmium that the authors have observed. However, the authors' assumption, at the bottom of p. 212, "that the two successive end-points are due to EDTA and NTA, respectively," is clearly wrong. The first end-point corresponds to the complexation of the last traces of metal ion by the total EDTA and NTA added. However, because EDTA gives the more stable complex, the shape of the inflection is governed by the NTA - metal ion equilibrium, and the first end-point is essentially due to NTA. Further titration results in decomposition of the metal - NTA complex by EDTA, the second inflection corresponding to the completion of this process, at which stage all the metal is present as the EDTA complex. The second inflection is therefore an EDTA end-point, the shape of the curve being modified by the presence of some NTA. The titration is analogous to that of a strong base by a mixture of strong and weak acids.

Calculation of the molarity of the solution from the volume used up to the first end-point gives the total NTA and EDTA concentration, while the concentration of EDTA alone may be calculated from the total volume used up to the second inflection.

Deductions by the authors on pages 213 and 215 concerning titrations with visual indicators are also in error as they are based on the erroneous conclusions from the potentiometric titrations.

In the authors' Fig. 1, the ordinate y is equal to—

$$\frac{\text{Complexometric normality of solution found by titration against metal M}}{\text{Complexometric normality of solution found by titration against zinc at pH 10}}.$$

Values of y greater than 1 can only be the result of a greater response of the indicator to NTA in the titration of M than in the titration of zinc at pH 10. It will be noticed that y is, in fact, greater than 1 for most of the metals titrated, and is particularly high for bismuth and cadmium. If the authors' contention were true that Solochrome Black T detects the EDTA end-point in the titration of cadmium and the NTA end-point in the titration of zinc, then values of y for cadmium would be less than 1 and the graph would slope downward from left to right.

The values of y may be compared with the ratio—

$$\frac{\text{Total (EDTA + NTA) normality of solution}}{\text{EDTA normality of solution}} = r$$

If EDTA (molecular weight, 372·2) contains x per cent. of NTA (molecular weight, 191·1), then $r = \dfrac{100 + 0{\cdot}95x}{100 - x}$. The corresponding values of x, r and y in the titrations of bismuth and cadmium are given below.

x	0·25	0·50	0·75	1·00
r	1·0049	1·0098	1·0147	1·0197
y (bismuth) ..	1·0041	1·0095	1·0140	1·0172
y (cadmium) ..	1·0048	1·0089	1·0134	1·0169

The maximum value attainable by y at each level of x is represented by r, and would occur if the precise NTA end-point were being detected in the titration of M and the precise EDTA end-point were being detected in the titration of zinc at pH 10. In fact, these figures do show that y approaches r quite closely in the titration of bismuth, and somewhat less so in the titration of cadmium. It is clear, therefore, that catechol violet is an effective indicator for NTA in the titration of bismuth, that Solochrome Black T does not respond appreciably to NTA in the titration of zinc at pH 10, and that the latter indicator does respond fairly well to NTA in the titration of cadmium. In the penultimate sentence of the Discussion, therefore, "EDTA" should be replaced by "NTA," and *vice versa*.

All the above deductions have been checked by titrations with NTA alone. The bismuth end-point obtained by using catechol violet is nearly as good with NTA as with EDTA. At pH 10, the titration of cadmium with NTA, with Solochrome Black T as the indicator, gives an end-point that is not very good, but which could probably be detected with a precision of 1 to 2 per cent. under optimum conditions. With zinc, under the same conditions, no end-point at all occurs; the solution becomes progressively bluer as excess of NTA is added, but only by virtue of mass-faction. Even with a several-fold excess of NTA the colour retains a violet tinge. This is to be expected, as the first set of figures shows that the complex between zinc and Solochrome Black T is more stable than that with NTA. The stability constant of the cadmium - Solochrome Black T complex is not available, but we may infer that it is considerably less than that of the cadmium - NTA complex.

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Mr. R. N. P. Farrow and Mr. A. G. Hill agree with Mr. Monk's comments, but feel that they do not affect the main aspect of the paper.

A Simple Multi-purpose Titrimeter

By W. JENNISON and M. L. CLARK

(*U.K.A.E.A., Analytical Group, Dounreay Experimental Reactor Establishment, Thurso, Caithness, Scotland*)

An inexpensive titrimeter has been designed for dead-stop, constant-current, photo-electric and potentiometric titration with any electrode system except glass. This instrument may also be a general-purpose galvanometer. Although of simple construction, the design may be of interest to others as the total cost of the components is less than £10, and the instrument can be built in a few hours. There is negligible short-term drift and a minimum of maintenance is required. The batteries used have shelf-life and no fault has developed after two years of continuous use.

Experimental

Description of instrument—

Two transistors are arranged as a common-emitter difference-amplifier mounted in an aluminium block and are so matched that they are independent of the resistance of the source to be measured. This amplifier has an input resistance of about 1·2 megohms for voltage measurement and 25,000 ohms for current measurement. A maximum sensitivity of 1·0 volt or 1·0 μA for full-scale deflection is obtained, but sensitivity controls that are normally off can be used to shunt the amplifier as required (Fig. 1). For calibration purposes, a voltage derived from B4, R6 and R7 can be fed through R3 to the detector input by depressing the push-switch (PS). A mark inscribed on the meter at 81 per cent. of full-scale deflection allows the detector to be set up to read 0 to 1 μA or 0 to 1·25-volt full scale. Five ranges are provided by the selector-switch and, when not in use, this switch is left in the "off" position, the "Set-mV" potentiometer at zero, the "Set-scale" potentiometer at maximum resistance, and the cell terminals disconnected.

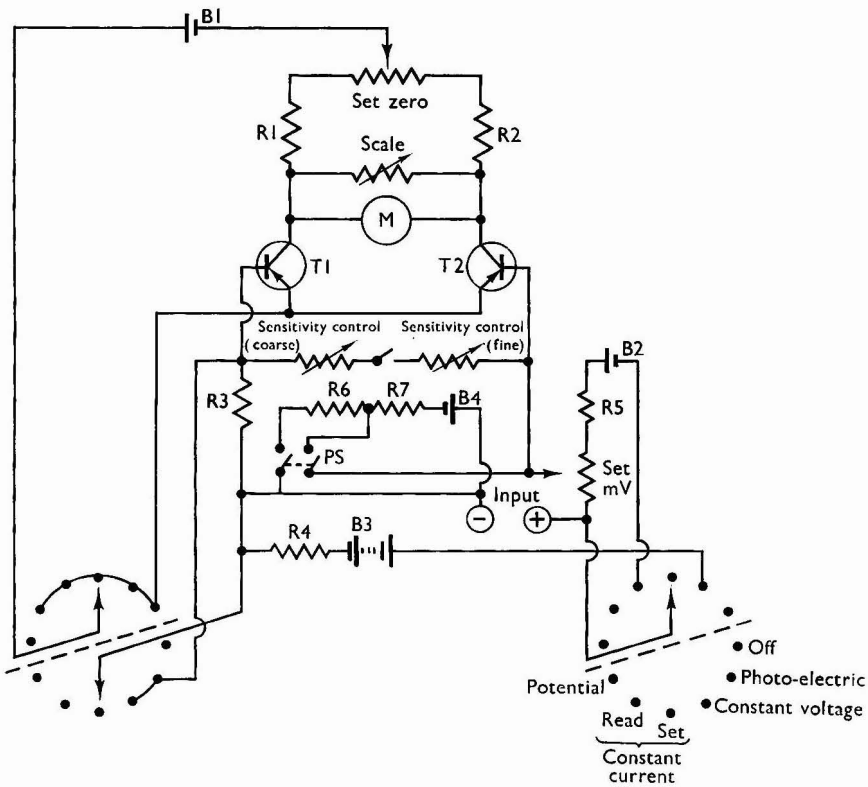

Fig. 1. Diagram showing circuit for instrument

Photo-electric titration—

Linearity of photocell response is normally unimportant in this type of titration, and the input resistance of the detector is varied in inverse proportion to the light falling on the cell. The selector-switch is turned to the "Photo-electric" position and a photo-voltaic-cell assembly with filter is connected. With the cell disconnected, the "Set-zero" potentiometer is adjusted to give zero on the meter scale. The cell is then connected, and the meter deflection is adjusted to a suitable value by the sensitivity controls. If light transmission through the titration solution is expected to rise at the end-point, the controls should be adjusted to give a meter deflection of 25 per cent. of full scale. Conversely, if the transmission is to fall, the meter is set to full-scale deflection. A titration may now be performed.

TITRATION AT CONSTANT VOLTAGE, DEAD-STOP TITRATION—

With the selector-switch in the "Constant voltage" position a known potential of 0 to 125 millivolts is impressed across the input terminals in series with the detector. The "Set-zero" potentiometer is adjusted, and the cell terminals are connected to the instrument. For dead-stop titrations, the sensitivity controls may be used to set the meter needle to full scale, whereas, in amperometric titrations of the "kick-off" type the sensitivity controls are switched off. The titrations may now be performed while observing the change in current between additions of titrant.

TITRATION AT CONSTANT CURRENT (POLARISED ELECTRODES)—

With the selector-switch in the "Constant-current Set" position the push switch is depressed and the meter needle is adjusted to give full-scale deflection by using the "Set-scale" potentiometer. The push-switch is released, the cell containing the solution to be titrated is connected across the instrument input terminals, and the meter zero is re-set. Finally, the selector-switch is moved to the "Constant-current Read" position, a polarising current of 2 μA flows through the cell, and the titration may be performed while observing the potential differences between additions of titrant. The meter reads 0 to 1·0 volt.

POTENTIOMETRIC TITRATIONS—

The selector-switch is put to the "Potential" position, the meter needle is set on zero, and the meter calibration is made as above. If a scale of 0 to 2·5 volt is required, the sensitivity controls can be adjusted accordingly. After calibration the appropriate electrode combination is connected across the input terminals and the titration is performed.

Appendix

COMPONENTS LIST

B_1, B_2	= 1·5-volt Exide T.20 batteries
B_3	= 15-volt Exide DH.521 battery
B_4	= 1·35-volt Mallory RM.3R Mallory cell
M	= 0 to 25 microammeter (internal resistance 1500 ohms)
Set zero	= 1000-ohm, 2-watt, wire-wound potentiometer
Scale	= 10,000-ohm, 2-watt, wire-wound potentiometer
Sensitivity control (Coarse)	= 25,000-ohm, 2-watt, C.T. potentiometer with "on - off" switch
Sensitivity control (Fine)	= 1000-ohm, 2-watt, wire-wound potentiometer
Set mV	= 100-ohm, 2-watt, wire-wound potentiometer
R_1	= 1800-ohm, $\frac{1}{8}$-watt resistor
R_2	= 1800-ohm, $\frac{1}{8}$-watt resistor
R_3	= 680,000-ohm and 560,000-ohm, $\frac{1}{8}$-watt resistors in series, with a tolerance of 1 per cent.
R_4	= 6·8-megohm, $\frac{1}{8}$-watt resistor with a tolerance of 1 per cent.
R_5	= 1000-ohm and 100-ohm, $\frac{1}{8}$-watt resistors in series, with a tolerance of 1 per cent.
R_6	= 1000-ohm, $\frac{1}{8}$-watt resistor with a tolerance of 1 per cent.
R_7	= 330-ohm, $\frac{1}{8}$-watt resistor with a tolerance of 1 per cent.
T_1	= OC75
T_2	= OC75
PS	= D.P.S.T. push-button switch spring-loaded "off"

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Some Anomalous Results given by Phase-solubility Analysis

BY J. V. WILKINSON AND J. S. WRAGG

(*Analytical Development Group, Standards Department, Boots Pure Drug Co. Ltd., Station Street, Nottingham*)

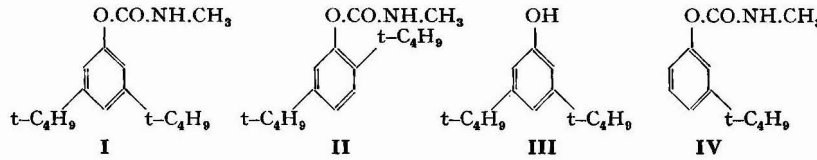
IN the accounts of phase-solubility analysis, given by Garratt, Johnson and King,[1] Higuchi and Connors,[2] Mader[3] and Outch,[4] each author has indicated some of the conditions in which the method is likely to fail. In all instances, it has been pointed out that the components of the solute must not form a solid solution, neither must they be present in the ratio of their solubilities. However, Higuchi is alone in emphasising that the most serious limitation to the application of phase-solubility analysis is the condition that the solubility of each component must not be affected by the presence of the other components.

When samples of an insecticide of commercial quality were recently examined by phase-solubility analysis, total impurity contents were obtained that were more than twice those found by gas - liquid chromatography, infrared spectrophotometry and isotope-dilution analysis. To investigate this occurrence, several mixtures were prepared containing the pure insecticidal compound together with known amounts of some of the impurities likely to be present in the commercial material, and these mixtures were assayed by the phase-solubility method.

Experimental

Preparation of mixtures—

Amounts of the insecticide, 3,5-di-t-butylphenyl methylcarbamate, (butacarb, **I**), were purified by re-crystallisation from light petroleum. Mixtures of known composition were prepared by adding to this re-crystallised material known amounts of 2,5-di-t-butylphenyl methylcarbamate, **II**, 3,5-di-t-butylphenol, **III**, and 3-t-butylphenyl methylcarbamate, **IV**, each of which had also been purified by re-crystallisation.



I **II** **III** **IV**

Prepare the mixtures by heating **I** to a temperature just above its melting-point of 100° C, and adding one or more of **II**, **III** and **IV** with stirring; continue stirring until the mixture has completely crystallised. When cool, grind the mixtures in a mortar.

Table I

Composition of prepared mixtures

	Percentage by weight			
Mixture	Re-crystallised insecticide **I**	Impurity **II**	Impurity **III**	Impurity **IV**
1	82	15	3	–
2	95	5	–	–
3	96	–	4	–
4	91	5	4	–
5	91	5	4	–
6	95	–	–	5
7	90	–	–	10
8	80	–	–	20

The compositions of the mixtures are shown in Table I. Two batches of re-crystallised insecticide (**I**) were used in the preparation of these mixtures; the first batch was used in mixtures 1 to 4, and the second was used in mixtures 5 to 8.

Phase-solubility procedure—

The technique described by Garratt was used in the phase-solubility determinations. In each instance, the equilibration temperature was 31° C and, except where otherwise stated, the period of equilibration was from 10 to 14 days. The solvents used were hexane, cyclohexane, methanol - water (5 + 2) and methanol - water (3 + 1).

To determine the solution concentration, the solvent was removed at room temperature under a stream of nitrogen and the residue dried at 100° C to constant weight.

After the assay of one of the process samples by phase-solubility analysis the solid phases were separated from the solution phases, washed with a limited amount of solvent and dried. The solid phases were examined by thin-layer chromatography on silica gel with hexane - t-pentyl alcohol (9 + 1) as the solvent. After drying, the chromatograms were sprayed with 5 N sodium hydroxide solution in methanol - water (1 + 1), heated at 100° C for 5 minutes, cooled and then sprayed with a 0·5 per cent. w/v solution of 2,6-dichloro-*p*-benzoquinone-4-chloroimide in methanol.

The solid phases were then combined and themselves assayed by the phase-solubility method.

Results

The results of assaying two process samples of the insecticide, the two re-crystallised samples of the insecticide and the prepared mixtures are shown in Table II.

Table II

Results of phase-solubility analysis

Sample	Equilibration period, days	Solvent	Percentage w/w of total compounds other than I added	Percentage w/w of total compounds other than I found
Process sample 1*	5	Hexane	—	32·2
	10	Hexane	—	32·4
	21	Hexane	—	33·0
Process sample 2, solid phases	10	Methanol - water (3 + 1)	—	17·6
	21	Hexane	—	23·3
	10	Hexane	—	0·0
Re-crystallised insecticide I used to prepare mixtures 1 to 4	14	Hexane	—	2·0
	14	Methanol - water (5 + 2)	—	0·6
Mixture 1†	10	Hexane	18	38·7
2	14	Hexane	5	11·7
2	14	Methanol - water (5 + 2)	5	6·1
2	14	Cyclohexane	5	9·7
3	14	Hexane	4	12·5
3	14	Methanol - water (5 + 2)	4	4·9
4	14	Hexane	9	22·8
4	14	Methanol - water (5 + 2)	9	11·2
Re-crystallised insecticide I used to prepare mixtures 5 to 8	14	Methanol - water (5 + 2)	—	0·0
Mixture 5	14	Methanol - water (5 + 2)	9	11·5
6	14	Methanol - water (5 + 2)	5	5·5
7	14	Methanol - water (5 + 2)	10	11·2
8	14	Methanol - water (5 + 2)	20	23·0

* Sample assayed for I by infrared = 88 per cent., and by isotope dilution = 86·6 per cent.

† Sample assayed for I by isotope dilution = 82·2 per cent.

A typical phase-solubility diagram is shown in Fig. 1. When the impurities are present in sufficient concentration, the diagrams show changes of slope. This is illustrated in Fig. 2.

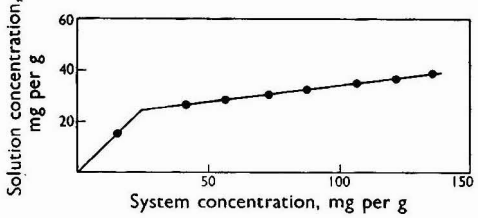


Fig. 1. Phase solubility of mixture 4, with hexane as solvent

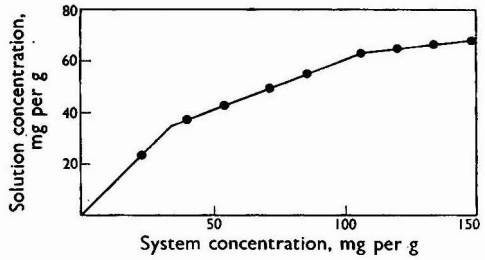


Fig. 2. Phase solubility of mixture 1, with hexane as solvent

No impurities were detectable in the solid phases after the assay of process sample 2, either when they were re-assayed or when they were examined by thin-layer chromatography.

Discussion

The following points arise from a consideration of these results—

(*i*) When hexane was used as the solvent, the contents of total impurity were more than twice those expected. When the more polar solvent methanol - water (5 + 2) was used, the results were still higher than theory, though only by 10 to 20 per cent.

A probable explanation of these results is that complexes are formed between the principal compound, **I**, and the other components of the solute. These complexes are probably the result of hydrogen bonding, though a simple 1 to 1 complexing would not fully explain the results when hexane was used as solvent. Such complex formation would be a reversible process and the position of the equilibrium would be influenced by the polarity of the solvent. The effect of complex formation on a phase-solubility determination would be to increase the apparent proportion by weight of the other components.

(*ii*) None of the phase-solubility diagrams showed any abnormality. The recovery of solute from the tube below saturation point each time was satisfactory. The solid phases were found to consist of pure **I**.

The point we particularly wish to make is one that we consider has not been adequately stressed by previous authors; when a substance is assayed by phase-solubility analysis with a given solvent, it is possible for the results to be in error, because of the formation of complexes, without the phase-solubility diagram giving any warning of the situation. While complex formation may be revealed by a change of solvent to one of different polarity, the use of a relatively polar solvent will not guarantee accurate results. It follows that, in any system where complex formation is a possibility, it is advisable to perform recovery experiments on suitable mixtures of known composition.

We thank Dr. D. A. Peak for his helpful advice and the provision of the compounds, and Dr. T. I. Watkins and Mr. M. W. Baker for the assays by the isotope-dilution method.

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A Simple Chromatograph for the Analysis of Air, Chlorine and Hydrogen Chloride

By D. M. RUTHVEN and C. N. KENNEY

(*Department of Chemical Engineering, University of Cambridge, Pembroke Street, Cambridge*)

The analysis of mixtures of air, chlorine and hydrogen chloride by gas chromatography has been studied in order to develop a rapid and accurate method suitable for kinetic measurements. Although the gas-chromatographic determination of chlorine has been reported several times, few workers appear to have used the method for hydrogen chloride.[1,2] The problems arising from the corrosive nature of these gases did not prove as great as expected, and the principal difficulty was to find a column which would give adequate resolution without causing excessive tailing of the hydrogen chloride peak. The instrument that was finally developed has been in operation for about a year and has proved satisfactory.

Experimental

Detector—

It has been generally assumed[2,3] that conventional katharometers are not suitable for use with these corrosive gases. Glass-covered katharometer elements which have been used by several workers[2,4] were tested and found to have the disadvantage of slow response. A rapid detector-response is essential for this analysis because it is necessary to work near to the limit of resolution of chlorine and air to minimise tailing of the hydrogen chloride peak. A conventional Gow-Mac katharometer, type 9285, fitted with high sensitivity W2 (tungsten - rhenium) elements, was tried and found to be completely satisfactory. Although the stainless-steel block became slightly corroded, there has been no sign of any deterioration of the elements.

CHOICE OF COLUMN—

Several column packings mentioned in the literature were tested before tri-tolyl phosphate, supported on Celite, was chosen. This stationary phase has the great advantage that the normal order of elution is reversed so that chlorine is eluted before hydrogen chloride. The chlorine peak is therefore unaffected by any tailing of the hydrogen chloride. Chlorine appears to react slowly with tri-tolyl phosphate, and the material that was actually used was first saturated with chlorine for several hours until the reaction had ceased. Although this packing material proved far superior to the others tested, details of column construction and packing required careful attention.

To reduce tailing, it was found necessary to keep both the liquid loading and the column length to the minimum consistent with resolution. A high carrier-gas flow-rate is desirable as this reduces tailing without appreciably affecting efficiency. In fact, the carrier-gas flow was limited to 75 ml per minute by the detector. With hydrogen, which proved to be perfectly satisfactory, the pressure drop across the column at this flow-rate is half an atmosphere. The column finally selected consists of 220 cm of 3 mm i.d. PTFE tube packed with 60 to 80-mesh acid-washed Celite having a liquid loading of 4 per cent. by weight of tri-tolyl phosphate. The presence of traces of moisture increases the tailing of the hydrogen chloride peak, but this effect is much less pronounced than with any of the other columns tested. The column was operated at room temperature and has shown no sign of deterioration with use. A comparison of several different types of column is given below.

Supports—

Crushed firebrick causes severe tailing of hydrogen chloride.

Powdered PTFE[1,5] is soft and difficult to pack properly into the column.

Celite is quite satisfactory.

Stationary phases—

Silica gel[6,7,8] is satisfactory for chlorine but hydrogen chloride is absorbed.

Chloro-fluorocarbon polymer oils (Kel-F No. 10)[1,2,5] require a very long column and cause appreciable tailing of both hydrogen chloride and chlorine.

Hexadecane[3] gives excellent peak shape for hydrogen chloride and chlorine. It is completely unsatisfactory as it reacts with, and is destroyed by, chlorine.

Tri-tolyl phosphate causes chlorine to be eluted before hydrogen chloride. Minimal tailing of hydrogen chloride occurs and the column does not deteriorate; it has no obvious disadvantage.

SAMPLING VALVE AND CARRIER-GAS ARRANGEMENT—

Because of its availability a standard stainless-steel Perkin-Elmer gas-sampling valve was used. Slow corrosion makes re-facing necessary every few weeks, and a similar valve constructed of a high nickel alloy would probably be preferable.

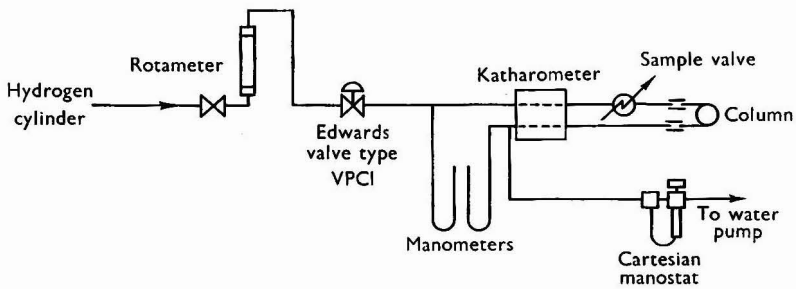

Fig. 1. Flow diagram for the chromatograph

To obtain accurately reproducible sample size it was found necessary to keep the pressure difference between sample gas and carrier gas to a few millimetres of mercury. This was achieved by using a water-pump to draw the carrier gas through the column. Accurate pressure control at both column inlet and outlet is essential as the detector is fairly sensitive to small pressure changes. An Edwards diaphragm valve VPC1 is used at the inlet, and an Edwards Cartesian manostat controls the outlet to the water-pump (Fig. 1).

The electrical circuit followed conventional lines. Power (12 volts) is supplied by a mains power-pack and the output signal is displayed on a 0 to 1-mV Honeywell recorder fitted with an integrator.

CALIBRATION AND ACCURACY—

The calibration was carried out with gas mixtures and also by sampling the pure gases under reduced pressure. The latter method, though simple, is limited in accuracy by leakage at the sample valve, particularly at low pressures.

A large glass syringe (capacity, 5 litres), fitted with a PTFE piston, was used for the preparation of the gas mixtures which were made up to an approximately known composition from cylinder gases and analysed with a gas burette. The calibration was carried out mainly with the two-component mixtures, nitrogen and chlorine, and nitrogen and hydrogen chloride, as the analysis of these mixtures with a gas burette is simpler and more accurate than that of mixtures containing both chlorine and hydrogen chloride. Checks on the three-component system agreed well with this calibration. The large syringe had the advantage that a large gas burette (100 ml) could be used and several samples could be taken from each gas mixture. However, this method of calibration was adopted largely because the syringe was already available and other equally satisfactory methods could probably be devised.

Nitrogen *plus* oxygen, and chlorine gave sharp peaks with very little tailing, and the calibration proved to be linear with peak height. Nitrogen and oxygen have slightly different calibration constants, therefore, as these two gases are eluted as one peak, a small correction is necessary for samples containing both these gases. Determination of accuracy is difficult because this varies with gas composition. However, over most of the range, the accuracy and reproducibility for nitrogen, oxygen or chlorine is about 0·25 per cent. The minimum amount of chlorine that can be detected is about 0·3 per cent. and the minimum amount of oxygen or nitrogen is less.

A calibration in terms of peak height would not be satisfactory for hydrogen chloride. The peak shows considerable tailing so that such a calibration would obviously be non-linear. For a given sample gas there are appreciable random fluctuations in the height of the hydrogen chloride peak, although the area remains approximately constant. A calibration based on peak area proved satisfactory as this was found to be closely linear. Traces of moisture, although increasing the tailing and having a pronounced effect on the peak height, have little effect on the area. Thus, provided the sample gas is above the dew-point, the presence of traces of moisture does not affect the area calibration. However, hydrogen chloride cannot be determined to the same degree of accuracy as the other components. The minimum amount that can be detected is about 2 per cent. and the accuracy over most of the composition range is about 1 to 2 per cent. There is a slight column-conditioning effect with hydrogen chloride and several samples are required each day before consistent values are obtained.

The linearity of the calibration proved to be a great advantage as a single sample of each pure gas is sufficient to check the calibration. If all of the operating conditions are kept exactly constant the calibration does not change appreciably with time. However, in practice the calibration was found to drift by a few per cent., principally owing to changes in the detector block temperature. No special precautions were taken to eliminate this effect as it proved simpler to check the calibration every few hours.

Under the conditions specified above, the retention times measured to peak maximum are given in Table I.

TABLE I

RETENTION TIMES OF THE GASES

Component	Retention time, minutes	Retention time, relative to air
Nitrogen + oxygen	0·262	1·0
Chlorine	0·354	1·35
Hydrogen chloride	0·67	2·56

The time required for a complete analysis is about 1½ minutes.

Although this instrument was developed for a kinetic study of the catalytic oxidation of hydrogen chloride to chlorine, it could, with slight modification, be applicable to the study of organic chlorination reactions in which the analysis of gases containing wet chlorine and hydrogen chloride can frequently arise.

A chromatogram for a typical gas mixture is shown in Fig. 2.

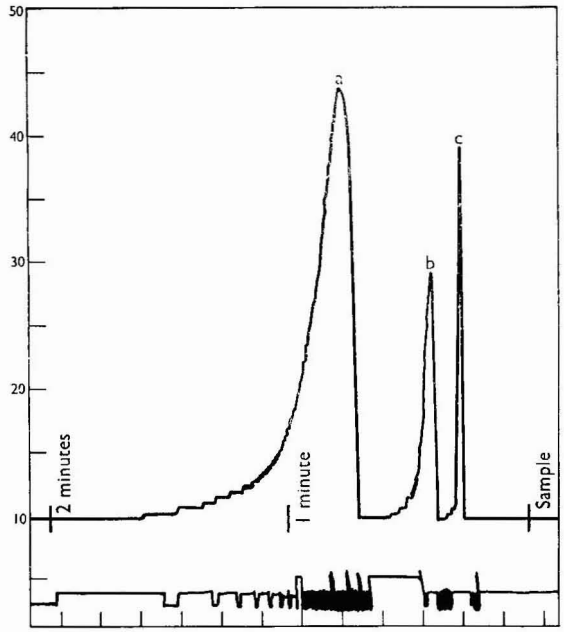

Fig. 2. Chromatogram for typical gas mixture; peak a, hydrogen chloride, 69·0 per cent.; peak b, chlorine, 18·2 per cent.; peak c, oxygen, 12·8 per cent.

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Book Reviews

DEVELOPMENTS IN APPLIED SPECTROSCOPY. Volume 4. Proceedings of the fifteenth Annual Mid-America Spectroscopy Symposium held in Chicago, Illinois, June 2–5, 1964. Edited by ELWIN N. DAVIS. Pp. xii + 546. New York: Plenum Press. 1965. Price $18.50.

The range of techniques covered by the 45 papers in this volume, out of the 110 papers presented at the Symposium, is too wide to allow for any detailed comment. The papers cover X-ray spectroscopy (pp. 3 to 117; 9 papers); infrared and Raman spectroscopy (pp. 121 to 212; 8 papers), ultraviolet and visible spectroscopy (pp. 215 to 251; 4 papers), gas chromatography (pp. 255 to 402; 11 papers) and emission, flame and atomic-absorption spectroscopy (pp. 419 to 546; 12 papers), together with a single paper on nuclear magnetic resonance spectroscopy.

Individual papers range in scope from rather trivial instrumental or practical matters to detailed instrumental design and construction, and from fairly obvious applications of the various techniques to extensive reports on novel analytical problems. Of the longer papers, one on soft and very soft X-ray fluorescence analysis, and another on gas-chromatography methods for the study of controlled-temperature polymer degradation may be noted. There are good discussions,

with theoretical background, of the absorbance ratio method for the infrared analysis of multi-component systems, and on diffuse reflectance spectroscopy. The odder topics include a paper on the study of the cathodo-luminescence of minerals in the petrographic microscope, and another on the gas-chromatographic analysis of gaseous contaminants from space-craft materials.

As with previous volumes in this series, the main value of the present book will be to reveal the way in which instrumental methods of analysis, at their contemporary level of design and performance, are being applied to the solution of new analytical problems. G. H. BEAVEN

ABSORPTION SPECTRA OF MINOR BASES. THEIR NUCLEOSIDES, NUCLEOTIDES AND SELECTED OLIGONUCLEOTIDES. By TAT'YANA VLADIMIROVNA VENKSTERN and ALEKSANDR ALEKSANDROVICH BAEV. Authorised translation from the Russian. Pp. vi + 86. New York: Plenum Press Data Division. 1965. Price $10.50.

In a brief introductory section the authors of this little book point out that the minor bases, already known to occur in very small proportions in ribonucleic (RNA) and deoxyribonucleic (DNA) acids, make larger contributions to the nucleotide sequences of the transfer or *s*-RNA's of much lower molecular weight, which have the function of transporting amino-acids to the ribosomes for the process of protein synthesis. The remainder of the book provides the spectrophotometric data required for the identification and determination of these minor bases and their nucleosides and nucleotides. These data take the form of fairly large (15 × 15 cm) spectra measured in 0·1N hydrochloric acid, 0·1N potassium hydroxide and after bromination in acid solution, all at the same concentration. The spectra of the brominated products assist in distinguishing compounds of the adenyl series, which remain largely unaltered, from those of the uracyl and other series of base derivatives, which are grossly altered by bromination.

The spectra are accompanied by brief tables of $\lambda_{max.}$, $\epsilon_{max.}$ and intensity ratios at selected pairs of wavelengths, all taken from the literature (119 references are listed). The minor bases, and their nucleotides and nucleosides, comprise the methylated derivatives of adenine, guanine, uracil and cytosine, together with pseudo-uridine, some oligonucleotides and data on the major nucleotides of RNA and DNA.

In view of the labour involved in obtaining reasonably pure reference samples of the minor base derivatives for the determination of standard spectra, the present compilation may be expected to have considerable practical value for workers in this particular field of biochemical analysis.

G. H. BEAVEN

POLAROGRAPHIC TECHNIQUES. By LOUIS MEITES. Second Edition. Pp. xviii + 752. New York, London and Sydney: Interscience Publishers, a division of John Wiley & Sons. 1965. Price 147s.

This revised edition of Professor Meites' well known introductory monograph on the theory and practice of polarography, the first edition of which appeared 10 years ago (*Analyst*, **1955**, **80**, 779), is virtually a new book. Every paragraph has been re-written and, in order to cover recent developments in polarography, the text has been extended to three times its original length.

The book follows the original plan, but fresh material has been introduced into every section. The section on commercially available polarographs now contains useful tables comparing, in some detail, the relative merits of 9 American, 1 Czechoslovakian, 2 Danish, 1 German, 1 Swiss and 3 Japanese (but no British) instruments. The practical exercises that formerly appeared at the end of each chapter have been omitted, but the appendix on trouble-shooting in polarographic circuits has been enlarged to include additional practical information.

The newer polarographic techniques and electrode systems, which were not discussed previously, are now surveyed and adequate references for further reading provided. It is, however, unfortunate that British made, square-wave, pulse and derivative-pulse polarographs should be dismissed as being too expensive and too unreliable for routine use.

The polarography of organic compounds is, as in the first edition, not discussed at length, but an appendix listing the polarographic characteristics of 600 representative organic compounds has been added.

The new edition can be strongly recommended as an introduction to modern polarography and to allied techniques, such as solid and stationary-electrode voltammetry, amperometry, chronopotentiometry, coulometry and stripping analysis. Polarographers, who possess a copy of the first edition, will certainly want a copy of this fuller and more up-to-date volume.

J. E. PAGE

TECHNIQUES OF OSCILLOGRAPHIC POLAROGRAPHY. By ROBERT KALVODA. Second Edition. Pp. 214. Amsterdam, London and New York: Elsevier Publishing Company. 1965. Price 60s.

This book is somewhat difficult to place in so far as its intended audience is concerned. In the introduction, the principles of oscillographic polarography are covered with the expertise that one would expect from Dr. Kalvoda, but the detailed instructions on the use of a particular commercial instrument appear to have little place in a work of this type.

The second chapter of the book, which is by far the largest of the four chapters, is devoted to the analytical applications of oscillo-polarography, and it is unlikely that the polarograms shown and the results quoted would persuade an analyst, used to the analytically biased polarographic techniques in use in the Western world, to consider oscillo-polarography as a very useful technique. For example, a copper concentration "as small as 10^{-6} M" is used as an example of the high sensitivity obtainable, and later zinc "as small as $1{\cdot}5 \times 10^{-4}$ M." I was disappointed to see an author of Dr. Kalvoda's standing still suggesting the use of polarography as a normal method for determining alkali metals and alkaline earths, and the section on aluminium makes no mention of the Willard and Dean Solochrome-violet reaction.

Section 3 on practical exercises begins with some very elementary work indeed, but the descriptions of the use of the technique for the study of reaction kinetics and capacity effects, where the method comes into its own, are good. Section 4 on the maintenance of apparatus and construction of auxiliary circuits is only 5 pages long, and contains far too little information to help the chemist, who does not specialise in electronics, whilst containing nothing that the electronics engineer would not have learned in the early stages of his course.

Some 263 references are given, by far the majority of which are, as one would expect, by Czech authors. The English in places shows signs of its continental origin, and this is probably the reason for the rather strange statement on page 47 that "if the polarograph is connected whilst nitrogen is being passed through the cell, the capillary drops irregularly." The book, on the whole, is well produced and presented; the only defect which I detected was an instance of faulty typesetting in a table heading on page 32. R. C. ROONEY

MÖSSBAUER EFFECT METHODOLOGY. Volume 1. Proceedings of the First Symposium on Mössbauer Effect Methodology. New York City, January 26th, 1965. Edited by IRWIN J. GRUVERMAN. Pp. viii + 200. New York: Plenum Press. 1965. Price $12.50.

This book records the proceedings of the First Symposium on Mössbauer Effect Methodology, which was held at the Sheraton Atlantic Hotel in New York on January 26th, 1965. It must be said immediately that the editor must be congratulated on the speed of the production of the book, which has taken less than a year to produce.

The papers given in the symposium review the state of the art (at that time), in terms of apparatus and instrument systems, and applications together with the ancillaries needed to produce environments for use in Mössbauer studies.

With the expansion of the applications of the Mössbauer effect, into a number of scientific research fields, there is a need for the complete description of the instrumentation systems, especially the transducer parts. These descriptions usually have been given as cryptic messages by a relatively small group of physicists, and in journals to which the inexperienced, or chemists, do not have easy access. This book answers many of these problems, and Section II on Spectrometers is to be recommended. Other problems that are discussed are the data-handling aspects, including computation of Mössbauer spectra.

Section IV is on environment ancillaries, especially the application of superconducting magnets to the Mössbauer effect, surely a field which will undergo very rapid development.

This book is, therefore, to be recommended most highly to any of us who have an interest in Mössbauer spectroscopy, although for its size it is expensive. One major fault of the book is the gross printing error in Chapter 1, where the heading on each page should read "Recent Applications to Chemical Problems," not "Recent Applications to Clinical Problems." G. NICKLESS

METALLKUNDLICHE ANALYSE: ZUSAMMENSETZUNG, STRUKTUR UND HABITUS DER PHASEN IN HETEROGEN LEGIERUNGEN. By Professor D. phil. WALTER KOCH and Dipl.-Physikerin. HELGA KOLBE-ROHDE. Pp. 497. Dusseldorf: Verlag Stahleisen, M.B.H.; Verlag Chemie, G.M.B.H. 1965. Price DM 135.

"Metallkundlich" has no exact English equivalent, but the scope of this book is defined by the sub-title as the determination of the composition, structure and appearance of the phases in

heterogeneous alloys. As long ago as 1938 a special laboratory was set up by Friedrich Krupp in Essen to study this problem, and the author was its leader for some years; he has since continued the work at the Max Planck Institut für Eisenforschung in Stuttgart.

While the spectrograph and, nowadays, the micro-probe analyser are powerful tools in phase identification they are subject to strict limitations: the spectrograph cannot resolve anything less than about 20 μ in extent and X-ray fluorescence does not encompass the lightest elements. Although Dr. Koch and his co-workers use these techniques where appropriate, the greater part of the work is concerned with chemical and electro-chemical means of isolating the phases (oxides, carbides, nitrides, silicates, sulphides and intermetallic compounds) present in ferrous alloys, and with their subsequent identification. Non-ferrous alloys are only briefly considered, and indeed this field seems to have been little studied.

The second part of the work consists of detailed tables of the properties of more than fifty phases occurring in ferrous and a few non-ferrous alloys; these are accompanied by photomicrographs and reproductions of X-ray diffraction patterns. This is undoubtedly the finest compilation of its kind in the literature, and does not require a knowledge of German for its use. It must, however, have been expensive to produce, and probably accounts for the high price of the book (about £12).

This is likely to be the standard work on the subject for many years to come, and should be in the library of every research laboratory concerned with the structure of iron, steel and ferrous alloys. G. M. HOLMES

L'ANALYSE CHIMIQUE ET PHYSICO-CHIMIQUE DE L'EAU. EAUX NATURELLES—EAUX USEES. By J. RODIER. Third Edition. Pp. xiv + 412. Paris: Dunod. 1966. Price F 72.

Water is analysed chemically in order to determine its suitability for a variety of purposes, as well as to evaluate its content of polluting matter; in this respect the chemical results supplement information provided by the bacteriological examination which is necessary before the microbial quality of a water may be ascertained. Dr. Rodier's first book on water analysis appeared in 1950. Most of it was re-written in its second edition in 1960. In this third edition considerable revision has taken place and more specific methods of analysis have been introduced. Radioactive methods of examining waters and limits for the radioactive content of the elements now occupy a large part of the book.

The book bears many of the marks of having been written from years of practice and experience, especially the chapter dealing with the preliminaries of sampling and instructions for labelling and for tracing underground water movement. The chapter on the interpretation of results is especially welcome and succeeds in being comprehensive without verbosity. The book is all-embracing in that it includes methods of analysis of all anions and cations commonly found in natural waters, besides 15 elements, including free cyanide, that are regarded as toxic, and those that are described as undesirable.

An allowance of 4 pages to the composition of buffer solutions and 22 pages to radioactivity is, in a book of this kind, excessive and quite out of proportion to the 40 pages allotted to the whole of the subject of waste-water analysis, which is almost tagged on as an afterthought. It is difficult to know where to leave off when one sets out to describe the use of instruments in water analysis, but if instruments such as the polarograph are mentioned repeatedly the reader should at least be given some indication of the circumstances in which they would offer advantages over the more traditional, colorimetric or titrimetric methods of analysis. The reviewer found the publisher's habit of omitting page numbers from pages that begin a chapter most exasperating; in one section of the book there are 5 unnumbered pages in sequence. But these are minor defects in a book that is well produced and laid out, comprehensive in its treatment of the subject of the physical and chemical examination of water and superbly illustrated by well drawn diagrams and carefully arranged tables. S. H. JENKINS

THE IDENTIFICATION OF ORGANIC COMPOUNDS. A MANUAL OF QUALITATIVE AND QUANTITATIVE METHODS. Sixth Edition (Third English Edition). By STIG VIEBEL, Dr.Phil. Pp. xvi + 452. Copenhagen: G.E.C. Gad Publisher. 1966.

One's first impression of this book is the size, and the absence of the tables usually considered an essential feature of such works. Larger by far than "Clarke," twice as large as "Campbell," three times as long as "Linstead and Weedon"—and these consist largely of tables! There are two main reasons for the size. Firstly, a quarter of the book is devoted to quantitative analysis

of the various functional groups, and secondly, the book was written for students at Copenhagen where far more laboratory time is devoted to qualitative analysis than in most English schools of chemistry. (The preface says 40 working days each of 5 hours; also in the final test the student has six 6-hour days to identify two substances.)

Chapter I (18 pages) deals with Purity, and includes methods for the determination of melting and boiling points (Siwoloboff's method is described, wrongly, and the gentleman not mentioned by name), refractive index, density, etc., and also deals with chromatography and ion exchange.

Chapter II, the Elements (35 pages), deals with the detection and determination of the commonly encountered elements, including fluorine. Active hydrogen (Zerevitinof) and active oxygen are discussed briefly with no practical details but with references to the literature.

Chapter III (9 pages) deals with preliminary examination, including taste, smell and the usual solubility tests, and concludes with half a page of references to works on absorption spectra.

The remaining 360 pages are occupied by Chapter IV, "Detection and Estimation of Functional Groups," and is completely comprehensive. There are 23 sections, and in addition to the common groups the following each have a section to themselves: Nitriles and Isocyanides, Diazo Compounds, Azo Compounds, Azoxy Compounds, Hydrazine, Nitroso Compounds, Substituted Hydroxylamines, The Triple Bond, etc.

Each section is further sub-divided and, under "20 Sulphur-containing Substances," we meet mercaptans, mustard oils, thioureas and thio-ethers, as well as the more common compounds. There are plenty of derivatives for each class of compound, *e.g.*, for carboxylic acids details are given for *S*-benzylisothiuronium salts, methyl esters (with boron trifluoride - methanol reagent), *p*-bromophenacyl esters and *p*-nitrobenzyl esters, amides, anilides, toluidides and hydrazides.

References to the literature are liberally scattered through the text and mistakes, as would be expected for a sixth edition, are few. An experienced student should, with care and plenty of time, be able to identify almost any compound under the sun by means of this book.

Can we, however, afford to spend so much time on the classical methods of analysis of any but the simpler types of compound? Would any examiner expect a candidate to identify, say, an isocyanide or unsymmetrical hydrazine by these methods in a single practical examination? How would one use this approach when examining, say, an alkaloid, a modern synthetic analgesic, a steroid hormone, or even caffeine or methyl-cellulose? The truth is—one would not! Students, because of the demands of other branches of the subject, have time for only the essentials of classical qualitative analysis, while professional analysts rarely have to identify an unknown substance without additional information as to origin and circumstances. Finally, any compound, except for the simplest, would be subjected directly to ultimate elemental analysis followed by nuclear magnetic resonance, ultraviolet and infrared spectroscopy, and all the other instrumental aids now at our disposal. Classical qualitative analysis may not be quite as obsolete as the old "5-radical inorganic mixtures" of our youth but it *is* obsolescent.

However, factual chemistry is still learned at the bench, and the properties of the functional groups are best studied through the simplest representatives of each class. A student who has the time and application to study this book, not only at the bench, but also at the fireside, will certainly learn a lot of real chemistry, and the quantitative determinations are not only of interest to students but of great value to practising analysts. LOUIS K. SHARP

Errata

JANUARY (1966) ISSUE, p. 28, 6th line. *For "approximately 0·6* N *(Procedure A)" read "approximately* 0·04 N *(Procedure A)."*

AUGUST (1966) ISSUE, p. 496, 3rd. line. *For* "tin(II) thiocynate" *read* "sodium thiocyanate."