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The Determination of Iron(II) Oxide in Silicate and Refractory Materials

Part I. A Review*

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THE determination of iron(II) oxide presents a number of difficulties. Two of special importance are the method of bringing the sample into solution and the prevention of oxidation of iron(II) in the process. With materials that are readily soluble in sulphuric or hydrochloric acids, the determination should present no great difficulty because it is possible to carry out decomposition in glass apparatus, and maintain an atmosphere of a suitable gas (carbon dioxide or nitrogen) to prevent oxidation. This is not so with materials resistant to a simple acid attack, and some procedures designed to overcome the difficulties associated with the determination of iron(II) oxide in such materials are dealt with here.

A third difficulty inherent in all methods is the possibility of the oxidation of iron(II) oxide during sample preparation, especially if grinding in air is used. According to Hillebrand et al.,[1] if on simple crushing of the material a product is obtained that leaves little or no residue when gently boiled with hydrofluoric acid, then no further preparation of the sample is needed. If, on the other hand, there is considerable residue, then the sample should be ground under alcohol only long enough to yield a powder that leaves little or no residue.

The main characteristics of each of the methods considered here are used as the basis for the arbitrary headings under which the methods are discussed. Usually the differences in the methods relate to the initial decomposition stage.

ACID DECOMPOSITION IN A SEALED TUBE

The method described by Mitscherlich[2,3] involves the decomposition of the material by heating with sulphuric acid in a sealed glass tube at temperatures up to 200° C. In this procedure, exclusion of oxygen (air) should present no difficulty, but complete dissolution of some of the materials is not always achieved. In Hillebrand's modification[1] of the original method, a weaker sulphuric acid solution is used and the air in the tube is replaced with carbon dioxide.

The determination of iron(II) oxide is subject to errors arising from the presence of sulphides and carbonaceous matter. With sulphides the error can be quite large because of the reduction of the iron(III) that may be present. Pyrite is normally resistant to boiling dilute acids, but under the conditions of increased temperature and pressure that prevail in the sealed-tube method, and in the presence of iron(III), it will go into solution with reduction of the iron(III) to iron(II). These difficulties with the method were fully discussed by Hillebrand, who concluded that, in the absence of sulphides and when the material is fully decomposed, the sealed-tube method is almost ideal; otherwise its application is limited.

* Reprints of this paper will be available shortly. For details see Summaries in advertisement pages.

For the decomposition of resistant silicate minerals, Riley[4] proposed a method that can be regarded as an extension of the sealed-tube method of decomposition. The resistant mineral is decomposed with hydrofluoric and perchloric acids in a Teflon bomb, which is placed in a steel vessel fitted with safety valves and containing water. The apparatus is maintained at 150° C in an oil-bath for 3 to 4 hours. Riley and Williams[5] suggested that this method of decomposition is suitable for the micro determination of iron(II) oxide in resistant minerals.

ACID DECOMPOSITION IN AN INERT ATMOSPHERE

This method generally (but not always) implies the use of hydrofluoric acid to decompose the sample. As originally devised by Cooke[6] it involved decomposition of the sample with hydrofluoric and sulphuric acids in a platinum crucible heated in a specially designed water-bath. The crucible was surrounded by an atmosphere of carbon dioxide to prevent aerial oxidation of the iron(II) during decomposition of the sample. The iron(II) was titrated with permanganate.

One of the difficulties associated with Cooke's method is the tendency for iron(II) to be readily oxidised in air in hydrofluoric acid solution, and this can occur during the titration. Another is the oxidation of bivalent manganese by permanganate in the presence of hydrofluoric acid. For reliable results the latter difficulty must be overcome by removal of the hydrofluoric acid or the elimination of its effect. Barnebey[7] used boric acid to eliminate the effect of the hydrofluoric acid by the formation of undissociated fluoroboric acid. Pratt[8] dispensed with Cooke's apparatus, and, to prevent oxidation of iron(II), carried out the decomposition in a large platinum crucible fitted with a perforated cover under which carbon dioxide was introduced. Hillebrand modified the method by introducing carbon dioxide only at the beginning, relying upon the evolution of steam from the boiling acid mixture to prevent entry of air during the dissolution of the sample.

Various other modifications designed to prevent oxidation have been proposed. Treadwell[9] carried out the decomposition in a platinum crucible supported in a lead box in which an atmosphere of carbon dioxide was maintained. The box was heated in a paraffin-bath, and the temperature at the end of the decomposition period could be raised to 120° C to remove hydrofluoric acid and so prevent it from interfering in the permanganate titration. Barnebey[10] dispensed with carbon dioxide as used in the Cooke apparatus and relied upon the generation of steam to displace air.

Instead of permanganate, Sarver[11] used dichromate to titrate iron(II), with diphenylamine as internal indicator and a platinum crucible fitted with a tight transparent bakelite lid for the sample decomposition. The lid was provided with a bakelite tube and funnel to enable the carbon dioxide and acid to be introduced into the crucible. The decomposition procedure used was as in Pratt's method. After cooling in a current of carbon dioxide, a measured excess of dichromate was added; the solution was then transferred to a beaker containing boric acid, and the excess of dichromate back-titrated with iron(II) sulphate with diphenylamine as indicator. Schollenberger[12] applied the technique developed by Sarver.

Soule[13] dispensed with the use of platinum ware and carried out the decomposition with hydrofluoric and sulphuric acids in a Pyrex flask in a current of carbon dioxide. He applied a correction for the glass dissolved by conducting a blank determination on ammonium iron(II) sulphate. In a later paper[14] he discussed possible errors in the determination arising from the presence of arsenic in the glass, and claimed that these could be avoided by titrating with cerium(IV) sulphate instead of permanganate.

In considering the effects of sulphides in methods in which hydrofluoric acid decomposition is used, Hillebrand suggested that hydrogen sulphide evolved from soluble sulphides would probably be expelled without reducing iron(III), but with sulphides containing iron the iron(II) oxide content would be in error by the amount of iron(II) they contained. This would be especially significant for pyrrhotite. The insoluble sulphide, pyrite, is resistant to attack by hydrofluoric acid in the absence of air, but Stokes[15] showed that in the presence of iron(III) it is attacked, with the oxidation of the sulphide and reduction of iron(III). The extent of this reaction depends on the amount of iron(III) present, and on the degree of fineness of the pyrite. Hillebrand considered the influence of pyrite on the iron(II) oxide determination in most rocks to be negligible, a view that was shared by Dittler.[16] The latter maintained that carbon dioxide should, however, be introduced into the bottom of the crucible to remove any trace of hydrogen sulphide that might be formed.

The effect of organic matter can be minimised if the titration of iron(II) is carried out with dichromate instead of permanganate, because, according to Sarver,[11] dichromate reacts much less readily with organic matter.

Densem,[17] in modifying Pratt's method, developed a rather complex apparatus designed to eliminate air during sample decomposition and during the subsequent titration of iron(II). This apparatus was subsequently modified by Harris.[18]

In an attempt to overcome oxidation during sample decomposition, Smirnov and Aidinyan[19] proposed dissolution of the sample with hydrofluoric and sulphuric acids while it was under a layer of toluene. They claimed that this could be achieved without "bumping" and without forming reaction products that would affect a permanganate titration of iron(II).

Modification of procedures in which hydrofluoric and sulphuric acids are used for sample decomposition has been made to enable micro or semi-micro determinations to be made of iron(II) oxide. Titrimetric methods were developed by Shioiri and Mitsui,[20] Das Gupta[21] and Meyrowitz.[22] Riley and Williams[5] used a spectrophotometric method in which the sample is decomposed in a stoppered Teflon tube heated in a boiling water-bath, the iron(II) being determined with dipyridyl. In a similar procedure, Shapiro[23] decomposed the sample with hydrofluoric and sulphuric acids in the presence of o-phenanthroline in a small plastic bottle. This reagent was designed to react with the iron(II) as it was released, with the formation of the iron(II) - o-phenanthroline complex, and so minimise aerial oxidation. The fact that heating affects the stability of the colour seems to militate against obtaining reliable results.

Nicholls[24] determined iron(II) oxide in carbonaceous shales by decomposing the sample in sulphuric acid - hydrofluoric acid solution, adding boric acid, and pouring the resulting solution into hydrochloric acid containing iodine monochloride. The iodine liberated was extracted into carbon tetrachloride and titrated with potassium iodate. It is claimed that the method is applicable in the presence of organic matter equivalent to up to 4 per cent. of carbon.

The decomposition of materials that cannot be brought into solution with hydrofluoric acid has been achieved with phosphoric acid. Konopicky and Caesar[25] determined iron(II) oxide in chrome ore by decomposing the sample by heating it with phosphoric acid in an atmosphere of carbon dioxide, the resulting solution being titrated with permanganate. For the determination of iron(II) oxide in ferrites Kleinert and Funke[26] applied a similar method in which nitrogen was used to prevent oxidation.

Clemency and Hagner[27] departed from the general practice of determining iron(III) as the difference between iron(II) and total iron; instead they determined it by using coulometrically generated titanium(III) ion as titrant, with an automatic spectrophotometric apparatus devised by Malmstadt and Roberts.[28] For the determination of iron(III) in the rocks G–1 and W–1, dissolution of the sample was achieved by heating it to just below the boiling-point with hydrofluoric and concentrated sulphuric acids. Initially, this decomposition was carried out in a specially constructed box in which an inert atmosphere could be maintained. The results were erratic and always low, but this was evidently not because of the incomplete dissolution of the sample, as a total-iron analysis showed a virtually complete recovery of iron. The low results were presumed to be caused by reduction of iron(III), perhaps by some constituent of the rocks. Subsequently the decomposition was carried out in a covered crucible. The heating time proved critical, 3 to 4 minutes being the optimum. With heating times in the range of 4 to 5 minutes results were low and erratic, and reduction of iron(III) appeared to occur. Heating times in excess of 5 minutes gave gradually increasing iron(III) values, presumably because of the predominance of air oxidation over the reduction observed.

Evidently, therefore, in this instance the problems associated with the determination of iron(II) are not overcome by carrying out a determination of iron(III) instead.

ACID DECOMPOSITION IN THE PRESENCE OF AN OXIDANT

To overcome difficulties with iron(II) oxide determinations arising from aerial oxidation, many methods have been proposed for decomposing the sample with acids in the presence of an oxidising agent. A known amount of oxidant is used, which oxidises the iron(II) as it is brought into solution, and the excess of oxidant, or the product of its reduction, is then determined by a suitable (usually titrimetric) method. This was suggested (but never tried) by Hackl,[29] who proposed decomposing the sample with hydrofluoric acid in the presence

of potassium dichromate and subsequently titrating the excess of dichromate. The first practical application of such a procedure was carried out by Shein,[30] who used it for the analysis of iron(II) oxide in chromite. He decomposed the sample by heating (360° to 380° C) with phosphoric and sulphuric acids in the presence of a weighed amount of vanadium pentoxide, and then titrating the excess of vanadate with iron(II) sulphate.

Modifications of Shein's method have been used by many other workers for determining iron(II) oxide, generally in chromite and chrome - magnesite materials. These materials are not decomposed by hydrofluoric and sulphuric acids but can be brought into solution with phosphoric acid.[31 to 38]

Most of the applications of acid decomposition in the presence of an oxidant reported in the literature are based on the use of phosphoric acid and vanadium pentoxide, as in Shein's method. However, other oxidants have been used. Goswami[39] claimed that a considerable amount (16 to 20 per cent.) of vanadium pentoxide underwent decomposition during sample dissolution, and proposed the use of cerium(IV) sulphate as oxidant. Although cerium(IV) sulphate undergoes some decomposition, Goswami obtained consistent and reproducible results for the determination of iron(II) oxide in chromite. The volume of iron(II) sulphate required for the cerium(IV) sulphate added was multiplied by a derived factor, which presumably allows for the slight decomposition of cerium(IV) sulphate under the conditions used. It must be assumed that the extent of decomposition is reproducible. Ingamells[40] based his method on the stability of both bivalent and tervalent manganese in phosphoric acid - pyrophosphate mixtures. A measured excess of potassium permanganate is incorporated in the phosphoric acid reagent containing bivalent manganese, which is oxidised by the permanganate to tervalent manganese. After addition and dissolution of the sample, the excess of tervalent manganese is titrated with iron(II) sulphate.

Nagato[41] developed a spectrophotometric method based on the decomposition of the sample with phosphoric acid in the presence of manganese(III) oxide; the optical density of the resulting solution containing tervalent manganese was read at 525 mμ.

More recently Cheng[42] used an oxidant prepared from ammonium cerium(IV) nitrate treated with phosphoric acid. He termed the resulting product "phosphatocerate." For the determination of iron(II) oxide in ferrites he decomposed the sample with phosphoric acid containing a measured amount of the phosphatocerate reagent and determined the excess of reagent by titration with iron(II) sulphate. Cheng claimed that phosphatocerate was stable at the temperature and for the time recommended for sample decomposition.

For samples decomposed by cold hydrochloric acid or hydrochloric - hydrofluoric acids mixture, Hey[43] used iodine monochloride as the oxidant. The iodine formed by reaction with iron(II) was determined by titration with potassium iodate.

Ishibashi and Kusaka[44] decomposed silicates by heating with hydrofluoric and sulphuric acids in the presence of ammonium metavanadate, and titrated the excess of vanadate with iron(II) sulphate. Wilson[45] used the same approach but carried out the decomposition at room temperature, extended periods being required for the dissolution of some samples. Under these conditions vanadate was considered to be stable in hydrofluoric acid. Later,[46] he adapted the method for micro-scale work by titration, as well as by introducing a colorimetric method based on the regeneration at pH 5 of iron(II) when the reaction $V^{4+} + Fe^{3+} \rightleftharpoons V^{5+} + Fe^{2+}$ proceeds to the right. The removal of iron(II) by the formation of a complex with dipyridyl assisted the reaction to proceed in the desired direction. The colour developed was then measured spectrophotometrically. Wilson's volumetric method has been adapted by Jackson[47] on a semi-micro scale for determining iron(II) in the ash and slag from pulverised-fuel boilers.

Potassium dichromate as the *in situ* oxidant was used by Reichen and Fahey[48] for determining iron(II) oxide in rocks and minerals. Decomposition was effected with hydrofluoric and sulphuric acids, at a temperature between 65° and 70° C. However, under these conditions dichromate was found to decompose slightly by reaction with hydrofluoric acid. This effect could be minimised by adding an iron(III) salt. The destruction of dichromate appeared to be proportional to the amount in excess, and so a correction based on the simple subtraction of a blank could not be applied. The alternative procedure was therefore adopted of multiplying the volume of iron(II) solution used to titrate the excess dichromate by the ratio of the volume of dichromate to iron(II) solution used in the blank determination.

The determination of iron(II) oxide in materials that can be decomposed by acids in the presence of an oxidant, although attractive in principle, may in practice give rise to a

number of difficulties, the most important of which is the stability of the oxidant under the conditions used. If decomposition of oxidant occurs it must be reproducible, and therefore rigid control of the conditions is required to ensure the same degree of decomposition in both sample and blank determinations. In addition, the possible effect of other constituents in the sample on the decomposition of an oxidant should not be overlooked. Some workers have tried to eliminate the possibility of oxidant decomposition by carrying out the acid treatment at room temperature. The disadvantage here is that while some materials are readily dissolved, others require long periods of digestion. A possible advantage is that oxidation of organic matter, such as carbonaceous material, may be less likely at room temperature.

TABLE I

SUMMARY OF METHODS IN WHICH ACID DECOMPOSITION IS USED IN THE PRESENCE OF ADDED OXIDANT

Material	Acid decomposition	Oxidant	Method of determination	Reference
Silicates, including rocks and minerals	Heating with HF - H_2SO_4	$K_2Cr_2O_7$	Titration of excess dichromate	Hackl[29]
	HCl or HCl - HF at room temperature	ICl	Iodine formed titrated with potassium iodate	Hey[43]
	Heating with HF - H_2SO_4	V_2O_5	Excess vanadate titrated with iron(II) sulphate	Ishibashi and Kusaka[44]
	HF at room temperature	NH_4VO_3	Excess vanadate titrated with iron(II) sulphate	Wilson[45]
	HF at room temperature	NH_4VO_3	Micro titration of excess vanadate, and also spectrophotometric	Wilson[46]
	Heating with H_3PO_4 - NaH_2PO_4	Tervalent manganese	Titration of excess manganese with iron(II) sulphate	Ingamells[40]
	Heating with HF - H_2SO_4, 65° to 70° C	$K_2Cr_2O_7$	Titration of excess dichromate with iron(II) sulphate	Reichen and Fahey[48]
Chromite (and chrome-magnesite refractories)	Heating with H_3PO_4 - H_2SO_4, 360° to 380° C	V_2O_5	Excess vanadate titrated with iron(II) sulphate	Shein[80]
	H_3PO_4 - H_2SO_4 heated at 360° C	V_2O_5	Titration of excess vanadate	Samanta and Sen[36]
	Heating with H_3PO_4 - H_2SO_4	V_2O_5	Titration of excess vanadate	Balyuk and Mirakyan[81]
	Heating with H_3PO_4 - H_2SO_4	V_2O_5	V^{4+} formed titrated with permanganate	Nagaoka and Yamazaki[35]
	H_3PO_4 - H_2SO_4 heated at 290° to 300° C	Cerium (IV) sulphate	Excess cerium(IV) titrated with iron(II) sulphate	Goswami[39]
	H_3PO_4 - H_2SO_4	V_2O_5	Titration of excess vanadate	Sasuga and Iida[37]
	Heating with H_3PO_4 - H_2SO_4	V_2O_5	Amperometric titration of excess vanadate with iron(II) sulphate	Kondrakhina et al.[34]
	Heating with H_3PO_4	V_2O_5	Titration of excess vanadate	Dippel[32]
Ash and slags	HF at room temperature	$NaVO_3$	Excess vanadate titrated with iron(II) sulphate	Jackson[47]
Tourmaline	Heating with H_3PO_4 - H_2SO_4	V_2O_5	Titration of excess vanadate with iron(II) sulphate	Gekht and Putok[33]
Ferrites	H_3PO_4 at 250° C	Mn_2O_3	Spectrophotometric determination of excess tervalent manganese	Nagato[41]
	H_3PO_4 at 250° to 300° C	Phosphato-cerate	Excess cerate titrated with iron(II) sulphate	Cheng[42]
Transition metal oxides	H_2SO_4	V_2O_5	V^{4+} titrated with permanganate	Wickham and Whipple[38]

The use of phosphoric acid enables many refractory materials to be brought into solution, but at the temperatures used (up to 380° C) there may be extensive decomposition of the oxidant as well as oxidation of organic matter.

Another difficulty that can affect iron(II) oxide determinations is the presence of sulphides, which may result in consumption of oxidant and thus in high values for the iron(II) oxide content.

A summary of methods involving acid decomposition in the presence of an oxidant is given in Table I.

EVOLUTION METHOD

Seil[49] proposed a completely different approach to the determination of iron(II) oxide; he suggested decomposing the finely ground material in phosphoric and sulphuric acids in a stream of carbon dioxide, and passing the resulting vapours through absorption tubes containing a measured amount of standard dichromate solution. According to Seil, sulphur dioxide was evolved by reaction of iron(II) oxide and the acids. This reduced the dichromate, and the excess of dichromate could then be determined by titration. More recently Tikhomirova *et al.*[50] applied this method to the determination of iron(II) in chromium ores and slags. They thought that the reduction of dichromate was due to absorption of sulphur dioxide and phosphine formed by reaction of iron(II) oxide with the acid mixture. It is apparent that other substances such as sulphides and organic matter, which could result in the reduction of sulphuric or phosphoric acid with the evolution of the gases mentioned above, must be absent.

FUSION METHOD

For the decomposition of refractory silicates and the subsequent determination of iron(II) oxide Rowledge[51] investigated the application of a number of fluxes, the most satisfactory being a fluoroborate $(NaF)_2.B_2O_3$. He heated the sample which was mixed with the flux in a sealed glass tube at 900° C, dissolved the melt in dilute sulphuric acid and titrated the iron(II) with permanganate. He applied the method to refractory silicates such as staurolite, tourmaline, axinite and garnet, and compared the values obtained by decomposition in hydrofluoric and sulphuric acids with those obtained by using the fluoroborate fusion procedure. Because these refractory materials were only partly decomposed by the acid treatment, the iron(II) oxide contents were determined by weighing the undecomposed material and calculating from the weight of mineral decomposed (obtained by difference). This procedure may be acceptable for homogeneous minerals, but it is unsatisfactory because of doubt about the composition of the undecomposed material. The comparisons made by Rowledge are rather confusing. With tourmaline the results obtained by the two methods agreed; for garnet the fusion method gave a value of about 1 per cent. higher than that by acid decomposition, and for staurolite and axinite the method gave a value of about 1 per cent. lower.

In an attempt to apply the Rowledge fusion method on a micro scale, Hey[43] encountered difficulties arising from the oxidation of iron(II) by air enclosed in the fusion tube, as well as those arising during the slow process of dissolving the melt in sulphuric acid. He overcame the former by conducting the fusion in an evacuated tube, and the latter by dissolving the melt in an excess of iodine monochloride in hydrochloric acid in a stoppered flask and titrating the liberated iodine with potassium iodate.

By carrying out the fusion in a platinum boat placed in a silica tube at 950° C through which a current of carbon dioxide was passed, Groves[52] excluded the possibility of oxidation. The iron(II) was then determined by permanganate titration or, better still, by the iodine monochloride method of Hey. Groves regarded the combination of fusion in carbon dioxide, followed by dissolution of the melt in hydrochloric acid containing iodine monochloride, as probably the best method available for determining iron(II) oxide in refractory materials.

More recently Mikhailova *et al.*[53] used the method of fluoroborate fusion in an atmosphere of carbon dioxide as developed by Groves for determining both bivalent and tervalent iron in rocks that are difficult to decompose. The solution obtained by dissolving the melt in a mixture of 10 per cent. sulphuric acid and saturated sodium oxalate was subjected to polarographic reduction. The heights of the waves corresponding to Fe^{2+} and Fe^{3+} were measured and the content of each form calculated. Subsequently[54] the fusion was carried out in a platinum crucible into which carbon dioxide was introduced.

COMBUSTION METHOD

This is based on the oxidation of iron(II) oxide to iron(III) oxide when the former is heated in a current of oxygen. In the method originally developed by Shein,[55] the sample was heated in a current of dry oxygen or air in an electric furnace maintained at 1000° C. The water and carbon dioxide evolved were collected in an absorption tube and weighed. The oxygen used in the oxidation of iron(II) oxide was then calculated from the increase in weight of the ignited residue. In a later modification the sample was heated first in pure nitrogen to expel volatile matter; it was then weighed, and finally re-ignited in oxygen. The iron(II) oxide content could then be calculated from the increase in weight.

An investigation of Shein's method as applied to chrome ores by Sasuga and Iida[37] with a thermobalance showed that, for samples low in aluminium and magnesium, the increase in weight reached a maximum at 800° to 900° C and that the weight decreased at higher temperatures. They concluded that the temperature at which the weight increase is recorded must be changed according to the sample analysed.

De Wet and van Niekerk[56] adapted the method on a semi-micro scale for the analysis of iron(II) oxide in South African chromites. In a study of the rapid analysis of chromite and chrome ores, Dinnin[57] compared iron(II) oxide determinations made by the Shein combustion method, the Shein method with vanadium pentoxide as oxidant, and the evolution method of Seil.

In a different approach, Habashy[58] determined the iron(II) oxide content of material that is difficult to dissolve, by ignition at 1000° C in a limited, known volume of oxygen. He then calculated the iron(II) oxide content from the volume of oxygen used in the oxidation of iron(II) oxide to iron(III) oxide. He claimed that interferences could be overcome by applying suitable corrections.

CONCLUSION

The diversity of publications on the determination of iron(II) oxide in silicate and refractory materials is indicative of the extent of the problem, and it would be unwise to assume that it has been solved. There does not appear to be any one method applicable to all types of materials. All methods suffer from some disadvantages, but perhaps the one most closely approaching the ideal is that of Rowledge, as modified by Groves. However, its application to some materials would amount to the use of unnecessarily severe conditions for sample decomposition that could be readily achieved by some simpler method.

The extent of interference arising from the presence of organic matter in a sample would depend on the method of sample decomposition used. It could be quite serious with decomposition in the presence of an added oxidant, especially if heating were required. With other titrimetric methods the degree of interference would be dependent on the type of organic matter and the oxidising power of the titrant used for the iron(II).

With materials containing sulphides, particularly iron sulphides, errors in the determination of iron(II) oxide seem unavoidable. Their magnitude is dependent on several factors, such as type and amount of sulphide, the iron(III) content and the method of sample decomposition. Apart from the likely sources of errors mentioned above, interference with iron(II) oxide determination could occur because of the presence of oxidising substances such as manganese dioxide, which would lead to low results arising from the oxidation of iron(II) during sample dissolution. On the other hand, Hillebrand has cited the interference of tervalent vanadium, which would consume the titrant used for iron(II) and lead to high results. These two sources of error are unlikely to be of common occurrence.

However, given an awareness of the problems involved, and a knowledge of the range of methods available for the determination of iron(II) oxide, it should be possible to select the method best suited to the type and number of samples to be analysed.

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The Determination of Iron(II) Oxide in Silicate and Refractory Materials

Part II. A Semi-micro Titrimetric Method for Determining Iron(II) Oxide in Silicate Materials

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A description is given of an apparatus of simple construction that can be made from plastic, in which the decomposition of silicate materials by hydrofluoric acid can be carried out. The procedure for the titration of iron(II) with dichromate under conditions that eliminate the risk of oxidation is outlined.

A comparison has been made between dichromate titres obtained potentiometrically and with diphenylamine sulphonate as indicator. Errors associated with the latter in the titration of small amounts of iron(II) are discussed. The method of sample decomposition and titration of iron(II) has been assessed by determining the iron(II) oxide content of the diabase W–1, and has been applied to the determination of iron(II) oxide in slags from a boiler fired with pulverised fuel.

In Part 1 of this paper methods are reviewed for determining iron(II) oxide in refractory materials. A simplified analytical procedure comprising dissolution of the sample followed by direct titration of iron(II) is now considered. Its application is restricted to silicate materials that are decomposed by hydrofluoric acid.

The method described here was developed after consideration had been given to the methods of Wilson[1] and Reichen and Fahey,[2] both of which involve sample decomposition in the presence of an oxidant. The former method,[1] in which vanadate is used as added oxidant with subsequent colorimetric determination of iron(II), gives satisfactory results when complete decomposition of the sample can be achieved at room temperature. However, most of the samples studied decomposed only very slowly under these conditions. In the method proposed by Reichen and Fahey,[2] in which dichromate is used as oxidant with subsequent titration of the excess, reproducible results could not be obtained for the titration of dichromate in the blank determination and the method was, therefore, not examined further.

DEVELOPMENT OF THE METHOD

APPARATUS—

The apparatus for sample decomposition was required to be of simple construction, to have provision for maintaining an inert atmosphere, to be suited to operation at temperatures above room temperature and to be capable of being used as the titration vessel. The construction of an all-plastic apparatus satisfying most of these requirements is fully described under Experimental, and is illustrated in Fig. 1.

This apparatus is similar in application to that devised by Sarver[3] and further described by Schollenberger,[4] who used a platinum crucible, which enabled the solution to be boiled, for sample decomposition; an inert atmosphere was maintained by carbon dioxide introduced above the solution through a tube in a bakelite lid.

DICHROMATE TITRATION OF IRON(II)—

The titration of iron(II) with dichromate was examined in the presence of phosphoric acid, of hydrofluoric acid after addition of boric acid and phosphoric acid, and of hydrofluoric acid. In all titrations sulphuric acid was present and sodium diphenylamine sulphonate was used as indicator, the titrations being carried out in an atmosphere of nitrogen. The same dichromate titre was obtained each time. As the titration in hydrofluoric acid solution gave sharp and distinct end-points, removal of hydrofluoric acid by forming complexes with boric acid appeared to be unnecessary. Accordingly all subsequent titrations were performed
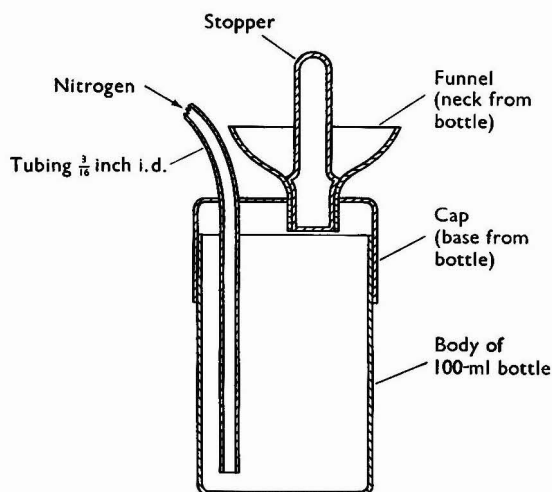
Fig. 1. Sample decomposition apparatus (poly-thene)

directly in solutions containing hydrofluoric acid. This acid acts in the same way as phosphoric acid in lowering the redox potential of the iron(II) - iron(III) system.

These observations confirm those of Schollenberger[4] who examined the titration of iron(II) with dichromate, with diphenylamine as indicator. He noted that the presence of hydrofluoric acid sharpened the end-point; and maintained that in the determination of iron(II) oxide in minerals it was preferable to perform a direct titration with dichromate, and that the addition of boric acid to inactivate the hydrofluoric acid used in the decomposition of a silicate should be omitted.

TITRATION OF DIFFERENT VOLUMES OF IRON(II) SOLUTION—

The next aspect studied was the effect of varying the amount of iron(II) titrated. Meyrowitz[5] observed that the reverse titration was disproportional. There was an apparent change in the normality of an iron(II) solution as the volume of dichromate was changed; the normality gradually increased as the volume of dichromate titrated increased. This disproportion was found to be more pronounced in large volumes (300 ml) than in small volumes (100 ml). To overcome this effect in an iron(II) oxide determination, a sample weight was chosen that required about the same volume of dichromate as was used in the standardisation of ammonium iron(II) sulphate.

TABLE I

TITRATION OF APPROXIMATELY 0·02 N IRON(II) SULPHATE WITH 0·02 N POTASSIUM DICHROMATE UNDER NITROGEN IN A HYDROFLUORIC - SULPHURIC ACID SOLUTION
(10 ml of 1 + 3 H_2SO_4 and 5 ml of HF diluted to 25 ml)

	Potentiometric titration		Diphenylamine sulphonate		Ferroin	
Iron(II) solution, ml	Potassium dichromate, ml	Apparent iron(II) normality	Potassium* dichromate, ml	Apparent iron(II) normality	Potassium† dichromate, ml	Apparent iron(II) normality
1·00	0·97	0·01940	1·01	0·02020	0·98	0·01960
2·00	1·95	0·01950	2·00	0·02000	1·96	0·01960
3·00	2·93	0·01953	2·95	0·01967	2·94	0·01960
4·00	3·91	0·01955	3·93	0·01965	3·93	0·01965
5·00	4·88	0·01952	4·89	0·01956	4·90	0·01960
6·00	5·85	0·01950	5·86	0·01953	5·88	0·01960
8·00	7·80	0·01950	7·80	0·01950	7·84	0·01960
10·00	9·76	0·01952	9·77	0·01954	9·80	0·01960

* Corrected for blank of 0·02 ml of 0·02 N potassium dichromate.
† Corrected for blank of 0·08 ml of 0·02 N potassium dichromate.

A similar effect has been reported by Rodden[6] and De Sesa[7] in the determination of small amounts of uranium, in which quadrivalent uranium is reacted with iron(III) ions, and the iron(II) ions formed are titrated in sulphuric acid - phosphoric acid solution with dichromate. No satisfactory explanation was offered for this disproportion. Toni,[8] in a study of the disproportion in uranium determinations, showed that the indicator, diphenylamine sulphonate, was responsible, and overcame the problem by determining the end-point potentiometrically.   Earlier, Kolthoff and Sarver[9] had discussed the possibility of side reactions occurring when diphenylamine was used as indicator in dichromate titrations.

In the present study the titration of iron(II) solutions in the presence of hydrofluoric acid with dichromate, with diphenylamine sulphonate as indicator, also proved to be disproportional for small amounts of iron(II).   In view of the findings of Toni, comparisons were made between titres obtained when the end-point was determined potentiometrically, with diphenylamine sulphonate, and with ferroin.   If the disproportion observed in titrations in which diphenylamine sulphonate was used arose from side reactions occurring on oxidation of this indicator, it was thought that this would not occur with ferroin with which the colour change on oxidation is caused by the oxidation of iron(II) to iron(III) within the o-phenanthroline complex.   The apparent iron(II) normality, as well as the dichromate titres, obtained for different volumes of iron(II) solution are shown in Table I.

The disproportion observed in the titration of small amounts of iron(II) with dichromate, with diphenylamine sulphonate as indicator, is not apparent when the end-point is detected potentiometrically or when ferroin is used as indicator.   In the latter case an apparently higher normality for the iron(II) solution was obtained.   It would appear that, as the potential of the iron(II) - iron(III) - o-phenanthroline system is about 1·06 volts, the indicator ferroin is not really suitable for use with dichromate as oxidant at the acidity used (3·6 N) when the chromium(III) - dichromate potential is about 1·11 volts (Smith and Richter[10]).   Because of this the solutions were possibly over-titrated in reaching the point of colour change of the indicator, which would account for the apparently higher normality of the iron(II) solution.   Disadvantages of ferroin as indicator are the large indicator blank and the gradual colour change from the orange of the iron(II) form through a virtually colourless stage to the blue of the iron(III) form.

The change in potential during a potentiometric titration of iron(II) with dichromate in the presence of hydrofluoric acid is particularly large and abrupt.   A typical titration graph is shown in Fig. 2.   End-points were calculated from the second derivative.
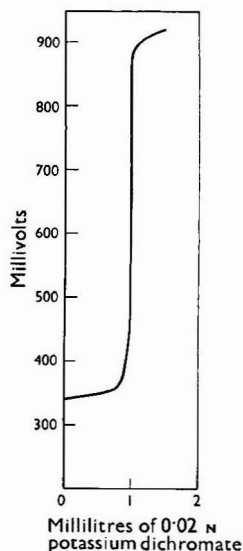

Fig. 2.   Typical potentiometric titration curve

Plotting dichromate titres, obtained by using potentiometric end-point detection, against millilitres of iron(II) solution (about 0·02 N) gives a linear relationship. A similar procedure in which dichromate titres, obtained with diphenylamine sulphonate as indicator, are used shows a change of slope occurring at approximately 5 ml of iron(II) solution.

By using the titres shown in Table I the following equations may be derived—

$$P = 0.9758\,M - 0.001 \quad .. \quad (1) \quad \text{(For the range 0 to 10 ml of iron(II) solution.)}$$
$$P = 0.9780\,M - 0.006 \quad .. \quad (2) \quad \text{(For the range 0 to 5 ml.)}$$
$$P = 0.9761\,M - 0.004 \quad .. \quad (3) \quad \text{(For the range 5 to 10 ml.)}$$
$$D = 0.9690\,M + 0.049 \quad .. \quad (4) \quad \text{(For the range 0 to 5 ml.)}$$
$$D = 0.9756\,M + 0.007 \quad .. \quad (5) \quad \text{(For the range 5 to 10 ml.)}$$

where P = dichromate titres obtained potentiometrically;
       D = dichromate titres obtained with diphenylamine sulphonate; and
       M = millilitres of about 0·02 N iron(II) solution.

From equations (2) and (4) the relationship between potentiometric and indicator titres for volumes of iron(II) solution in the range up to 5 ml is given by—

$$P = 1.0093\,D - 0.055 \quad .. \quad .. \quad .. \quad .. \quad (6)$$

Similarly, from equations (3) and (5) for volumes of iron(II) solution in the range 5 to 10 ml, the relationship is—

$$P = 1.0005\,D - 0.011 \quad .. \quad .. \quad .. \quad .. \quad (7)$$

Thus in this range titres obtained with the indicator diphenylamine sulphonate tend to be about 0·01 ml higher than those obtained potentiometrically. This is probably because the potentiometric end-point is more easily detected than the indicator end-point, slightly more titrant being required with the latter to develop a perceptible colour change. However,

TABLE II

IRON(II) OXIDE DETERMINED IN W–1

Nitrogen flow-rate	Sample weight, mg	Time, minutes	0·02 N Potassium dichromate, ml	Iron(II) oxide, per cent.	Iron(II)* oxide, per cent.
100 ml	75·95	60	4·44	8·40	8·38
per	72·75	50	4·30	8·49	8·47
minute	74·75	50	4·36	8·42	8·40
	75·00	60	4·55	8·72	8·70
	71·22	60	4·30	8·68	8·66
	69·10	60	4·06	8·44	8·40
	70·10	30	4·19	8·59	8·55
500 ml	74·85	60	4·51	8·66	8·64
per	71·05	60	4·28	8·66	8·64
minute	72·15	30	4·36	8·68	8·66
	77·75	60	4·69	8·67	8·65
	79·95	30	4·89	8·79	8·77
	80·80	30	4·93	8·77	8·75
	79·25	30	4·83	8·76	8·74
	69·30	60	4·18	8·67	8·63
	68·70	15	4·19	8·76	8·72
	70·18	60	4·28	8·76	8·74
	68·00	70	4·14	8·75	8·71
	70·50	60	4·28	8·72	8·70
	69·80	60	4·25	8·75	8·73
	67·22	120	4·06	8·68	8·64
	73·40	60	4·47	8·75	8·73
	71·91	30	4·36	8·71	8·69
	71·76	30	4·34	8·69	8·67
	73·57	30	4·48	8·75	8·73
	72·84	30	4·43	8·74	8·72
	73·43	30	4·44	8·69	8·67
Average				8·72	8·70
Standard deviation				0·04	0·04
95 per cent. confidence limits for average ..				8·72 ± 0·02	8·70 ± 0·02

* Calculated from the relationship between potentiometric and diphenylamine sulphonate titres according to equation (6).

within the limits of experimental error it is considered that for titrating volumes of iron(II) solution in the range 5 to 10 ml, the difference in dichromate titres between the two methods is not significant.

Equation (6) shows that for up to 5-ml volumes of iron(II) solution the difference between the dichromate titres is significant.

Accepting potentiometric titration as the reference method, equation (6) may be used to correct titres obtained with diphenylamine sulphonate as indicator and thus overcome the disproportion. In practice, the error in determining an iron(II) oxide content will only be significant for low titres and low sample weights. In Table II, in which iron(II) oxide determined in W–1 is shown, titres were in the range of 4 to 5 ml for the sample weights taken. These titres, corrected on the basis of equation (6), are on the average 0·01 ml less than un-corrected titres, and accordingly the percentage of iron(II) oxide is lower by 0·02. There-fore, the correction at this level is no greater than the probable experimental error. For practical purposes titres greater than 4 ml need not be corrected, but to ensure wider applica-tion of the method it is desirable to establish the relationship between potentiometric and diphenylamine sulphonate titres.

<center>EXPERIMENTAL</center>

APPARATUS—

Sample decomposition apparatus—This was constructed from 100-ml polythene bottles, and its structure may be clearly seen in Fig. 1. For potentiometric titrations the cap shown was replaced by one with holes for the insertion of the platinum electrode and salt bridge, and for the addition of nitrogen.

Platinum electrode—This consisted of thin-sheet platinum, 15×5 mm, welded to a platinum wire and sealed into a glass tube, which was covered with thin-walled plastic tubing to protect it from hydrofluoric acid vapours (see Fig. 3).

Salt bridge—A length of polythene tubing, to one end of which a plug of tightly rolled filter-paper had been fitted, was filled with saturated potassium chloride solution. A dip-type calomel electrode could be inserted tightly into the other end of the tube (see Fig. 3).

Potentiometer—Potentiometric measurements were made with a pH meter (supplied by W. G. Pye and Co., England) in conjunction with a dip-type calomel reference electrode.

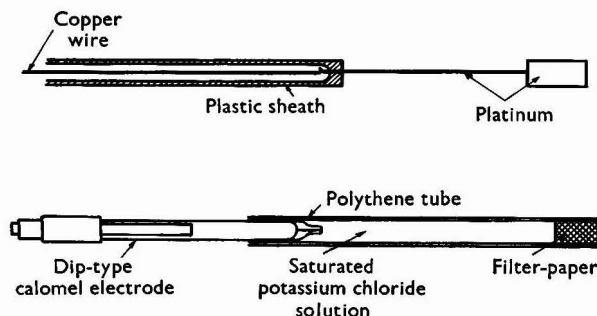*Burette*—A 10-ml Class A burette graduated at 0·02 ml intervals.

Fig. 3.   Platinum electrode and "salt bridge"

REAGENTS—

Hydrofluoric acid, analytical-reagent grade, 40 per cent.

Sulphuric acid, 1 + 3—One volume of concentrated sulphuric acid poured into three volumes of distilled water.

Potassium dichromate, 0·02 N—Prepare by powdering analytical-reagent grade potassium dichromate and drying it at 120° C for 2 hours; cool it in a desiccator. Weigh 0·9807 g, dissolve it in distilled water and dilute the solution to 1 litre.

Diphenylamine sulphonate solution, 0·3 per cent.—Dissolve 0·3 g of sodium diphenylamine sulphonate in 100 ml of distilled water.

Ferroin indicator, 0·025 M—Dissolve 1·485 g of o-phenanthroline monohydrate in 100 ml of a solution containing 0·695 g of iron(II) sulphate, $FeSO_4.7H_2O$.

PROCEDURE—

Weigh, to the nearest 0·01 mg, a 20 to 100-mg sample depending on the expected iron(II) oxide content. Transfer to the decomposition vessel, assemble the apparatus and flush with nitrogen at a rate of about 100 ml per minute. Carefully add 10 ml of freshly boiled, 1 + 3, sulphuric acid solution, and increase the nitrogen flow to 500 ml per minute. Make sure that the nitrogen inlet is below the surface of the acid. Swirl the apparatus to disperse the sample, then add 5 ml of hydrofluoric acid from a small plastic vial. Place the stopper loosely in the funnel and immerse the apparatus to about half its depth in a water-bath maintained at 80° C. During sample dissolution swirl the flask occasionally. When dissolution is complete (generally in about 30 minutes) remove the apparatus from the hot water-bath and place it in a cold water-bath. Remove the funnel, and wash down the inner surface of the vessel with freshly boiled and cooled distilled water until the volume is about 25 ml. Allow the apparatus to cool for 10 to 15 minutes. Remove it from the cooling bath, add one drop of diphenylamine sulphonate indicator and immediately titrate with dichromate until a definite purple colour is reached. Correct the volume of dichromate used by subtracting the titre obtained in a blank determination. The percentage of iron(II) oxide in the sample is given by—

$$\frac{\text{Dichromate (ml)} \times 143\cdot7}{\text{Sample weight (mg)}}.$$

For dichromate titrations of less than 4 ml, correct the titre on the basis of equation (6).

If a potentiometric titration is desired, the cap fitted with platinum electrode and salt bridge is used in place of the normal cap. The electrode and bridge are withdrawn to the top of the cap during sample dissolution and lowered into the solution before titration.

TABLE III

EFFECT OF SAMPLE WEIGHT ON THE DETERMINATION OF IRON(II) OXIDE IN W–1

End-point detection	Sample weight, mg	0·02 N Potassium dichromate, ml	Iron(II) oxide, per cent.	
Potentiometric	19·62	1·19	8·72	
	38·14	2·30	8·67	8·70
	73·73	4·47	8·71	Average
	78·47	4·74	8·68	
Ferroin	20·29	1·24	8·78	
	38·58	2·33	8·68	8·71
	74·59	4·50	8·67	Average
	79·30	4·82	8·73	
Diphenylamine sulphonate	20·20	1·26	8·96	8·68*
	37·63	2·31	8·82	8·71*
	77·17	4·68	8·71	8·70*

* Corrected on basis of equation (6).

RESULTS AND DISCUSSION

INFLUENCE OF NITROGEN FLOW-RATE—

To prevent oxidation of iron(II) during decomposition of a silicate material, "oxygen-free" nitrogen was used to displace air from the apparatus. On testing the method for the determination of iron(II) oxide in the diabase W–1, it was found that the variation in results initially obtained occurred because the nitrogen flow-rate of 100 ml per minute was insufficient, especially when, during the course of sample dissolution, the apparatus was swirled to ensure dispersion of the sample. Consistent results were obtained when the flow-rate was increased to 500 ml per minute, which ensured a good flushing of the apparatus and in addition served to keep the sample dispersed. Passing the nitrogen through a saturated iron(II) sulphate solution and then through a bubbler containing reduced anthraquinone β-sulphonic acid did not significantly change the results as compared with those obtained by using nitrogen directly from the cylinder.

Tests on an iron(II) sulphate solution treated in the apparatus in the same way as a sample for periods up to 1 hour gave dichromate titres averaging about 99·5 per cent. of those obtained by immediate titration.

The results obtained for the iron(II) oxide content of W–1 under numerous different conditions are shown in Table II.

VARIATION IN SAMPLE WEIGHT—

In view of the disproportion evident in the titration of iron(II) in amounts equivalent to less than 4 ml of 0·02 N dichromate when diphenylamine sulphonate is used as indicator, various weights of W–1 were taken for the determination of the iron(II) oxide content, with three methods of end-point detection. The results of these determinations are shown in Table III. The iron(II) oxide contents of 8·96 and 8·82 per cent. obtained for sample weights of approximately 20 and 38 mg, respectively, with diphenylamine sulphonate are significantly higher than the average value of 8·70 per cent. obtained for 20 determinations with sample weights averaging about 72 mg. However, if the dichromate titres found in these determinations with the lower sample weights are corrected on the basis of equation (6), iron(II) oxide contents become 8·68 and 8·71 per cent., respectively, in agreement with the average value.

In the determination of iron(II) oxide in a sample with diphenylamine sulphonate as indicator, it is necessary to take a sample weight which will require a dichromate titre of not less than 4 ml of 0·02 N solution, or alternatively to correct the titre on the basis of the relationship between potentiometric and diphenylamine sulphonate titres. The use of ferroin for end-point detection does not suffer from this limitation, but the tendency towards over-titration and the difficulty of observing the exact colour change are disadvantageous. Potentiometric titration is by far the most reliable method of end-point detection, as it is not subject to the disproportion effect when small volumes of iron(II) are titrated, and because the potential break is so distinct.

APPLICATION OF THE METHOD

The method of sample decomposition and subsequent direct titration of iron(II) with dichromate, with diphenylamine sulphonate as indicator, has been applied to the determination of iron(II) oxide in a number of slags from a boiler fired with pulverised fuel. The results of the determinations shown in Table IV are compared with iron(II) oxide contents obtained by Wilson's colorimetric method. In general, the titration procedure gave slightly higher contents than the colorimetric method. Two samples allowed to stand at room temperature for 4 hours did not give complete decomposition, and a large increase in iron(II) oxide contents was obtained in repeat determinations left overnight.

CONCLUSION

The determination of iron(II) oxide in silicate materials decomposed by hydrofluoric acid can be easily performed in an inexpensive apparatus under conditions that prevent oxidation. Difficulties associated with the titration of small amounts of iron(II) with dichromate, with diphenylamine sulphonate as indicator, can be avoided by using adequate sample weights or, if this is not possible, by application of a correction where necessary, based on the relationship between potentiometric and diphenylamine sulphonate titres of an iron(II) solution. For the most precise determinations potentiometric titration is recommended. In the determination of iron(II) oxide in W–1, an average of 8·70 per cent. was

TABLE IV

DETERMINATION OF IRON(II) OXIDE IN SLAGS FROM PORT AUGUSTA POWER
STATION, SOUTH AUSTRALIA

Sample	Proposed method	Wilson's colorimetric method	
		4 hours	16 hours
A	5·17*	—	5·01
	5·26*	—	5·01
B	9·48	—	9·12
	9·45	—	9·18
C	9·74	—	9·62
	9·67	—	9·47
D	10·99	9·25	10·69
	10·92	8·74	10·10
E	7·78	7·07	7·89
	7·85	6·67	7·60

* Corrected on basis of equation (6).

found in 20 determinations, with a standard deviation of $\pm 0\cdot04$ per cent. The 95 per cent. confidence limits for the average are $8\cdot70 \pm 0\cdot02$ per cent. The preferred value for the iron(II) oxide content of W–1 as given by Fleischer and Stevens[11] is $8\cdot74$ per cent., whereas Ingamells and Suhr[12] give a preferred value of $8\cdot71$ per cent.

It should be possible to extend the application of the method by using an apparatus constructed of PTFE, to enable the acid to be boiled during decomposition.

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The Determination of Fluorine or Phosphorus in Organic Compounds by a Micro-titrimetric Method

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A method is described for determining fluorine or phosphorus in organic compounds synthesised for medical research. After combustion of the compound in an oxygen flask and absorption of the decomposition products in water, the contents are diluted with isopropanol, and titrated with thorium nitrate solution with a Solochrome cyanine R screened indicator. The addition of buffers is unnecessary. The removal of elements that interfere in the titration of fluorine is also described.

IN the determination of the elements in organic compounds by micro-analytical techniques it is desirable that the methods used should be both accurate and simple. This is not achieved in the determination of fluorine or phosphorus; known methods are accurate, but there is a need for greater simplicity. With the oxygen-flask[1] method, the decomposition of organic compounds has been simplified, but the final determination of the ionised fluorine or phosphorus is elaborate.

The hydrofluoric acid formed by combustion of fluorine compounds that contain no other acid-forming elements can be titrated directly with 0·01 N sodium hydroxide.[2] An alternative method that was found to give good results is to add potassium iodate and then potassium iodide to the acid solution and titrate the liberated iodine with a standard solution of sodium thiosulphate.

Attempts made by the author, when using the oxygen flask for combustion, to find a titration finish applicable to organic compounds containing fluorine or phosphorus that have been synthesised for medical research are described in this paper.

No reports in the literature could be found relating to the titration of orthophosphate with thorium, but, as thorium forms an insoluble phosphate, it was thought that a thorium solution could be used as a titrant for phosphorus and the same method used as that used for fluorine, especially as phosphate was found to interfere with the determination of fluorine. Sulphate also interfered, but as the interference was not quantitative it could not be used as a method for the determination of sulphur.

EXPERIMENTAL

Metals that form insoluble fluorides and phosphates were chosen for examination. These are: thorium, zirconium, lanthanum, cerium, yttrium, bismuth and iron. Indicators that form coloured complexes with one or more of them are: Alizarin red S, acid Alizarin black S, Solochrome (Eriochrome) cyanine R,* pyrocatechol violet, methylthymol blue, xylenol orange, thoron, PAN, SPADNS, Zincon, Tiron, phenylfluorone, arsenazo and purpurin.

Complexes of these metals and indicators have all been used in the spectrophotometric determination of either the metal or fluorine, but reports on them in the literature are too numerous to quote. The majority of these failed in the present work for one or more of the following reasons: some required very close pH control as the indicators are also acid - base indicators; end-points with some were slow; and in many instances the colour change was gradual from one tone to another with no sharp change taking place. In nearly all of these combinations the metals formed highly insoluble lakes that separated out, and end-points were masked.

The most promising results, however, were obtained by titrating fluoride and orthophosphate ions with thorium nitrate and Solochrome cyanine R as indicator. Willard and Horton[3] state that the preferred order of indicators for the titration of fluoride with thorium nitrate is (*i*) purpurin sulphonate, (*ii*) Alizarin red S, and (*iii*) Solochrome cyanine R.

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* The indicator is referred to by the suppliers as "Solochrome cyanine R" and this designation will be used throughout.

Alizarin red S is the most widely mentioned indicator used for the determination of fluorine. In no instance, however, when used on the micro-scale in the author's laboratory, was it possible to detect a reliable end-point with this indicator by straight titration, and at all times it was necessary to exercise strict pH control by the use of buffers. The recommended method was that of Dahle *et al.*,[4] and is based on colour matching by comparison and described in full by Clark,[5] and Milton and Waters.[6]

It was therefore decided to investigate more fully the use of Solochrome cyanine R. Supplies of this indicator were obtained from three sources, A, B and C. A 0·25 per cent. aqueous solution of each indicator was prepared and 3 drops of indicator and 1 drop of 2 N nitric acid were added to three separate flasks, each of which contained 25 ml of distilled water. The colour of each solution was golden brown and changed, for the solutions containing A and B, to red - purple on the addition of 1 drop of 0·01 N thorium nitrate, although with C, 0·1 ml of the thorium solution was required to give a colour change.

Visual inspection of the solid indicators showed that a second compound was probably present in each. Any such compound was water-soluble as no residue was left when the solutions were filtered.

A 0·25 per cent. solution of each indicator was then prepared in 96 per cent. ethanol and, on filtering, A and B left a small amount of a white organic crystalline material, while C left an inorganic residue amounting to about 66 per cent. of the dye. This was shown to be sodium sulphate. A and B were used for further investigations.

A standard solution of 0·01 N sodium fluoride was used for the titration with thorium nitrate, but the colour change at the end-point was not considered satisfactory.

Cheng[7] has shown that in the determination of chlorine and bromine, the colour of the end-point is greatly enhanced if the determination is carried out in a mixture of water and an organic solvent such as ethanol, methanol or isopropanol. Experiments on these lines were then conducted for the titration of fluoride with thorium nitrate, and, as isopropanol was the solvent chosen by Cheng for determining chlorine and bromine, this solvent was used in the present work. To each of two flasks, one of which contained 25 ml of isopropanol and water (4 + 1), and the other, 25 ml of distilled water, were added 3 drops of indicator (A or B) and 1 drop of 2 N nitric acid. The isopropanol solution turned yellow, but the aqueous solution showed no change. Both solutions turned purple on adding 1 drop of thorium nitrate solution, but the colour of the propanol solution was much more intense. Screening the indicator with methylene blue made the end-point even more distinctive, and in the titration of fluoride the colour change was from green to blue - purple. Successive titrations of 2 ml of 0·01 N hydrofluoric acid in a mixture of 3 ml of water and 20 ml of isopropanol with 0·02 N thorium nitrate required 0·98, 0·99, 0·98, 0·98 and 0·99 ml.

Similar experiments conducted with orthophosphoric acid solutions gave excellent end-points. In the titration of sulphate solutions, however, the end-points were poor, and as they were not quantitative this titration was not investigated further. The presence of phosphorus and sulphur interferes with the determination of fluorine and their removal is described later in this paper.

METHOD

All combustions were carried out in 250-ml silica oxygen flasks.[8]

REAGENTS—

Solochrome cyanine R—Prepare a 0·25 per cent. solution in 95 per cent. ethanol and filter into a bottle. The solution is quite stable.

Methylene blue solution, 0·05 per cent., w/v, aqueous.

Thorium nitrate, 0·02 N (0·005 M)—Dissolve 2·7610 g of thorium nitrate tetrahydrate, $Th(NO_3)_4.4H_2O$, in distilled water and make up to 1 litre.

Isopropanol—Use AnalaR grade.

DETERMINATION OF FLUORINE—

Weigh accurately sufficient of the compound to give approximately 1 mg of ionised fluorine. Wrap it in a square of filter-paper and place it in the platinum spiral. Transfer 4 ml of distilled water into a 250-ml silica flask and flush it with oxygen. Light the tab and plunge the spiral into the flask. When the combustion is complete, shake the flask well and allow it to stand for 10 minutes. Wash down the spiral and stopper with the minimum amount of water (1 to 2 ml) and gently boil the contents of the flask for about 10 seconds

to expel carbon dioxide. Cool the flask and add 20 ml of isopropanol. Then add 0·3 ml of Solochrome cyanine R indicator and 3 drops of methylene blue. The colour should now be green; if not, add 1 drop of 2 N nitric acid. Titrate with 0·02 N thorium nitrate to a blue - purple end-point (when the green colour begins to darken, the thorium nitrate should be added slowly with vigorous shaking of the flask). The thorium nitrate is standardised empirically by combusting standard organic fluorine compounds.

The results obtained by this method are shown in Table I.

TABLE I

DETERMINATION OF FLUORINE IN THE ABSENCE OF SULPHUR AND PHOSPHORUS

		Fluorine		
Compound	Weight, mg	found, per cent.	required, per cent.	Difference, per cent.
Trifluoroacetanilide M.A.S.	2·983	30·25	30·14	+0·11
	3·125	30·33	30·14	+0·19
	3·068	29·92	30·14	—0·22
	3·130	30·04	30·14	—0·10
	2·979	30·20	30·14	+0·06
	3·111	29·95	30·14	—0·19
p-Fluorobenzoic acid M.A.S.	5·300	13·64	13·56	+0·08
	4·857	13·62	13·56	+0·06
Trifluoromethylbenzoic acid M.A.S.	3·155	29·93	29·98	—0·05
2,5-Di-(2-fluorophenyl)-oxadiazole 1,3,4 ..	5·879	14·92	14·72	+0·20
2-(2-Fluorophenyl)-5-phenyloxadiazole 1,3,4 ..	7·082	7·55	7·92	—0·37
	8·290	7·72	7·92	—0·20
Fluoxymesterone	9·237	5·75	5·65	+0·10
$C_{14}H_{10}NO_2F_3$	3·735	20·32	20·25	+0·08
$C_{21}H_{27}N_2OF_3$	5·264	15·0	15·0	Nil
$C_{21}H_{29}NO_3F_3$	5·278	14·38	14·2	+0·18
$C_{10}H_{12}NO_2F$	6·100	9·62	9·64	—0·02
$C_9H_9NO_2F_2$	3·922	18·9	18·9	Nil

REMOVAL OF PHOSPHORUS AND SULPHUR—

Soluble salts of barium, lead, zinc, magnesium and silver were used for the attempted removal of phosphorus (assumed to be present as orthophosphate), and barium and lead for the removal of sulphur as sulphate. Only silver gave complete recovery of fluoride in the presence of phosphorus, and neither barium nor lead gave complete recovery of fluoride in the presence of sulphur. However, complete removal of sulphate was obtained by the use of benzidine.

REMOVAL OF PHOSPHORUS—

The method of Colson[9] for the removal of phosphorus in the determination of sulphur was used, except that the ion-exchange resin procedure was omitted as excess of silver ions has no effect on the titration with thorium nitrate.

REAGENT—

Silver oxide—Prepare as described by Colson.

After the contents of the flask have been briefly boiled, add about 50 mg of silver oxide sludge and boil for about 1 minute. Cool the flask and filter through a small Hirsch funnel into a titration flask. Wash out the flask with the minimum amount of water and add isopropanol to give a final strength of 70 to 80 per cent. Carry out the titration as for fluorine. The addition of 1 drop of 2 N nitric acid is necessary to neutralise the slight solubility of the silver oxide. Determine the blank value.

REMOVAL OF SULPHUR—

Bring the contents of the flask to the boil and add 2 ml of 1 per cent. w/v solution of pure benzidine in 96 per cent. ethanol; boil for a further 15 seconds. Allow the flask to cool under running tap water for a minimum of 1 hour, and then filter into a titration flask as above and carry out the titration as for fluorine.

The excess benzidine will need to be neutralised with 2 N nitric acid.

Table II shows the results obtained with some compounds that contain fluorine and either phosphorus or sulphur.

TABLE II

DETERMINATION OF FLUORINE WHEN SULPHUR OR PHOSPHORUS IS PRESENT

					Fluorine		
Compound				Weight, mg	found, per cent.	required, per cent.	Difference, per cent.
Trifluoroacetanilide ..	..	..	..	3·160	29·7	30·14	−0·44
(+ phenylthiourea) ..	..	..	..	3·288	30·65	30·14	+0·51
				3·362	30·0	30·14	−0·14
$C_{10}H_{12}F_3SN_3HCl.H_2O$	..	..	..	4·848	18·16	17·94	+0·22
				5·444	18·0	17·94	+0·06
$C_{16}H_{15}N_2O_2SF_3$	..	..	..	4·485	15·78	16·00	−0·22
$C_{15}H_{15}N_2O_2SF$	..	..	..	8·317	6·18	6·20	−0·02
$C_{22}H_{40}NPF_6$ ..	..	..	..	3·504	25·1	24·6	+0·50
				3·890	24·88	24·6	+0·28

DETERMINATION OF PHOSPHORUS—

The method and reagents are the same as those described under Determination of Fluorine. Standardise the thorium nitrate empirically with standard organic phosphorus compounds.

Owing to the lack of organic phosphorus standards only triphenylphosphine can be quoted, although research compounds have been successfully analysed. Table III shows the results obtained with triphenylphosphine.

TABLE III

DETERMINATION OF PHOSPHORUS IN THE ABSENCE OF FLUORINE AND SULPHUR

					Phosphorus		
Compound				Weight, mg	found, per cent.	required, per cent.	Difference, per cent.
Triphenylphosphine ..	..	..	..	4·068	11·70	11·80	−0·10
				4·614	11·85	11·80	+0·05
				3·977	12·06	11·80	+0·26
				4·893	11·70	11·80	−0·10

DISCUSSION

The method described gives excellent results for fluorine or phosphorus when no interfering elements are present. It is not necessary to add buffers to maintain a definite pH.

Although phosphorus and sulphur can be removed when the determination of fluorine is required, it has not been found possible to remove fluorine or sulphur when requiring to determine phosphorus.

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The Colorimetric Determination of Boron in Soils, Sediments and Rocks with Methylene Blue

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A method is presented for the determination of boron in soils, sediments and rocks. The sample is decomposed by a mixture of hydrofluoric and sulphuric acids, and the complex formed between fluoroborate ions and methylene blue is extracted with 1,2-dichloroethane. Boron is determined either by visual colour of the blue complex, or by spectrophotometry. The method is rapid and sensitive.

THE reaction of the fluoroborate ion with methylene blue has been applied to the determination of boron in soils, sediments and rocks, following its use in the analysis of iron and steel,[1,2] and the procedure described below has been developed.

METHOD

REAGENTS—

Sulphuric acid, 10 N—Prepare from analytical-reagent grade acid.
Hydrofluoric acid, 40 per cent. w/w, analytical-reagent grade.
Methylene blue, 0·08 per cent. w/v, aqueous.
1,2-Dichloroethane.
Sodium tetraborate—Decahydrate, analytical-reagent grade.
Standard boron solutions—Dissolve 0·4408 g of sodium tetraborate in 10 N sulphuric acid, and dilute with this acid to 500 ml in a calibrated flask to give a solution containing 100 μg of boron per ml. Dilute further with 10 N sulphuric acid to give solutions containing 2, 5 and 10 μg of boron per ml.

PROCEDURE—

Weigh 0·1 g of sample into a polythene beaker, and add 2·5 ml of 10 N sulphuric acid and 0·5 ml of 40 per cent. hydrofluoric acid. Stir the solution with a polythene rod, cover and leave to stand at room temperature for 2 hours. Add 2 ml of water, mix and leave to stand for 15 minutes. Transfer 1 ml of the clear solution by pipette into a test-tube calibrated at 15 ml. Add 1 ml of 0·08 per cent. methylene blue and dilute to 15 ml with water. Add 5 ml of 1,2-dichloroethane, stopper the tube and shake it vigorously for 30 seconds. Allow the phases to separate and compare the intensity of colour in the organic layer with a standard series.

A reagent blank must be determined.

PREPARATION OF THE STANDARD SERIES—

Into eleven polythene beakers transfer by pipette, from the dilute standard solutions, 0, 1·0, 2·0, 3·0, 4·0, 5·0, 7·5, 10·0, 15·0, 20·0 and 25·0 μg of boron, respectively. Add sufficient 10 N sulphuric acid to each beaker to give a total volume of 2·5 ml. Add 0·5 ml of 40 per cent. hydrofluoric acid, mix well and leave to stand for 2 hours. Then add 2 ml of water and mix. Remove 1 ml from each solution with a pipette and treat as described for a sample solution in Procedure.

DISCUSSION OF THE METHOD

This method was developed for geochemical research studies in which the boron was derived from the mineral colemanite, and was readily attacked in the cold by a mixture of dilute hydrofluoric and sulphuric acids. Consequently, application of the method to other materials may be restricted by this method of sample decomposition.

TABLE I

COMPARISON BETWEEN CRUSHED AND UNCRUSHED SAMPLES

Sample No.	Boron, p.p.m.		Sample No.	Boron, p.p.m.	
	−20 mesh	−80 mesh		−20 mesh	−80 mesh
1	38	45	5	340	340
2	105	100	6	280	200
3	220	240	7	200	180
4	320	330	8	180	190

Formation of fluoroborate takes place as the sample is being attacked. The acid concentrations are not critical, but must be kept at the same constant level for both samples and standards, and 10 N sulphuric acid is used so that alkaline samples produce no significant variation in acidity. The volume of 40 per cent. hydrofluoric acid is kept low to minimise the colour extracted from the zero standard. The time allowed for this stage is not critical; some samples went completely into solution within 1 hour, whereas others appeared unaltered, even after standing overnight. Nevertheless, boron was always completely converted into the soluble fluoroborate within 2 hours. Samples have been left in contact with the acids for up to 3 days without ill effect, although there is great danger of volume loss by evaporation. Complete recovery was achieved when boron was added to samples as sodium tetraborate; it was also found possible to increase the sample weight to 250 mg without any other alteration to the procedure.

As it was necessary for geological reasons to analyse the −20-mesh fraction of many samples, a comparison was made between results obtained when this fraction was crushed to pass 80 mesh and on the uncrushed material. From the results given in Table I it will be seen that, in general, there is no significant difference, although analysis of the coarser material may introduce greater sampling errors.

There is always a slight colour extracted from the zero standard that is dependent upon the final aqueous phase acidity, decreasing with decreasing acidity while the efficiency of

TABLE II

EFFECTS OF VARIOUS ELEMENTS ON THE DETERMINATION OF BORON

Element	Amount added, mg	Boron present, μg	Boron found, μg	Element	Amount added, mg	Boron present, μg	Boron found, μg
Aluminium	5·0	0	<0·1	Manganese	1·0	0	<0·1
	5·0	1·0	1·0		1·0	1·0	1·0
Arsenic (as	0·1	0	3·5	Mercury(II)	0·1	0	0·4
$Na_2HAsO_4.7H_2O$)	0·1	1·0	4·5		0·1	1·0	1·2
Barium	0·1	0	<0·1	Molybdenum (as	0·1	0	<0·1
	0·1	1·0	1·0	$Na_2MoO_4.2H_2O$)	0·1	1·0	1·0
Calcium	5·0	0	0·1	Nickel	0·1	0	<0·1
	5·0	1·0	1·0		0·1	1·0	1·0
Chloride	1·0	0	0·2	Nitrate	1·0	0	3·0
	1·0	1·0	1·0		1·0	1·0	4·0
Chromium (as	0·04	0	<0·1	Potassium	1·0	0	<0·1
$CrCl_3$)	0·04	1·0	1·0		1·0	1·0	1·0
Chromium (as	1·0	0	⩾5·0	Silicon (as	2·5	0	<0·1
$K_2Cr_2O_7$)	1·0	1·0	⩾5·0	SiO_2)	2·5	1·0	1·0
	0·1	0	⩾5·0	Silver	0·1	0	<0·1
	0·1	1·0	⩾5·0		0·1	1·0	1·0
Cobalt	0·1	0	0·2	Sodium	5·0	0	<0·1
	0·1	1·0	1·0		5·0	1·0	1·0
Copper	0·1	0	<0·1	Titanium	1·0	0	<0·1
	0·1	1·0	1·0		1·0	1·0	1·0
Iron(II)	5·0	0	<0·1	Tungsten (as	0·1	0	<0·1
	5·0	1·0	1·0	$Na_2WO_4.2H_2O$)	0·1	1·0	1·0
Iron(III)	5·0	0	<0·1	Vanadium	0·1	0	<0·1
	5·0	1·0	0·9		0·1	1·0	1·0
Magnesium	5·0	0	<0·1	Zinc	0·1	0	<0·1
	5·0	1·0	1·0		0·1	1·0	1·0

extraction of the fluoroborate complex increases. It is therefore important that the final acid concentration should be constant, and if more than 1 ml of sample solution is used for analysis, the preparation of the standard series must be adjusted to obtain similar conditions. Likewise, when less than 1 ml is used, compensating amounts of hydrofluoric and sulphuric acids must be added with the aliquot. The standard series shows an increasing intensity of blue from a slightly blue zero.

The molarity of methylene blue in the final aqueous phase must not be less than four times that of the boron, an increase of 50 per cent. having no effect upon the intensity of colour in the organic phase. The complex is readily extractable, agitation for 15 seconds is probably adequate, and it is stable for 2 days. Benzene, carbon tetrachloride, chloroform, isopentyl acetate, isopentanol, toluene, various petroleum spirits and white spirit were tried unsuccessfully as alternative solvents.

It was found convenient to use polythene ice-cube trays for the acid treatment of the sample, a second tray being used as a cover; pipettes were made from quartz glass or polythene. Borosilicate glassware must, of course, be avoided, and even soft glass can contain a few per cent. of boron and so cause contamination. Quartz glass test-tubes were used for the colorimetry, although lead glass is also satisfactory and such test-tubes are much cheaper. Bark corks were unsatisfactory as they absorbed a considerable amount of methylene blue which did not wash out and could be liberated during a later test to give high results; the use of silicone rubber stoppers eliminated this source of error.

A spectrophotometric finish could be adopted, the boron - methylene blue complex exhibiting an absorption maximum at 640 mμ. A blank determination should be used as reference.

INTERFERENCE FROM OTHER ELEMENTS—

The effects of various ions are shown in Table II. Dichromate, nitrate, arsenate and mercury (II) ions have adverse effects, but the concentrations at which interference occurs are unlikely to be obtained in normal samples.

RESULTS

The reproducibility of the colorimetric stage was tested by using several aliquots from one sample solution. A mean value of 177·5 p.p.m. was obtained, with a standard deviation of ±6·1 p.p.m.

Replicate analyses on three samples gave mean values of 32·5, 177·5 and 233 p.p.m., with standard deviations of ±2·8, ±6·1 and ±12·1 p.p.m., respectively.

Several samples, including the standard rocks G1, W1 and Sy1, were analysed by the proposed method and by the official A.O.A.C. method.[3] The results are shown in Table III,

TABLE III

COMPARISON OF RESULTS FROM VARIOUS METHODS

| | Boron, p.p.m. | | | |
Sample No.	Proposed procedure	A.O.A.C. method	Spectrography	Mass spectrometry
G1	<2	<10	—	2·8
W1	12	23	17	18
Sy1	72	69	70	—
1	184	173	—	—
2	80	80	—	—
3	92	91	—	—
4	122	137	—	—
5	134	137	—	—
6	166	160	—	—
7	188	182	—	—
8	186	182	—	—
9	182	182	—	—
10	186	182	—	—
11	44	46	—	—

together with some spectrographic[4] and mass spectrometric[5] determinations on the standard rocks. The low values for G1 and W1 are likely to be caused by the inadequacy of the acid treatment of the particular mineral containing the boron.

The work described forms part of the programme of the Applied Geochemistry Research Group under the direction of Professor J. S. Webb.

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The Analysis of the Organophosphorus Pesticide, Fenitrothion, by an Infrared Method

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An infrared method is described for determining the fenitrothion [*OO*-di-methyl-*O*-(3-methyl-4-nitrophenyl)-phosphorothioate] content of technical material after chromatography over silica gel. The fenitrothion is eluted with methylene chloride, and after evaporating the eluate to dryness the residue is dissolved in carbon disulphide to give a concentration of fenitrothion in the range 0·8 to 1·1 per cent. w/v. Measurements taken at three absorption peaks in conjunction with related minima are used to calculate the fenitrothion content from previously prepared calibration graphs that relate absorbance to fenitrothion content.

The standard deviation of this method, based on ten samples of technical material, is 0·53 per cent.

THE increasing use of organophosphorus pesticides necessitates the development of more specific methods of analysis. Non-specific methods, such as those based on the reduction of the nitro group, for example the method of O'Keeffe and Averell[1] for technical parathion, tend to give erroneously high results owing to the presence of "related impurities." A search of the literature has revealed some general papers on fenitrothion [*OO*-dimethyl-*O*-(3-methyl-4-nitrophenyl)-phosphorothioate][2,3,4,5] and two concerned with its analytical determination.[6,7] One of these analytical methods involves polarography after separation of technical material by thin-layer chromatography, and the other utilises gas - liquid chromatography for residue analysis. Neither method was considered suitable for our purposes, and the object of our work has been to develop a rapid, specific infrared method for determining the fenitrothion content of technical and formulated materials (Notes 1, 2 and 3).

EXPERIMENTAL

In an attempt to isolate fenitrothion from associated impurities, preliminary separations were carried out by loose-layer chromatography. Examination of several solvent systems on the adsorbents, alumina, silica gel and Florisil, showed that a good separation could be obtained on silica gel by using methylene chloride as developing solvent.

Chromatography over a column of silica gel (50 g) with methylene chloride as developing solvent isolated the major component from technical material (0·5 g). After elution of this material, which represented about 95 per cent. w/w recovery, a clear fraction was obtained

NOTES—

1. This method of determination can also be applied to liquid formulations as follows—

Make a slurry of 50 g of silica gel with petroleum spirit (boiling range 40° to 60° C) and methylene chloride (1 + 1 v/v) and place it in the chromatographic column; drain off excess solvent.

Take a sufficient formulation to contain 0·2 g of fenitrothion and transfer it quantitatively with a minimum amount of petroleum spirit - methylene chloride to the top of the silica gel column. Place a beaker under the column and allow the solution to percolate into the adsorbent. Add small amounts of solvent to ensure complete adsorption of the fenitrothion into the silica gel. Continue to elute with petroleum spirit - methylene chloride (about 150 ml) until the formulation solvent is removed.

Elute with 250 ml of methylene chloride and continue from this point as directed under Chromatographic Separation of Fenitrothion.

2. Normally, formulation ingredients and decomposition products (if any) that are formed on storage will be retained on the adsorbent.

3. Possible interference will be detected if the concentration of fenitrothion with any one of the peaks differs by more than ±3 per cent. (relative) of the mean. If this occurs, the particular value should be discarded and the remaining concentrations averaged.

before the first impurity was eluted. The other impurities remained on the column as four separate zones.

PROOF OF STRUCTURE OF THE MAJOR COMPONENT—

The major component isolated by chromatography was examined by (*a*) mass spectrometry with an A.E.I. Ltd. M.S. 2H mass spectrometer, (*b*) infrared spectrometry with a Grubb Parsons spectrometer and (*c*) thin-layer chromatography, by using a thin layer (275 μ) of silica gel G and chloroform or methylene chloride - benzene (1 + 1, v/v) as developing solvents. The chromatoplate was sprayed with 2,6-dichloro-*p*-benzoquinone-4-chlorimine, which appears to be specific for the P → S group by forming a red coloured derivative.[8]

The mass spectrometer showed a parent peak at *m/e* 277 (fenitrothion) and intense fragment ions at *m/e* 260, *m/e* 125 and *m/e* 109. The fragment ion at *m/e* 260 was attributed to the elimination of OH from the 3-methyl-4-nitro substituents of fenitrothion. Beynon *et al.*[9] have reported that *o*-nitrotoluene eliminates OH and this is accompanied by ring closure. This evidence confirms that the methyl and nitro groups are *ortho* to one another in the aromatic ring of fenitrothion. The fragment ion at *m/e* 125 was formed by the cleavage of the P–O bond in the P–O–aromatic group to give a substituted phenoxy ion. The intensity of the ion at *m/e* 109 was comparable to that of the ion at *m/e* 125. It appears that there is a re-arrangement within the mass spectrometer followed by elimination of the phosphorus moiety to produce an ion at *m/e* 109.

The infrared spectrum showed the presence of P–O–aromatic, P–O–methyl and probably P → S groupings. P → O and P–S–aromatic groupings were absent. The P → S group was confirmed by thin-layer chromatography, which showed one component only to be present.

Infrared analysis of the phenol isolated after hydrolysis of the major component showed this to be identical with 4-nitro-*m*-cresol (Sadtler infrared spectrogram No. 23685). This confirmed that the methyl and nitro groups are in the 3 and 4 positions, respectively.

Thus, the analytical evidence obtained by mass spectrometry, infrared spectrometry and thin-layer chromatography shows that the major component is fenitrothion, the structure of which is—
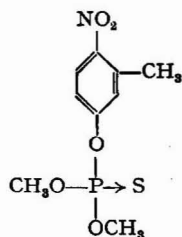
METHOD

APPARATUS—

Infrared spectrophotometer—An instrument capable of quantitative analysis in the 2 to 15-μ region is required.

Sealed liquid absorption cell—0·4-mm path length.

Hypodermic syringe—Glass, Luer type, 2-ml capacity.

Chromatographic columns—15 × 500 mm, fitted with a glass tap and a solvent reservoir of approximately 500 ml.

REAGENTS—

Silica gel—Whatman Chromedia SG31.

Benzene, general-purpose reagent grade.

Methylene chloride, general-purpose reagent grade.

Carbon disulphide, B.D.H. Ltd. general-purpose reagent grade or equivalent.

Fenitrothion—Analytical standard material of purity greater than 99 per cent. Prepare material suitable for analytical calibration purposes by using the chromatographic separation method.

CHROMATOGRAPHIC SEPARATION OF FENITROTHION—

Prepare a slurry of 50 g of silica gel with methylene chloride and transfer it to a chromatographic column with a cotton-wool plug to retain the adsorbent. Allow the solvent to pass through the column until the meniscus reaches the top of the adsorbent.

Weigh, to the nearest 0·1 mg, an amount of sample that contains approximately 0·2 g of fenitrothion. Dissolve the fenitrothion in a minimum of benzene (3 ml) and transfer the solution quantitatively to the top of the column by using methylene chloride. Allow the solvent to percolate into the silica gel, then wash the inside of the chromatographic column with three 5-ml portions of solvent, allowing each portion to penetrate the adsorbent independently.

Elute with 250 ml of methylene chloride and collect the eluate in a tared 400-ml beaker. When the solvent reaches the top of the adsorbent, stop the flow of solvent, wash the tip of the column with solvent and replace the 400-ml beaker with a 100-ml beaker. Evaporate the solvent contained in the 400-ml beaker at room temperature by using a forced draught. This is the fenitrothion residue.

Collect a further 50-ml fraction and evaporate to dryness in a forced draught to confirm the complete elution of fenitrothion by the absence of residue. In the unlikely event of a residue being present, examine it by infrared spectroscopy as it may be a related impurity.

Re-weigh the tared 400-ml beaker and obtain the weight of the extract. Calculate the percentage extract (to give a guide to the fenitrothion content) and submit the extract to quantitative infrared analysis.

INFRARED ANALYSIS

CALIBRATION OF APPARATUS—

Into each of several 20-ml calibrated flasks, weigh (to the nearest 0·1 mg) 160, 180, 200 and 220-mg amounts of the standard sample of fenitrothion. Dissolve each in carbon disulphide, dilute to the marks, and mix thoroughly. The strengths of these solutions will therefore be 0·8, 0·9, 1·0 and 1·1 g per 100 ml.

Fill the 0·4-ml cell with the most dilute of these standard solutions by means of the hypodermic syringe. Adjust the spectrophotometer to the optimum instrument settings with respect to gain, slit width, balance, response, chart speed and wavelength-scanning speed. Make duplicate scans over the 7·0 to 9·0-μ region.

Flush out the cell with carbon disulphide, dry, re-fill, in turn, with each of the remaining calibration solutions and, without changing instrument conditions, repeat the duplicate scans over the 7·0 to 9·0-μ region.

For each of the scans of the calibration solutions measure the transmitted radiant power, as a proportion of the incident radiant power, at the following wavelengths—

(*i*) From the absorption peak at about 7·45 μ to the minimum, *i.e.*, reference point at about 7·60 mμ.

(*ii*) From the absorption peak at about 8·05 μ to the minimum, *i.e.*, reference point at about 7·95 μ.

(*iii*) From the absorption peak at about 8·05 μ to the minimum, *i.e.*, reference point at about 8·30 μ.

(*iv*) From the absorption peak at about 8·55 μ to the minimum, *i.e.*, reference point at about 8·50 μ.

Construct a calibration graph for each absorption peak by plotting the percentage transmission on a logarithmic ordinate *versus* the corresponding concentration (g per 100 ml) on a linear abscissa. Construct the line of best fit through each set of four points.

PROCEDURE—

Dissolve the material obtained from the chromatographic procedure in a volume of carbon disulphide sufficient to give a concentration of fenitrothion in the range 0·8 to 1·1 per cent. w/v.

Fill the same 0·4-mm path-length cell that was used in calibrating the instrument with each sample solution in turn, and make duplicate scans over the 7·0 to 9·0-μ region. The same instrument conditions must be used as for the calibration.

Calculate the percentage transmission of the three specified peaks for each sample.

CALCULATION—

Read from the appropriate calibration graph the concentration, in g per 100 ml, of fenitrothion in each solution.

Take the average of the four concentrations thus obtained. Concentrations should not differ by more than 4 per cent. of the mean.

Calculate the fenitrothion content by using the following equation—

$$\text{Fenitrothion content per cent. by weight} = \frac{C \times V}{W}$$

where *C* is the average concentration as read from calibration graphs, g per 100 ml;
V is the volume of sample solution, ml; and
W is the original weight of sample taken for chromatographic "clean-up," g.

RESULTS AND CONCLUSIONS

The results obtained by using the method described are shown in Table I.

TABLE I

FENITROTHION CONTENT OF TECHNICAL MATERIAL

Technical fenitrothion, sample number	Fenitrothion content, per cent. w/w		Standard deviation, per cent.
	Individual	Mean	
1	92·4, 93·4, 92·6, 93·4, 92·5, 92·8, 92·7, 93·8, 93·8, 93·1	93·1	0·53
2	96·9, 97·5	97·2	—
3	96·4, 97·3	96·8	—
4	96·9, 97·3	97·1	—

The method described is specific and is used routinely for determining the fenitrothion content of technical material and its formulations. It has proved to be satisfactory. The standard deviation of the method, which includes adsorption chromatography and infrared spectroscopy, is 0·53 per cent.

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The Thermal Analysis of Lichens Growing on Limestone

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In an investigation of the pedogenic activities of lichens growing on limestones it was found that they varied greatly in their calcium oxalate content. As chemical methods of determining calcium oxalate in such material are somewhat tedious, detailed qualitative and quantitative studies of oxalate in specific lichen species were made by using differential thermal and thermogravimetric analysis. The results show that in a controlled atmosphere of oxygen-free nitrogen, thermal methods provide a rapid means of identifying and determining calcium oxalate in lichens, and also enable an assessment to be made of the non-oxalate derived carbonate in the sample.

IT has been recognised for some time that there is a correlation between the amounts of calcium and oxalate in many plants, and it has been claimed that calcium oxalate is abundant in all plant species inhabiting limestone. However, in the course of an investigation of the pedogenic activity of lichens, Syers[1] found that those growing on limestone varied widely in their oxalate content.

Chemical methods of determining oxalate in plant materials are tedious, especially for small amounts. Recently, Mitchell and Knight[2] demonstrated that oxalate in higher plants, for example, *Beta vulgaris* and *Epilobium lanceolatum*, may be conveniently monitored by thermal methods, and in view of this a detailed study of calcium oxalate in specific lichen species was conducted by using thermal methods.

Because the energy changes occurring in a material when heated may vary according to the atmosphere enveloping it, the thermal characteristics of lichen species were determined in oxygen, nitrogen and carbon dioxide to ascertain under which of these atmospheres the calcium oxalate reactions were distinct from other pyrolysis reactions occurring in the lichens. Further, as the area of a peak in a thermal curve is indicative of the amount of reactant, it was necessary to establish which of the thermal peaks associated with the decomposition of calcium oxalate was the most satisfactory for its quantitative determination.

METHODS

MATERIALS—

Seven lichen species were examined, namely, *Aspicelia calcaria, Caloplaca heppiana, Physcia adscendens, Physcia caesia, Rhizocarpon calcareum, Verrucaria nigrescens* and *Xanthoria parietina*. The samples, which were obtained by scraping the surface of lichen-colonised limestone, were dried at 40° C and ground to pass through a 0·5-mm sieve.
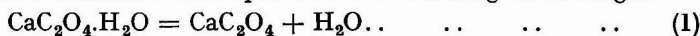
PROCEDURES—

The differential thermal analysis curves were determined under controlled-atmosphere conditions; equipment and experimental details have been described elsewhere by Stewart, Birnie and Mitchell.[3] The thermogravimetric characteristics of the lichens in a nitrogen atmosphere were also determined by using a TR 01 Stanton thermobalance from which the differential thermogravimetric curves were derived.

RESULTS

The complete-combustion curve for calcium oxalate monohydrate (Fig. 1, curve D) exhibits an endothermic peak at 243° C, indicating the loss of water of hydration. The series of exothermic effects between 450 and 500° C traces the oxidation of the oxalate to carbonate, and the decomposition of the latter to calcium oxide is shown by the 900° C endothermic peak.

In an inert atmosphere calcium oxalate decomposes in the following three stages—

$$CaC_2O_4.H_2O = CaC_2O_4 + H_2O.. \qquad .. \qquad .. \qquad .. \qquad (1)$$

$$CaC_2O_4 = CaCO_3 + CO \quad .. \qquad .. \qquad .. \qquad .. \qquad (2)$$

$$CaCO_3 = CaO + CO_2 \quad .. \qquad .. \qquad .. \qquad .. \qquad (3)$$

All three reactions are endothermic and the heats of reaction ($H°$ 298° K, kcal. per mole) are 13·3, 15·5 and 42·5, respectively (Simons and Newkirk[4]). Under the dynamic heating conditions of differential thermal analysis, the peaks associated with these reactions occur at about 235°, 500° and 900° C (Fig. 2, curve D). Thus, in the interpretation of the thermal curves of lichens, endothermic effects in the 150° to 250° C region are regarded as indicative of the dehydration of calcium oxalate, effects between 400° and 600° C of oxalate decomposition, and in the 700° to 900° C range of carbonate decomposition. Reasons for the fluctuations between the peak temperatures of calcium oxalate alone and the peaks on the lichen curves due to the presence of calcium oxalate will be dealt with later.

The thermal characteristics of the seven lichen species fall into three well defined groups, depending on the amount of calcium oxalate and calcium carbonate present. Thus *Physcia adscendens*, *Physcia caesia* and *Xanthoria parietina*, which contain small amounts of calcium oxalate, have closely similar thermal curves, as do the oxalate-rich species *Rhizocarpon calcareum*, *Aspicilia calcaria* and *Caloplaca heppiana*. The *Verrucaria nigrescens* sample is unique in that it contains an appreciable amount of calcium carbonate.

EFFECTS IN AN OXYGEN ATMOSPHERE—

The complete-combustion curves for *Physcia adscendens* and *Verrucaria nigrescens* (Fig. 1, curves A and B) exhibit exothermic peaks at about 300°, 430° and 500° C and the peak areas decrease with increasing temperature. Complete-combustion patterns of this type are common for fresh plant material and are thought to reflect concentrations of carbohydrates of high molecular weight.[2] The combustion curve of *Rhizocarpon calcareum* (curve C) is
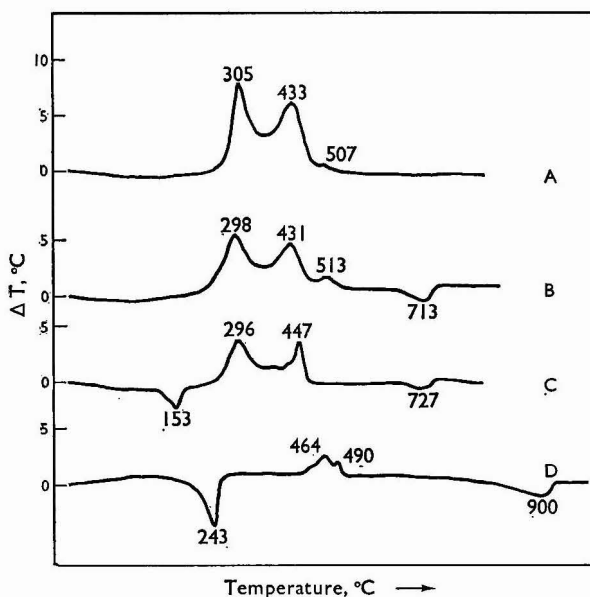

Fig. 1. Differential thermal analysis curves in an oxygen atmosphere (10 ml per minute) for: A, *Physcia adscendens*; B, *Verrucaria nigrescens*; C, *Rhizocarpon calcareum*; and D, calcium oxalate monohydrate, (15-mg samples diluted with 50 mg of calcined kaolin, loosely packed; recording sensitivity, 170 μvolts per inch; heating rate, 10° C per minute)

markedly different. The endothermic peak at about 150° C indicates the dehydration of calcium oxalate, and the exothermic system is limited to peaks at 296° and 447° C.   The latter peak, from a comparison with that of curve D, is affected by the combustion of calcium oxalate, but the complete-combustion curves are obviously not satisfactory for the un-ambiguous detection of calcium oxalate.

EFFECTS IN A NITROGEN ATMOSPHERE—

Temperature range 150° *to* 250° *C*—The small endothermic peaks at 175° and 177° C on the pyrolysis curves of *Physcia adscendens* and *Verrucaria nigrescens* indicate that these materials contain only small amounts of oxalate (Fig. 2, curves A and B).   The dehydration of the oxalate in these two lichen species occurs 50° to 60° C lower than that in *Rhizocarpon calcareum* (curve C) and that of pure calcium oxalate (curve D).   Further, the dehydration of the oxalate in *Physcia* and *Verrucaria* occurs in a single stage.   The reason for the doubling of the dehydration peak, which is particularly marked in the curve for *Rhizocarpon* (Fig. 2,
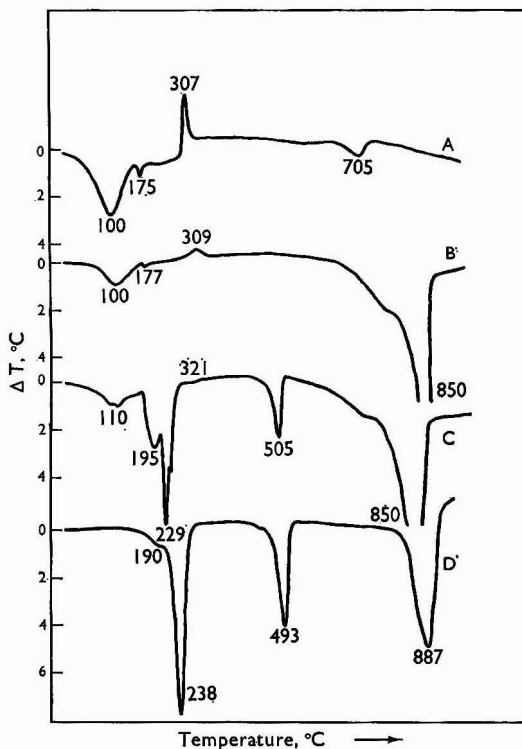

Fig. 2. Differential thermal analysis curves in a nitrogen atmosphere (200 ml per minute) for: A, *Physcia adscendens*; B, *Verrucaria nigrescens*; C, *Rhizocarpon calcareum*; and D, calcium oxalate monohydrate, (50-mg samples diluted with 120 mg of calcined kaolin, hard packed; recording sensitivity, 70 μvolts per inch; heating rate, 10° C per minute)

curve C), is not absolutely clear.   Substantial amounts of magnesium oxalate dihydrate are known to accumulate in certain plant tissue.   This magnesium salt dehydrates in the 200° C region.   The presence of magnesium oxalate in the lichens would result also in an endothermic peak between 600° and 700° C, marking decomposition to magnesium carbonate, and there is no indication of such a peak.   Further, the magnesium contents of these lichens were determined chemically and found to be very low, indeed that of *Rhizocarpon* was the lowest, being 149 p.p.m.   *Physcia* and *Verrucaria* contained 500 and 974 p.p.m., respectively.   De-hydration of oxalic acid dihydrate occurs at 100° C and the monohydrate decomposes between

180° and 190° C.   Free oxalic acid, however, cannot be present in the lichens because the exchangeable calcium is high: *Rhizocarpon*, 898 milli-equivalents per 100 g; *Physcia*, 120 milli-equivalents per 100 g; and *Verrucaria*, 946 milli-equivalents per 100 g.

From evidence of infrared-absorption spectroscopy, not only is calcium oxalate the only oxalate present, but it appears to be exclusively in the monohydrate form.   Although the energy changes shown on a differential thermal analysis curve that indicate the loss of sorbed moisture in the sample are not necessarily reflected exactly by the differential thermogravi-metric curve, it is nevertheless significant that the differential thermogravimetric curves of *Rhizocarpon calcareum* and calcium oxalate (Fig. 3, curves C and D) give no indication of a two-stage dehydration of oxalate.   The variation in the dehydration pattern of oxalate, as reflected by the differential thermal analysis curve, may be related to the dilution and hard-packing technique developed for examination of the sample under inert-atmosphere conditions.   Hard packing would tend to restrict the egress of water vapour and the effect would be enhanced by increased concentration, particularly in the initial stages.
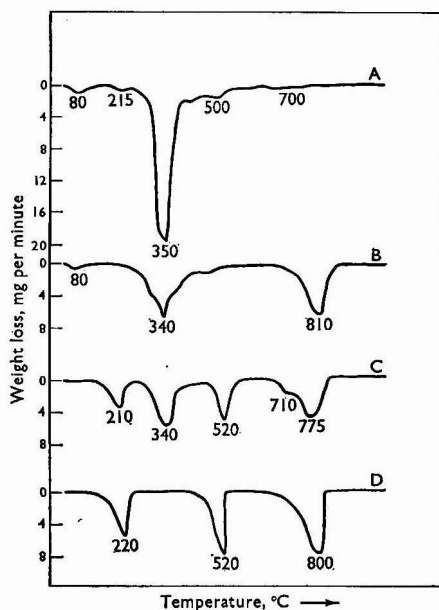

Fig.   3.   Differential   thermogravimetric curves in a nitrogen atmosphere (200 ml per minute) for: A, *Physcia adscendens*; B, *Verrucaria nigrescens*; C, *Rhizocarpon calcareum*; and D, calcium oxalate monohydrate, (sample weight, 100 mg; heating rate, 4° C per minute)

Temperature range 250° *to* 400° *C*—Substantial weight losses between 300° and 400°C are shown on the differential thermogravimetric curves of lichens determined in an inert atmos-phere (Fig. 3, curves A to C), *Physcia adscendens* losing 52 per cent., and *Rhizocarpon calcareum*, 21 per cent.   There is no weight loss in this region for calcium oxalate (curve D).   Infrared spectroscopy indicated that the *Physcia* species contained considerably larger amounts of carbohydrate than the *Rhizocarpon* species.   The pyrolysis curves of cellulose and carbo-hydrates of low molecular weight exhibit well defined endothermic peaks about 300° C,[5] and attributing, therefore, the weight loss in the 350° C region to carbohydrate decomposition, it is noteworthy that no endothermic effect is recorded in this region of the differential thermal analysis curves of lichens under inert atmosphere conditions (Fig. 2, curves A to C).   There

is, however, an exothermic reaction, whose intensity decreased with reduction in carbohydrate content.   As a peak on a differential thermal analysis curve need not necessarily represent a single reaction, but may be the record of the summation of simultaneous reactions, it is conceivable that the endothermic effect associated with the initial decomposition of the carbohydrate component is completely masked by an exothermic reaction involving the products of decomposition.   As care was taken to exclude oxygen in these determinations, the exothermic peaks probably resulted from auto-oxidation by organic components of the lichens.

Temperature range 400° to 600° C—The absence of an endothermic peak in the region of 500° C on the differential thermal analysis curves of *Physcia adscendens* and *Verrucaria nigrescens* (Fig. 2, curves A and B) is in accord with a low calcium oxalate content.   However, the differential thermogravimetric curve of the former (Fig. 3, curve A) shows a weight loss about this temperature which, by comparision with that for calcium oxalate, corresponds to a content of less than 5 per cent.   The corresponding weight loss for *Rhizocarpon* (curve C) is equivalent to a calcium oxalate content of 60 per cent.

Temperature range 700° to 950° C—The pyrolysis curves of *Verrucaria nigrescens* and *Rhizocarpon calcareum* in a nitrogen atmosphere (Fig. 2, curves B and C) exhibit a large endothermic peak at 800° C resulting from the decomposition of calcium carbonate, which in the latter species is derived from calcium oxalate.   However, for the *Verrucaria* sample the calcareous substratum is probably the principal source.   The carbonate decomposition peak on the curve of *Physcia adscendens* (curve A) is much smaller and occurs at a considerably lower temperature (705° C), both features being in accord with a lower calcium oxalate content.
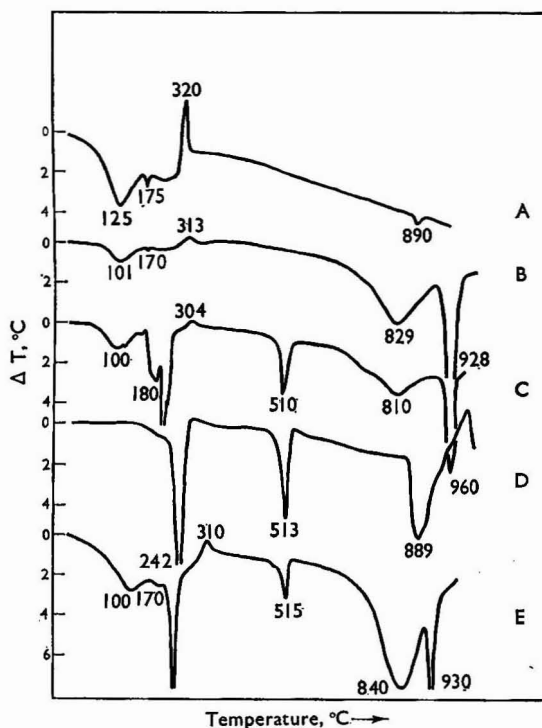

Fig. 4.   Differential thermal analysis curves in a carbon dioxide atmosphere (200 ml per minute)   for: A, *Physcia adscendens*; B, *Verrucaria nigrescens*; C, *Rhizocarpon calcareum*; and D, calcium oxalate monohydrate; E, 1 + 1 mixture of *Physcia adscendens* and calcium oxalate monohydrate, (50-mg samples diluted with 120 mg of calcined kaolin, hard packed; recording sensitivity, 70 μvolts per inch; heating rate, 10° C per minute)

EFFECTS IN A CARBON DIOXIDE ATMOSPHERE—

The differential thermal analysis curves of lichens determined in a carbon dioxide atmosphere (Fig. 4, curves A to C) are similar to those observed in a nitrogen atmosphere, except in the 700° to 950° C range of curves B and C for *Verrucaria* and *Rhizocarpon*, respectively, where a doubling of the peak reflecting carbonate decomposition occurs. Although the curves for *Physcia* and calcium oxalate monohydrate (curves A and D) do not show this doubling phenomenon, that of a mixture (1 + 1) of these materials (curve E) does possess a complex high temperature - peak system, but only when the lichen and oxalate have been intimately mixed (ground for 1 minute in a vibratory ball mill). The lichen samples on reaching 700° C consist essentially of a carbonaceous residue *plus* calcium carbonate and, as the temperature is raised, increasing amounts of carbon dioxide will be produced as decomposition of the latter proceeds. The high temperature - peak pattern could arise from the oxidation of the carbon residue by carbon dioxide, and consequently would represent an endothermic effect with a superimposed exothermic reaction. This effect, however, only occurs under a dynamic carbon dioxide atmosphere when an additional and substantial amount of carbon dioxide is produced *in situ*, as in the decomposition of samples containing appreciable amounts of calcium carbonate. Under these conditions, the voids in the sample well are completely filled with carbon dioxide. The doubling effect is not observed when nitrogen is the controlling atmosphere, but in this instance the carbon dioxide from carbonate decomposition would diffuse more easily from the reaction site.

DISCUSSION

The results show that calcium oxalate in naturally occurring materials such as lichens may be unambiguously identified on the differential thermal analysis curve, but only when determined in a nitrogen atmosphere. Comparison of the weight loss in the 500° C region of the differential thermogravimetric curves of the lichens with the corresponding weight loss on the differential thermogravimetric curve for a known quantity of calcium oxalate provides a simple and direct estimate of the oxalate in the lichens. The oxalate content of these plants was also determined chemically by a micro method[6] and it will be noted (Table I) that while agreement between thermogravimetry and the chemical method is good for high concentrations of oxalate, at low levels the former method is less satisfactory.

TABLE I

AMOUNTS (PER CENT.) OF CALCIUM OXALATE AND CALCIUM CARBONATE IN LICHENS DETERMINED BY DIFFERENTIAL THERMOGRAVIMETRIC, DIFFERENTIAL THERMAL ANALYSIS AND CHEMICAL METHODS

	Calcium oxalate, per cent., determined by—			Calcium carbonate, per cent., determined by—	
	differential thermo-gravimetric method	differential* thermal analysis method	chemical method	differential† thermo-gravimetric method	chemical‡ method
Physcia adscendens	5	2·7	2·2	2·3	<5
Verrucaria nigrescens	nil	0·8	1·4	52	54
Rhizocarpon calcareum	60	60	56	7·0	11
Aspicelia calcarea	42	48	39	21	24

* Determined from 170° C peak area for *Physcia* and *Verrucaria* (Fig. 2, curves A and B) and from 500° C peak area for *Rhizocarpon* (Fig. 2, curve C).
† Obtained by subtracting the calculated weight loss for calcium oxalate derived carbonate from total weight loss between 700° and 800° C (Fig. 3, curves A to C).
‡ Obtained by treating the dry matter with 0·1 N hydrochloric acid overnight, filtering, and then back-titrating the unused acid with alkali.

Differential thermal analysis can also provide a quantitative result as the area enclosed by a peak is proportional to the amount of reactant. As the 500° C peak on the differential thermal analysis curve of calcium oxalate resulting from the oxalate - carbonate reaction is, unlike the dehydration peak, invariably free from doubling, it may be used for the quantitative determination of oxalate by simply measuring the area of this peak on the lichen curve and comparing this with the peak area for the known amount of calcium oxalate monohydrate (see, *e.g.*, *Rhizocarpon calcareum*, Table I). For low levels of oxalate this 500° C peak is either

absent or poorly defined (*Physcia adscendens* and *Verrucaria nigrescens*, Fig. 2, curves A and B). Here, however, oxalate dehydration is recorded as a single peak (170° C), and integration of this provides a good assessment of the oxalate content of the lichen (Table I). Indeed, the agreement between differential thermal analysis and the chemical determinations of oxalate is remarkably close. Finally, consideration of the weight losses on the differential thermogravimetric curves associated with carbonate decomposition in relation to the effects on these curves resulting from the first and second stages of calcium oxalate decomposition, enables an assessment to be made of the non-oxalate derived carbonate (Table I), which is in good agreement with the chemical determination of this component.

Ability to distinguish these two forms of calcium in the sample has shown that the three lichen species containing appreciable amounts of calcium oxalate, namely, *Aspicelia calcaria*, *Caloplaca heppiana* and *Rhizocarpon calcareum* are confined to calcareous rocks (*i.e.*, obligate calicoles), whereas *Physcia adscendens*, *Physcia caesia* and *Xanthoria parietina*, which contain 5 per cent. or less of calcium oxalate, are ubiquitous.

These results, supported by other unpublished evidence, show that thermal techniques provide a rapid and reliable means of determining oxalate in plant materials. Magnesium oxalate commonly occurs with calcium oxalate in plants; however, there is no indication of the presence of magnesium salt in the lichen species examined, all the evidence pointing to the oxalate being present exclusively in the form of the calcium oxalate monohydrate.

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The Proportion of 2-Methylbutanol and 3-Methylbutanol in Some Brandies and Whiskies as Determined by Direct Gas Chromatography

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Stationary phases suitable for the separation of 2-methylbutanol ("active" pentanol) and 3-methylbutanol (isopentanol) are discussed. The most suitable for the determination of these alcohols in potable spirits by direct injection of samples are diethyl tartrate and polyethylene glycol 200. Polyethylene glycol 200 is preferred because other congeners can be determined at the same time. With n-pentanol as an internal standard, 65 samples of cognac brandies, Scotch and other whiskies have been examined on one or other of these stationary phases. The sum of the two pentanol isomers determined separately agrees well with their determination as a single peak on polyethylene glycol 1500. The ratio of the concentrations of the isomers appears to be characteristic of the type of spirit.

IN a previous paper[1] it was demonstrated that the principal higher alcohols (n-propanol, isobutanol and "isopentanol") in cognac brandies and Scotch whiskies occur in relative proportions that are, within a narrow range, characteristic of each type of spirit. It was suggested that determination of these proportions might be of use in identifying spirits analytically if sufficient results covering a wider range of spirituous products were available. The precise biochemical mechanism of formation of these alcohols and other congeners[2] has been reviewed recently, and is still a matter for controversial discussion and further investigation. From a practical standpoint however, it is reasonable to suppose that under the precisely maintained conditions of fermentation of closely controlled starting materials and with the subsequent careful distillations that are carried out by producers of high quality spirituous beverages, many congeners will occur in consistent proportions to each other, according to the type of product. The actual amounts of the congeners may be expected to vary, for example, as the proportion of starch to protein varies in the wort or must, and according to the fractionation used during distillation. Consistency in any one type of spirit is aided by the judicious blending which tends to even out random changes.
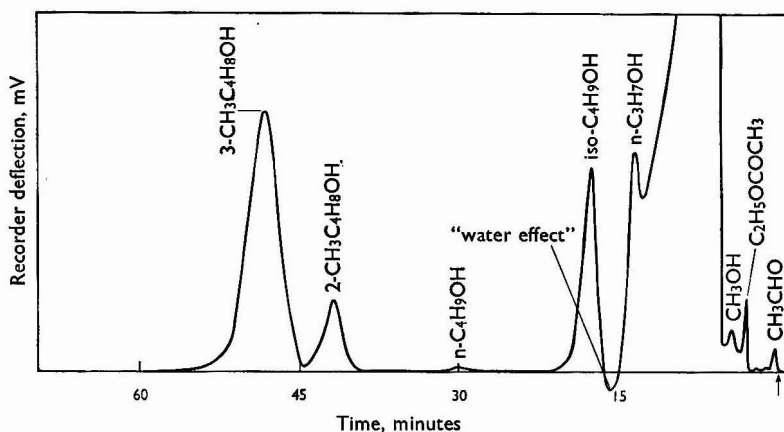

Fig. 1. Chromatogram of a ~~Scotch all malt whisky on polyethylene glycol 200~~ cognac brandy on diethyl tartrate

The analyses reported previously did not distinguish between 2-methylbutanol and 3-methylbutanol. These isomers are often considered together as "isopentanol," although this name is sometimes applied colloquially to the 3-methyl isomer to distinguish it from the 2-methyl isomer. The latter is sometimes referred to as "active" pentanol because of its asymmetric carbon atom. These isomers have close boiling-points, and are unlikely to be separated in the distillation of spirituous beverages. It was therefore thought of interest to investigate their presence individually in spirits of the type already examined.

EXPERIMENTAL

Stationary phases suitable for the separation of the isopentyl isomers have been discussed by Dinsmoor and Webb[3] with special reference to the analysis of sherry concentrates. Glycerol,[4] "Tide"[5] and triethanolamine[6] have also been used. These stationary phases had disadvantages. Efficient columns were seldom obtained, and packing the columns was sometimes difficult because of the stickiness of the prepared Celite. Even when efficient columns were obtained, long retention times were necessary for good separation and the base-line disturbance attributed to the water injected, and the large ethanol peak, both overlapped the pentanol peaks.

All the stationary phases capable of separating the isomers are polar, and therefore several polar esters were investigated. Only partial separation was obtained on 6 foot × ¼ inch, 10 per cent. dibutyl tartrate and diethyl citrate columns, but excellent separation was obtained with a similar column of 10 per cent. diethyl tartrate at 55° C. As the column aged, the earlier peaks of n-propanol and isobutanol merged with the ethanol peak, but the separation of the pentanol isomer remained satisfactory over 400 hours of use. Since this work was started, Prabucki and Pfenninger[7] have also described this use of diethyl tartrate for the study of extracts of spirits and beers. More recently in this laboratory in an investigation of the most volatile congeners, it was found that a 20 per cent. column of polyethylene glycol 200 gives excellent separation of the pentanol isomers at 70° C. The "bleed" of stationary phase is lower than with the diethyl tartrate column, and other congeners may be determined on the same chromatogram. The column lasts for more than 600 hours. Chromatograms on the two selected stationary phases are shown in Figs. 1 and 2, and the relative retention of a number of compounds on polyethylene glycol 200, as determined in solution in 40 per cent. v/v ethanol, are given in Table I.

TABLE I

RELATIVE RETENTION OF SOME COMMON COMPOUNDS ON POLYETHYLENE GLYCOL 200

Substance	Relative retention*	
Methanol	0·366	
Ethanol	0·446	
Isopropanol	0·434	Obscured by ethanol
s-Butanol	0·702	
Isobutanol	1·000	
n-Butanol	1·481	
2-Methylbutanol	1·98	
3-Methylbutanol	2·12	
n-Pentanol	2·70	
Acetaldehyde	0·078	Tends to tail as column ages
n-Propionaldehyde	0·154	Not separated from acetone
Isobutyraldehyde	0·262	
Isovaleraldehyde	0·89	
Acetal	0·169	Obscured by ethyl acetate
Acetone	0·150	Not separated from propionaldehyde
Ethyl formate	0·121	
Ethyl acetate	0·172	
Ethyl isobutyrate	0·26	
Ethyl isovalerate	0·47	Obscured by ethanol
Ethyl hexanoate	1·23	
n-Propyl acetate	0·29	
Isobutyl acetate	0·36	Obscured by methanol
Isopentyl acetate	0·66	

* Isobutanol = 1·00.

METHOD

This was similar to that previously described.[1]   The samples, adjusted to exactly 40 per cent. by volume of ethanol by addition of water or pure ethanol, were mixed with one-tenth of their volume of pure 40 per cent. ethanol containing 500 mg of pure n-pentanol per ml as an internal standard.   Columns were either a 6 foot × ¼-inch copper column of diethyl tartrate, 10 parts and Celite (80 to 100 mesh), 90 parts, or a 9 foot × ⅛-inch (0·08 i.d.) stainless-steel column of polyethylene glycol 200, 20 parts and Celite (100 to 120 mesh), 80 parts. Operating conditions for diethyl tartrate were: carrier gas, nitrogen at a flow-rate of 70 ml per minute at 55° C, and for polyethylene glycol 200, nitrogen at a flow-rate of approximately 18 ml per minute at 70° C.   The polyethylene glycol 200 was obtained directly from Messrs. Union Carbide Ltd. (samples obtained from chemical wholesalers were not always satisfactory and sometimes had an opaque semi-solid appearance).   For analysis, 2 or 5 μl of the sample mixture were used and the results obtained compared with standards prepared in 40 per cent. v/v ethanol.
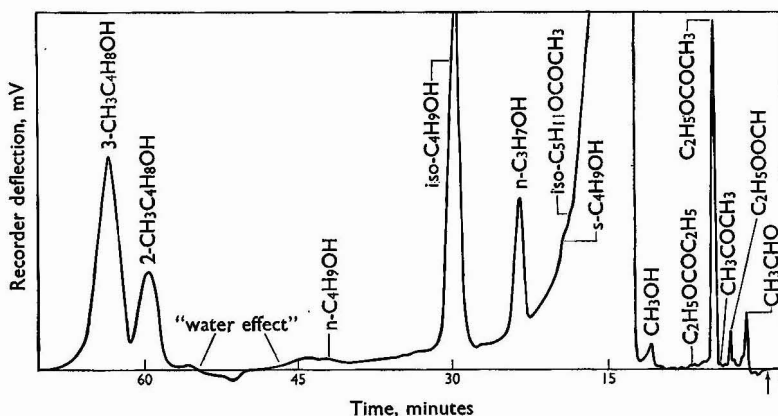

Fig. 2.   Chromatogram of a ~~cognac brandy on diethyl tartrate~~ *Scotch all-malt whisky on polyethylene glycol 200.*

RESULTS

The results are shown in Table II.   The spirits are of a similar type to those previously analysed for the principal higher alcohols.[1]   The whiskies, type A, are blended Scotch whiskies, and are all popular blends consumed in Great Britain.   The malt whiskies are types used for blends and are all "single" whiskies, *i.e.*, the product of an individual "batch."   Some of

TABLE II

2-METHYLBUTANOL AND 3-METHYLBUTANOL IN BRANDY AND WHISKY

2-Methylbutanol, parts per 100,000 of absolute ethanol	3-Methylbutanol, parts per 100,000 of absolute ethanol	Sum	Sum, as determined on polyethylene glycol 1500	Ratio, 2-Methylbutanol / 3-Methylbutanol
A. *Blended whiskies*—				
22	57	79	83	0·38
25	62	87	79	0·41
26	60	86	89	0·44
21	59	80	79	0·36
21	57	78	75	0·37
24	62	86	84	0·39
22	56	78	77	0·38
20	53	73	75	0·37
22	56	78	75	0·41
24	60	84	87	0·39
24	58	82	80	0·41
25	65	90	92	0·38
24	67	91	95	0·36
21	54	75	74	0·39

TABLE II—*continued*

	2-Methylbutanol, parts per 100,000 of absolute ethanol	3-Methylbutanol, parts per 100,000 of absolute ethanol	Sum	Sum, as determined on polyethylene glycol 1500	Ratio, 2-Methylbutanol / 3-Methylbutanol
B. *Malt whiskies*—					
	63	162	225	218	0·39
	64	166	230	224	0·39
	67	152	219	217	0·44
	64	162	226	220	0·39
	81	188	269	265	0·43
	56	169	225	220	0·33
	63	142	205	200	0·44
	55	146	201	202	0·38
	52	137	189	185	0·38
	51	118	169	164	0·43
	52	134	186	180	0·39
	63	149	212	213	0·42
	71	171	242	233	0·41
	61	164	225	219	0·37
C. *Other whiskies*—					
U.S. Bourbon	137	354	491	500	0·39
U.S. Bourbon	130	356	486	475	0·37
Canadian ..	22	61	83	88	0·36
Canadian ..	16	36	52	—	0·44
Dutch ..	10	31	41	46	0·32
Dutch ..	10	27	37	39	0·37
Dutch ..	12	33	45	42	0·36
D. *Cognac brandies, good quality*—					
	15	77	92	90	0·20
	37	169	206	210	0·22
	45	195	240	237	0·23
	38	170	208	210	0·22
	40	166	206	202	0·24
	35	166	201	196	0·21
	42	201	243	251	0·21
	43	197	240	250	0·22
	39	195	234	240	0·20
	34	189	215	211	0·19
	58	278	336	329	0·21
	34	164	198	202	0·21
	15	65	80	85	0·23
	38	174	212	207	0·22
	33	142	175	775	0·23
	29	126	155	162	0·23
	37	176	213	207	0·21
	26	110	136	141	0·24
	32	154	186	195	0·21
	36	162	198	190	0·22
E. *Brandies, inferior quality*—					
	32	121	153	156	0·26
	34	134	168	162	0·25
	28	113	141	134	0·25
	34	128	162	159	0·24
	36	145	181	177	0·25
F. *"Marc de Bourgogne" brandies*—					
	67	201	268	261	0·33
	58	199	257	252	0·29
	62	220	283	290	0·29
	53	174	227	231	0·30
	60	194	254	259	0·31

the malt whiskies had been matured for 6 years in sherry casks at the time of analysis and some had been bottled almost immediately after distillation, a few months before analysis. Re-examination 18 months after bottling revealed no change in composition during this time. No significant difference was found in the ratio of the pentanol isomers in the new and the old spirits, which are not therefore distinguished from each other here. The cognac brandies, "D," bear a good commercial reputation and are blends of matured spirits. The cognac

brandies, "E," possess less commercial esteem, and, as reported previously,[1] contain appreciable proportions of s-butanol and a larger proportion of methanol than usually found in higher quality spirits.

The grain whisky used for blending with Scotch malt whisky is the product of efficient fractional distillation, and contains a negligible amount of pentanols. As a result, there is little difference in the ratio of the pentanol isomers between the blends and the all-malt types. The whiskies produced outside Scotland are also the products of fermentation of unmalted grain, but have not been highly fractionated, and therefore contain pentanols. The ratio of the isomers in the few examples of these latter spirits examined falls within the range of that found for the Scotch whiskies and may be significant of a grain ferment, whether malted or not. The lowest proportion of the 2-methyl isomer is found in the good quality cognacs (0·19 to 0·24), and is significantly different from that of the inferior cognacs (0·24 to 0·26), and the "Marc de Bourgogne" (0·29 to 0·33).

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An Instrument for the Continuous Determination of Carbon Dioxide in High Purity Water*

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An instrument for the continuous determination of carbon dioxide in high purity water is described. The instrument consists essentially of a device for transferring the carbon dioxide from the water to a gas stream, which is then passed through an aqueous suspension of calcium carbonate. The pH of this suspension, which is proportional to the concentration of carbon dioxide in the gas stream and hence the original water, is recorded continuously.

Precision tests at levels of 20 and 60 μg per litre had a standard deviation of about 4 μg per litre.

THE corrosive effect of carbon dioxide in boiler feed-waters is largely suppressed by the addition of volatile amines to produce a high pH. Under conditions of high temperature and pressure, however, the carbon dioxide could react with the protective oxide film on the metal surfaces of the feed system, increasing the transport of iron and copper to the boiler and accelerating high temperature corrosion.

Manual methods of analysis for carbon dioxide of adequate precision and sensitivity are available,[1,2,3] but few continuous methods have been described. Bartley and Moult[4] proposed a method for the determination of carbon dioxide in steam by using an infrared gas analyser to determine the carbon dioxide present in the gases evolved from a conventional Straub de-gasser. Ehrenberg and Smit[5] have described a method involving continuous acidification of the sample, followed by equilibration with carbon dioxide-free air which is subsequently equilibrated, in turn, with water. Conductivity measurements are then made on the water. Neither of these methods has been applied to boiler feed-water.

The instrument described here is robust, reliable and does not require the continuous addition of reagents.

DESCRIPTION OF THE INSTRUMENT

A diagram of the instrument is shown in Fig. 1. The first part is based on a design proposed by Burdon[6] for stripping a "selected compound" from a sample of water by boiling and sweeping it into a measuring device. Component parts of the glassware are shown in Figs. 2, 3, 4 and 5.

The sample is passed through a cation-exchange resin to remove ammonia, which would interfere. It is then fed to the primary heating coil (1·45 kW) at a flow-rate of 220 ml per minute. This flow-rate is adjusted by altering the height of the constant head. The heated sample passes into the fractionating column, which is vacuum jacketed and filled with glass Raschig rings, where it is scrubbed by steam produced by the secondary heating coil. The steam and carbon dioxide then pass into the stripping column, which is also vacuum jacketed and filled with Dixon gauze rings. Carbon dioxide-free air is sucked in through a non-return valve by a small pump, thus sweeping the carbon dioxide into the final stage.

Toren and Heinrich[7] and Lodge *et al.*[8] have shown that the hydrogen-ion concentration of a suspension of calcium carbonate through which air is passing is proportional to the carbon dioxide content of the air. This system is used in the instrument, the pH being recorded continuously. The pH meter is a Pye Dynacap set to read from pH 8 to 10; the output of the meter is fed into a 500-μA current recorder, so that the effective chart width covers pH 8 to 9.

The flow-rate of the sweep gas is controlled at 60 ml per minute by an automatic flow regulator.

* Presented at a meeting of the Automatic Methods Group on Thursday, November 17th, 1966.
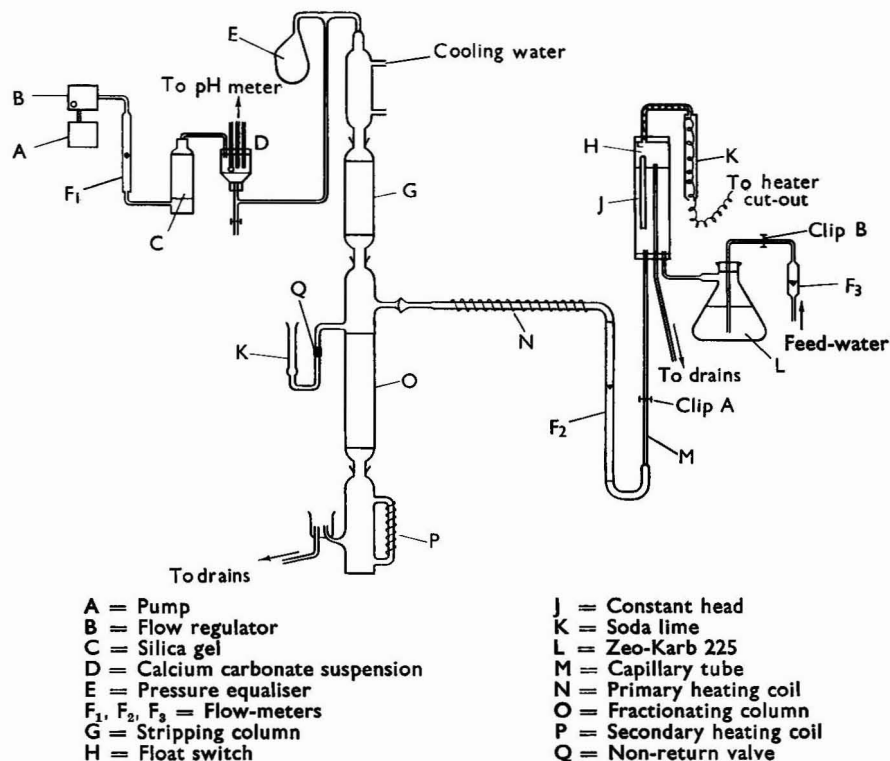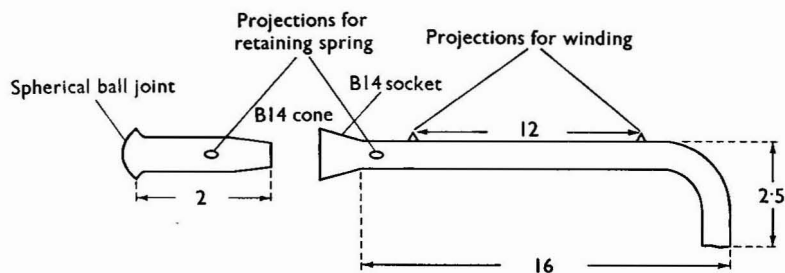
A = Pump
B = Flow regulator
C = Silica gel
D = Calcium carbonate suspension
E = Pressure equaliser
F_1, F_2, F_3 = Flow-meters
G = Stripping column
H = Float switch

J = Constant head
K = Soda lime
L = Zeo-Karb 225
M = Capillary tube
N = Primary heating coil
O = Fractionating column
P = Secondary heating coil
Q = Non-return valve

Fig. 1.   Diagram of the instrument

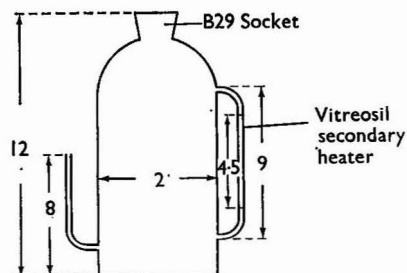Fig. 2.   Primary heater (Vitreosil) and connecting piece (Pyrex), (dimensions in inches)

Fig. 3.   Details of the re-boiler (dimensions in inches)
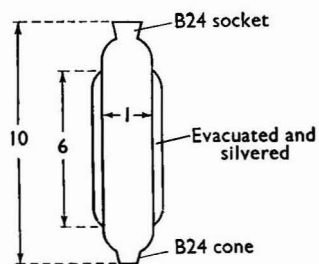
Fig. 4.   Details of stripping column (dimensions in inches)
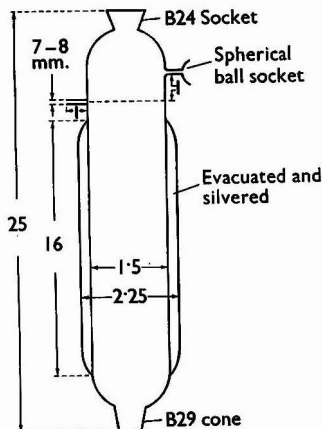
Fig. 5.   Details of fraction-
ating column (dimensions in inches)

In the event of loss of sample, the secondary heating coil will merely boil the water in
the re-boiler under reflux.   The primary heating coil, however, will become red hot.   For
this reason, a safety device, which consists of a float switch in the constant head, de-activates
a relay, switching off the primary heater when the water level falls.   The circuit diagram is
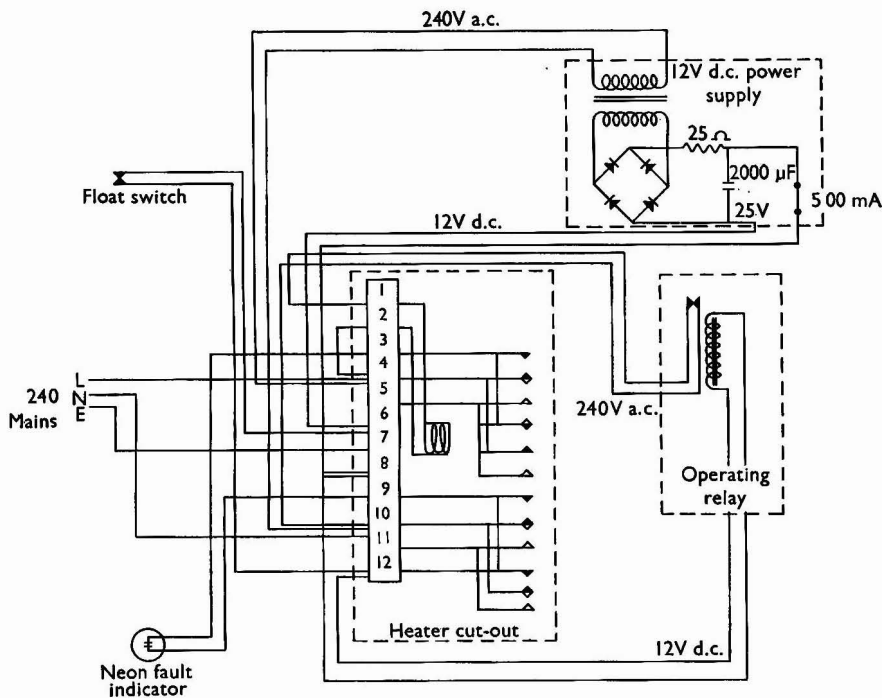shown in Fig. 6.

Fig. 6.   Circuit diagram of the automatic cut-off safety device

DISCUSSION

According to Henry's law, the mass of gas absorbed by a fixed volume of liquid at a given temperature is proportional to the (partial) pressure. As the air above the liquid in the instrument is free from carbon dioxide and is continually being renewed, the carbon dioxide will be removed from the water and transferred to the sweep gas. Heating to boiling accelerates this process.

The reactions involved when carbon dioxide dissolves in water are well known—

$$CO_2 + H_2O = H_2CO_3 = H^+ + HCO_3^-$$
$$HCO_3^- = H^+ + CO_3^{2-}$$
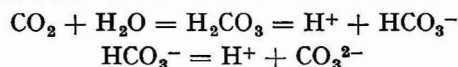
The equilibrium constants for these reactions are—

$$K_1 = \frac{(H^+)(HCO_3^-)}{(CO_2)}, \text{ and } K_2 = \frac{(H^+)(CO_3^{2-})}{(HCO_3^-)}.$$

Several workers have used a solution of sodium hydrogen carbonate to determine carbon dioxide in gas streams, *e.g.*, Maxon and Johnson,[9] and each group has found that the hydrogen-ion concentration is proportional to the carbon dioxide content. Assuming the activity coefficients to be unity and the hydrogen carbonate concentration to be high enough to be effectively constant, as K_2 is much smaller than K_1—

$$(H^+) = \frac{K_1}{C}(CO_2) \text{ where } K_1 \text{ and } C \text{ are constants.}$$

Toren and Heinrich[7] and Lodge *et al.*[8] also found a linear relationship by using a suspension of calcium carbonate in water, the hydrogen carbonate ion presumably being formed from the reactions—

$$CaCO_3 = Ca^{2+} + CO_3^{2-}$$
$$CO_3^{2-} + H_2O = HCO_3^- + OH^-$$
$$OH^- + CO_2 = HCO_3^-$$
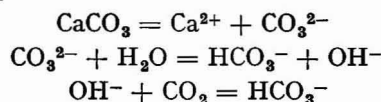
The advantage of the calcium carbonate suspension is that a decrease in the carbon dioxide content of the gas stream reduces the carbon dioxide content of the suspension to give a higher pH, whereas a sodium hydrogen carbonate solution does not readily lose carbon dioxide.

Johnson and Williamson[10] calculated the solubility of calcium carbonate with decreasing carbon dioxide concentration, and concluded that the solubility decreases to a minimum, after which the carbonate begins to decompose to calcium hydroxide and the calcium-ion concentration increases again. The pH will, of course, rise rapidly when this occurs. According to these authors, the minimum occurs at a partial pressure of 4×10^{-7} atmospheres of carbon dioxide. If this figure is calculated back to a water sample flowing through the instrument, the corresponding carbon dioxide concentration in the water is about 0.2 μg per litre. Below this concentration, therefore, the pH of the suspension can be expected to rise more rapidly.

EXPERIMENTAL

The original Burdon design of the fractionating columns proposed blowing the sweep gas through to the measuring device, but it was found that the system rapidly became unstable, the rather vigorous boiling of the water blowing the sample back out of the primary heater. A suction arrangement through a non-return valve with a rubber bag to act as as a pressure stabiliser proved satisfactory.

In early trials with the instrument a sample flow-rate of 220 ml per minute and a sweep gas flow-rate of 30 ml per minute were used. It was necessary to have a vent to the atmosphere on starting to allow the gases expanded from the system to escape. Increasing the gas flow-rate to 60 ml per minute removed the necessity for a vent, but reduced sensitivity. However, the starting procedure is simplified.

It was originally intended to obtain a zero by inserting a soda-lime tube in the gas stream immediately before the pH cell. When this was done, the reading obtained was much higher than expected. This was obviously caused by the effect predicted by Johnson and Williamson when the partial pressure of carbon dioxide fell below 4×10^{-7} atmospheres. An alternative method of establishing the zero is to remove carbon dioxide from the sample before it enters

the instrument. However, attempts to remove carbon dioxide from condensate with a small, mixed bed de-mineralisation column were not successful. A column, 40 cm high and 2·5 cm in diameter only removed about half of the carbon dioxide from water containing 70 μg per litre. Further tests with other resins showed that the best result, a reduction to less than 1 μg per litre, could be obtained by using two columns in series, the first filled with mixed bed and the second with De-Acidite FF. The zero of the instrument is therefore obtained by installing two such columns in the sample line. Zero tests have not been carried out at values higher than 70 μg per litre.

The effect of ambient temperature changes on the response of the instrument was not investigated in detail. However, variations from 15° to 25° C produced no noticeable change in the results obtained during tests. The automatic temperature compensation facility of the pH meter was used to correct for glass electrode temperature effects.

CALIBRATION—

The difficulties involved in making and storing large quantities of water of a known carbon dioxide content made a dynamic method of calibration preferable. The instrument was installed in a mixed bed de-mineralisation plant, the water produced by the plant being used as the dilution water for standard solutions of sodium carbonate that were introduced by a small pump. Volumes, ranging from 1 to 5 ml per minute, of 0·005, 0·002 and 0·001 N sodium carbonate solutions were added to de-mineralised water flowing at 300 ml per minute. To confirm that the sodium carbonate was satisfactorily converted into carbon dioxide by the cation-exchange resin, samples of the effluent from the instrument were analysed with a flame photometer. No sodium was detected at any of the concentrations used.

The calibration procedure was to obtain a steady reading with de-mineralised water as the sample, and then pump in standard sodium carbonate solutions in random order, noting the pH obtained at each level. Steady pH readings were normally obtained about 20 minutes after adjusting the flow-rate. The carbon dioxide content of the de-mineralised water was determined by a standard manual method,[2] and this value subtracted as a blank.

The results obtained (Table I) show a straight line relationship between carbon dioxide and hydrogen-ion concentration. A regression analysis gave the equation of the regression line as $x = 82·26y - 140·5$, where $x = \mu$g of carbon dioxide per litre and $y = H^+ \times 10^{-9}$. The standard deviation about the regression line is 2 μg per litre, and the standard error of the regression coefficient is $\pm 0·9$ μg per litre.

TABLE I

CALIBRATION RESULTS

Carbon dioxide content of de-mineralised water, μg per litre	Carbon dioxide added, μg per litre	pH	$H^+ \times 10^{-9}$
5	152	8·44	3·63
5	285	8·28	5·25
5	50	8·62	2·40
5	183	8·40	3·98
5	73	8·57	2·69
5	146	8·45	3·55
5	102	8·53	2·95

PRECISION TESTS—

A knowledge of the precision obtainable at the two main levels encountered in modern power stations was desirable, i.e., 20 μg per litre, a typical economiser inlet value and 60 μg per litre, a typical condensate value.

Sodium carbonate solutions were added, as before, to a basic supply of de-mineralised water. Table II shows the results obtained, the standard deviations being 4·1 μg per litre at the 20 μg per litre level and 3·6 μg per litre at the 60 μg per litre level.

Further tests were then carried out in a power station with water taken from the condenser of a 120-mW generator. Samples were taken simultaneously for manual determinations by the standard method. The results, which are detailed in Table III, have a standard deviation of 3·0 μg per litre. For this series, the zero was obtained by using the combined de-ionisation columns recommended.

The standard deviation obtained with the instrument at the levels likely to be found in feed water is about ± 4 μg per litre. This compares with a figure of better than ± 2 μg per litre for the manual method.[2] However, the instrument requires little attention, and provides a continuous record of carbon dioxide concentrations.

TABLE II

PRECISION RESULTS IN LOGICAL ORDER

Carbon dioxide content of de-mineralised water, μg per litre	Carbon dioxide added, μg per litre	Carbon dioxide found, μg per litre	Difference, μg per litre
5	23	23	−5
3	23	30	+4
2	24	22	−4
2	20	22	0
2	20	16	−6
3	52	56	+1
2	57	60	+1
2	55	52	−5
2	53	60	+5
2	53	56	+1

TABLE III

SIMULTANEOUS DETERMINATIONS ON CONDENSATE

Manual determination, μg per litre	Instrument, μg per litre	Difference, μg per litre
22·6	24	+1·4
28·8	24	−4·8
25·4	28	+2·6
18·7	20	+1·3
25·0	24	−1·0

OPERATION—

The instrument is started by switching on the pump, pH meter and recorder, and opening clips A and B. The sample flow is adjusted to between 300 and 500 ml per minute and the heaters are switched on.

When closing down, the heaters are switched off and clips A and B closed. It is convenient to leave the pump and pH meter switched on to keep the interior free from carbon dioxide and to reduce the response time on starting. It is possible to check for leaks in this way as the pH should rise above 9·2.

Once started, the instrument needs no further attention, providing the sample flow exceeds 220 ml per minute. Below this value the float switch operates to switch off the primary heater. If indicating soda lime is used for the guard-tubes the colour change will show when they should be replaced. The life of the cation-exchange resin will depend on the ammonia content of the water. Unusually low results are an indication that regeneration is required, but it is preferable to replace or regenerate the resin on a routine maintenance basis, the frequency being determined by the known ammonia content and the flow through the instrument. The calcium carbonate should be replaced about once a month as it becomes slimy, although no adverse results have been obtained. The drift of the instrument is governed by the stability of the pH meter. Variations of less than $\pm 0·005$ pH units over 2 hours have been obtained when making zero checks, but it is advisable to check the zero daily.

APPLICATIONS AND PERFORMANCE

The instrument can be used for the continuous determination of carbon dioxide in high purity waters of the type used for feeding boilers in power stations. It is installed in a cabinet to prevent mechanical damage, and it has been used in power stations for about 2 years, proving to be robust and reliable in operation. The instrument has functioned satisfactorily on samples varying in temperature from 15° to 32° C.

Appendix

CONSTRUCTIONAL DETAILS OF THE INSTRUMENT

The framework of the instrument is made of aluminium angle and is about 5 feet high, 3 feet wide and 2 feet deep.

Most of the internal parts are glass with the exception of the heaters, which are Vitreosil wound with nichrome wire and covered with aluminous cement. Flexible connections are made in neoprene. The dimensions of the heaters and columns are not thought to be critical. However, details are as follows. Primary heater (1·45 kW), length 16 inches, bore 13 mm nominal; secondary heater (132 W supplied by a 50-V transformer), length 4·5 inches, bore 13 mm nominal; re-boiler, height 12 inches, internal diameter 2 inches with B29 socket; fractionating column (vacuum jacketed), height 25 inches, internal diameter 1·5 inches with B29 cone and B24 socket; stripping column, height 10 inches, internal diameter 1 inch with B24 cone and socket.

The pH cell consists of a sintered-glass crucible (35-ml capacity) containing 1 g of analytical-reagent grade calcium carbonate, and sufficient distilled water to cover the electrodes. The glass electrode is a screened general-purpose electrode, E.I.L., type GHS33. Other items are an Austen Dymax Mark 1 pump, a Platon Flostat Minor flow regulator, a Pye Dynacap pH meter, a Record 500-μA recorder and a Rotameter flow-meter.

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The Determination of Salt in Bacon by Using a Sodium-ion Responsive Glass Electrode

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Rapid determination of the salt content of cured meat products with a sodium-ion responsive electrode is described and discussed. The method enables the percentage of salt on water content to be measured directly on the meat in a few minutes. Many determinations can be made cheaply and accurately enough for purposes of routine factory control.

INCREASING amounts of bacon are now being retailed, ready sliced, in vacuum packs. This bacon may be produced by automatic or semi-automatic production methods.[1] To obtain a product that has a satisfactory shelf life and is organoleptically acceptable it is necessary to control the salt content of the bacon within close limits. This control is aided by rapid and accurate analysis soon after production.

Existing chemical methods are fairly slow and demand experience to reach the required accuracy. An electrochemical method for determining the sodium chloride concentration directly in bacon would give a rapid result in terms of salt on water without weighing. A recent publication[2] describes a method for the determination of the chloride content of cured meat products, but this method requires the weighing and dissolution of the sample.

Sodium-ion responsive glass electrodes are now available commercially and have been used in many analytical applications.[3] It was considered worthwhile to attempt to determine the sodium-ion concentration in bacon by simple contact of the muscle with the electrode. This type of electrode in conjunction with a standard electrode has a potential, E, varying with the sodium ion activity, A_{Na}, according to the equation—

$$E = \text{Constant} + \frac{2.3 \, RT \log A_{Na}}{F}.$$

This potential can be determined in the same way as the potential of a hydrogen-ion sensitive glass electrode, by using a calomel reference electrode and a high impedance voltmeter. The salt content of lean bacon expressed as a percentage of the water content can vary from 2 to 10 per cent., and over this concentration range the sodium-ion activity coefficient varies considerably. To use this electrode analytically, it is therefore necessary to use a calibration curve prepared from salt solutions of known concentration. Because of the temperature dependence of the potential, the calibration curve should be determined at the same temperature as the actual determination.

The membrane of the type of glass electrode used, GEA33,* is made from B104 glass. This electrode is very specific for sodium ions, and can tolerate a 50-fold concentration of potassium ions without affecting the sodium ion response. The potential of this electrode is affected by a pH of less than 7, but at the concentration of sodium ions found in bacon, this effect is not significant.

This paper reports the use of this electrode system for the determination of salt in back, half gammon and streaky bacon.

EXPERIMENTAL

APPARATUS AND CALIBRATION—

*Sodium-ion responsive glass electrode, type GEA33.**

*Saturated calomel electrode, type GRJ23.**

pH meter—A very stable pH meter is required, the No. 48B Blood pH meter* being suitable.

A calibration curve was prepared by using solutions of analytical-reagent grade sodium chloride ranging in concentration from 1 to 10 g of sodium chloride per 100 g of water.

* E.I.L. Ltd., Richmond, Surrey.

PREPARATION OF SAMPLE—

The simplest method appeared to be to wrap portions of sliced bacon round the electrodes, but this proved to be too irreproducible for routine use. Mincing, macerating, or grinding the sample gave reproducible results, but was too time consuming for production control.

Two methods of sample preparation were finally adopted for routine use after experience with many hundreds of determinations.

(a) Two slices of bacon were placed together and rolled into a cylinder. Two lateral holes were bored into the roll by No. 1 and No. 3 cork borers, and the electrodes inserted into the holes. This method will be referred to as the "roll method."

(b) The sample was rapidly chopped with a multiblade knife (Fig. 1). A sample of macerated lean bacon was ready for analysis in half a minute.
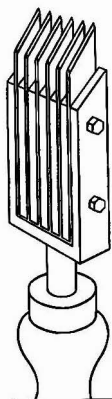

Fig. 1. Diagram of a six-bladed cutter, comprising a $\frac{1}{2} \times \frac{1}{8}$-inch M/S flat bar, six TY - Linker blades and a file handle

PROCEDURES—

Glass electrode method—

When not in use the electrodes were kept immersed in a 6 per cent. salt solution, and before being inserted into a bacon sample they were lightly wiped with paper tissue but were not allowed to dry off. Repeated washing with distilled water prolonged the equilibration time and was avoided. After insertion into the sample, the electrodes were left until a steady millivolt or pH reading was obtained (usually 3 to 5 minutes), after which the electrodes were removed, wiped with paper tissue, and either inserted into another sample or replaced in the salt solution. The percentage of salt, based on the water content of the sample, was then read directly from the calibration curve.

Chemical method—

The results obtained by the glass electrode method were compared with those obtained on the same samples by the A.O.A.C. Volhard assay, as previously used in bacon analysis.[4] The published method, which gives the percentage of salt in bacon, was modified by adding sucrose to the reaction mixture after oxidising with the nitric acid and potassium permanganate and boiling until the solution was colourless.

For comparison with the glass electrode method the results were converted to percentage salt on the moisture content of the bacon. The latter was determined by drying the sample overnight in an air-oven with a fan at $100° C \pm 1° C$.

RESULTS AND DISCUSSION

Results obtained on macerated samples of the lean of back bacon are shown in Table I.

TABLE I

SALT DETERMINATIONS ON MACERATED *LONGISSIMUS DORSI* MUSCLES OF BACK BACON

| Sample | Time of reading, minutes | Meter reading, mV | Percentage of salt on water, w/w | |
			Electrode method	Chemical method
1	10	897·5	6·0	6·3
2	0·5	896·0	6·5	6·8
	2	895·0		
	5	895·0		
	10	894·5		
3	0·5	907·0	4·4	4·5
	2	907·0		
	5	907·5		
	10	907·0		
4	0·5	909·0	5·0	5·2
	2	904·4		
	5	903·5		
	10	903·5		
5	0·5	896·5	7·0	7·3
	2	893·5		
	5	893·5		
	10	893·0		
6	0·5	897·0	6·5	7·0
	2	895·0		
	5	895·0		

These results show a satisfactory correlation, although the percentage salt on water given by the electrode is slightly lower than that given by the chemical method.   The response time of the electrode is seen to be about 3 minutes.

Table II shows the results obtained by the roll method of sample preparation.

TABLE II

SALT DETERMINATIONS ON VARIOUS TYPES OF BACON SAMPLES PREPARED BY
USING THE ROLL METHOD OF PREPARATION

| Type of sample | Percentage of salt on water, w/w | | Agreement |
	Electrode method	Chemical method	
Longissimus dorsi.. ..	3·3	3·1	good
	4·7	4·3	good
	5·9	6·0	good
	5·0	5·8	poor
	5·8	5·9	good
	8·3	7·8	good
	9·6	8·1	poor
Half gammon lean ..	4·0	4·6	poor
	5·9	6·5	poor
	8·0	7·0	poor
	9·6	8·5	poor
Streaky	5·7	5·7	good
	6·3	6·6	good
	7·0	7·2	good
	8·2	8·5	good
	9·8	9·5	good
	10·5	10·3	good

Reproducibility when using the roll method was not always satisfactory for the leaner samples of *longissimus dorsi* muscle and half gammon.   Often a slight movement of the electrodes caused fluctuations in the meter reading.   It is, however, evident from Table II that streaky bacon can be analysed satisfactorily by this method.   The reason for this is not clear, but is probably owing to the lower moisture content of this type of bacon.

Table III shows the regression of the values, y, determined by the chemical methods on those obtained by the electrode, x. The electrode method has been used for many thousands of analyses. To estimate the reliability of this method compared with the chemical method a statistical analysis has been performed; 50 samples each of lean and streaky bacon were used.

TABLE III

SUMMARY OF STATISTICAL ANALYSIS OF SALT DETERMINATIONS ON BACON
BY CHEMICAL AND SODIUM ELECTRODE METHODS

	Longissimus dorsi and half gammon (macerated sample)	Streaky bacon (roll method)
Regression equation relating percentage of salt by chemical method (y) to electrode method (x) ..	$y = 0.526 + 0.996 x$	$y = 0.424 + 0.938 x$
Confidence limits for slope ($P = 0.95$)	0.940, 1.052	0.852, 1.024
Residual standard deviation of individual points	0.42	0.46

For both methods the slope of the regression line did not differ significantly from 1. The regression equations show that the electrode gives a lower value than the chemical method. This can also be seen by inspection of the results shown in Table I. It is not serious for production control purposes, and is probably caused by the activity coefficient of the sodium ion in the aqueous environment of the sample being different from that in aqueous solution.

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SHORT PAPERS

Mobile Laboratory Methods for the Determination of Pesticides in Air

Part I. Phosphorothiolothionates

By G. A. LLOYD AND G. J. BELL

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THE determination of pesticides in air affords a means of studying the effects on human exposure to these products when used in agriculture and horticulture. For such determinations, a mobile laboratory[1] is used for collecting and analysing air samples. Although elaborate analytical instruments cannot be used in the field to make these analyses, colorimetric procedures are practicable as portable spectrophotometers are available commercially.

Procedures are required that will determine the more toxic pesticides, chiefly the organo-phosphates, at concentrations in air of generally less than 0.2 mg per m³. Devices are used to sample the air, normally at 10 litres per minute, and the sampling period is adjusted commensurately with the sensitivity of the available colorimetric procedure.

In general, these field methods should have few, brief manipulations, and should allow less than 10 μg to be determined normally in 20 ml of solution with an error of not more than 10 per cent. Contamination of samples by inorganic phosphates is likely on farms and in glasshouses and, therefore, well known procedures cannot be used to determine organophosphorus pesticides by decomposition to the phosphate ion. Hazardous or unstable reagents, lengthy refluxing and the use of complex apparatus should be avoided, but techniques requiring prolonged incubation at less than 50° C, or boiling for short periods, are practicable in a mobile laboratory. Few published methods satisfy these requirements; suitable procedures are described in this paper and in ensuing papers.

Disulfoton,[2] diethyl *S*-[2-(ethylthio)ethyl] phosphorothiolothionate, and phorate,[2] diethyl *S*-(ethylthiomethyl) phosphorothiolothionate, are highly toxic insecticides and occupational hygiene studies required their determination to less than 0.1 mg per m³ in air. Published methods[3] did not meet the requirements for use in the field. It was found, however, that certain phosphoro-thiolothionates or phosphorothiolates could be readily hydrolysed to yield thiols, which can be determined by a suitably modified colorimetric procedure of extremely high sensitivity.[4]

METHOD

APPARATUS—

Spined glass bubblers[5]—These are to contain 20 to 30 ml of solvent for operation at an air flow of 10 litres per minute for vapour sampling.

Glass-fibre filters—Whatman GF/A, 5·5 cm in diameter,[6] for sampling dusts or droplets.

Filter holders—These can be based on the design of the Ministry of Labour test-paper holder,[7] the intake orifice being 12 mm in diameter for operation at an air flow of 10 litres per minute.

Stoppered tubes—25-ml graduated, capacity, 40 ml to stopper.

REAGENTS—

Trapping solution—Dilute 1 volume of N aqueous sodium hydroxide containing 0·05 per cent. w/v of sodium cyanide to twice its volume with isopropanol.

Sulphuric acid, 50 per cent. v/v and 1 per cent. v/v.

Sodium nitrite stock solution—Dissolve 3·5 g of sodium nitrite and 0·5 g of sodium hydroxide in 1 litre of water.

Sodium nitrite working solution, 0·005 N—Dilute 1 volume of stock solution with 9 volumes of 1 per cent. v/v sulphuric acid.

Ammonium sulphamate, 10 per cent. w/v, aqueous.

Sulphanilamide—Prepare a 1·5 per cent. w/v solution in 6 N hydrochloric acid.

N-1-*Naphthylethylenediamine dihydrochloride*—Prepare a 0·4 per cent. w/v solution in 6 N hydrochloric acid. Replace with a fresh solution after 1 week.

Mercuric acetate—Prepare a 5 per cent. w/v solution in 75 per cent. v/v acetic acid.

Chromogenic reagent—Prepare by mixing 8 volumes of sulphanilamide solution, 1 volume of N-1-naphthylethylenediamine solution and 1 volume of mercuric acetate solution. Replace with a fresh solution every hour.

SAMPLING LAYOUT—

Set up the sampling devices in the breathing zones of operators working with the pesticide and connect each device to a metered source of vacuum.

Each filter holder must be placed with the opening pointing downwards to simulate, as far as is practicable, the mechanics of inhalation through a nostril. In our experience the exposure indicated agrees reasonably well with that whereby the operator himself provides the air flow.[8]

Significant amounts of pesticides might be vaporised when applied in the form of a dust or spray, in which event, sample the air through a glass-fibre filter backed by a bubbler at each sampling position. At the air sampling-rate of 10 litres per minute, a glass-fibre filter will normally trap a proportion of pesticide in vapour form, but this may be offset by evaporation of droplets or vaporisation from dust particles trapped on the filter. Hence, the amount found in the backing bubbler cannot reliably be used to calculate the vapour concentration in air. Normally, a precise separation of the two forms is not required for protection purposes and, to date, no air filter has been found that completely separates airborne particles and vapours.

AIR SAMPLING—

(i) *Vapours*—Charge each bubbler with 20 ml of trapping reagent and add more isopropanol, according to the intended period of sampling, as indicated below.

Intended sampling time, minutes	5	5 to 10	11 to 15	16 to 21	22 to 26	27 to 31	32 to 36	36 to 40
Added isopropanol, ml	2	4	5	7	8	9	10	11

Draw air through each bubbler at 10 litres per minute, preferably for 10 to 30 minutes. Under these conditions the trapping efficiency is 80 per cent. for disulfoton and phorate; all results must be multiplied by the factor 5/4.

Determine the collection efficiency for all pesticide vapours by connecting two or more charged bubblers in series. Draw air, at normal temperature and humidity, over a sample of the pesticide at 10 litres per minute for periods of 10 to 30 minutes. Repeat the test with different concentrations of pesticide in air, generally in the range 0·01 to 0·2 μg per litre; this can be done by varying the exposed surface area of pesticide.

Transfer the bubbler contents to a graduated tube, adjust the volume to 20 ml with isopropanol and replace the stopper. Allow it to stand for 24 hours at about 15° C to complete the hydrolysis of the phosphorothiolothionates to the corresponding thiols.

(ii) *Droplets or dusts*—Draw air at 10 litres per minute through a glass-fibre filter. Transfer an exposed filter to a graduated tube containing 25 ml of the trapping reagent. Stopper the tube and shake it well to break up the filter. Allow it to stand for 24 hours at about 15° C to complete the extraction and hydrolysis of the phosphorothiolothionate. Filter, if necessary (Whatman GF/A glass-fibre filter), transfer an aliquot (normally 20 ml) to a similar graduated tube and adjust the volume as required to 20 ml with the trapping reagent. The recovery of 5 to 50 μg of disulfoton or phorate from a glass-fibre filter is better than 80 per cent., but for protection purposes it is better to over-estimate the risk, and for this reason the amounts found should be multiplied by the factor 5/4.

PROCEDURE—

Add 5·0 ml of 50 per cent. v/v sulphuric acid to 20 ml of hydrolysed extracts. Stopper the tube, mix the solution carefully by inversion and cool to about 15° C. Add 1·0 ml of 0·005 N sodium nitrite solution, mix and allow the solution to stand for 1 minute. Add 0·5 ml of ammonium sulphamate solution and shake the solution vigorously for 1 minute, occasionally releasing the stopper; allow it to stand for a further 1 minute. Add 5·0 ml of chromogenic reagent. Mix well and allow it to stand for 2 hours avoiding direct sunlight. Measure the absorbance of the solution at 540 mμ, with a light path of not less than 4 cm. Deduct the reagent or air blank reading and refer to a calibration graph prepared similarly from standard amounts (5 to 50 μg) of toxicant in 20-ml volumes of trapping reagent.

The accuracy of the colorimetric method is within $\pm$10 per cent. but the over-all accuracy with which airborne toxicants can be determined is limited partly by the accuracy with which the volumes of air samples are measured and controlled.

Demeton-S-methyl, oxydemeton-methyl, dimethoate, mecarbam and morphothion[2] have been successfully determined by this procedure and it is probable that demeton-S, thiometon, prothoate and vamidothion[2] can be determined similarly.

Phosphorothionates and 50-μg amounts of the following phosphorothiolothionates, azinphos-methyl, azinphos-ethyl, dioxathion, malathion and menazon[2] give little or no reaction when analysed by this method.

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Mobile Laboratory Methods for the Determination of Pesticides in Air

Part II.* Thionazin

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THIONAZIN,[1] diethyl *O*-2-pyrazinyl phosphorothionate, is a highly toxic pesticide and, following its recent introduction into the United Kingdom,[2] a study of the risks in its use to agricultural workers was required. In part, this involved its determination in the form of vapour or dust in air, at concentrations of about 0·1 mg per m³.

Thionazin can be determined spectrofluorimetrically[3] but this is impracticable in the field. A colorimetric procedure is described which, in general, meets with the requirements of a field method.[4] It involves hydrolysis of thionazin to hydroxypyrazine, which is then reacted with bromine to form a glutaconic aldehyde. Condensation of the latter with *p*-phenylenediamine hydrochloride yields a reddish brown polymethine dye that is soluble in acetone.[5]

METHOD

APPARATUS—

Spined glass bubblers.
Glass-fibre filters—Whatman, GF/A, 5·5 cm in diameter, and filter holders for air sampling.
Stoppered tubes—Glass, 25-ml graduated, capacity, 40 ml to stoppers.
Water-bath at 40° C.

REAGENTS—

Trapping solution—Dilute 1 volume of 0·2 N aqueous sodium hydroxide to 2 volumes with isopropanol.
Acetic acid, glacial.
Bromine water, saturated—Store over bromine.
Phenol - bromide solution—Dissolve 2 g of phenol and 5 g of potassium bromide in 100 ml of water.
p-*Phenylenediamine hydrochloride*—Dissolve 0·5 g in 100 ml of 2 per cent. v/v hydrochloric acid.
Standard thionazin—Prepare a solution that contains 10 μg of thionazin per ml in isopropanol.
Alternative standard—Prepare hydroxypyrazine from glycinamide and glyoxal,[6] and dissolve it in isopropanol (4 μg per ml ≡ 10·5 μg of thionazin per ml).

* For details of Part I of this series, see reference list, p. 809.

AIR SAMPLING—

Set up the sampling devices in the breathing zones of operators as described for the determination of phosphorothiolonothionates in air.[4]

(i) *Vapours*—Charge each bubbler with 20 ml of the trapping reagent. Add more isopropanol according to the intended period of sampling[4] and draw air through the solution at about 10 litres per minute. At the end of the sampling period (10 to 30 minutes), transfer the contents of each bubbler to a graduated tube, adjust the volume to 20 ml with isopropanol, stopper and allow the solution to stand for 17 to 21 hours in a water-bath at 40° C. Adjust the level of water in the bath so that it does not greatly exceed that of the solution in the tube, thus allowing the upper part of the tube to remain relatively cool.

(ii) *Droplets or dusts*—Draw air at 10 litres per minute through a glass-fibre filter. Transfer the exposed filter to a graduated tube containing 25 ml of the trapping reagent. Stopper and shake the tube well to break up the filter. Allow it to stand for 17 to 21 hours in a water-bath at 40° C so that the upper part of the tube remains relatively cool.

PROCEDURE—

Cool the hydrolysed extract to about 15° C. Add isopropanol to replace any lost by evaporation and spin the extract in a centrifuge or filter it from the glass-fibre filter by passing it through a similar filter. Add 1·0 ml of acetic acid to 20 ml of the hydrolysed extract, mix well, and then add 1·0 ml of bromine water, stopper, mix and allow to stand for 1 minute. Add 0·5 ml of phenol - bromide solution, stopper, mix well, and allow to stand for 1 minute, ensuring that no free bromine remains around the stopper. Finally, add 1·0 ml of the *p*-phenylenediamine hydrochloride solution and 10 ml of acetone. Mix and allow the stoppered tube to stand in a water-bath at 40° C for 30 minutes.

Measure the absorbance of the solution at 465 mμ, with a light path of not less than 4 cm. Deduct the reagent or air blank reading and refer to a calibration graph prepared similarly from 10 to 200-μg amounts of thionazin and which has preferably been standardised against hydroxypyrazine by this method. Make the appropriate corrections when aliquots have been taken from the original extract and multiply the amount of thionazin found in a bubbler and on a glass-fibre filter by the factors 4/3 and 5/4, respectively, to allow for the collection efficiency of the bubbler (75 per cent.) and a minimum recovery of 80 per cent. from the glass-fibre filters.

The accuracy of the over-all procedure is about 10 per cent., the measurement and control of air volumes being the major limiting factor. However, the accuracy of the colorimetric procedure is such that liquid formulations and their dilutions, as used in the field, can be analysed normally to within the limits of ± 10 per cent. by diluting suitable aliquots with the trapping solution.

Formaldehyde, alkyl or aryl organomercury fungicides may be used with thionazin for horticultural purposes, but 100-μg amounts of these pesticides cause negligible interference when this method of analysis is used.

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NOTE—Reference 4 is to Part I of this series.

Determination of Thiourea in Sewage and Industrial Effluents

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THE determination of thiourea in industrial sewage and effluents is important because of the strong inhibitory power of thiourea on the biological oxidation of nitrogen. There is some doubt about the concentration of thiourea that would be damaging to a purification plant, but it is desirable to be able to determine the compound in concentrations of about 1 p.p.m., and to detect it at a much lower level.

Methods have been described for detecting and determining thiourea in certain fruits.[1] These methods involve the use of Grote's reagent in some modification, the preparation of which is extremely tedious. For *detection* of thiourea in low concentrations the A.O.A.C. recommends the use of pentacyano-ammonio ferroate. It has now been found that a 0·1 per cent. solution of this compound reacts quantitatively and reproducibly with thiourea in slightly acid solution (pH up to 4·5) to give a clear blue colour that can be used to detect 0·07 mg of thiourea and to determine 0·02 to 0·7 mg in 50 ml of solution (see second paragraph of Standardisation).

METHOD

PREPARATION OF PENTACYANO-AMMONIO FERROATE—

Dissolve 2 g of sodium nitroprusside in 8 ml of 0·88 ammonia solution and allow the solution to stand in a refrigerator at about 0° C for 24 hours. The mass may then appear solid. Bring the mixture to room temperature, add about 1 ml of distilled water, mix and filter. To the filtrate add absolute ethanol until no further precipitate (yellow) forms. Filter with suction, wash with ethanol to remove ammonia, and dry the solid in a vacuum desiccator. Reduce to powder and store over calcium chloride in a desiccator in the dark.

For use, prepare a 0·1 per cent. solution in water. It is necessary to expose this solution to daylight for it to become sensitive; it is sufficient to prepare the solution in the morning when it becomes satisfactory for use after about 4 hours. It remains usable for at least 2 days thereafter, if stored in the dark.

STANDARDISATION—

Prepare standards containing up to 0·75 mg of thiourea in 50 ml. Add 0·1 ml of dilute acetic acid (10 per cent. v/v) and 3 ml of the pentacyano-ammonio ferroate reagent. Mix the solutions. Allow 30 minutes for the blue colour to develop. Measure the absorption at 610 mμ with a 1-cm or a 4-cm cell depending upon the sensitivity desired. The range may be extended upwards by using more reagent. The colour develops rapidly after about 10 minutes and is stable between 20 and 40 minutes after mixing.

The plot of absorptiometer reading against thiourea concentration is a straight line, provided the amount of thiourea present does not exceed 0·7 mg. If the concentration expected is below 2 p.p.m. (0·1 mg) it is advisable to use 4-cm cells.

TABLE I

DETERMINATION OF THIOUREA IN MIXED SOLUTIONS

Composition of mixture, mg per 50 ml			Thiourea found, mg			
Ammonium thiocyanate	Sodium thiosulphate	Thiourea	Total	Correction for ammonium thiocyanate	Net	Recovery, per cent.
1·0	0	0·27	0·315	0·07	0·245	91
0·5	0	0·54	0·515	0·05	0·49	91
1·0	0	0·54	0·585	0·07	0·515	95
0·5	1·5	0·27	0·35	0·05	0·30	111
1·0	3·0	0·27	0·36	0·07	0·29	107
0·5	1·5	0·54	0·655	0·05	0·605	112
1·0	3·0	0·54	0·60	0·07	0·53	98
0	3·0	0·27	0·34	0	0·34	126
1·0	3·0	0	0·07	0·07	0	—
0	0	0·22	0·22	0	0·22	100

The above determinations were made with an E.E.L. Spectra instrument fitted with a 1-cm cell.

Fearon[2] investigated the reactions of pentacyano-ammonio ferroate with many compounds and laid the foundation for the present method. He concluded that the only substances capable of producing stable blue colours in acid solution are the thioureas and related sulphur compounds. These include ammonium thiocyanate and the thiosulphate ion. Under the conditions now described it requires about ten times the amount of ammonium thiocyanate to give a blue colour comparable with that given by thiourea. Provided that ammonium thiocyanate is not present in very much greater concentration it is therefore practical to determine both compounds in a mixture. The thiocyanate is determined by the pyridine - pyrazolone method[3] and the equivalent absorption deducted from the total to obtain a net value due to thiourea. The reaction with

thiosulphate is much less sensitive and is probably different in its mechanism; 1·5 mg gave the same absorption as 0·02 mg of thiourea. Consequently, thiosulphate must be present in amounts that can be determined by titration before it is likely to interfere, and a correction can then be made.

Recovery tests with mixtures of thiourea, ammonium thiocyanate and sodium thiosulphate were carried out. The results in Table I were obtained.

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Absorptiometric Determination of Fenitrothion Residues in Cocoa Beans

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FENITROTHION, dimethyl 3-methyl-4-nitrophenyl phosphorothionate, the active ingredient of the commercial formulation, Sumithion, has a low mammalian toxicity, *viz.*, acute oral LD_{50} to rats, 673 mg per kg, and is effective against a wide range of insects.[1] It has shown promise for the control of cocoa capsids, *Distantiella theobroma*, and there was a need for a method for determining residues of the insecticide in treated cocoa beans at harvest. Dawson, Donegan and Thain[2] described a gas - liquid chromatographic method for determining residues of fenitrothion and related compounds in cocoa beans, after extracting them into benzene and purifying by solvent partitioning. Kovac and Sohler[3] extracted fenitrothion residues from fruits and vegetables with light petroleum and separated them by thin-layer chromatography, with subsequent ultraviolet absorptiometry of the hydrolysed product, 3-methyl-4-nitrophenolate. Similar methods were used for determining traces of fenitrothion in milk,[4] bananas[5] and cocoa beans.[6]

The colorimetric method, developed by Averell and Norris[7] for determining residues of parathion (diethyl 4-nitrophenyl phosphorothionate), may be adapted for determining fenitrothion. The method depends on reduction of the insecticide to the corresponding aromatic amine and the formation of a coloured compound by diazotisation and coupling with *N*-1-naphthylethylene-diamine. When applied to cocoa beans, it suffers from interference by plant pigments and fat, which comprise about 50 per cent. of the bean.[8] To overcome these difficulties, the clean-up procedure and the effect of several variables on the development of the colour were investigated. In the method described below, the insecticide is extracted into chloroform. The extract is decolorised with activated charcoal, and the fat removed from the hot reduced solution by adding paraffin wax,[9] which collects the oily substance and the zinc powder as a solid mass on cooling. The final colour is developed in a hydrochloric acid - isopropanol solution, and reaches the approximate maximum intensity 1 hour after adding the coupling reagent.

METHOD

APPARATUS—

Macerator—A top-drive macerator with adaptors for fitting a 200-ml macerator flask, obtainable from Townson and Mercer, was used.

Spectrophotometer—A Unicam SP600.

REAGENTS—

Hyflo Super-cel—Obtainable from Johns-Manville Co. Ltd.

Activated charcoal—For decolorising purposes. Obtainable from British Drug Houses Ltd.

Hydrochloric acid, 0·5 N - isopropanol, 60 per cent. solution—Dilute 45 ml of concentrated hydrochloric acid (sp.gr. 1·18) and 600 ml of isopropanol to 1 litre with water.

Zinc powder.

Paraffin wax, congealing point about 60° C.

Sodium nitrite solution, 0·25 per cent. w/v—Prepare a fresh solution daily.

Ammonium sulphamate solution, 2·5 per cent. w/v—Prepare a fresh solution weekly.

N-1-*Naphthylethylenediamine solution, 1 per cent. w/v*—Dissolve 0·5 g of N-1-naphthylethylene-diamine dihydrochloride in 50 ml of hydrochloric acid - isopropanol solution; prepare a fresh solution every 3 days.

STANDARD SOLUTIONS OF FENITROTHION—

(i) *Stock solution*—Dissolve 0·050 g of pure fenitrothion in chloroform, and dilute to 250 ml with chloroform.

1 ml of fenitrothion ≡ 200 μg of fenitrothion.

(ii) *Working solution*—Dilute 10·0 ml of stock solution to 200 ml with chloroform.

1 ml of solution ≡ 10 μg of fenitrothion.

PROCEDURE—

Preparation of calibration graph—Transfer 0, 2·0, 4·0, 6·0, 8·0 and 10·0 ml of working standard solution, equivalent to 0, 20, 40, 60, 80 and 100 μg of fenitrothion, respectively, to six 250-ml round-bottomed flasks with B24 1-inch necks. Half immerse the flasks in turn in a water-bath at about 60° C, and evaporate off the chloroform under reduced pressure. Add to each flask 25 ml of hydrochloric acid - isopropanol solution and 1 g of zinc powder, and boil under reflux for 15 minutes on a heating mantle. Add 2 g of paraffin wax, and boil for a further ½ minute. Filter the cooled contents through a cotton-wool pledget in a 2-inch diameter funnel into a 50-ml calibrated flask. Wash the reaction flask and filter twice with 10-ml portions of hydrochloric acid - isopropanol solution, and collect the washings in the calibrated flask.

Add to the calibrated flask 1 ml of sodium nitrite solution, mix, and set aside for 10 minutes. Add 1 ml of ammonium sulphamate solution, mix, and set aside for 10 minutes. Add 1 ml of N-1-naphthylethylenediamine solution, make up to the mark with hydrochloric acid - isopropanol solution, mix, and set aside for 1 hour. Filter the solution through a fluted Whatman No. 5 filter-paper, discard the first few millilitres of filtrate, and measure the optical density of a further 10 ml at 550 mμ in a 2-cm optical cell, with water in the reference cell. Draw the calibration graph relating optical densities of the standards to concentrations of fenitrothion in micrograms. The graph was linear, and optical densities of 0·04 and 0·22 were obtained by the reagent blank and 20 μg of fenitrothion, respectively.

Determination—Take 250 g of cocoa beans at random from the sample provided, and grind to coarse particles. Weigh 50 g of the ground sample into a macerator flask, add 100 ml of chloroform, and macerate for 3 minutes. Prepare a Hyflo Super-cel filter as follows: moisten a Whatman No. 5 filter-paper with water in a 7-cm Buchner funnel supported on a 500-ml filter flask, and suck it dry. By using a 1-inch rubber bung, gently press 5 g of Hyflo Super-cel to form a pad on the Buchner funnel, wash it with a small amount of chloroform under suction, and discard the filtrate. Filter the macerated material through the prepared filter. Wash the macerator flask with 100 ml of chloroform, and pass the washings through the same filter into the filter flask. Transfer the filtrate to a 200-ml calibrated flask, dilute to the mark with chloroform, and mix.

Transfer 100 ml of the filtrate (equivalent to 25 g of sample) to a 250-ml conical flask, add 1 g of activated charcoal, and mix. Filter the contents through a Whatman No. 40 filter-paper into a 250-ml round-bottomed flask with a B24 1-inch neck. Wash the conical flask with a small amount of chloroform, and pass this through the same filter into the round-bottomed flask.

Evaporate off the chloroform under reduced pressure at 60° C. Complete the reduction of fenitrothion, and the development and measurement of the colour as directed in the "Preparation of calibration graph," commencing from "Add to each flask 25 ml of hydrochloric acid - isopropanol solution . . ."; read off from the calibration graph the fenitrothion content of the sample solution in micrograms.

Conduct a similar determination with 50 g of untreated cocoa beans, as described above.

$$\text{Fenitrothion content, p.p.m.} = \frac{Y}{25} \times \frac{100}{\text{Recovery, per cent.}}$$

where Y = the weight of fenitrothion found (corrected for the blank), μg.

RESULTS AND DISCUSSION

Recoveries by the proposed method were determined by analysing untreated cocoa beans, to which 0·4 to 3 p.p.m. of fenitrothion had been added. Results are shown in Table I.

TABLE I

RECOVERY EXPERIMENTS ON UNTREATED COCOA BEANS

Fenitrothion added, p.p.m.	Fenitrothion found, p.p.m.	Recovery, per cent.
0·4	0·30	75
0·8	0·53	66
	0·60	75
	0·52	65
	0·52	65
	0·65	81
	0·53	66
	0·47	59
	0·56	70
1·6	0·89	56
	1·18	74
	0·94	59
	1·16	72
	1·06	66
	0·94	59
2·0	1·45	72
	1·42	71
	1·28	64
	1·34	67
3·0	2·73	91
	2·38	79
	1·98	66

The percentage recoveries ranged from 56 to 91 with a mean of 69 (standard deviation, ±8·0). The average apparent fenitrothion content obtained on 12 samples was 0·22 p.p.m., with a standard deviation of ±0·12 p.p.m., on which basis the limit of detection for the method is considered[10] to be 0·2 p.p.m.

During 1962, field trials were conducted in Tafo and Bunso, Ghana, by Plant Protection Limited with the co-operation of the National Research Council of Ghana, for the control of cocoa capsids. In these trials each acre was treated with 5 gallons of 0·5 per cent. fenitrothion in water. Thirteen samples of cocoa beans that were harvested 1 to 30 days after applying the treatment were analysed for fenitrothion residues. No residue was detected in any of the samples treated.

With minor modifications, the method has been satisfactorily applied to tomatoes grown on soils drenched with fenitrothion for controlling nematodes. The low dry matter content of tomatoes permits a 100-g sample to be taken for each determination, and its insignificant content of fatty materials renders the paraffin-wax treatment unnecessary. However, more colour from plant pigments appears in the chloroform extract, and rather more activated charcoal, i.e., 4 g, is required to decolorise the extract. The modified method is simpler, has the limit of detection of 0·1 p.p.m., and is considered to be generally applicable to various fruit and vegetable crops.

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Spectrophotometric Determination of Complexed Dibenzoylmethane

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IT is useful when studying metal β-diketonates to have a rapid method for determining the β-diketone bound to the central ion.

Bonner and Thorne[1] have described an absorptiometric method for determining acetylacetone that is based on the formation of a complex with the iron(III) ion. To aid our investigation of some dibenzoylmethides, we have developed a similar method for determining dibenzoylmethane.

EXPERIMENTAL

INFLUENCE OF VARIOUS FACTORS ON THE ABSORPTION SPECTRUM OF THE IRON(III) - DIBENZOYL-METHANE COMPLEX—

If a solution of ammonium iron(III) sulphate in 0·1 N sulphuric acid is added to an equal volume of a solution of dibenzoylmethane in ethanol, a red - violet coloured complex is obtained, whose absorption spectrum in the range 400 to 700 mμ is shown in Fig. 1. The absorption maximum lies at 518 mμ, and the extinction coefficient at this wavelength is 1085 litres per mole per cm.

A 1 + 1 water - ethanol mixture proved to be a good solvent for dibenzoylmethane, its iron(III) complexes and also for iron(III) alum. This solvent has, therefore, been used for our tests in preference to other possible solvents.

The concentration of the sulphuric acid influences the wavelength and optical density at the absorption maximum of the iron(III) complex, as shown in Fig. 2. It is necessary, therefore, to fix a value for the sulphuric acid concentration. We found that 0·1 N sulphuric acid is excellent
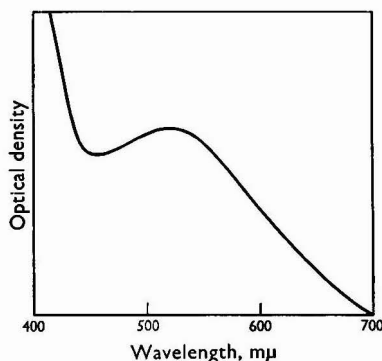

Fig. 1. Absorption spectrum of the iron(III) - dibenzoylmethane complex in ethanol - 0·1 N sulphuric acid mixture
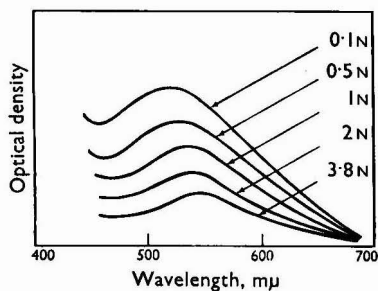

Fig. 2. Influence of the sulphuric acid concentration on the iron(III) - dibenzoylmethane complex absorption

for this purpose, that is, for hydrolysing most of the metal dibenzoylmethides.

The colour intensity obtained changes with time, but not markedly; the greatest intensity is observed 40 minutes after adding the iron(III) alum. The absorption was measured with a Beckman DK2 spectrophotometer, with 1-cm quartz cells.

It is necessary to use acetone to dissolve some dibenzoylmethides. As acetone reduces the molar extinction coefficient of iron(III) dibenzoylmethide, it is necessary to use the same amount of acetone in the calibration solutions.

There is no interference from the metal ions obtained from the dibenzoylmethides hydrolysed, at least when present in amounts up to that of the dibenzoylmethane determined.

The method has been applied to the dibenzoylmethides of iron(III), chromium(III), cobalt(III), copper(II), nickel(II), titanium(IV), zirconium(IV), cobalt(II), tin(IV) and uranium(VI). Good results were obtained for all of the complexes, except for the chromium(III) and cobalt(III) derivatives. These complexes are not hydrolysed by the 0·1 N sulphuric acid added to their ethanol - acetone solutions.

Probably the high stability of the β-diketonates of d_3 and d_6 ions, as found by Collman[3] for chromium, cobalt and rhodium acetylacetonates, may be the reason for the inapplicability of this method to these complexes. Perhaps they could be more easily hydrolysed after previous reduction of the ions.

The following procedure is suggested.

PROCEDURE—

Dissolve the dibenzoylmethide in acetone, or preferably in ethanol, and place a volume (A) of the solution, which should contain at most 7 mg of dibenzoylmethane, in a 25-ml calibrated flask and add $(12 \cdot 5 - A)$ ml of ethanol and 5 ml of $0 \cdot 1$ N sulphuric acid. After shaking the mixture, add 5 ml of iron(III) alum ($8 \cdot 2$ per cent. w/v in $0 \cdot 1$ N sulphuric acid) and then make up the volume with $0 \cdot 1$ N sulphuric acid. Read the absorption at 518 mμ, 40 minutes after adding the iron(III) alum, against a blank containing the same volumes of solvents as were used for the sample solution.
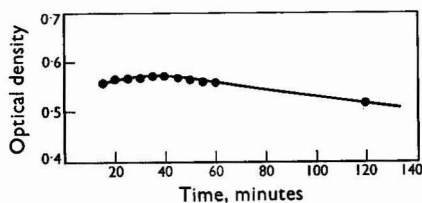

Fig. 3. Absorption variation at different times for a solution containing 3 mg of dibenzoylmethane per 25 ml

Inorganic anions that could precipitate or give complex ions with iron(III), such as phosphate, molybdate, fluoride and citrate, must be eliminated because of their interference in the determination. The presence of chlorides and sulphates in concentrations higher than that of the iron(III) added could interfere. It is, therefore, evident that the calibration graph must be made with solutions containing the same amounts of these salts, in the same way as separate calibration is required for different amounts of acetone.

TABLE I

DETERMINATION OF DIBENZOYLMETHANE IN DIBENZOYLMETHIDES

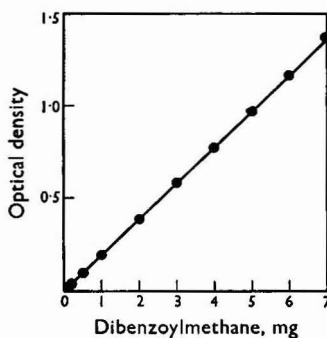|			Theoretical percentage of dibenzoylmethane	Dibenzoylmethane found, per cent.
Ti(dibenzoylmethane)$_2$Cl$_2$	..	..	79·0	79·6
Zr(dibenzoylmethane)$_3$Cl	..	..	84·1	84·0
Sn(dibenzoylmethane)$_2$Cl$_2$	..	..	70·1	69·5
Ni(dibenzoylmethane)$_2$	..	..	88·3	88·5
Cu(dibenzoylmethane)$_2$	..	..	87·5	88·1
Fe(dibenzoylmethane)$_3$	..	..	92·3	92·5
Co(dibenzoylmethane)$_2$	..	..	88·4	88·3
UO$_2$(dibenzoylmethane)$_2$	..	..	62·5	62·1

Fig. 4. Calibration graph for the iron(III) - dibenzoylmethane complex

All other β-diketones must be absent or separated beforehand.

With the above procedure, and over a range of concentration (0·1 to 7 mg of dibenzoylmethane per 25 ml) we obtained a straight-line graph (Fig. 4) passing through the origin, according to the relationship—

$$d = 1085\,l\,c$$

where d is the optical density, l is the cell width, and c is the molar concentration of dibenzoylmethane.

The results obtained in the determination of dibenzoylmethane in some dibenzoylmethides are given in Table I.

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Detection of Some 2-Hydroxy and 2-Methoxy Estrogens and Other Phenolic Compounds by a Modified Folin - Ciocalteu Test

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IN the routine determination of estrogen metabolites on paper-chromatographic strips with the Folin - Ciocalteu reagent to locate the compounds,[1] it was found that some compounds gave the characteristic blue colour before the addition of a base. As King[2] had speculated that such an anomalous compound might be 2-hydroxyestriol, it was decided that an investigation of the specificity of the phenomenon might prove of value.

The compounds tested included a variety of substituted phenols and related compounds. These compounds were dissolved in a suitable solvent and spotted on Whatman No. 1 paper strips. The Folin - Ciocalteu reagent[3] diluted (1 + 2) with water was applied by putting about 10 ml in a watch-glass and passing the strip through it. Ammonia solution was applied by the same method, whether or not a reaction was noticed after dipping the paper in the reagent. Care was taken to exclude ammonia fumes from the working area during the application of the reagent.

RESULTS—

The results of the Folin - Ciocalteu test on phenolic compounds are given in Table I.

TABLE I
RESULTS OF FOLIN - CIOCALTEU TEST ON PHENOLIC COMPOUNDS

Compound	Before application of ammonia solution	After application of ammonia solution
Catechol	+	+
Coumarin	−	−
Diethylstilbestrol	+	+
2,3-Dimethoxyestrone	−	−
Epinephrine	+	+
L-Epinephrine bitartrate	−	−
Estradiol	−	+
Estradiol diacetate	−	−
Estriol	−	+
Estrone	−	+
Gallic acid	+	+
Guaiacol	+ (violet)	+
2-Hydroxyestradiol	+	+
6-Ketoestradiol	−	+
16-Ketoestradiol	−	+
2-Methoxyestradiol	+	+
2-Methoxyestrone	+	+
Phenol	−	+
Piperonal	−	−
Vanillin	−	+
Veratraldehyde	−	−
Veratrole	−	−

Of all the compounds tested, vanillin was the only *o*-hydroxy or *o*-methoxy phenol that failed to give a positive reaction before the addition of ammonia solution, while diethylstilbestrol was the only compound other than *o*-hydroxy or *o*-methoxy phenols that did give such a reaction. Each time a reaction occurred before the addition of ammonia solution, its subsequent addition resulted in a more intense blue colour. It was also noted that *o*-hydroxy phenols gave a stronger reaction than *o*-methoxy phenols. 2,3-Dimethoxyestrone and veratrole (1,2-dimethoxybenzene) gave no reactions at all. With 2-methoxyestrone the sensitivity of the reaction was approximately $2 \mu g$ per cm^2; for 2-hydroxyestradiol the sensitivity was $1 \mu g$ per cm^2. A commercial preparation of the reagent gave identical results.

These observations should be of use in the preliminary classification of estrogen metabolites. This work was supported by the USPHS Training Grant ITI GM 1301.

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A Rapid Infrared Spectrophotometric Method for the Analysis of *pp'*-DDT in Formulations of Technical DDT

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ROUTINE analysis of dichlorodiphenyltrichloroethane (DDT) formulations demands a rapid and reliable method for determining the insecticidally active constituent. The insecticidal properties of the material depend on the concentrations of the *pp'*-isomer present. The principal difficulty to overcome is that of determining *pp'*-DDT in the presence of related compounds, particularly the *op'*-isomer.

Infrared spectrophotometry has been used by many analysts to determine DDT in pesticide formulations. Most workers[1,2,3] have used the band centred on 1015 cm^{-1}. This band is also present in the spectrum of *op'*-DDT and, therefore, its intensity is related to the total amount of DDT present. McDonald and Watson[2] overcame the effect of *op'*-DDT by using a compensating solution in the reference beam of a double-beam spectrophotometer. Downing, Freed, Walker and Patterson[4] obtained *pp'*-DDT concentrations by difference, after determining the impurities at other wavenumbers. A comparison of spectra of *op'*-DDT and *pp'*-DDT suggested that the *pp'*-isomer peak at 808 cm^{-1} would be relatively free from interference from the *op'*-isomer. Subsequently, our attention was drawn to a paper by Henry, Colas and Prat[4] in which this peak (808 cm^{-1}) was used for determining DDT. These workers, however, applied their method only to the analysis of technical DDT.

EXPERIMENTAL

APPARATUS—

Double-beam infrared spectrophotometer—A Unicam SP100, fitted with a sodium chloride prism, was used.

Chromatography tubes, 30 cm × 1·6 cm i.d.—The tubes incorporate stopcocks to control the flow-rate of solvent.

Spectrophotometer cells—1-mm path length potassium bromide or sodium chloride cells were used.

REAGENTS—

Florisil, 60 to 100 mesh—In the batch used no heat treatment was found to be necessary.

Carbon disulphide, analytical-reagent grade.

Light petroleum - diethyl ether developing solution—Mix 6 volumes of diethyl ether (peroxide free) with 94 volumes of light petroleum (b.p. 60° to 70° C).

Acetone, laboratory-reagent grade.

Sodium sulphate, anhydrous crystalline analytical-reagent grade.

pp'-*DDT*, *m.p.* 108·5° to 109° C—Recrystallise from 75 per cent. ethanol, then from light petroleum.

op'-*DDT*, *m.p.* 77·5° *to* 77·6° C.

PROCEDURE—

Pack a chromatographic column with Florisil, slurried in hexane, to a height of 9 cm. By sucking solvent up through the column, air bubbles are easily removed. When the Florisil has settled, place a 1·5-cm layer of anhydrous sodium sulphate on top of the adsorbent. Allow the solvent to flow until it is level with the top of the sodium sulphate.

Make an appropriate dilution of the sample of DDT formulation in acetone so that about 0·1 g of *pp*'-DDT is contained in a volume of less than 5 ml of solution. Transfer this volume quantitatively to the top of the column.

Begin collecting the eluate from the time of introducing the sample to the column. When the sample solution has just entered the column, add the ether - light petroleum (6 + 94) solution to the tube, and develop the chromatogram with this solvent. Adjust the flow-rate to 2 to 3 drops per second.

Discard the first 10 ml of eluate as containing no DDT (see Note). The next 20 ml should contain all of the DDT. Transfer this fraction to a 50-ml Erlenmeyer flask and evaporate the solution to dryness on a steam-bath under a stream of dry nitrogen. Continue this evaporation for a period of 1½ minutes, after visible traces of volatile substances have disappeared.

Take up the residue in carbon disulphide, transfer the solution quantitatively to a 10-ml calibrated flask and dilute to volume. Dry the solution with a small amount of anhydrous sodium sulphate.

Determine the absorbance of the solution in a 1-mm cell in the wavenumber region from $750 cm^{-1}$ to $830 cm^{-1}$, with carbon disulphide in the 1-mm reference cell.

Draw a horizontal base-line from the trough at $798 cm^{-1}$ and, from this line, measure the absorbance of the peak at $808 cm^{-1}$. Determine the concentration of *pp*'-DDT in the solution from a standard curve of concentration against absorbance.

The standard curve can be prepared by measuring the absorbance of *pp*'-DDT in carbon disulphide in the concentration range of 2 to 20 mg per ml. The resulting relationship between concentration and absorbance is linear, but there is a small positive intercept (absorbance ≈ 0·01) on the absorbance axis when the line is extrapolated back to zero concentration.

[NOTE—The batch of Florisil should be checked to determine at what volume the DDT leaves the column.]

RESULTS AND DISCUSSION

PREPARATION OF SAMPLE—

DDT dusts containing only insecticide and inert fillers require only extraction with carbon disulphide and filtration before measurement.

The presence of surface-active agents and solvents in formulations precludes direct determination on the sample after simple dilution in carbon disulphide, as it has been found that the surface-active agents and the solvents present in most concentrates interfere. The surface-active agents may be removed by a chromatographic step while the solvents may be evaporated.

Acetone is used for dilution of the sample before chromatography. This solvent will dissolve both formulations based on organic solvents as well as the aqueous "mayonnaise" types. Further, no non-volatile contaminant in laboratory-reagent grade acetone interferes with the subsequent infrared determination.

RETENTION VOLUME ON THE COLUMN—

Technical DDT was applied to the column and the non-volatile residue, after evaporation, weighed for each 5 ml of eluate collected. The peak of the DDT eluted from the column occurred in the fourth 5-ml fraction, when the conditions of the chromatography were those described under "Procedure." By discarding the first 10 ml of eluate and collecting the next 20 ml, all of the DDT is recovered from the sample.

LOSS OF DDT DURING HEATING ON THE STEAM-BATH—

On heating 0·0375 g of DDT in a 50-ml Erlenmeyer flask on the steam-bath under a stream of dry nitrogen, the weight of the DDT decreased by 0·15 per cent. per minute. The DDT was

weighed at intervals of 5 minutes for a total time of 25 minutes. Thus, under the conditions of the analysis, heating for 1 to 2 minutes has little effect on the recovery of the DDT.

PRECISION OF THE METHOD—

Eight determinations, as described in the procedure, were made on a commercial DDT formulation, the same volume of the formulation being taken each time.

The results provide the following values—

Mean absorbance	..	..	0·293
Standard deviation	..	..	0·00316
Coefficient of variation	..	..	1·1 per cent.

It should be noted here that these determinations were made with the one column of adsorbent. It is this that makes determinations rapid, as only one column need be packed for eight to ten determinations.

RECOVERIES OF pp'-DDT—

Various amounts of pp'-DDT were weighed and 4 ml of an acetone solution of a commercial DDT sample were added to each. The resulting fortified solutions were analysed.

The absorbances at 808 cm^{-1} were measured and the corresponding pp'-DDT concentrations were read from the standard curve. As the solutions had been diluted to 10 ml with carbon disulphide, the amount of pp'-DDT present in the solutions was found by multiplying the concentration by ten. The last column then shows the percentage recovery.

These results are shown in Table I.

TABLE I

RECOVERY OF pp'-DDT

Solution	pp'-DDT added, g	Absorbance at 808 cm^{-1}	pp'-DDT found, from standard curve, g	pp'-DDT recovered (pp'-DDT found, less 0·099 g), g	Recovery, per cent.
1	0	0·296	0·0990	0	—
2	0	0·298	0·0990	0	—
3	0·0188	0·357	0·1190	0·0200	106
4	0·0295	0·386	0·1290	0·0300	102
5	0·0465	0·432	0·1455	0·0465	100
6	0·0512	0·446	0·1505	0·0515	100
7	0·0568	0·453	0·1530	0·0540	95
8	0·0889	0·551	0·1870	0·0880	99

EFFECT OF op'-DDT ON THE PEAK AT 808 CM^{-1}—

A similar experiment to the recovery of pp'-DDT was carried out with various weights of op'-DDT instead of pp'-DDT.

A constant volume of an acetone solution of a technical DDT formulation containing 0·1 g of pp'-DDT was added to the column for each run. To this volume amounts of op'-DDT, from 0 to 0·07 g, were added. Calculation of a coefficient of variation for the five figures obtained gives a value of 1·1 per cent. This figure, on comparison with the precision of the analysis, indicates that op'-DDT has little effect on the peak at 808 cm^{-1}.

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Book Reviews

HIGH RESOLUTION NUCLEAR MAGNETIC RESONANCE SPECTROSCOPY. Volume II. By J. W. EMSLEY,
J. FEENEY and L. H. SUTCLIFFE. Pp. xxii + 665–1154. Oxford London, Edinburgh,
New York, Paris and Frankfurt: Pergamon Press. 1966. Price 105s.

This is the second of a two-volume work covering the whole field of high resolution nuclear
magnetic resonance spectroscopy to date, and is a worthy successor to the first volume. This
second volume is devoted to showing how information concerning molecular structure may be
deduced from n.m.r. spectral parameters, and is not concerned with the theoretical basis of n.m.r.
spectra, which was the main subject of Volume I. The first 200 pages of the book are accordingly
devoted to correlations of ^{1}H resonance spectral parameters with molecular structure. The treat-
ment is systematic throughout. The alkanes receive first consideration. Correlations of chemical
shifts with electronegativities of substituent groups are prominent here. Magnitudes of spin -
spin coupling constants in alkanes often provide information concerning bond angles. Thus, from
geminal coupling constants, H–C–H bond angles can be deduced, while from *vicinal* coupling
constants, magnitudes of dihedral angles can be determined. Included in this section on alkanes
are the spectra of various alkyl compounds as well as cycloalkanes and their derivatives. For
cyclohexane and its derivatives, the importance of n.m.r. in distinguishing between *axial* and
equatorial hydrogen atoms, thus greatly aiding configuration studies, is made particularly apparent.
The next section of the book is devoted to the spectra of alkenes. The spectra of compounds
containing vinyl groupings are first considered. The three magnetically non-equivalent hydrogen
atoms in a vinyl group normally give rise to a highly coupled ABC spectrum. Full analysis of
this system provides the chemical shifts of the three nuclei, together with the magnitudes and signs
of the *geminal, cis* and *trans* coupling constants. Many correlations with these parameters have
been made. For instance, chemical shifts have been related to Hammett σ-constants and group
dipole moments, *geminal* coupling constants have been related to H–C–H bond angles and *cis*
and *trans* coupling constants to the electronegativity of a substituent X in $CH_2 = CHX$ type
compounds. Di-substituted and tri-substituted olefins make up the next two sections of the book.
Again, the treatment is comprehensive and a considerable amount of data is tabulated. On
account of the near constancy of the *geminal, cis* and *trans* coupling constants towards different
substituents in di-substituted olefines, n.m.r. spectra can often clearly distinguish between different
geometrical isomers of the same compound.
After a short chapter devoted to acetylenic molecules, the important subject of the n.m.r.
spectra of aromatic molecules is considered. The effects of substitution on the *ortho, meta* and
para hydrogen chemical shifts are fully discussed in terms of the relative contributions of inductive,
resonance, diamagnetic anisotropy and electric-field effects. Good linear correlations of *para* ^{1}H
and ^{13}C chemical shifts with Hammett σ-constants are found but only when all intermolecular
effects on shielding have been eliminated by examining samples in very dilute solutions in inert
solvents. In di-substituted benzenes the substituent effects on the chemical shifts of the ring
hydrogen atoms can, in general, be shown to be additive. The authors then discuss the spin -
spin coupling constants in aromatic molecules. These, unlike the chemical shifts, are fairly
independent of the substituents and, as might be intuitively expected, are such that $J_{ortho} >$
$J_{meta} > J_{para}$, and all have the same relative sign. Heterocyclic molecules are the subject of the
next section of the book and this is followed by a chapter devoted to "large complex organic
molecules." Included under this heading are alkaloids, steroids, fatty acids, amino-acids, peptides
and other miscellaneous compounds. Here the authors have been content simply to summarise
briefly the most important work on these species of compounds. This is reasonable in view of
the large amount of data now available, but the book does suffer a little in this section from its lack
of detail. The first half of the book ends with a chapter devoted to miscellaneous ^{1}H resonance
studies, which includes spectra of paramagnetic species, polymers and studies of medium effects
in liquids.
The second half of the book deals with n.m.r. studies of nuclei other than hydrogen. Pride
of place is inevitably given here to ^{19}F resonance studies. The widespread interest in fluorine
chemistry is made abundantly clear from the vast amount of ^{19}F spectral data now available.
The large ^{19}F chemical shifts compared to ^{1}H shifts cause ^{19}F spectra, in general, to be less strongly
coupled than ^{1}H spectra, thus making spectral analyses less arduous. Spin coupling constants
for F–F coupling are also considerably larger in general than the corresponding H–H couplings.
However, their variation with spatial proximity and inter-bond angles are far less predictable

than the H–H coupling constants. This is apparent in the discussion of the many types of fluorine compounds where numerous apparently anomalous F–F coupling constants arise. The treatment here is very comprehensive and reflects the authors' particular interest in ^{19}F resonance work. The various types of fluorine compounds are treated systematically and the important rôle that n.m.r. has played in establishing the structures of fluorine compounds is well in evidence. The remainder of the book is devoted to spectra of other magnetic nuclei apart from hydrogen and fluorine. The varying amounts of space devoted to these nuclei reflect, to a large extent, the relative importance of these nuclei from the point of view of chemists who use the n.m.r. technique. Thus, ^{11}B, ^{13}C and ^{31}P receive detailed coverage, whereas other nuclei such as ^{14}N, ^{17}O, ^{29}Si and ^{59}Co are mentioned much more briefly.

This volume will most certainly be used as an important reference book on n.m.r. spectral data. Its usefulness in this rôle is particularly enhanced by the inclusion of six appendices setting out further data in the form of tables and charts. Included in these appendices are a table of τ-values of many organic compounds based on the data of Tiers, charts of ^{1}H chemical shifts and coupling constants of a wide variety of chemical groupings, a table of substituent shielding effects in benzenes and a comprehensive table of ^{31}P chemical-shift values. The amount of data contained within the covers of this book is vast, and it comes as a surprise to find that the book is only 490 pages in length. In order to achieve such compression of subject matter the authors have organised their material well and presented much of the data clearly in tabular form. Another good feature of this book (and also Volume I) is the very large number of spectral traces reproduced. This certainly is a great aid to reading a book of this type.

The price of the book may seem rather high for its moderate length, but the amount of data presented therein, and the great use to which this book will inevitably be put by all practising n.m.r. spectroscopists, make it well worth the money.   K. G. ORRELL

ANALYSIS OF THE NEW METALS—TITANIUM, ZIRCONIUM, HAFNIUM, NIOBIUM, TANTALUM, TUNGSTEN AND THEIR ALLOYS. By W. T. ELWELL and D. F. WOOD. Pp. xii + 275. Oxford, London, Edinburgh, New York, Toronto, Paris and Braunschweig: Pergamon Press. 1966. Price 60s.

This monograph represents a considerable expansion of Messrs. Elwell and Wood's earlier publication "The Analysis of Titanium and Zirconium" as it includes methods for the analysis of hafnium, niobium, tantalum, tungsten and their alloys, and revised methods for titanium and zirconium.

The authors state in the preface that their book is intended as a laboratory compendium containing essential information for the satisfactory analysis of titanium, zirconium, hafnium, niobium, tantalum, tungsten and their alloys, rather than a treatise on the analytical chemistry of these metals. However, the authors do go to the trouble of giving a useful summary of the analytical considerations involved with each determination, including comments that should pilot the analyst clear of most of the pitfalls associated with new techniques. For example, it is extremely useful for the practising analyst to receive such detailed guidance, as in the case of the polarographic determination of tin and lead, that "The addition of 2 ml of potassium ferro-cyanide solution (2%) prevents the interference of more than 200 ppm of copper or 500 ppm of molybdenum." Much invaluable detail is also provided for spectrographic procedures. The very full description of the oxide - resin technique for the determination of the impurities in zirconium is typical. All equipment is fully described and the treatment of samples and preparation of standards is treated with unusual thoroughness. Weights, volumes and all information necessary for the prosecution of a successful analysis are present.

Sampling, a subject often omitted from books of this type, is covered in the introduction and includes not only methods of sampling for massive metals but also for special materials such as titanium sponge and granules.

Recommendations are made for machining and drilling samples of these reactive metals with a minimum of contamination.

The authors are to be congratulated on the meticulous manner in which this monograph has been written and arranged. It is obvious that a tremendous amount of thought and care has gone into the making of this book. This effort is apparent in every aspect of the book and extends to the important but often neglected index and contents list.

All analysts with an interest in these less common metals will appreciate the efforts of their two colleagues in making available such useful and important knowledge.   G. L. MILLER

POLYSACCHARIDES, PEPTIDES AND PROTEINS. Volume 4. By R. T. COUTTS and G. A. SMAIL. Pp. x + 209. London: William Heinemann Medical Books Ltd. 1966. Price 30s.

This small volume, well bound in soft-back style, forms the fourth of a series of *Pharmaceutical Monographs* under the general editorship of Professor J. B. Stenlake of the University of Strathclyde. It is in two parts: the first 72 pages by R. T. Coutts of the University of Saskatchewan deal with the polysaccharides of pharmaceutical interest, and the remainder of the book, by G. A. Smail, gives, in an admirably concise way, an up-to-date outline of the chemistry of polypeptides and proteins.

The authors handle their subject matter in tutorial style, and the book is primarily intended as an adjunct to honours undergraduate courses in pharmacy. However, it provides an extremely readable introduction to protein and polysaccharide chemistry and is to be recommended, not only to undergraduates, but also to workers in related fields who may wish to acquire an understanding of the basic chemical and physical properties of these biological polymers, without going too deeply into detail.

The five chapters on polysaccharides provide an admirable summary of the properties of most of the common polysaccharides likely to be of interest to the pharmacologist, but it is a little surprising to find that although hyaluronic acid, chitin and the blood group substances are described, there is no mention of chondroitin sulphate or keratan sulphate. The fifth chapter on the somatic antigens of gram-negative bacteria is so short as to be of little value except as a reminder of the existence of this important class of compounds.

The section (120 pages) dealing with the proteins and peptides is quite excellent, and provides an adequate introduction to general protein chemistry, as well as concise and useful accounts of the polypeptide and protein hormones of pharmaceutical value. The last chapter contains a useful summary of information about the more important polypeptide antibiotics.

S. M. PARTRIDGE

THE SEPARATION OF BIOLOGICAL MATERIALS. BRITISH MEDICAL BULLETIN. Volume 22, No. 2. May, 1966. Edited by R. A. KEKWICK. Pp. 103–194. London: Medical Department; The British Council. 1966. Price 30s.

Although the title suggests that this number of the British Medical Bulletin will be mainly of importance to the biochemist and medical research worker, this volume will, in fact, be of inestimable value to the analyst, reviewing, as it does, major advances that have been made in the separation of the materials of which the cell is composed. Fifteen to twenty years ago the powerful separations possible by chromatography were replacing the relatively simple methods of precipitation, partition and other physical and chemical techniques that were the main armamentation of the analyst, and which usually demanded analytical methods specific for the substances to be measured. With the newer methods described in this monograph, many substances closely resembling one another in their chemical properties may be separated so completely that they can be determined by non-specific means. This is particularly clearly illustrated by the modern techniques of analysis of amino-acids, which are determined by the ninhydrin reaction after separation on columns. In gas chromatography, there is separation of micro or millimicrogram amounts of material which are detected and determined by wholly non-specific detectors such as by argon or flame ionisation, electron capture or katharometry. Such methods are already widely used by the clinical chemist, and their application to the problems of other analysts is already in progress.

This volume provides concise, critical reviews of the theory and practice underlying all of the recently developed methods of separation. After a theoretical account of the physical chemistry of porous systems by Ogston, the practical applications are dealt with by Andrews in his account of gel filtration (or exclusion chromatography), and by McDougall and Synge who discuss electrophoresis in gels. Ultra-centrifugation in a solution with a density gradient has provided an important technique for separating sub-cellular particles, viruses, proteins, nucleoproteins and nucleic acids. This technique has played an important rôle in the purification and concentration of viruses for vaccine production, and is of particular importance when used in conjunction with sensitive assays or isotopic labelling experiments. These techniques have thrown light on our knowledge of the structures of the haemoglobins and various peptides and protein hormones, and also on the mechanism whereby nucleoproteins provide the basis of heredity. By such methods genes have been found to control synthesis of proteins and enzymes and indirectly, therefore, of all biological materials. There are also excellent accounts of the separation of the nucleic acids, of viruses, of proteins and protein sub-units, and of peptides and of amino-acids. The reviews

of gas - liquid chromatography by Scott, of the separation of lipids by Nichols, Morris and James, of peptides by C. J. O. R. Morris, and of carbohydrates and related substances by Northcote are likely to be of the most practical importance to the practising analyst. Mathias has reviewed especially well the application of a variety of these methods to the separation of sub-cellular particles.

These methods are primarily of concern to biochemists investigating the nature and behaviour of highly labile substances and structures under dynamic conditions, and for them this new volume of the British Medical Bulletin will be unequalled. To the practising analyst there may well be much that is new, and it can confidently be recommended that this volume should be on the shelves of all analysts. The scientific editor, Professor R. A. Kekwick, and the British Council are to be congratulated on this excellent volume, which is a bargain at 30 shillings.

C. H. GRAY

INFRARED BAND HANDBOOK SUPPLEMENTS 3 AND 4. Edited by HERMAN A. SZYMANSKI. Pp. xvi + 261. New York: Plenum Press Data Division. 1966. Price $15.00.

Supplements 3 and 4 contain nearly 5000 bands over the range 2 to 25 μ, bringing the total number listed in the original Handbook and its supplements to over 18,000. All the results collected for the present volume have been published between 1962 and 1964, and the assumption is made that very accurate, high resolution instruments have been used to obtain such data. Although this assumption may not be fully justified, the suggestion that spectral bands of an unknown compound need only be compared with published data within 5 cm^{-1} is probably a fair one.

The mode of presentation of the bands in decreasing wavenumbers is convenient, and information concerning the structural formula of the compound, its physical state and the more important instrumental conditions is easily accessible.

The main criticism of the book (and its predecessors) is its possible superfluity in a field where spectral documentation is already in an advanced state. Dr. Szymanski has undertaken a mammoth task in preparing these volumes and doubtless feels committed to the publication of many more in the years to come, but the question which surely must be in the minds of most practising spectroscopists is "Will it all have been worthwhile?"   D. A. ELVIDGE

SUBMICRO METHODS OF ORGANIC ANALYSIS. By RONALD BELCHER, D.Sc. Pp. x + 173. Amsterdam, London and New York: Elsevier Publishing Company Ltd. 1966. Price 55s.

This latest contribution by Professor Belcher to the analytical chemists' library is a neat little book—nicely dedicated to Sir Harry Melville and Professor Maurice Stacey, who did so much to encourage the development of the Birmingham School of Analytical Chemistry (would that there were others similarly inclined in British Universities!).

Written in Belcher's customary clear, concise style, the text is strongly practical in outlook. Indeed, its synopsis claims that "workers will find here, for the first time in book form, all they need in order to apply the techniques in their own laboratories"—with what initial degree of success is, regrettably, not specified: experience with manipulations on the submicro scale is necessary for success. Nevertheless, for students and analysts of all ages, anxious to set about starting to acquire the necessary experience, this is *the* book; filled with useful practical detail, it is an essential distillate of the practical experiences of Professor Belcher and his co-workers in this field.

An interesting introduction is followed by useful sections dealing with the submicro balance and with the general apparatus required for submicro work. Chapters are then devoted to the methods recommended for determining each of the following elements or functional groups: nitrogen; carbon and hydrogen; chlorine; bromine and iodine; fluorine; sulphur; phosphorus and arsenic; carboxyl; organic bases in non-aqueous media; alkoxyl and *N*-methyl; acetyl; carbonyl; olefinic unsaturation; nitro; nitroso; and thiol; and other chapters deal with periodate oxidation and with the cryoscopic determination of molecular weight.

Much of the work described in this text-book has already been published and, to those of us who have followed the published accounts of Professor Belcher's studies as they progressed, this book must, at first sight, appear to be rather an expensive way of acquiring reprints. The text, however, incorporates improvements to some of the earlier procedures and gives additional details that have not been published; to anyone starting off on the submicro scale this is the sort of book that is cheap at any price.

It is to be hoped that this timely compilation by Professor Belcher will lead to increased interest in the methods he has pioneered—methods that deserve more interest than has been shown in recent years. The importance of the submicro scale and Belcher's contributions have yet to be realised in full.

                                                                D. M. W. ANDERSON

NOBEL LECTURES: CHEMISTRY, 1922–1941. Pp. xii + 508. Published for the Nobel Foundation. Amsterdam, London and New York: Elsevier Publishing Company Ltd. 1966. Price 160s.

As is known, by the publishing arrangement between the Elsevier Publishing Company and the Nobel Foundation, the Nobel lectures are now becoming available in the English language. This present volume includes the presentation addresses, the Nobel lectures given by, and the biographies for, each Nobel Laureate in Chemistry from 1922 to 1941, for each year in which the award was given.

It is difficult to review this book because these lectures are often the foundation on which the author's chemical education is based. To the analytical chemist this period includes three lectures that should hold his attention. They are Aston's lecture on Mass Spectra; Pregl's lecture on Quantitative Micro-analysis of Organic Substances; and Debye's lecture on Methods to Determine the Electrical and Geometrical Structure of Molecules.

Richard Kuhn, the Nobel Prize winner in 1938, was unable to accept his medal until 1949, and we are, therefore, denied the honour of his lecture. Of great interest are the many contributions that Kuhn and his school made to the perfection of chromatographic methods for the isolation of carotenoids.

It is of interest to speculate on the use that modern methods of analysis, *e.g.*, gas - liquid chromatography, would have been to, say, Winhaus in the constitution of the sterols, or his pupil Butenandt on sex hormones.

The book is recommended to all those who have interest in learning how chemistry has evolved in the last 4 decades. All of life is here, because often a few discreet words describe the tragedies of our civilisation.

The volume is beautifully printed, and exquisitely produced, although to the ordinary reader its price is prohibitive. However, we should expect to find it on the shelves of all good libraries.

                                                                G. NICKLESS

ANALYSIS INSTRUMENTATION—1965. Proceedings of Eleventh Annual Analysis Instrumentation Symposium held May 26–28, 1965, at Montreal, P.Q., Canada. Edited by L. FOWLER, R. G. HARMON and D. K. ROE. Pp. viii + 240. New York: Plenum Press. 1966. Price $12.50.

The pattern of previous years (*Analyst*, 1965, **90**, 639) is repeated in this volume, in which 21 papers presented at the Montreal Symposium are printed. The topics are very diverse: of the eight sections the largest contains four papers, while in one section a dissertation on writing instruction manuals for instruments adjoins a description of a miniaturised gas chromatograph - mass spectrometer for a space mission to analyse the soil on Mars. Although this diversity certainly reflects the breadth of interest in instrumentation it is unlikely to encourage compulsive buying of the volume by laboratories with restricted interests. Printed by photolitho from typescript, the volume contains rather less material than a single issue of a normal journal, and so must be rated as extremely expensive. Publication time has lengthened from the not unreasonable 9 months of the previous issue to 12 months. No discussion—often held to justify this form of publication—of the papers is reported.

To quote from the foreword, "The editors of this volume, Messrs. Harmon and Roe, spent many long hours of tedious labour." They have done a good job, but to what end? Good papers which should be in the open literature, accessible to all, are obscured in a publication that few can afford and fewer would read. Ephemera and trivia need not be recorded. It is said that this form of publication provides a necessary end product to a symposium, one which a conferee can take back to his employer as an earnest of his attendance and the benefits arising therefrom, and a return for the payment of his expenses. A costly memento. Continuing the previous quotation from the foreword, "Without the willingness of the authors to prepare the reports of their work, neither the symposium nor the book would have been possible. Our first and strongest thanks must go to them." There should also be added the willingness of the authors to deny themselves the appearance of their work in the acknowledged journals and its dissemination in the abstracting journals. The present volume contains fewer papers but of a more uniform quality than the previous volume. I can find nothing to justify publication in the present form.

                                                                E. BISHOP

ANALYTICAL CHEMISTRY OF SELECTED METALLIC ELEMENTS. By JAMES J. LINGANE. Pp. xiv + 143. New York: Reinhold Publishing Corporation; London: Chapman & Hall Ltd. 1966. Price 28s.

The elements discussed in this booklet are the alkali metals and alkaline earths, aluminium, antimony, arsenic, bismuth, cadmium, chromium, cobalt, copper, iron, lead, magnesium, manganese, mercury, molybdenum, nickel, silver, tin, titanium, tungsten, uranium, vanadium and zinc. The author, who has been concerned for many years with the teaching of analytical chemistry at the University of Harvard, explains in his preface that it is his express aim to provide an introduction rather than a comprehensive account of the analytical chemistry of these metals. In it he hopes to provide a feeling for the individual characteristics of each metal by providing a factual account of the chemistry and physical properties upon which all of the classical and physico-chemical analytical properties are based. Unquestionably he has succeeded. This is a book that will be found useful by all students of analytical chemistry in the age group 16 to 60.

In each individual monograph the text usually offers information on the physical properties of the metal itself, on its various ionic species, its electrochemical potentials, tendency towards complex formation, oxides and common salts, methods of separation from other elements, reactions with organic reagents, recommended method of analysis, gravimetric, titrimetric and physico-chemical (most frequently electrochemical). The facts are well marshalled and set out.

There are many flashes of humour in the text, *e.g.*, in describing the Marsh test . . . "the very sensitive test for arsenic immortalised by A. Conan Doyle with the able assistance of Sherlock Holmes," . . . followed by references to "Arsenic and Old Lace" and "who dunit" literature. In another chapter we find, "Aluminium hydroxide exists only on the blackboards of chemistry classes," . . . and so on. These leaven the text well when it might otherwise become indigestibly heavy because of its concentrated texture. At the same time it must be observed that there is sometimes a looseness of terminology about the text that gives the impression that we are dealing with transcribed lecture notes rather than conventionally produced text, *e.g.*, ions are generally written as M^{+++} but occasionally as M^{+n} and even M^{n+} (page 71). Similarly, the terminology "homogeneous precipitation" is placed in cautionary quotes on page 15 whilst on page 16 it is correctly used as "precipitation from homogeneous solution" and then a few lines subsequently as (the physically impossible) "homogeneous precipitation," this time without quotes. The author defends his use of " $+5$ uranium" and so on in place of "uranium(V)" etc., in his preface, but this does go against international usage, and although one appreciates the difference between "cc" and "ml," use of the former again goes against modern practice. Similarly, the use of the word "complexone," defined expressly as "a complexing agent," on pages 46 and 47 is not generally accepted. English students will also have to resist the author's use of the split infinitive though it is frequently rather attractive.

These are, however, subjective minor criticisms of an attractive and worthwhile booklet, that will undoubtedly be of considerable service to students of analytical chemistry in all walks of the profession.   T. S. WEST

PROBLEMS FOR GENERAL CHEMISTRY AND QUALITATIVE ANALYSIS. By C. J. NYMAN and G. B. KING. Pp. x + 274. New York, London and Sydney: John Wiley and Sons. 1966. Price 23s.

This book is a compilation of problems in general chemistry and qualitative analysis, aimed initially, it is supposed, at students of Washington State University. Frankly, this book is suitable only for use in schools in this country, to advanced and scholarship levels. What is interesting is the approach to the topic, and it provides a comparison with the books used in this country for this level of teaching.

The book is divided into chapters; in each chapter the principles for solving each given type of problem are given first, followed by examples of problems in which these principles are used. Each chapter is completed by a series of examples, the answers being given in an Appendix. Naturally, problems are arranged in order of increasing difficulty.

A detailed discussion of significant figures is given in another Appendix, as well as a review of fundamental mathematical operations. This book can be recommended as an interesting facet of the educational system in chemistry of the United States.   G. NICKLESS

THE SOCIETY FOR ANALYTICAL CHEMISTRY

FOUNDED 1874.   INCORPORATED 1907.

THE objects of the Society are to encourage, assist and extend the knowledge and study of analytical chemistry and of all questions relating to the analysis, nature and composition of natural and manufactured materials by promoting lectures, demonstrations, discussions and conferences and by publishing journals, reports and books.

The Society includes members of the following classes:—(a) Ordinary Members who are persons of not less than 21 years of age and who are or have been engaged in analytical, consulting or professional chemistry; (b) Junior Members who are persons between the ages of 18 and 27 years and who are or have been engaged in analytical, consulting or professional chemistry or *bona fide* full-time or part-time students of chemistry.   Each candidate for election must. be proposed by three Ordinary Members of the Society.   If the Council in their discretion think fit, such sponsorship may be dispensed with in the case of a candidate not residing in the United Kingdom.   Every application is placed before the Council and the Council have the power in their absolute discretion to elect candidates or to suspend or reject any application.   Subject to the approval of Council, any Junior Member above the age of 21 may become an Ordinary Member if he so wishes.   A member ceases to be a Junior Member on the 31st day of December in the year in which he attains the age of 27 years. Junior Members may attend all meetings, but are not entitled to vote.

The Entrance Fee for Ordinary Members is £1 1s. and the Annual Subscription is £4 for Ordinary Members taking *The Analyst* and *Proceedings of the Society for Analytical Chemistry* only, or £9 for Ordinary Members taking *Analytical Abstracts* in addition to *The Analyst* and *Proceedings*.   Junior Members are not required to pay an Entrance Fee and their Annual Subscription is £1 1s.   No Entrance Fee is payable by a Junior Member on transferring to Ordinary Membership.   The Entrance Fee (where applicable) and first year's Subscription must accompany the completed Form of Application for Membership.   Subscriptions are due on January 1st of each year.

Scientific Meetings of the Society are usually held in October, November, December, February, April and May, in London, but from time to time meetings are arranged in other parts of the country.   Notices of all meetings are sent to members by post.

All members of the Society have the privilege of using the Library of The Chemical Society.   Full details about this facility can be obtained from the Librarian, The Chemical Society, Burlington House, Piccadilly, London, W.1.

The Analyst, the Journal of the Society, which contains original and review papers, information about analytical methods and reviews of books, and has a world-wide distribution, and *Proceedings of the Society for Analytical Chemistry*, in which are reported the day-to-day activities of the Society, are issued monthly to all Ordinary and Junior Members.   In addition, any Ordinary Member may receive *Analytical Abstracts*, providing a reliable index to the analytical literature of the world, on payment of higher Annual Subscription.

Forms of application for membership of the Society may be obtained from the Secretary, The Society for Analytical Chemistry, 14 Belgrave Square, London, S.W.1.

LOCAL SECTIONS AND SUBJECT GROUPS

THE North of England, Scottish, Western, Midlands and North East Sections were formed to promote the aims and interests of the Society among the members in those areas.

Specialised Groups within the Society are concerned with the study of various branches of analytical chemistry of specialised or topical interest.   Groups dealing with such topics as Microchemical Methods, Biological Methods, Thin-Layer Chromatography, Atomic-Absorption Spectroscopy, Thermal Analysis, Automatic Methods, Particle Size Analysis, Radiochemical Methods and a Special Techniques Group, covering very new developments and specialised physical methods, are at present active—and further Groups are formed from time to time as the need arises.

Non-members of the Society may participate in the activities of a Group.

The Sections and Groups hold their own meetings from time to time in different places. Members of the Society pay no extra for membership of a Section or Group.   Application for registration as a member should be made to the Secretary of the Society.