83. *A New Technique for the Ultimate Micro-analysis of Organic Compounds.*

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In a new technique for the determination of carbon, hydrogen, sulphur, and the halogens in organic compounds on a micro-scale, combustion is effected at 800° by a rapid stream of oxygen (50 ml. per min.) in a tube containing no catalyst or contact material. Carbon and hydrogen are determined gravimetrically, a roll of silver gauze being inserted near the exit end of the combustion tube to absorb the interfering acid gases. Sulphur is quantitatively absorbed and retained as silver sulphate; by extracting this with water the amount of sulphur can also be determined gravimetrically. With compounds containing nitrogen, Elving and McElroy's external absorbents were used to remove nitrogen oxides which would otherwise cause error in the determination of carbon. The sulphur can also be absorbed, and the halogens always are, in external absorbents and determined titrimetrically. Complete absorption occurs at the fast rate of flow used, thus greatly reducing the time for the determinations.

The methods to be described differ from those based on Pregl's micro-methods for the determination of carbon, hydrogen, sulphur, and the halogens in that the combustion is effected by a rapid stream of oxygen in a tube maintained at 800° by means of an electric furnace and containing no catalyst or contact material. The apparatus is similar to that usually adopted in micro-combustion technique.

In the method for the determination of carbon and hydrogen a roll of silver gauze is inserted near the exit end of the combustion tube to absorb interfering gases. If the beak end of the combustion tube is attached by means of a ground joint, the roll of silver gauze may be removed, and the sulphur, if present, can then be determined by estimating the silver sulphate produced.

For the titrimetric determinations of sulphur and the halogens the usual absorbents are used, together with a combustion tube without a catalyst packing. The oxygen flow is much faster than that usually employed in these determinations, but complete absorption occurs. This is in marked contrast to the standard technique, which employs a slower oxygen stream to ensure complete absorption, the combustion often requiring up to one hour as compared with less than 10 mins. with the new methods.

We originated this unpacked tube technique for the ultimate analysis of coal (*Fuel*, 1941, **20**, 130) and, with 0·5 g. of sample, combustion was completed in 10 mins. So far as we are aware this was the first use of an unpacked tube for this purpose, and we have now adapted it to the micro-scale.

For the analysis of organic compounds the conditions are somewhat different from those used for coal: a high temperature is necessary to ensure complete removal of sulphur from the ash in coal, whereas with organic compounds the sulphur can be removed at a much lower temperature unless the substance contains an inorganic element which may form a sulphate only decomposable at a high temperature. The latter type of compound represents a special case and the method has been standardised for the more usual type of compound, a temperature of 800° being used.

EXPERIMENTAL.

Apparatus.—The apparatus is illustrated in the figure, most of the component parts being conventional in micro-chemical laboratories. The oxygen is led from a cylinder or gas-holder to a White–Wright flowmeter adjusted to measure a rate of oxygen flow of 50 ml. per min. From here it passes first through a White–Wright preheater and then through a scavenging train packed with soda asbestos in the first tube and anhydrone in the second; then it is led to the side arm of the combustion tube. All the connections are made with matured rubber tubing, the glass tubes making "butt" joints.

The combustion tube is made of transparent silica of 9 mm. internal diameter and 55 cm. long. It passes through an electric furnace consisting of a lagged silica tube of 15 mm. bore and 25 cm. long with nichrome wire winding and metal body. The furnace is supported on two pillars mounted on an asbestos cement board and is connected in series with a sliding resistance and ammeter. The beak end of the combustion tube protrudes 7·5 cm. out of the furnace, and the inlet portion, which has a 7-cm. length of silica tubing sliding over it, protrudes 22·5 cm.

To calibrate the apparatus, the electric furnace is heated so that the temperature inside the combustion tube measured with a thermocouple is 800°. The current necessary to maintain this temperature is noted, and in all subsequent work is kept at this value. To carry out a blank determination on the assembled apparatus, the oxygen flow is adjusted to 50 ml. per min., the absorption tubes, which are of the standard Pregl type packed with anhydrone and soda asbestos backed with anhydrone, are connected, and finally the Marriotte bottle. After being connected in the apparatus for 10 mins., the absorption tubes should not have increased in weight by more than 0·02 mg.

The fast rate of flow of oxygen obviates the need for a baffle behind the boat, since any tendency to back diffusion is minimised. Also, the fact that no catalyst or contact material is present in the combustion tube simplifies the setting up of the apparatus.

Gravimetric Methods.—*Method of combustion when only carbon and hydrogen (and oxygen) are present.* To carry out a determination of carbon and hydrogen, weigh 3—5 mg. in a platinum boat, adjust the oxygen flow to 50 ml./min., and attach the weighed anhydrone and soda asbestos absorption tubes. Insert the platinum boat and contents to a distance of 17·5 cm. into the combustion tube, and connect the Marriotte bottle. Carefully decompose the sample by heating the 7-cm. silica tube surrounding the combustion tube with a Bunsen burner, and gradually slide it over that portion of the combustion tube containing the platinum boat, and up to the electric furnace. The whole operation should occupy about 10 mins., whereupon the Marriotte bottle should be disconnected and the absorption tubes detached and weighed.

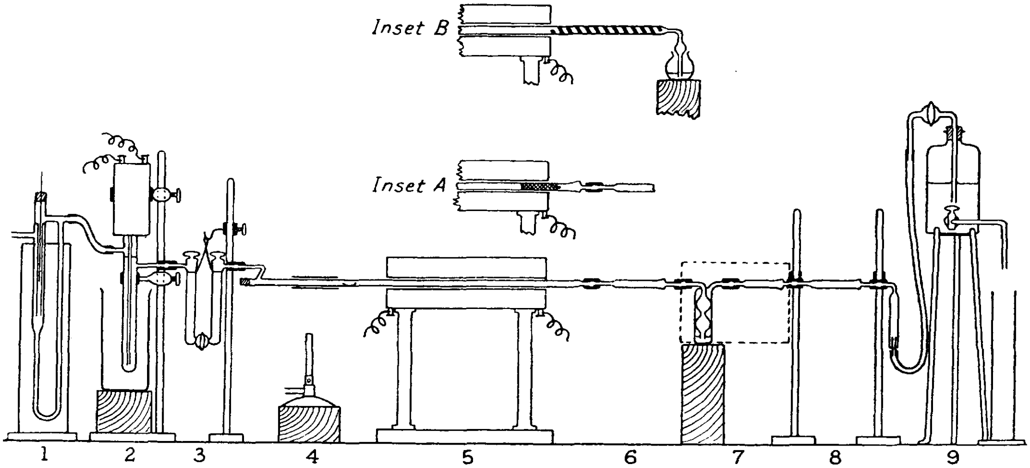
	Calc.		Found.	
Substance.	C, %.	H, %.	C, %.	H, %.
Methylglucoside	43·3	7·21	43·4, 43·4, 43·5, 43·4	7·32, 7·33, 7·24, 7·37
Benzoic acid	68·9	4·95	69·0, 68·9, 68·7	5·15, 4·90, 5·01
Naphthalene	93·7	6·29	93·5, 93·5	6·35, 6·26
Sulphonal	36·8	7·02	36·8, 36·9	6·98, 7·00
Chlorobenzoic acid	53·7	3·19	54·0, 53·7	3·28, 3·22
Chlorophthalic acid	47·9	2·49	47·9	2·55

Some of the results obtained are shown on p. 313, together with results for carbon and hydrogen in some sulphur- and chlorine-containing compounds (*q.v.*) : they are comparable in accuracy with those obtained with the general standard methods of micro-analysis.

Method of combustion when sulphur is present. If, in addition to carbon, hydrogen, and oxygen, the substance contains sulphur, this element may be determined simultaneously with the carbon and hydrogen. A modified combustion tube is used having the beak end attached by means of a ground joint. The absorbing material consists of a roll of pure 20-mesh silver gauze, 2½ in. wide, with the edges folded so as to give a width of 2 in., and rolled to fit snugly into the combustion tube (see Inset A). The roll must not be wound too tightly, for only the outer layers will then come in contact with the combustion gases, and the central portion is pulled out at one end to form a grip for crucible tongs. When suitably placed in the combustion tube, this roll absorbs all the oxides of sulphur and the silver sulphate formed can be used to determine the sulphur. The suitable placing is best done by attaching a delivery tube to the exit end of the combustion tube so as to form a bubbler and passing this into a flask containing a few ml. of neutral 1% hydrogen peroxide. Approximately 5 mg. of sulphonal or other compound containing sulphur are then burnt, and the exit gases bubbled through the hydrogen peroxide. When the combustion has been completed, the hydrogen peroxide is titrated with 0·01N-sodium hydroxide, and if any signs of acidity are found the procedure is repeated, the roll of silver gauze being moved ¼ in. further into the tube. With the electric furnace used and a roll of silver gauze approximately 2 in. long, it was found that complete absorption of the oxides of sulphur occurred when the outer end of the roll was ½ in. outside the furnace or at any position further inside the furnace.

To carry out the simultaneous determination of carbon, hydrogen, and sulphur, insert the silver gauze roll into the combustion tube to the position previously fixed as most suitable, attach the weighed absorption tubes, and carry out the combustion on 3—5 mg. of substance as described previously. When the combustion is complete, withdraw the roll of silver gauze from the tube, cool it on a metal block, weigh it, and extract it with 200 ml. of boiling water for 2 mins. Wash the roll thoroughly with water, dry it by immersion in alcohol followed by ether, and burn off the residual ether in a Bunsen flame until the roll begins to glow dull red; cool the roll on the metal block and weigh it. The loss in weight is silver sulphate, and when multiplied by 10·28, it gives the percentage of sulphur in the substance. The determination of carbon and hydrogen is completed as usual.

General Arrangement of Apparatus.



1, White–Wright flowmeter. 2, White–Wright preheater. 3, Scavenging train. 4, Bunsen burner for heating the silica tube sliding over that part of the combustion tube containing the sample in a platinum boat. 5, Electric furnace. 6, Water-absorption tube. 7, Arnold bubbler containing N/50-$KMnO_4$ in conc. H_2SO_4 backed with anhydrone for absorbing oxides of nitrogen. 8, Carbon dioxide absorption tube. 9, Marriotte bottle with guard tube.

Inset A. Modification for gravimetric sulphur determination showing position of roll of silver gauze.

Inset B. Modification for titrimetric determinations.

With compounds containing more than 30% of sulphur, a roll 2 in. long did not absorb all the oxides of sulphur formed, but a roll 3½ in. long gave a satisfactory result for rubeanic acid (see table). The results for sulphur so determined are recorded below for a number of compounds.

Substance.	S, %, calc.	S, %, found.	Substance.	S, %, calc.	S, %, found.
Sulphanilic acid	18·5	18·5, 18·4	Thiocarbanilide	14·1	14·4
Sulphonal	28·1	28·0, 28·1, 28·1	Atropine sulphate	4·7	4·7, 4·9
Cystine	26·7	26·7, 26·8	Rubeanic acid	53·4	54·0

If only the carbon and hydrogen are required, a permanent packing of silver wool can be used in the exit end of the combustion tube (Kirner, *Ind. Eng. Chem. Anal.*, 1935, **7**, 363). In this case a beak-ended tube of the conventional type may be employed.

Method of combustion when halogens are present. For substances containing halogens alone or in addition to sulphur, the silver roll still effects removal of interfering gases, and the same technique is adopted as when sulphur is the only interfering element. The estimation of the weight of silver halide formed cannot be recommended for determination of the halogen present, probably owing to the fact that silver halides tend to volatilise off the silver roll and condense on the cooler parts of the combustion tube. This in no way affects the carbon and hydrogen figures (see, *e.g.*, first table), but it causes the halogen figures to be up to 1% low when determined by loss in weight of the silver roll on extraction in ammonia for silver chloride, and by potassium cyanide solution for the bromide and iodide.

The silver halides formed do not interfere with the gravimetric determination of sulphur as above : the small solubility of the silver halides in the extracting water is depressed beyond interference by the solubility of the silver sulphate.

In this case, however, care is necessary in burning off the residual drying ether so as not to volatilise any silver halide in the flame. This is done by heating the silver gauze roll in the Bunsen flame only until it just begins to glow dull red.

Results for the gravimetric determination of sulphur in the presence of halogens are recorded below: the accuracy of the sulphur determination is unimpaired. Also included are results for the gravimetric determination of sulphur in the presence of oxides of nitrogen: the accuracy of the sulphur figure is again unimpaired because the silver roll is too hot to permit the formation of silver nitrate. Known mixtures of sulphonal with varying amount of chlorobenzoic acid, bromodinitrobenzene, iodoacetic acid, and dinitrobenzoic acid were used in these tests.

Substance.	Radical present.	S, %, calc.	S, %, found.
Sulphonal	Cl	28·1	28·3, 27·7
Sulphonal	Br	28·1	28·0, 28·1
Sulphonal	I	28·1	27·8
7-Iodo-8-hydroxyquinoline-5-sulphonic acid	I	9·12	9·18
Sulphonal	NO_2	28·1	28·1, 27·9, 27·6, 27·9, 27·9

Method of combustion when nitrogen is present. In the case of nitrogenous substances, a two-bulb Arnold bubbler containing either (i) N/50-potassium permanganate in concentrated sulphuric acid or (ii) a saturated solution of potassium dichromate in the same acid, suitably backed with anhydrone, is attached between the anhydrone and the soda asbestos used to absorb water and carbon dioxide. This is to absorb the oxides of nitrogen which may be formed during the combustion and would otherwise cause an error in the carbon figure. The combustion is then carried out in the usual manner. Although it depends on how the nitrogen is combined whether or not oxides of nitrogen are formed, it is probably better to use the bubbler for all compounds containing nitrogen until information is available as to which compounds do and which do not form oxides of nitrogen. In our experiments the bubbler was only necessary for compounds containing nitro- and nitroso-groups.

The usual methods of removing oxides of nitrogen are with a reduced copper spiral or lead peroxide maintained at 180°. Neither of these methods was adopted by us, since the copper spiral would have oxidised rapidly under our conditions and the lead peroxide would have necessitated the introduction of a packing into the combustion tube and a special heating mortar. Elving and McElroy (*Ind. Eng. Chem. Anal.*, 1941, **13**, 660) recommended the use of the above absorbing solution (i) or (ii) in a specially designed bubbler placed between the water and the carbon dioxide absorber. There is a diversity of opinion as to whether or not any absorption of oxides of nitrogen occurs in the water absorber; we have certainly noted an increase in weight of the water absorption tube when using calcium chloride with the Friedrich method. It was claimed by Elving and McElroy, however, that oxides of nitrogen were not absorbed by anhydrone, and our results (below) confirm this. There was negligible increase in weight of the anhydrone tube beyond that required for the hydrogen content, and the carbon figures showed that complete absorption of the oxides of nitrogen by solution (i) was occurring. When the bubbler was not used, the soda asbestos tube increased in weight by an amount corresponding to 4% above that required for the carbon in the one case tested.

	Calc.		Found.	
Substance.	C, %.	H, %.	C, %.	H, %.
Dinitrobenzoic acid	39·6	1·89	40·0, 39·8, 39·8, 39·7, 39·8	2·00, 2·00, 1·90, 1·85, 1·96
Nitroaniline	52·2	4·35	52·5, 52·1	4·44, 4·49
Nitroacetanilide	53·3	4·44	53·5	4·39
Azobenzene	79·1	5·49	78·9	5·64

Titrimetric Methods.—The general set-up of apparatus used in the titrimetric methods for the determination of sulphur and the halogens is the same as that used in the gravimetric methods. A standard spiral combustion tube is used but with a slight modification in that a capillary delivery tube is fused on to the beak end and bent so as to act as a bubbler (see Inset B).

Sulphur. Place in a 50-ml. silica flask 10 ml. of neutral 6% hydrogen peroxide, together with a measured volume of 0·01N-sodium hydroxide, estimated to be slightly in excess of the amount required to neutralise all the sulphuric acid formed. Wet the spiral with the mixture and allow the draining to run back into the flask into which the delivery tube is inserted to act as a bubbler. Adjust the oxygen flow to 50 ml. per min. and insert a 3—5-mg. sample of the compound weighed into a platinum boat. Decompose the sample and complete the combustion by the method previously described. When the tube is cool, wash it out into the flask, make the solution acid with a measured amount of 0·01N-sulphuric acid, boil, and back titrate with 0·01N-sodium hydroxide. Calculate the sulphur from the total acidity produced. The determination may be expedited if a combustion tube is used which allows the glass spiral to be washed without waiting for the furnace to cool, such as that described by Beazley (*Ind. Eng. Chem. Anal.*, 1939, **11**, **229**).

If chlorine or bromine is present, the same procedure of absorption in 6% hydrogen peroxide is recommended, followed by the determination of the total acidity. The halogen is determined simultaneously by Gibson aud Caulfield's method (*Analyst*, 1935, **60**, **522**), mercuric oxycyanide being used. With compounds which produce oxides of nitrogen, the same authors' barium chromate procedure is recommended.

Chlorine. When chlorine is present alone it is determined by the procedure adopted for sulphur, the chlorine being calculated from the hydrogen chloride absorbed by the alkali.

Iodine. The rapid combustion technique is applied to the Leipert iodine method without further modification. The oxygen flow is maintained at 50 ml./min., and the combustion takes very little longer than 5 mins., but the results show that all the iodine is retained by the spiral wetted with sodium hydroxide. No excess alkali is required in the bubbler when this method is used: if excess is added it causes trouble with the end-point in the subsequent titration.

Some of the results obtained by these methods are recorded below: the accuracy is as good as that given by the general standard methods of titrimetric micro-analysis.

Substance.	Calc., %.	Found, %.	Substance.	Calc., %.	Found, %.
Sulphonal	S, 28·1	27·8, 28·0, 28·3, 27·8, 28·0	Chlorophthalic acid	Cl, 17·7	17·8, 17·9, 17·9, 18·0
Atropine sulphate	S, 4·7	4·6	Iodobenzoic acid	I, 51·2	51·2, 51·1
Sulphanilic acid	S, 18·5	18·8, 18·7, 18·4	Iodoacetic acid	I, 68·3	68·3, 68·2
Chlorobenzoic acid	Cl, 22·7	22·5, 22·5	Iodoform	I, 96·9	96·7

Appendix.—With the method originally developed for the analysis of coal, which used 0·5 g. of sample, an empirical factor of 0·1065 was necessary when converting the silver sulphate into sulphur. The decrease in weight of the silver roll after extraction was always less than that required by the actual sulphur content of the coal. We were at a loss to account for this, since no sulphur was found to pass the roll. When we applied our technique to the micro-scale we were surprised to note that the correct factor was applicable. Later, in applying the technique to organic compounds on the macro-scale, it was again found that the correct factor gave low results although all the sulphur oxides had been retained by the roll, but the empirical factor worked out for coals when applied gave good results. We now believe this is due to the formation of a small amount of a more complex silver salt, such as silver pyrosulphate, and the mixture of this with the silver sulphate gives rise to the empirical factor 0·1065 as against the theoretical factor for silver sulphate of 0·1028.

The use of an empirical factor for the macro-scale tests was not due to oxides of sulphur passing the silver roll, since by attaching a bubbler containing neutral hydrogen peroxide at the end of the combustion tube negligible acidity was produced. It was not due to the retention of sulphuric acid in the combustion tube, as was shown by washing it out and titrating the washings. It was not due to loss of silver sulphate by the roll, either mechanically or by volatilisation, since the sulphur calculated from the increase in weight as SO_4'' gave the same result as that calculated from the loss in weight as silver sulphate; this would not have been the case had some silver sulphate been lost at the same time as the SO_4 was being absorbed. It was not due to the formation of silver sulphite along with the sulphate since it would have necessitated the formation of two-thirds of the sulphur as silver sulphite and one-third as silver sulphate to satisfy the factor used. Moreover, such an excess of silver sulphite would not have been soluble in the extraction water. It can, therefore, only be due to the formation of some complex sulphur compound, such as silver pyrosulphate, and it would necessitate only 6% of the loss in weight on extraction in water to consist of silver pyrosulphate to account for the empirical factor of 0·1065.

Since silver pyrosulphate, if present, would be hydrolysed by water to give silver hydrogen sulphate, it may be determined in the extraction water by precipitation as barium sulphate. The fact that the whole of the sulphur is retained by the silver and can yield the amount of sulphur ion demanded was shown by two series of experiments.

Direct precipitation by barium chloride cannot be used because silver chloride would be precipitated simultaneously, so in the first series of tests barium nitrate was used. Since it is well known that adsorption occurs with this precipitant, solutions containing equivalent known amounts of sulphate ion and silver ion were precipitated under the same standard conditions as the solutions containing the extracted silver salts. With the comparison solutions the results were always higher than demanded by the amount of sulphate known to be present. This amount was deducted from the figure obtained by precipitating the extraction solution with barium nitrate. The percentage of sulphur calculated from the barium sulphate after deduction of the amount due to adsorption corresponded to that obtained from loss of weight of the silver roll using the empirical factor.

In the second series of tests the silver was precipitated from the extraction water with hydrochloric acid and filtered off. The sulphate ion was then determined in the filtrate by precipitation with barium chloride. In this case also the results obtained by barium sulphate and loss of weight of the silver roll using the empirical factor corresponded.

Although this did not prove the presence of silver pyrosulphate directly, the above experiments indicate that probably some silver salt of a complex sulphur acid is formed which is hydrolysed to sulphate ion. The only one which seems likely to us is the pyrosulphate, and it suggests that in the later stages of the combustion the inner portion of the silver roll already covered with silver sulphate reacts with sulphur trioxide to give this complex compound. On the micro-scale there is probably always sufficient free silver surface to cope with all the oxides of sulphur produced.

Conclusions.—We have shown that our unpacked tube combustion technique can be adapted for the micro-analysis of organic compounds. This technique is simpler and much more rapid than previous methods which rely on a slow stream of oxygen and a tube packed with complex catalysts or oxidants to ensure complete combustion.

We have greatly extended the usefulness of silver gauze in the ultimate analysis of organic compounds, and have shown that it can be used to absorb all the interfering gases except oxides of nitrogen, for which external absorbents are recommended. At the same time we have made it the basis of a gravimetric sulphur determination, thus determining all three elements from a single combustion.

In addition, we have shown that the usual reagents will absorb effectively the combustion products at the fast rate of flow we recommend, thus disproving the idea that in the determination of sulphur and the halogens a slow rate of oxygen flow is necessary. No attempt has been made to find the limiting rate below 50 ml. per min., where quantitative combustion in the same time will occur, since this rate of flow was found to be easy to control.

This technique was first applied to the analysis of coal, and since the chlorine content of coal is usually less than 0·25% for such small amounts the volatility of the silver halide does not prevent the chlorine figure being given to within 0·02%, which is often all that is required.

Since we have already adapted this technique to the macro-analysis of organic compounds, there should be little difficulty in evolving a similar semi-micro technique. Obviously a point will be reached where salts other than silver sulphate are formed, but it should be possible to obtain a suitable factor over a limited range of sulphur contents.

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