515. *Submicro-methods for the Analysis of Organic Compounds. Part VII.* The Determination of Nitrogen in Heterocyclic Compounds and Azo-, Hydrazo-, and Nitro-compounds, and in the Presence of Other Elements.*

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The submicro-method for the determination of nitrogen has been extended and modified for nitro-, azo-, and hydrazo-compounds, heterocyclic bases, and nitrogenous compounds which contain sulphur and the four halogens. The modified procedure is as accurate as the original one.

Our submicro-determination of nitrogen [1] in organic compounds has now been extended to its determination when present in forms which normally require special treatment by the Kjeldahl method, and to the analysis of nitrogenous compounds which contain other elements which might interfere.

In our previous work [1] a lower limit of 380° on the central thermometer of the heating block was specified to allow a sufficient safety factor for random temperature variations throughout the rest of the block. In the experiments now described a redesigned electrical heating block with superior insulation was used and more accurate measurement of temperature was achieved by using a calibrated thermocouple. Consequently it was found possible to work at 350°.

1. *Determination of Nitrogen in Compounds which contain Sulphur or Halogens.*—The original method was applicable to the determination of nitrogen in compounds which also contained sulphur, fluorine, or chlorine without loss in accuracy. Some etching of the glass tubes occurred with fluorine compounds. Sulphur dioxide and hydrochloric acid were formed when sulphur and chlorine were present, but had no adverse effect.

Erratic results were obtained when bromine- or iodine-containing compounds were analysed; similarly ammonium bromide and iodide gave low recoveries. Since bromate, iodate, iodide, iodine, and bromine interfere with the iodometric titration, close attention was paid to the nature of the halogen products formed during decomposition. In no case were traces of iodate or bromate found in the digest. Iodine compounds yielded free iodine and iodide ion and we concentrated on overcoming the interference of the iodide ion. Various attempts to mask or remove iodide were unsuccessful, but reduction of the digestion temperature had a beneficial effect on the recovery of nitrogen, and when this was investigated further it was found that nitrogen could be quantitatively recovered at 350° ± 5°. At this temperature the iodine in the digest existed entirely as element; it was readily removed at the same time as the sulphur dioxide when the opened tube was placed in an oven at 90° for 10 min. Low recoveries, due to incomplete decomposition of the compound, were obtained at 320—325° and also at 375—380°, owing to iodide formation.

Bromine no longer interfered when the digestion temperature was maintained in the range 350—380°. Table 1 shows the results obtained in the analysis of nitrogenous compounds containing sulphur and the halogens. As in the previous work samples of between 30 and 80 μg. were taken for analysis. The accuracy of the modified method is as good as that previously reported.

2. *Determination of Nitrogen in Heterocyclic Compounds.*—Low results were obtained in the analysis of 8-hydroxyquinoline (a typical heterocyclic compound) when digested at 350° for 30 min. as described above, or for 1 hr. Recovery was much better at 400° for 30 min. and improved when the time was extended to 1 hr. At 420° for 30 min., however,

* Part VI, preceding paper.

[1] Belcher, West, and Williams, *J.*, 1957, 4323.

recovery was quantitative in the analysis of this and five other heterocyclic compounds (Table 2).

For iodine-containing heterocyclic compounds, *e.g.*, 8-hydroxy-7-iodoquinoline-5-sulphonic acid, low results were obtained at 420° owing to formation of iodide ion, and

TABLE 1.

Compound	Range of wts. (μg.)	Nitrogen (%) Calc.	Found	Error (%)	No. of detns.	Range of errors (%) Max.	Min.
Trifluoroacetanilide	36·39—68·93	7·40	7·21	0·19	4	−0·31	+0·08
p-Chloroacetanilide	43·90—78·94	8·30	8·20	0·10	5	−0·30	−0·02
Phenylthiourea	39·66—58·32	18·41	18·49	0·08	2	+0·16	−0·01
S-Benzylthiuronium chloride	43·79—63·77	13·82	13·70	0·12	2	−0·14	−0·11
Bromomethyltriphenyl pyrrolone	42·43—48·42	3·47	3·39	0·08	3	−0·45	−0·18
p-Bromoacetanilide	50·97—73·53	6·54	6·45	0·09	3	−0·35	−0·04
2 : 4 : 6-Tribromoaniline	64·71—67·61	4·24	4·42	0·18	2	+0·21	+0·15
2-(4 : 5-Dibenzyloxy-2-bromophenyl)-propionamide	41·67—50·81	3·18	3·33	0·15	4	+0·41	+0·02
p-Iodoacetanilide	43·14—76·81	5·36	5·46	0·10	4	+0·28	−0·03
Phenyltrimethylammonium iodide	51·09—66·19	5·32	5·41	0·09	4	+0·31	+0·01
Methyl 2-benzyloxycarbonylamino-2 : 6-dideoxy-6-iodo-α-D-glucopyranoside	43·79—67·82	3·20	3·20	0·00	4	−0·12	+0·01
o-Iodohippuric acid	48·59—73·43	4·59	4·57	0·02	4	+0·17	+0·04
2-(3 : 4-Dimethoxyphenyl)ethyl-*N*-methyl-3 : 4-methylenedioxybenzyl-ammonium iodide	45·26—53·04	3·06	2·93	0·13	3	−0·26	+0·08

TABLE 2.

Compound	Range of wts. (μg.)	Nitrogen (%) Calc.	Found	Error (%)	No. of detns.	Range of errors (%) Max.	Min.
8-Hydroxyquinoline	42·97—47·28	9·64	9·64	0·00	3	+0·33	−0·12
1 : 10-Phenanthroline	42·49—63·39	14·12	14·03	0·09	4	−0·28	−0·01
Cocaine	37·76—60·43	4·62	4·53	0·09	4	+0·25	+0·05
Codeine	37·04—63·11	4·41	4·34	0·07	4	−0·20	−0·03
Morphine	37·10—60·70	4·62	4·64	0·02	4	+0·24	−0·08
Nicotinic acid	39·22—61·30	11·39	11·36	0·03	7	+0·41	−0·06
8-Hydroxy-7-iodoquinoline-5-sulphonic acid *	47·33—65·24	3·99	4·08	0·09	6	+0·39	+0·05
Nicotinic acid *	41·18—67·50	11·39	11·43	0·04	6	+0·31	−0·15
8-Hydroxyquinoline *	50·80—53·31	9·64	9·54	0·10	2	−0·06	−0·15

* These analyses were effected at 350° with mercuric sulphate catalyst.

also at 350° owing to incomplete decomposition. Mercuric sulphate was then tried as catalyst; at 420° low recoveries were still obtained, but at 350° the recovery was quantitative (Table 2). Heterocyclic compounds not containing iodine also yielded excellent results.

3. *Determination of Nitrogen in Nitro-, Azo-, and Hydrazo-compounds.*—Recovery was low when the original method [1] was applied to azo- and nitro-compounds. The addition of mercuric sulphate had an adverse effect. When glucose was added as a reductant [2,3] with double the usual amount of sulphuric acid (to decompose the extra organic matter) excellent results were obtained at a digestion temperature of 420° This slight modification also permitted the successful analysis of a hydrazine compound which contained fluorine and chlorine. The results of several analyses are shown in Table 3.

Discussion and Conclusions.—Nitrogen in organic compounds which contain sulphur, chlorine, and fluorine can be determined without modification, but if bromine or iodine is present the digestion temperature should be controlled at 350°, or low recoveries may

[2] Elek and Sobotka, *J. Amer. Chem. Soc.*, 1926, **48**, 501.
[3] Baker, *Analyst*, 1955, **80**, 481.

result. For the analysis of nitro- and azo-compounds it is necessary to add a small amount of glucose to the digestion mixture before mineralisation in order to reduce the nitrogen before decomposition. It is advisable to maintain the temperature at 420° for this determination. Heterocyclic compounds require the addition of mercuric sulphate to the

TABLE 3.

Compound	Range of wts. (μg.)	Nitrogen (%) Calc.	Found	Error (%)	No. of detns.	Range of errors (%) Max.	Min.
p-Nitroaniline	46·76—62·38	20·28	20·34	0·06	4	+0·14	0·00
p-Nitroacetanilide	43·87—43·92	15·55	15·54	0·01	2	−0·24	+0·21
p-Nitrobenzoic acid	44·74—52·72	8·38	8·10	0·28	3	−0·51	−0·02
m-Nitrobenzoic acid	48·54—62·38	8·38	8·47	0·09	2	+0·10	+0·08
p-Nitrophenol	41·81—53·87	10·07	10·00	0·07	4	−0·24	−0·12
m-Dinitrobenzene	40·61—56·74	16·67	16·43	0·24	5	−0·34	+0·02
Azobenzene	48·17—55·93	15·38	15·30	0·08	2	−0·26	+0·10
Azonaphthalene	46·80—55·21	9·93	10·00	0·07	3	−0·29	+0·22
Azotoluene	49·04—59·29	13·33	13·38	0·05	4	+0·38	−0·08
Pentafluorophenylhydrazine hydrochloride	28·51	11·94	12·05	0·11	1	—	—

digestion medium as a catalyst, but if the compound contains iodine the digestion temperature must be controlled at 350°.

EXPERIMENTAL

1. *Determination of Nitrogen in Halogen- and Sulphur-containing Compounds.—Reagents and apparatus.* These were the same as those described previously [1] except that 0·01N-sodium hypochlorite and thiosulphate were used.

Procedure. As before except that with iodine- or bromine-containing compounds the temperature of digestion was maintained at 350° ± 5°. The only additional precaution was to ascertain (iodine compounds) that the iodine was completely eliminated when the digest tube was removed after 5 min. in the oven at 90°.

2. *Determination of Nitrogen in Heterocyclic Compounds.—Procedure.* As before [1] except that 1 or 2 crystals (*ca.* 100—200 μg.) of solid mercuric sulphate (M.A.R.) were added before the digestion tube was sealed. The digestion temperature was maintained at 350° ± 5° for 30 min., although higher temperatures could be used when bromine and iodine were absent. It was still necessary to add the 2% mercuric sulphate reagent to the digest as indicator for the adjustment of pH since the mercuric sulphate added as catalyst did not always dissolve.

3. *Determination of Nitrogen in Nitro-, Azo-, and Hydrazo-compounds.*—The previously described apparatus and procedure [1] was used except that 20 μl. of concentrated sulphuric acid were used instead of 10, and 0·5—0·6 mg. of glucose (M.A.R.) was added to the tube before sealing and heating at 420° for 30 min.

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