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DETERMINATION OF NITROGEN IN SOILS

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Estimation of nitrogen is perhaps the most important item of analytical procedure in agricultural chemistry and, indeed, modern chemistry in general. The subject has naturally attracted much attention and, in recent years, several attempts have been made either to develop new methods of estimation or to improve on the old ones so as to increase their accuracy and speed of execution. Much useful progress has been made in both the directions and will be considered briefly in the present communication.

Bal [1925] first showed that certain types of soils—such as those of the black cotton areas of the Central Provinces in India—offer considerable resistance to the usual Kjeldahl method of acid digestion and yield invariably low and inconsistent estimates of total nitrogen. He showed that this was due to the inadequate penetration of the soil by the concentrated ('dry') sulphuric acid used for the digestion and recommended pre-treatment with water to overcome this difficulty. He showed that by adopting the 'wet' method, as he designated the modifications, not only smoother digestion but also higher and more consistent estimates of total nitrogen can be obtained.

Although this was a highly important finding, its significance was not generally realized,* presumably, due to the impression that it related only to a particular type of soil. The later observations of Sreenivasan [1932] showed, however, that it applied to other types of soils as well. 'Dry' digestion generally yielded low estimates of nitrogen, whereas pre-treatment with water or, preferably, dilute acid led to smoother digestion and to higher and more consistent values being obtained.

The significance of pre-treatment with water was discussed by Sreenivasan and Subrahmanyan [1933] who showed that in presence of concentrated ('dry') sulphuric acid the soil silicates tend to form impenetrable protective coats of silica around undigested soil particles. Even prolonged boiling does not break open these coats, so that a small part of the soil always goes undigested and is only partially attacked by alkali during subsequent distillation. On the other hand, pre-treatment with water facilitates easy penetration so that the soil is fully digested by the acid which follows. Those authors also showed that 'wet'

* Some recent workers (Martin and Griffith, *J. S. C. I.*, 1935, **54**, 234T; Walkley, *J., Agric. Sci.*, 1935, **25**, 398) have recognised the advantages of pre-treatment with water.

digestion can be greatly accelerated by small quantities of hydrogen peroxide. By adopting that procedure, digestion of a resistant type of soil which may normally occupy five to six hours by the usual Kjeldahl method, can be completed in under ninety minutes. Commercial samples of hydrogen peroxide often contain some nitrogen, but the necessary correction can be easily applied by a previous blank determination.

In his later researches, Sreenivasan [1934, 1] extended his observations to different types of nitrogenous materials. He showed that 'wet' digestion is always quicker and smoother and, in many cases, yields higher and more consistent estimates than those obtained by the 'dry' method. The beneficial effects of wet digestion are best seen in the case of materials containing silicates. Sreenivasan [1934, 2] also showed that addition of small amounts of an oxidising agent—peroxide, perchlorate, permanganate or dichromate—greatly increases the rate and general efficiency of digestion.

The use of oxidising agents naturally precludes nitrates from the estimate of total nitrogen. This is rather disadvantageous, because although most soils and biological materials in general contain no more than traces of nitrates, there are yet others which contain useful quantities of that constituent, which cannot therefore be ignored. In view of this, Sreenivasan [1935] subsequently modified his method to include nitrates. The procedure consists in pre-treating the soil with dilute alkali and Devarda's alloy. The reaction, which proceeds in the cold, effectively reduces even large quantities of nitrates into ammonia. Subsequent treatment with sulphuric acid and small quantities of an oxidising agent followed by the usual digestion yields accurate estimates of total nitrogen including nitrates.

The foregoing modifications, though constituting distinct improvements on the existing methods, are still based on the same principle as the original Kjeldahl method. They also share some of its defects, the chief among which is the emission of objectionable acid fumes. These defects are sought to be remedied and the digestion conducted at a very much faster rate according to the method of oxidative digestion proposed by Narayanayya and Subrahmanyan [1935]. The procedure consists in heating the material to be digested with a mixture of dichromate and sulphuric acid (2 : 1)when the organic carbon is rapidly oxidised, leaving only the nitrogen in the residue, chiefly as ammonium sulphate. A small part of the nitrogen remains in combination with the chromium in the digest, but this can be released by treatment with a suitable reducing agent such as zinc. The small quantities of nitric acid found during oxidation can also be reduced to ammonia by that treatment. After reduction, the digest is distilled with excess of alkali in the usual way.

The only important condition to be observed is that the vapours should be cooled (with a air or water-cooled condenser) during digestion. Otherwise, there will be a small loss of nitrogen through volatilisation of nitric acid. Chlorides

(and halides in general) interfere with the digestion and cause loss of nitrogen in the elementary form, but this can be prevented by adding small quantities of a mercury salt, preferably, the oxide.

The products of chromic acid digestion of organic nitrogen were studied by Shewan [1935], Acharya [1935, 1] and Harihara Iyer *et al.*, [1935]. Shewan drew attention to loss of nitrogen when different nitrogenous substances are heated with the sulphuro-chromic acid mixture in the usual way. Acharya, who studied the products of digestion at low temperatures and under reduced pressure, showed that a considerable part of the nitrogen is converted into nitrate. He also traced [1935, 2] a relation between the structure of the starting material and extent of loss of nitrogen. While agreeing with these findings, Harihara Iyer, Rajagopalan and Subrahmanyan [1935] showed that the loss of nitrogen —at any rate in the case of soils and the commoner biological materials—can be entirely avoided by adding chromic acid (saturated aqueous solution) or solid dichromate to a boiling mixture of the material to be digested with 2 : 1 sulphuric acid. They adduced evidence to show that the loss of nitrogen observed by Shewan was due to the intermediary formation of ammonium dichromate which undergoes partial decomposition on heating the digesting mixture. The loss occurs between 130° and 150°c. On adding chromic acid to the boiling mixture (temp. 170°C.) with 2 : 1 acid (by volume), the intermediary formation of ammonium dichromate and the attendant loss of nitrogen are avoided. Based on these and other observations, Harihara Iyer *et al.* proposed a modified method of oxidative digestion which may be outlined as follows. The material to be digested (soil, 10 grms ; others in proportion) is weighed out into the distilling flask (cap. about 1,500 c.c.) and then treated with mercuric oxide (2 grms) water (20 c.c.) and sulphuric acid (40 c.c.). The flask is then fitted with an air or water cooled condenser and the mixture raised to boil. Saturated aqueous solution of chromic anhydride (about 5 c.c.) is then introduced through the condenser in small instalments at a time. The boiling is continued for thirty minutes after which the heating is stopped and the digest diluted with water. It is then treated with sodium sulphite (about 10 grms.) and raised to boil. (The sulphite need not be weighed out but may be added in small instalments at a time until there is a pronounced smell. When the reduction of unused chromic acid is complete, there is also a characteristic change in colour from bright green to pale blue). After boiling for about two minutes, pure zinc (2 grms.) is added and the heating continued for a further period of about ten minutes. The contents of the flask are then cooled and distilled with excess of alkali in the usual way.

Some of the chemicals used for the determination (*e.g.*, zinc, chromic anhydride) may contain minute quantities of nitrogen. The necessary correction can be applied by a blank determination.

The above procedure has several advantages over the Kjeldahl method. In the first place, it does away with fumes. The digestion proceeds very rapidly

and is, in fact, mostly complete within the first five minutes after addition of the oxidising agent. The extra boiling is only to ensure complete digestion. A further advantage is that the estimate includes nitrates, so that no special treatment is needed to include that form of nitrogen. The use of the condenser helps to retain not only the nitrate already present in the soil, but also any that may be added to it. Both the digestion and the distillation proceed smoothly and are in fact very much more satisfactory than similar operations in the Kjeldahl method. The related processes are so simple and, at the same time, so rapid that, given a number of distilling sets, a single worker can easily complete about thirty determinations a day.

It is well known that during chromic acid digestion the entire quantity of organic carbon is converted into carbon dioxide. Although the previous attempts to combine the wet combustion of carbon with the estimation of nitrogen in the residue have not been successful, it should still be possible, with the better knowledge of the products of reaction, to standardise conditions so as to facilitate both the determinations on the same sample. Some promising results in this direction have already been obtained by Bhaskaran, Harihara Iyer and Rajagopalan (private communication) who have shown that by using a modification of the apparatus originally designed by Subrahmanyan *et al.* [1934] for the wet combustion of soils and adding chromic acid to the boiling acid mixture, accurate estimates of both the constituents can be carried out. Using the same apparatus, carbonate carbon can also be separately estimated by preliminary distillation with phosphoric acid. By adding sufficient excess of that acid, alkali earth carbonates, can be completely decomposed, the corresponding acid phosphates (which are soluble) being formed. (Neither sulphuric acid nor hydrochloric acid is suitable for this purpose. The former forms the insoluble sulphate which forms protective coats around unattacked carbonate particles, so that correct estimates are not generally obtained when more than about 1 per cent. of alkali earth carbonates are present. Dilute hydrochloric acid yields accurate estimates of carbonate but the residue is not suitable for the estimation of organic carbon because of the presence of excessive amounts of chloride in the medium). Subsequent digestion with sulphuric and chromic acids, in the manner previously outlined, yields correct values for organic carbon and total nitrogen.

It is generally recognised that progress in scientific research is largely determined by the accuracy, ease and rapidity of the methods employed. In many lines of modern chemical research—especially in agricultural chemistry and biochemistry in general—estimation of carbon and nitrogen form the most important determinations. It may be hoped, therefore, that with the foregoing and other developments, the agricultural chemist will soon be in a position to undertake very many more new lines of enquiry than has hitherto been possible.

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