

## 〈461〉 NITROGEN DETERMINATION

Some alkaloids and other nitrogen-containing organic compounds fail to yield all of their nitrogen upon digestion with sulfuric acid; therefore these methods cannot be used for the determination of nitrogen in all organic compounds.

### METHOD I

#### Nitrates and Nitrites Absent

Place about 1 g of the substance, accurately weighed, in a 500-mL Kjeldahl flask of hard borosilicate glass. The material to be tested, if solid or semisolid, may be wrapped in a sheet of nitrogen-free filter paper for convenience in transferring it to the flask. Add 10 g of powdered potassium sulfate or anhydrous sodium sulfate, 500 mg of powdered cupric sulfate, and 20 mL of sulfuric acid. Incline the flask at an angle of about 45°, and gently heat the mixture, keeping the temperature below the boiling point until frothing has ceased. Increase the heat until the acid boils briskly, and continue the heating until the solution has been clear green in color or almost colorless for 30 minutes. Allow to cool, add 150 mL of water, mix the contents of the flask, and again cool. Add cautiously 100 mL of sodium hydroxide solution (2 in 5), in such manner as to cause the solution to flow down the inner side of the flask to form a layer under the acid solution. Immediately add a few pieces of granulated zinc, and without delay connect the flask to a Kjeldahl connecting bulb (trap), previously attached to a condenser, the delivery tube from which dips beneath the surface of 100 mL of boric acid solution (1 in 25) contained in a conical flask or a wide-mouth bottle of about 500-mL capacity. Mix the contents of the Kjeldahl flask by gentle rotation, and distill until about four-fifths of the contents of the flask has distilled over. Titrate with 0.5 N sulfuric acid VS, determining the endpoint potentiometrically. Perform a blank determination, and make any necessary correction. Each mL of 0.5 N sulfuric acid VS is equivalent to 7.003 mg of nitrogen.

When the nitrogen content of the substance is known to be low, the 0.5 N sulfuric acid VS may be replaced by 0.1 N sulfuric acid VS. Each mL of 0.1 N sulfuric acid VS is equivalent to 1.401 mg of nitrogen.

#### Nitrates and Nitrites Present

Place a quantity of the substance, accurately weighed, corresponding to about 150 mg of nitrogen, in a 500-mL Kjeldahl flask of hard borosilicate glass, and add 25 mL of sulfuric acid in which 1 g of salicylic acid previously has been dissolved. Mix the contents of the flask, and allow the mixture to stand for 30 minutes with frequent shaking. To the mixture add 5 g of powdered sodium thiosulfate, again mix, then add 500 mg of powdered cupric sulfate, and proceed as directed under *Nitrates and Nitrites Absent*, beginning with "Incline the flask at an angle of about 45°."

When the nitrogen content of the substance is known to exceed 10%, 500 mg to 1 g of benzoic acid may be added, prior to digestion, to facilitate the decomposition of the substance.

### METHOD II

#### Apparatus

Select an appropriate 300-mL Kjeldahl flask, from which the nitrogen is first liberated by acid digestion and then transferred quantitatively to the titration vessel by steam distillation.

#### Procedure

Place an accurately weighed or measured quantity of the material, equivalent to 2 to 3 mg of nitrogen, in the digestion flask of the apparatus. Add 1 g of a powdered mixture of potassium sulfate and cupric sulfate (10:1), and wash down any adhering material from the neck of the flask with a fine jet of water. Add 7 mL of sulfuric acid, allowing it to rinse down the wall of the flask, then, while swirling the flask, add 1 mL of 30 percent hydrogen peroxide cautiously down the side of the flask. (Do not add hydrogen peroxide during the digestion.)

Heat the flask over a free flame or an electric heater until the solution has a clear blue color and the sides of the flask are free from carbonaceous material. Cautiously add to the digestion mixture 70 mL of water, cool the solution, and arrange for steam distillation. Add through a funnel 30 mL of sodium hydroxide solution (2 in 5) in such manner as to cause the solution to flow down the inner side of the flask to form a layer under the acid solution, rinse the funnel with 10 mL of water, tightly close the apparatus, and begin the distillation with steam immediately. Receive the distillate in 15 mL of boric acid solution (1 in 25), to which has been added 3 drops of methyl red-methylene blue TS and sufficient water to cover the end of the condensing tube. Continue the distillation until the distillate measures 80 to 100 mL. Remove the absorption flask, rinse the end of the condensing tube with a small quantity of water, and titrate the distillate with 0.01 N sulfuric acid VS. Perform a blank determination, and make any necessary correction. Each mL of 0.01 N sulfuric acid VS is equivalent to 140.1 µg of nitrogen.

When a quantity of material containing more than 2 to 3 mg of nitrogen is taken, 0.02 N or 0.1 N sulfuric acid may be employed, provided that at least 15 mL is required for the titration. If the total dry weight of material taken is greater than 100 mg, increase proportionately the quantities of sulfuric acid and sodium hydroxide.