

METHOD 15.0

Effective 1st January 2018

HYDROCYANIC ACID

1. Scope and Field of Application

This method is for the determination of hydrocyanic acid, free and combined in the form of glycosides, in feeding stuffs and in particular in products derived from linseed, manioc flour and certain species of beans.

2. Principle

The sample is suspended in water. The hydrocyanic acid is released by the action of enzymes, separated by steam distillation and collected in a specific volume of acidified silver nitrate solution. The silver cyanide is separated by filtration and the excess silver nitrate is titrated with a solution of ammonium thiocyanate.

3. Reagents

3.1 Anti-foam (e.g. silicone).

3.2 Nitric acid (d=1.42g/ml).

3.3 Ammonia solution: Prepare by diluting one volume of ammonia (d=0.88g/ml) with two volumes of water.

3.4 Ammonium ferric sulphate, saturated solution.

3.5 Sweet almonds suspension: crush twenty blanched sweet almonds in 100ml water at 37 to 40°C. Check that there is no hydrocyanic acid in 10ml of the suspension using sodium picrate paper or by carrying out a blank test as described in the last paragraph of 5.

3.6 Sodium acetate solution, neutral to phenolphthalein: 10g sodium acetate, anhydrous, per 100ml.

3.7 Ammonium thiocyanate solution, 0.02N.

3.8 Silver nitrate solution, 0.02N.

4. Apparatus

4.1 Oven regulated at 37-38°C.

4.2 Apparatus for steam distillation fitted with a condenser with a curved extension piece.

4.3 1 litre flat-bottomed flasks with ground-glass stoppers.

4.4 Oil bath.

4.5 Burette graduated to 0.05ml.

5. Procedure

Weigh to the nearest 0.005g, approximately 20g of the prepared sample, place in a 1 litre flat-bottomed flask (4.3) and add 50ml of water and 10ml of sweet almond suspension (3.5). Stopper the flask and transfer to the oven (4.1) for sixteen hours at 37-38°C. Cool to room temperature, add 80ml of water, 10ml of sodium acetate solution (3.6) and a drop of anti-foam (3.1).

Connect the flask to the steam distillation apparatus (4.2) and place in the oil bath (4.4) which has first been brought to a temperature slightly above 100°C. Distil 200 to 300ml of liquid by passing a current of steam through the flask and gently heating the oil bath. Collect distillate in an Erlenmeyer flask protected from the light and containing exactly 50ml of silver nitrate solution 0.02N (3.8) and 1ml of nitric acid (3.2). Make sure that the

condenser's extension piece is immersed in the silver nitrate solution.

Transfer the contents of the Erlenmeyer flask to a 500ml graduated flask, make up to volume with water, mix and filter. Remove 250ml of the filtrate, add approximately 1ml ammonium ferric sulphate solution (3.4) and titrate the excess silver nitrate with the solution of ammonium thiocyanate 0.02N (3.7). A blank test may, if required, be carried out by applying the same procedure to 10ml of sweet almond suspension (3.5), omitting the sample.

6. Expression of the Results

6.1 Calculate the Hydrogen Cyanide (HCN) content using the formula

$$\text{HCN} = \frac{[(S-B) \times 0.54 / 1000] \times 100}{W}$$

Where:

- S = Sample titration (ml), where 1ml Silver Nitrate = 0.54mg HCN
BI = Blank titration (ml)
1000 = factor for conversion of mg to g
W = Mass of sample used (g)

If the blank test indicates that silver nitrate solution 0.02N has been consumed, subtract the value of this from the volume consumed by the distillate of the sample.

1ml of 0.02N AgNO₃ = 0.54mg of HCN.

Express the result as a percentage of the sample.

6.2 Take the result as the arithmetic mean of 2 determinations, if the requirement for repeatability (7.) is satisfied. If it is not, then the determinations should be repeated.

6.3 Report the result as a percentage to one decimal place.

7. Repeatability

The difference between results of two parallel determinations on the same sample must be less than 10% in relative value to the highest value obtained.

8. **NOTE:** If the sample (e.g. beans) contains a large quantity of sulphides a black precipitate of the silver sulphide is formed, which is filtered together with the silver cyanide deposit. The formation of this precipitate consumes silver nitrate solution and this effect must be allowed for in the calculation of the HCN content. To do this, proceed as follows. treat the deposit left on the filter with 50ml of ammonia (3.3) in order to dissolve the silver cyanide. Wash the residue in dilute ammonia and then determine its silver content. Convert the value obtained into ml of 0.02N silver nitrate solution and subtract this volume from the volume of silver nitrate solution consumed by the sample distillate.