METHOD SPECIFICATION
Department of Animal and Aquaculture Sciences, NMBU

Method name: Kjeldahl-N
BIOVIT No .: Msp1040

1. Method of analysis / Principle / Main instrument

The Kjeldahl method is used to determine the quantitative amount of nitrogen (NH_4 ^{x}) in a sample. It was originally developed in 1883 by the Danish chemist Johan Kjeldahl to determine the protein content of grains.

The method is an internationally recognized reference method for determining the protein content of feed. It can be used for almost all types of samples, such as food, raw materials, water, sludge, concentrates, roughage, fish feed, grain and artificial fertilizers. The method involves decomposing the amino acids in the protein using high temperature (420 ° C), strong acid (95% sulfuric acid) and a catalyst ($CuSO_4/K_2SO_4$).

It is important to be aware that the Kjeldahl method fails to measure the total nitrogen content of organic samples. Nitrogen atoms in oxidized nitrogen (NO_x) and nitrogen atoms in heterocyclic compounds (cyclic organic compounds containing at least one atom other than carbon in the ring chain) are not determined.

Decomposition

$$N(protein) + H_2SO_4\ (l) \xrightarrow{\frac{CuSO_4}{K_2SO_4}} CO_2\ (g) + SO_2\ (g) + H_2O\ (l) + NH_4^+\ (aq) + SO_4^+\ (aq)$$

Distilling

$$NH_4^+\ (aq) + OH^-\ (aq) \xrightarrow{\Delta} NH_3\ (g) + H_2O\ (l)$$

$$B(OH)_3\ (aq) + NH_3\ (aq) + H_20\ (l) \rightarrow NH_4^+\ (aq) + B(OH)_4^-\ (aq)$$

Titration

$$B(OH)_4^-\ (aq) + HCl\ (aq) \leftrightarrows B(OH)_3\ (aq) + H_20\ (l) + Cl^-\ (aq)$$

Equivalence

$$B(OH)_3\ (aq) + 2\ H_20\ (l) \leftrightarrows B(OH)_4^-\ (aq) + H_3O^+\ (aq)$$

Main instrument: Kjeltec 8400- automated distillation unit + support unit with scrubber (Foss, Denmark)

2. Reference and any modifications

AOAC Official method 2001.11 –Protein (crude) in animal feed, forage, grain and oilseeds. (Block Digestion with a Copper Catalyst and Steam Distillation into Boric Acid)

Modification: The samples are added with 15 mL of H_2SO_4 (> 95%) and boiled for 45 minutes. After cooling, 65 mL of RO water is added.

3. Requirements for the degree of grinding and temperature of the sample for storage before analysis

The samples must be homogenized before extraction - degree of grinding 1 mm

The analyzed analytical sample must contain between 1 - 200 mg N.

Sample amount: 0.5 - 1.0 g homogeneous sample

4. Contact persons

Lab leader: Hanne Kolsrud Hustoft

Responsible for analysis: Elin Kristoffersen / Heidi Askerud

5. Other literatur

1. Egli, H., Kjeldahl Guide, 1st edition, Büchi Labortechnik AG, Switzerland, 2008

2. Persson, J., Handbook for Kjeldahl Digestion, 4th edition, Sweden, 2008

3. Commission Regulation (EC) No 152/2009. 27 Jan 2009. Laying down the methods of sampling and analysis for the official control of feed. Annex III, P, Official Journal of the European Union L54 / 1 from 26/02/2009.