Crude Protein Analysis: Modified Kjeldahl Procedure

Materials:

250 ml glass digestion tubes
Digestion Block with exhaust manifold
Foss Kjeltec 2300

Reagents:

Concentrated (98%) Sulfuric Acid (VWR Cat # 2876-46, FW 98.08, CAS # 7664-93-9)

0.1142 N Sulfuric Acid (Ricca Cat # 8255-5, FW 98.08, CAS # 7664-93-9)

Sodium Hydroxide (40%) (Mallinckrodt Cat # SX059-1, FW 40.00, CAS # 1310-73-2 or Macron Cat # 770810, FW 40.00, CAS # 1310-73-2)

Boric acid, 4% Indicating (Ricca Cat # 1070-5, FW 61.83, CAS # 10043-35-3). Dilute to 2% by mixing 50:50 (v:v) 4% boric acid with 18 MOhm water.

Catalyst

Digestion Procedure:

Preheat digestion block to 420 °C.

1. Transfer 0.25-1 g (depending on protein content) sample in duplicate to digestion tube.

Note: At least one duplicate of chemical blank (acid and catalyst without sample) should be run per day to serve as a chemical blank.

2. Add 2 catalyst tablets to the tube. Add 15 ml conc. H_2SO_4.
3. Place full rack of tubes in preheated digestion block and put heat shields in place. Cover with exhaust manifold. Run water vacuum at high flow for 5 minutes, then reduce flow and close fume hood. Digest for 1 hour (samples should be clear green color).
4. Remove rack from digestion block, remove heat shields and allow to cool for 2-5 minutes with exhaust system running. Remove exhaust manifold and cool an additional 5 minutes.
5. While wearing safety glasses, carefully add 10-20 ml dd water with squeeze bottle to each tube to prevent solidification of sample.
6. Dilute each sample with approximately 90 ml dd water.

Distillation Procedure:

A technician is required to start up the Foss Kjeltec 2300.

1. Fill a digestion tube approximately one third full of water. Carefully seat tube in Kjeltec machine and begin distillation. Record machine readout and remove tube. Repeat this step until 2 values are reached that are within 0.007 or 0.008 of each other.
2. Place tubes, containing samples, on Kjeltec and distill as outlined above. Run chemical blanks first, these will serve as the correction factor for calculations. Record each value after distillation cycle is complete. These values indicate the milliliters of 0.1142 N H_2SO_4, which were titrated for each sample.

Calculations:

$$\% \, CP = \frac{[(ml \; acid \; titrated - ml \; acid \; in \; chemical \; blank) \times (0.1142 \times 1.401)]}{Sample \; weight \times DM} \times 6.25$$

0.1142 = Normality of Standardizing Acid

1.401 = mw N/10

Reference

AOAC official method 2001.11. Protein (Crude) in Animal Fee, Forage, Grains and Oilseeds, Official Methods of Analysis, 18th Ed, Revision 3, 2010