

AOAC PEPSIN INSOLUBLE NITROGEN
Up-dated September 2013

I. Reference:

AOAC. 1984. Official Methods of Analysis (14th ed). Association of Official Analytical Chemists. Washington, D.C.

II. Personal Protective Equipment:

- A. Lab coat
- B. Safety glasses/goggles
- C. Latex gloves

III. Reagents:

- A. Pepsin Solution – 0.2% pepsin (activity 1:10,000) in 0.075 N HCl. Sigma Catalog No. P 7000
 - 1. Prepare just before using
 - 2. Add 6.1 ml Conc. HCl too ~800 ml distilled H₂O
 - 3. Add 2.0 g pepsin and stir gently to dissolve (do not heat)
 - 4. q.s. to 1 liter

IV. Procedure:

- A. Weigh 0.3 g dried ground sample into 100 ml in vitro tube
- B. Add 50 ml freshly prepared pre-warmed (45°C) pepsin solution to each tube
- C. Be sure sample is completely wetted by pepsin solution. Incubate with constant agitation for 16 hours at 45°C.
- D. Filter with hot dH₂O through Whatman #541 filter paper. Wash thoroughly.
- E. Dry and run Kjeldahls on filter paper or scrape, mix, and weigh residue for LECO analysis

V. Calculations:

- A. Calculate as % pepsin-insoluble nitrogen or as % of total sample nitrogen