# Lecture 1

## PROXIMATE ANALYSIS OF FEEDSTUFF

### Sampling and Preparation for Analysis

Before undertaking an analysis the results of which are to be used to represent the composition of a consignment of a feedstuff, it is important that the sample is sufficient in amount and that it is selected properly from the bulk so as to be fairly representative of it. Sampling is however not a laboratory operation. The sample to be used, if necessary, is expected to be sufficiently dried to enable it to be finely ground.

## PROXIMATE ANALYSIS

This refers to the determination of the major constituents of feed and it is used to assess if a feed is within its normal compositional parameters or somehow being adulterated. This method partitions nutrients in feed into 6 components: water, ash, crude protein, ether extract, crude fibre and NFE.

### Moisture Determination

Moisture is determined by the loss in weight that occurs when a sample is dried to a constant weight in an oven. About 2g of a feed sample is weighed into a silica dish previously dried and weighed. The sample is then dried in an oven for $65^0$C for 36 hours, cool in a desiccator and weighed. The drying and weighing continues until a constant weight is achieved.

$$\%\text{Moisture} = \frac{\text{wt of sample + dish before drying - wt of sample+ dish after drying}}{\text{Wt of sample taken}} \times 100$$

Since the water content of feed varied widely, ingredients and feed are usually compared for their nutrient content on moisture free or dry matter (DM) basis.

%DM = 100 - %Moisture.

### Ether Extract

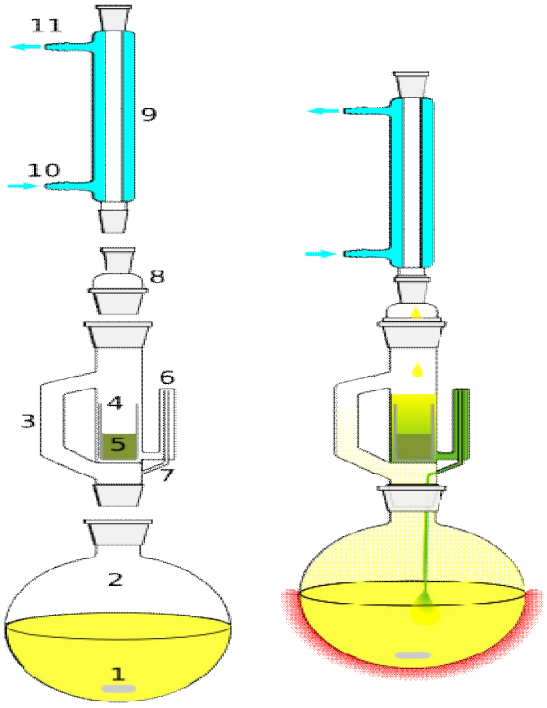
The ether extract of a feed represents the fat and oil in the feed. **Soxhlet apparatus** is the equipment used for the determination of ether extract. It consist of 3 major components

1. An extractor: comprising the thimble which holds the sample
2. Condenser: for cooling and condensing the ether vapour
3. 250ml flask

Procedure: about 150ml of an anhydrous diethyl ether (petroleum ether) of boiling point of 40-60$^0$C is placed in the flask. 2-5g of the sample is weighed into a thimble and the thimble is plugged with cotton wool. The thimble with content is placed into the extractor; the ether in the flask is then heated. As the ether vapour reaches the condenser through the side arm (distillation path) of the extractor, it condenses to liquid form and drop back into the sample in the thimble, the ether soluble substances are dissolved and are carried into solution through the syphon tube back into the flask. The extraction continues for at least 4 hrs. The thimble is removed and most of the solvent is distilled from the flask into the extractor. The flask is then disconnected and placed in an oven at 65$^0$C for 4 hrs, cool in desiccator and weighed.

$$\%\text{Ether extract} = \frac{\text{wt of flask + extract – tare wt of flask}}{\text{wt of sample}} \quad x100$$

**Soxhlet Apparatus**




***1:*** *Stirrer bar/anti-bumping granules* ***2:*** *Still pot (extraction pot) - still pot should not be overfilled and the volume of solvent in the still pot should be 3 to 4 times the volume of the soxhlet chamber.* ***3:*** *Distillation path* ***4:*** *Soxhlet Thimble* ***5:*** *Extraction solid (residue solid)* ***6:*** *Syphon arm inlet* ***7:*** *Syphon arm outlet* ***8:*** *Expansion adapter* ***9:*** *Condenser* ***10:*** *Cooling water in* ***11:*** *Cooling water out*

**Crude Fibre**

The organic residue left after sequential extraction of feed with ether can be used to determine the crude fibre, however if a fresh sample is used, the fat in it could be extracted by adding petroleum ether, stir, allow it to settle and decant. Do this three times. The fat-free material is then transferred into a flask/beaker and 200mls of pre-heated 1.25% $H_2SO_4$ is added and the solution is gently boiled for about 30mins, maintaining constant volume of acid by the addition of hot water. The buckner flask funnel fitted with whatman filter is pre-heated by pouring hot water into the funnel. The boiled acid sample mixture is then filtered hot through the funnel under sufficient suction. The residue is then washed several times with boiling water (until the residue is neutral to litmus paper) and transferred back into the beaker. Then 200mls of pre-heated 1.25% $Na_2SO_4$ is added and boiled for another 30mins. Filter under suction and wash thoroughly with hot water and twice with ethanol. The residue is dried at $65^0$C for about 24hrs and weighed. The residue is transferred into a crucible and placed in muffle furnace (400-$600^0$C) and ash for 4hrs, then cool in desiccator and weigh.

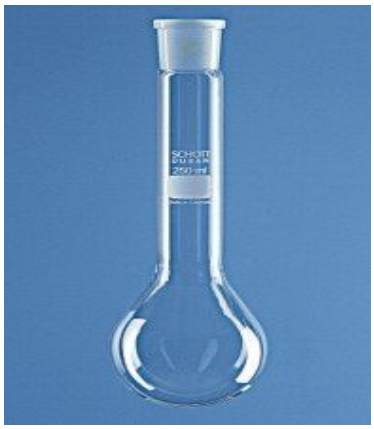
$$\%\text{Crude fibre} = \frac{\text{Dry wt of residue before ashing - wt of residue after ashing}}{\text{wt of sample}} \times 100$$

**Crude Protein**

Crude protein is determined by measuring the nitrogen content of the feed and multiplying it by a factor of 6.25. This factor is based on the fact that most protein contains 16% nitrogen. Crude protein is determined by **kjeldahl method.** The method involves: Digestion, Distillation and Titration.

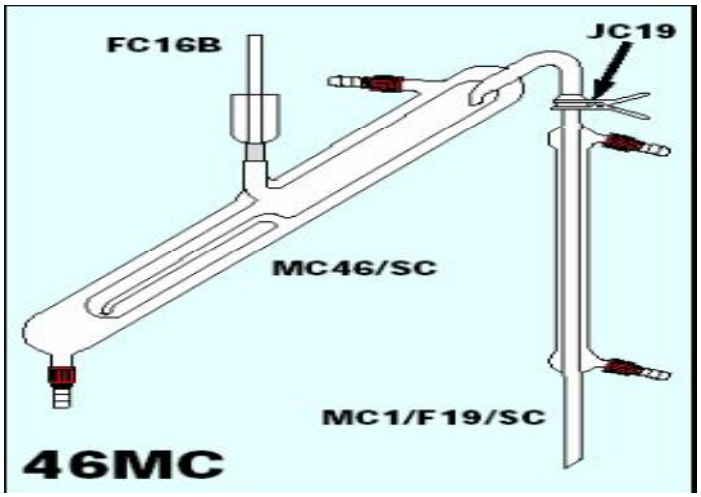
**Digestion**: weigh about 2g of the sample into kjeldahl flask and add 25mls of concentrated sulphuric acid, 0.5g of copper sulphate, 5g of sodium sulphate and a speck of selenium tablet. Apply heat in a fume cupboard slowly at first to prevent undue frothing, continue to digest for 45mins until the digesta become clear pale green. Leave until completely cool and rapidly add 100mls of distilled water. Rinse the digestion flask 2-3 times and add the rinsing to the bulk.

**Kjeldahl Flask**



**Distillation**: Markham distillation apparatus is used for distillation. Steam up the distillation apparatus and add about 10mls of the digest into the apparatus via a funnel and allow it to boil. Add 10mls of sodium hydroxide from the measuring cylinder so that ammonia is not lost. Distil into 50mls of 2% boric acid containing screened methyl red indicator.

**Markham Distillation Apparatus**




**Titration**: the alkaline ammonium borate formed is titrated directly with 0.1*N* HCl. The titre value which is the volume of acid used is recorded. The volume of acid used is fitted into the formula which becomes

$$\%N = \frac{14 \times VA \times 0.1 \times w \times 100}{1000 \times 100}$$

VA = volume of acid used w= weight of sample

%crude protein = %N x 6.25

**Ash**

Ash is the inorganic residue obtained by burning off the organic matter of feedstuff at 400-600$^0$C in muffle furnace for 4hrs. 2g of the sample is weighed into a pre-heated crucible. The crucible is placed into muffle furnace at 400-600$^0$C for 4hrs or until whitish-grey ash is obtained. The crucible is then placed in the desiccator and weighed

$$\%Ash = \frac{\text{wt of crucible+ash} - \text{wt of crucible}}{\text{wt of sample}}$$

**Nitrogen Free Extract (NFE)**

NFE is determined by mathematical calculation. It is obtained by subtracting the sum of percentages of all the nutrients already determined from 100.

$$\%NFE = 100-(\%moisture + \%CF + \%CP + \%EE + \%Ash)$$

NFE represents soluble carbohydrates and other digestible and easily utilizable non-nitrogenous substances in feed.