ALLAMA IQBAL OPEN UNIVERSITY, ISLAMABAD
(Department of Home & Health Sciences)

Course: Basic of Food Sciences (7511) **Semester: Autumn, 2012**
Level: M. Sc **Credit: 3(2+1)**

CONTENT LIST

Following items are included in the study pack.

Note: If any one of the above items is missing from your study pack, please contact:

The Mailing Officer
Mailing Section
Services & Operational Unit
AIOU, H-8, Islamabad

ALLAMA IQBAL OPEN UNIVERSITY, ISLAMABAD

(Department of Home and Health Sciences)

WARNING

1. **PLAGIARISM OR HIRING OF GHOST WRITER(S) FOR SOLVING THE ASSIGNMENT(S) WILL DEBAR THE STUDENT FROM AWARD OF DEGREE/CERTIFICATE, IF FOUND AT ANY STAGE.**
2. **SUBMITTING ASSIGNMENTS BORROWED OR STOLEN FROM OTHER(S) AS ONE'S OWN WILL BE PENALIZED AS DEFINED IN "AIOU PLAGIARISM POLICY".**

Course: Basic of Food Sciences (7511) **Semester: Autumn, 2012**
Level: Postgraduate **Credit: 3(2+1)**
Total Marks: 100 **Pass Marks: 40**

ASSIGNMENT No. 1

Q. 1 Write short notes on the following: **(16)**
- a) Food as a source of energy
- b) Reducing sugars
- c) Phytochemicals
- d) Organic fruits and vegetables

Q. 2 Discuss protein structure in detail. Explain various categories of amino acids. **(12)**

Q. 3 Describe important properties of fats and oils with view point of food processing. **(12)**

Q. 4 Explain chemistry of water. Discuss importance of water in food preservation and shelf life of food. **(12)**

Q. 5 Describe physical and chemical composition of meat in detail. **(12)**

Q. 6 Elaborate various steps and procedures involved in industrial milk processing. **(12)**

Q. 7 Explain effects of pigments on the value of different fruits and vegetables. **(12)**

Q. 8 Write down process of emulsion formation. Explain different types of emulsifiers used in food processing. **(12)**

ASSIGNMENT No. 2
(PRACTICALS)

Total Marks: 100
Pass Marks: 40

Practical No. 1
How to prepare different Solutions **(10)**

Practical No. 2
Measure the pH in a given sample **(10)**

Practical No. 3
Determination of moisture in wheat flour **(10)**

Practical No. 4
Determination of Total Ash **(10)**

Practical No. 5
Determination of total solids (Gravimetric Method) in milk **(10)**

Practical No. 6
Determination of protein by Kjeldahl Method **(10)**

Practical No. 7
Determination of Crude Fiber **(10)**

Practical No. 8
Determination of Fat Contents **(10)**

Practical No. 9

a) Viva voce of the experiments carried out during all sessions **(10)**
b) Submission of practical notebooks **(10)**

References

Annexure

STUDENTS GUIDE

The second assignment is of practical nature to be completed during the 3 days workshop, which will be held according to the schedule. The practicals will be demonstrated in one of the specified laboratories by AIOU. In order to complete these assignments, the students are asked to note the following points.

1. The practical based workshop will be held at the end of the semester.
2. There are 10 practical sessions in this course, which require duration of at least 3 days.
3. The students will have to take 3 days leave from their work place for this purpose.
4. The timings for the practicals will be as per workshop schedule sent to you.
5. Students will have to prepare a practical workbook to record the methodology; however practicals will be demonstrated during the workshops.
6. The completed workbook will be marked by the laboratory tutors.
7. Your text book and brief practical manual is being provided along with this assignment.
8. Wherever there is ambiguity in methods and procedures, the students are advised to consult their tutor in the practical laboratory.

INSTRUCTIONS REGARDING THE 2ND ASSIGNMENT

TUTOR GUIDE

This assignment will be completed under the close supervision of tutors. The tutors are requested to follow the following points.

1. These practicals will be conducted during 3 days workshop and will be demonstrated/conducted in a relevant laboratory arranged by AIOU.
2. Arrange the equipment, chemicals, culture media etc in advance, a day before the practical session is to be started.
3. The tutor should guide the students about the next day's practical and activities.
4. It will be better if the overlapping practical are conducted once for all practical sessions. In this way two practicals can be conducted simultaneously in order to save time.
5. A brief practical manual is also being provided to the tutors and students, which contains all procedures required to conduct the enlisted experiments. The tutor should also guide the students about the specific procedures and methods.
6. The timings for the workshop will be 8:30 am to 2:30 pm unless communicated by the regional office concerned. The students will also prepare an observation/practical workbook in which they will record the methodology and result of experiments, which will be demonstrated by tutors in the laboratory. This

book will be evaluated by the tutor and will be treated as 2nd assignment of the course 7511. After marking this book the tutor will submit the result to the concerned Regional Office.

GUIDE LINE FOR PRACTICALS

Course: Basic of Food Sciences (7511)

PRACTICAL No. 1

How to Prepare Different Solutions

In order to prepare different concentration solution we must have knowledge of some other commonly used terms.

Classification of Mixtures
Following mixtures of substances are far more common in Food Analysis.

Heterogeneous Mixtures
Heterogeneous Mixtures are those in which the mixing is not uniform and which therefore have regions of different compositions — i.e., there are observable boundaries between the components (e.g., ice-water, salad dressing, milk, dust in air).

Homogeneous mixtures
Heterogeneous Mixtures are those in which the mixing *is* uniform and which therefore have a constant composition throughout; there are no observable boundaries because the substances are intermingled on the molecular level (e.g., salt water, sugar water, air).
The composition of a mixture is variable, and it retains the properties of some of its components.

Types of Homogeneous Mixtures
Homogeneous mixtures can be classified according to the size of their constituent particles:
Solutions contain particles with diameters in the range of 0.1 to 2 nm (the typical size of ions or small molecules). Solutions are transparent and do not separate on standing. (ex.: salt water, sugar water, gasoline, air)
Colloids contain particles with diameters in range of 2 to 500 nm. They are often opaque, but also do not separate on standing. (ex.: milk, fog, soap in water)
Suspensions are mixtures having even larger particles; these are not truly homogeneous, because the particles separate on standing and are visible with microscopes. (ex.: blood, aerosol sprays).

Solutes and Solvents
- A solution consists of a *solute* and a *solvent*:
  - **solute**—the substance which is being dissolved.

– **solvent** —the substance (usually a liquid) that dissolves the solute (usually, the solvent is the most abundant component in the mixture).

• **Aqueous solution** are solutions in which the solvent is water.

Common Laboratory Solvents

Common Polar Solvents	Common Nonpolar Solvents
Water (H_2O)	Hexane (C_6H_{14})
Acetone (CH_3COCH_3)	Diethyl ether ($CH_3CH_2OCH_2CH_3$)*
Methanol (CH_3OH)	Toluene (C_7H_8)
Ethanol (CH_3CH_2OH)	Carbon tetrachloride (CCl_4)

Kinds of Solutions

Solutions are most commonly solids or liquids dissolved in another liquid, but other kinds of solutions are possible:

Gaseous solutions: All gases are infinitely soluble in one another (e.g., the atmosphere). Small amounts of non polar gases can dissolve in water due to dipole-induced dipole attractions.

– At 25°C and 1 atm. pressure, only 3.2 ml of O2 dissolves per 100 ml of water, but this small solubility is essential to life in aquatic environments.

– The solubility of oxygen and carbon dioxide in water is enhanced by some chemical processes.

Solid solutions: Gases, liquids, or other solids can be dispersed in solids. Waxes and metal **alloys** are types of solid-solid solutions.

Saturated and Unsaturated Solutions

A saturated solution contains the maximum amount of dissolved solute, and cannot (usually) dissolve any more of the solute.

An unsaturated solution contains less than the maximum amount of dissolved solute.

A supersaturated solution contains a greater-than normal amount of a solute; these solutions are unstable, and a slight disturbance causes the "extra" solute to precipitate out.

Units of Concentration

The **concentration** of a solution is the amount of solute dissolved in a certain amount of solution (or solvent).

– a **dilute solution** contains small quantities of solute relative to the amount of solvent.

– a **concentrated solution** contains large quantities of solute relative to the amount of solvent.

There are several common units which are used for expressing concentration:

Molarity (M)

$$\text{Molarity} = M = \frac{\text{moles of solute}}{\text{Liters of solution}} = \text{mol L}^{-1}:$$

- A solution of desired molarity is made by dissolving the required number of moles of solute ***to*** the desired volume (remember it's moles per liter of *solution*, **NOT** per liter of *solvent*).

- To prepare a 1.000 M solution of NaCl in water, you would dissolve 1.000 mol of NaCl in enough water to make 1 L of solution.
  - Molarity is temperature-dependent, since volume and density are affected by the temperature.
  - Solution volumes are not necessarily additive.

- **Molal concentration**, or **molality**, ***m*** (mol/kg):
  - To prepare a 1.000 *m* solution of NaCl in water, you would dissolve 1.000 mol of NaCl in 1 kg of water.
  - Since molality is temperature-independent, this unit is often used when describing the physical properties of a solution (especially colligative properties).
  - Molality is additive, unlike molarity.
  - Molarity and molality are similar for dilute aqueous solutions

Molality (m)

- **Molal concentration**, or **molality**, ***m*** (mol/kg):

$$\text{Molality} = m = \frac{\text{moles of solute}}{\text{kilograms of solvent}} = \text{mol kg}^{-1}$$

  - To prepare a 1.000 *m* solution of NaCl in water, you would dissolve 1.000 mol of NaCl in 1 kg of water.
  - Since molality is temperature-independent, this unit is often used when describing the physical properties of a solution (especially colligative properties).
  - Molality is additive, unlike molarity.
  - Molarity and molality are similar for dilute aqueous solutions.

Mass Percent (mass %)

Mass percent, **mass % (*w/w*)**:

$$\text{Mass percent} = \frac{\text{mass of solute}}{\text{mass of solution}} \times 100\%$$

Parts per million, **ppm**:

$$\text{ppm} = \frac{\text{mass of solute}}{\text{mass of solution}} \times 10^6$$

Parts per billion, **ppb**:

$$\text{ppb} = \frac{\text{mass of solute}}{\text{mass of solution}} \times 10^9$$

– temperature independent, but it's more difficult to measure liquids by mass.
– The density of a solution must be known to convert mass %, ppm, and ppb to molarity.

Volume percent, **% (*v/v*)**:

$$\text{Volume percent} = \frac{\text{volume of solute}}{\text{volume of solution}} \times 100\%$$

– Volume percent is used most often for mixtures of liquids or mixtures of gases (e.g., rubbing alcohol is 70% isopropyl alcohol in water by volume).

Mole fraction, ***X***:

$$\text{X} = \frac{\text{moles of solute}}{\text{moles of solution}} = \frac{n_{\text{solute}}}{n_{\text{solute}} + n_{\text{solvent}}}$$

$$\text{Mole percent (mol \%)} = \text{X} \times 100\%$$

Mole fractions are independent of temperature, and are useful in dealing with gas concentrations.

PRACTICAL No. 2

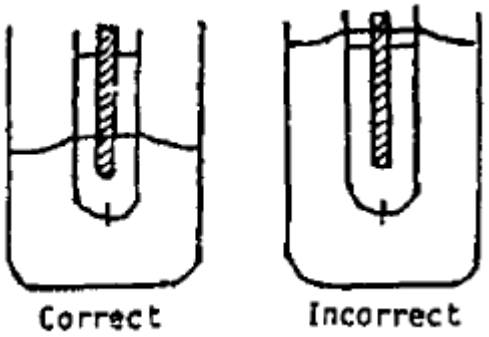
Measure the pH in a Given Sample

Theory and Guide Line

It is very important that the pH meter be operated and maintained properly. One should always follow the specific instructions provided by the manufacturer. For maximum accuracy, the meter should be standardized using two buffers (two-point calibration). Select two buffers of pH values about 3 pH units apart, bracketing that of the antidpated sample pH. The three standardization buffers used most widely in laboratories are a pH 4.0 buffer, a pH 7.0 buffer, and a pH *9.0* buffer (at 25°C). These are the typical pink, yellow, and blue solutions found adjacent to pH meters in many laboratories.

When standardizing the pH electrode, follow manufacturer's instructions for one-point

calibration; rinse thoroughly with distilled water and blot dry. Immerse electrode in the second buffer (pH 4, for example) and perform a second standardization. This time, the pH meter slope control is used to adjust the reading to the correct value of the second buffer. Repeat these two steps; if necessary, until a value within 0.1 pH unit of the correct value of the second buffer is displayed. If this cannot be achieved, the instrument is not in good working condition. Electrodes should be checked, remembering that the reference electrode is more likely in need of attention. One should always follow the electrode manufacturer's specific directions for storage of a pH electrode. In this way, the pH meter is always ready to be used and the life of the electrodes is prolonged. One precaution that should be followed pertains to a calomel reference electrode. The storage solution level always should be at least 2 em below the saturated KCl solution level in the electrode in order to prevent diffusion of storage solution into the electrode (Fig. 7-2).



Correct and incorrect depth of calomel electrodes in solutions.

Procedure:

1) Standardize the pH meter by using any two buffer mentioned above. When standardizing the pH electrode, follow manufacturer's instructions for one-point calibration; rinse thoroughly with distilled water and blot dry. Immerse electrode in the second buffer (pH 4, for example) and perform a second standardization. This time, the pH meter slope control is used to adjust the reading to the correct value of the second buffer. Repeat these two steps; if necessary, until a value within 0.1 pH unit of the correct value of the second buffer is displayed. If this cannot be achieved, the instrument is not in good working condition.
2) Take adequate amount of sample whose pH is needed to be determine
3) Then immerse the electrode in solution and to control the temperature you can use temperature probe.
4) When reading is stable, note that value and that is pH of your sample
5) One should always follow the electrode manufacturer's specific directions for

storage of a pH electrode. In this way, the pH meter is always ready to be used and the life of the electrodes is prolonged. One precaution that should be followed pertains to a calomel reference electrode. The storage solution level always should be at least 2 mm below the saturated KCl solution level in the electrode in order to prevent diffusion of storage solution into the electrode.

PRACTICAL No. 3
Determination of Moisture in Wheat Flour

Definition

The moisture content of a sample is the loss in weight when it is dried in accordance with these rules. It is expressed as a percentage of the weight of the original sample.

Apparatus

(a) Grinding mill- capable of grinding rapidly and uniformly without development of appreciable heat. The ground material should pass through 1.0mm I.S sieve

(b) Moisture dishes-made of aluminum or stainless steel approx 7.5mm wide and 2.5 mm deep with tight fitting lids and having an effective surface area enabling the test portion to be distributed so as to give a mass per unit area of not more than 0.3 g/cm².

(c) Constant-temperature oven, electrically heated, controlled in such a way that, during normal working, the temperature of the air and of the shelves carrying the test portions is within the range 130 to 133°C in the neighbourhood of the test portions. Desiccators- containing an effective desiccant

Procedure

(1) Mix the test sample and grind suitable quantity to give sufficient ground material for replicate determination. Ensure that the sample is neither too coarse not too fine and passes through 1.0 mm sieve.

(2) Weigh accurately 5gm of sample in previously dried and tarred dish and place the dish with its lid underneath in the oven for 2 hours. The time should be reckoned from the moment the oven attains 130 °C after the dishes have been placed.

(3) Remove the dish after 2 hours, cool in the desiccators and weigh

Calculations

Moisture percent = $(W_1-W_2) \times 100 / W_1-W$

Where

W_1= Weigh in gm of the dish with material before drying

W_2= Weigh in gm of the dish with material after drying

W = Weigh in gm of the empty dish

PRACTICAL No. 4
Determination of Total Ash

Ignite the dried material in dish left after the determination of moisture with flame of burner till charred. Transfer to a muffle furnace maintained at 550-600 °C and continue ignition till grey ash is obtained. Cool in desiccators and weigh. Repeat the process of

heating, cooling and weighing at half hour interval till the difference in weight in two consecutive weighing is less than 1mg. Note the lowest weight.

Calculations

Moisture percent = $(W_2\text{-}W) \times 100 / W_1\text{-}W$

Where

W_2= Weigh in gm of the dish with ash

W= Weigh in gm of the empty dish with material after drying

W_1= Weigh in gm of dish with dried material taken for test

PRACTICAL No. 5

Determination of Total solids (Gravimetric method) in Milk

Principle

Pre drying of a test portion on a boiling water bath and subsequent evaporation of the remaining water in a drying oven at a temperature of 102±2°C

Apparatus

a) Analytical balance

b) Desiccators provided with an efficient desiccant (for example freshly dried silica gel with a hydrometric indicator)

c) Boiling water bath provided with openings of adjustable size.

d) Drying oven, ventilated capable of being maintained thermostatically at 102±2°C throughout the total working space.

e) Flat bottomed dishes of height 20-25mm, dia 50-75 mm and made of appropriate material (stainless steel, nickel or aluminums) provided with well fitted readily removable lids.

f) Water bath capable of being maintained at 35°C-40°C

Preparation of sample

Transfer sample to a beaker, warm slowly to 35-40 °C on a water bath with careful mixing to incorporate any cream adhering to the sample. Cool the sample quickly to room temperature.

Procedure

Heat a dish with its lid alongside in the drying oven at least 1 hour. Place the lid on the dish and immediately transfer to desiccators. Allow to cool to room temperature (at least 30 mins) and weigh to the nearest 0.1mg. Add 5ml of prepared sample, place the lid on the dish and weigh again. Place the dish without the lid on the vigorously boiling water bath in such a way that the bottom of the dish is directly heated by the steam. Continue heating till most of the lid and transfer to the desiccators. Allow the dish to cool and weigh to the nearest 0.1mg. again heat the

dish with its lid alongside in the oven for 1 hour. Place the lid on the dish and immediately transfer to desiccators. Allow to cool and weigh again. Repeat the operation again until the difference in the two consecutive weighing does not exceed 1mg. record the lowest mass.

Calculation

Total solid content = m_2-m_0 x100/m_1-m_0

Where $m_{0=}$ mass in gm of dish+lid

PRACTICAL No. 6

Determination of Protein by Kjeldahl Method

The protein content is determined from the organic nitrogen by Kjeldahl method. The various nitrogenous compounds are converted into ammonium sulphate by boiling with concentrated sulphuric acid method. The ammonium sulphate formed is decomposed with an alkali (NaOH) and the ammonia liberated is absorbed in excess of standard solution of acid and then back titrated with standard alakali.

Appratus

a) Kjeldahl digestion flask- 500 or 800ml
b) Kjeldahl distillation apparatus, - same digestion flask fitted with rubber stopper through which passes lower end of efficient rubber bulb or trap to prevent mechanical carryover of NaOH during distillation.
c) Conical flask, 250ml
d) Burette 50ml.

Regents

a) concentrated sulphuric acid- sp gr 1.84
b) Sodium Hydroxide solution-45%. Dissolve 450 gm of Sodium Hydroxide in 1000ml water
c) Standard Sulphuric acid solution-0.1N
d) Standard Sodium Hydroxide solution-0.1N
e) Methl red indicator solution- dissolve 0.5gm methyl red in 100 ml of alcohol

Procedure

Weigh quickly about 5-8 g of the prepared ice cream sample and transfer to a 500 or 80ml Kjeldahl flask taking care to see that no portion of the sample clings to the neck of the flask. Add o.7gm of Mercuric oxide, 15 gm of Potassium Sulphate and 40ml of concentrated sulphuric acid. Add two to three glass beads. Place the flask in inclined position on the stand in the digestion chamber and digest. Heat the flask gently at low flame until the initial frothing ceases and the mixture boils steadily at a moderate rate. During heating rotate the flask several times. Continue heating for about an hour or more until the colour of the digest is pale blue. Cool the digest and add slowly 200 ml of water. Cool, add a piece of granulated Zinc or anti bump granules and carefully pour down the side of the flask sufficient Sodium Hydroxide

solution (45mgm/liter) to make the contents strongly alkaline (about 110ml) before mixing the acid and alkaline layer. Connect the flask to a distillation apparatus incorporating an efficient flash head and condenser. To the condenser fit a delivery tube which dips just below the surface of the pipetted volume on standard acid contained in a conical flask receiver. Mix the contents of the digestion flask and boil until 150 ml have distilled in to the receiver. Add 5 drops of methyl red indicator and titrate with 0.1 N Sodium Hydroxide solution. Carry out a blank titration.

1ml of 0.1 N H_2SO_4=0.0014gm N.

In case of dairy ice cream/kulfi calculate milk protein as N×6.38

PRACTICAL No. 7

Determination of Crude Fiber

Reagents

a) Dilute Sulphuric acid-1.25 percent(w/v) accurately prepared

b) Sodium hydroxide solution-1.25percent (w/v) accurately prepared

c) Ethyl alcohol- 95 percent by volume

Procedure

Weigh accurately about 2.5-3gm sample and transfer to an extraction apparatus (Soxhlet extractor) and extract with petroleum ether. Air dry the extracted sample and transfer to a dry 1 liter conical flask. Tak 200ml of sulphuric acid in a beaker and bring it to a boil. Transfer the whole of the boiling acid to the flask containing the defated material and immediately connect the flask with a water cooled refluxed condenso and heat so that the contents of the flask begin to boil within 1minute. Rotate the flask frequently taking care to keep material from rmaining on the sides of the flask and out of contact with the acid. Continue boiling for exactly 30 minutes. Remove the flask and filter through fine linen (about 18 threads to a cm) held in a funnel and wash with boiling water until the washings are no acid to litmus. Bring to boil some quantity of sodium hydroxide solution

Wash the residue on the linen in to the flask with 200ml of boiling sodium hydroxide solution. Immediately connect the flask to the reflux condenser and boil for exactly 30 minutes. Remove the flask and immediately filter through the filtering cloth. Thoroughly wash the residue with boiling water and transfer to a gooch crucible prepared with a thin compact layer of ignited asbestos. Wash the residue thoroughly first with hot water and then with about 15ml o ethyl alcohol. Dry the gooch crucible and contents at 105±2°C in an air oven until constant weight is achieved. Cool and weigh. Incinerate the contents of the gooch crucible in a muffle furnace furnace until all carbonaceous matter is burnt. Cool the gooch crucible containing ash in a dessicator and weigh.

Calculation

Crude fiber percent by wt=$(W_1\text{-}W_2) \times 100/ W$

W_1= wt in gm of gooch crucible and contents before ashing

W_2= wt in gm of gooch crucible containing asbestos and ash

W= wt in gm of dried material taken for the test

Calculate crude fiber on dry wt basis by giving correction for the moisture content

PRACTICAL No. 8

Determination of Fat Contents

Fat is important to all aspects of meat production and processing. Fresh and frozen meat prepared for manufacturing purposes is specified in terms of fat content (expressed as chemical lean). This is an important specification of commercial trading as well as being an important technical specification for product end-use. Manufacturing meat that is traded as a commodity on the international market is specified in terms of its fact content (expressed as chemical lean) and this is one of the primary product testing criteria for product imported by our overseas customers. Apart from the commercial importance of the fat content of unprocessed meat, especially manufacturing meat, fat content is an important technical and regulatory specification for almost all processed meat products. There are several rapid methods for determining the fat content of meat and meat products and these methods mostly produce results that are sufficiently accurate and reliable for routine product testing purposes. Given the importance of fat content however as a commercial and regulatory specification, it is necessary to have a method that is recognised as a standard and which can be referred to as a means of validating rapid methods and in dispute resolution processes. The "Soxhlet" method described here is recognised by the Association of Official Analytical Chemists (AOAC) as the standard method for crude fat analysis. In addition, some rapid instrumental methods are also approved by the AOAC.

Application

The Soxhlet method for determining crude fat content is a lengthy process requiring up to a day for a single analysis. The solvent extraction step alone takes six hours. The method is therefore not favoured for routine testing purposes in the meat industry; rather it is used as a standard reference method. As well as being used to determine the fat content of meat and meat products, the Soxhlet method can be used to determine the fat content of meat meal. In the case of meat meal, the Soxhlet method is often the method of choice as a routine test.

Outline of Method

Crude fat content is determined by extracting the fat from the sample using a solvent, then determining the weight of the fat recovered. The sample is contained in a porous thimble that allows the solvent to completely cover the sample. The thimble is contained in an extraction apparatus that enables the solvent to be recycled over and over again. This extends the contact time between the solvent and the sample and allows it time to dissolve all of the fat contained in the sample. In order for the solvent to thoroughly penetrate the sample it is necessary for the sample to be as finely comminuted as possible.

Before the solvent extraction step can begin the sample must be dried. Often a moisture analysis is required as well as a fat analysis and this can be achieved by accurately weighting the sample after drying and before extraction, as well as before drying. If a moisture analysis is not required the sample need only be weighed before drying and again after solvent extraction. In either case the sample must be weighed accurately on an analytical balance at each stage of the analysis.

When the sample is being weighed it is important not lose any part of it including any moisture that may weep from the sample during weighting. Loss of this moisture can be avoided by weighing the sample directly into a pre-dried extraction thimble or alternatively on to a pre-dried filter paper. If a moisture analysis is required, the dried extraction thimble or filter paper also has to be pre-weighed. After weighing, the sample (in the thimble or filter paper) can be placed in the oven for drying. After drying, the sample can be placed directly into the distillation apparatus for extraction.

Method

Equipment

- Analytical balance (at least 1 mg sensitivity).
- Electrical drying oven to be operated at 102ºC± 1ºC.
- Soxhlet extraction unit comprising:
  - Round bottom flask, 150 ml Soxhlet extractor with 60 ml siphoning capacity and condenser.
  - Cellulose extraction thimbles (28 x 80 mm)
- Fume cupboard
- Heat source, either electric heating mantle, or steam bath 100 ml beaker
- Desiccators with silica gel desiccant
- Glass rod

Reagents

- Petroleum spirit boiling point 60-80ºC
- Cotton wool free of fat
- Acid washed sand

Procedure

Note: Steps 8 – 12 are performed in a fume cupboard.

1. Rinse all glassware with petroleum spirit, drain, dry in an oven at 102ºC for 30 min. and cool in a desiccator.
2. Place a piece of cotton wool in the bottom of a 100 ml beaker. Put a plug of cotton wool in the bottom of an extraction thimble and stand the thimble in the beaker.
3. Accurately weigh 5 g of sample into the thimble. Add 1 - 1.5 g of sand and mix the sand and sample with a glass rod. Wipe the glass rod with a piece of cotton wool and place cotton wool in the top of the thimble. (Addition of sand is not required for analysis of meat meal). Dry the sample in an oven at 102ºC for 5 hours. The drying step may be omitted in the analysis of meat meal.

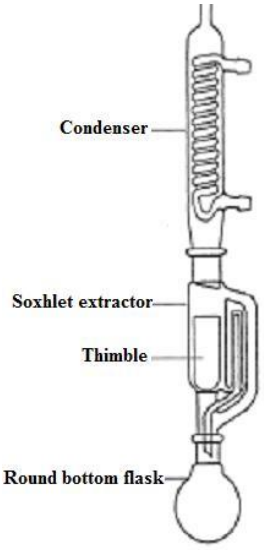


Figure 1: Soxhlet Extraction apparatus

4. Allow the sample to cool in a desiccators.
5. Take the piece of cotton wool from the bottom of the beaker and place it in the top of the thimble.
6. Insert the thimble in a Soxhlet liquid/solid extractor (Figure 1).
7. Accurately weigh a clean, dry 150 mL round bottom flash and put about 90 mL of petroleum spirit into the flask.
8. Assemble the extraction unit over either an electric heating mantle or a water bath.
9. Heat the solvent in the flask until it boils. Adjust the heat source so that solvent drips from the condenser into the sample chamber at the rate of about 6 drops per second.
10. Continue the extraction for 6 hours. For sausage meat and other emulsified products, the extraction should be performed in stages: Extract for about 4 hours, then remove the heat source and drain the solvent from the extractor in the flask. Remove the thimble from the extractor and transfer the sample to a 100 mL

beaker. Break up the sample with a glass rod. Return the sample to the thimble and replace the thimble in the extractor. Rinse the beaker with petroleum spirit and pour rinsings into the extract. Continue extraction for a further two hours.

11. Remove the extraction unit from the heat source and detach the extractor and condenser. Replace the flask on the heat source and evaporate off the solvent. (The solvent may be distilled and recovered).
12. Place the flask in an oven at 102ºC and dry the contents until a constant weight is reached (1-2 hours).
13. Cool the flask in a desiccator and weigh the flask and contents.
    Weight of empty flask (g) = W1
    Weight of flask and extracted fat (g) = W2
    Weight of sample = S
    % Crude fat = (W2 – W1) x 100 /S

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