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DIGESTIBILITY OF FEEDS IN FISH AND SHELLFISH  
AND METHODS TO DETERMINE DIGESTIBILITY

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Digestibility of a feedstuff can be described as the amount of feed that can be digested and absorbed by the animal in relation to consumption. Digestibility is measured as digestibility coefficient when it is expressed as percentage and as digestible energy which is most often used in warm water fishes. Knowledge of nutrient availability is necessary for effective substitution of ingredients for formulating low cost diets.

Factors affecting digestibility

Digestive coefficients are influenced by several factors and do not remain constant for a given feedstuff or species. Some of the factors affecting digestibility are:

- Nutrient composition of diet. This is a very important factor. Protein digestibility decreases with increase in carbohydrate in the diet in channel catfish. Similar observation made in some other fishes also.
- Gastric evacuation time. Lack of time for complete digestion and absorption which may result from increased food intake may affect digestibility. Amount of food consumed does not affect protein

digestion in fish but may have influence on digestion of lipids and carbohydrates.

- Increase in meal size reduces absorption in many fishes probably by reducing the surface area of the meal to be digested in comparison with small food particles.
- Crude fibre in diet. It has protective value for other nutrients and also help in easy passage of food through the alimentary canal.
- Microorganism in the diet.
- Temperature. Increase in temperature increases digestibility in some fish (channel cat fish). In carp, however, no change in protein digestibility over a range of temperature that supports growth, but digestibility reduces when temperature is reduced below this range.
- Species diversity

#### Nutrient digestibility in fish and shellfish

In general the digestible energy of fishes is 75-80% of the energy consumed and 96-98% of this digestible energy is metabolisable. Digestibility of proteins is affected by the presence of carbohydrates, both in quality and quantity, in fish and shellfish. In Penaeus japonicus 93% of digestion and absorption of glucose and galactose are completed in midgut but only 76-80% of sucrose and glycogen are digested and absorbed in this region. The process is completed in the hindgut. Starch digestibility is very good in warmwater fish and prawns. In common carp 85% of starch is absorbed at a dietary level of 10-60%.

Most animal proteins appears to be highly digestible in fish and prawns, as much as 99% of protein can be digested in some fishes.

Lipid digestibility in fishes varies from 70-94% depending on the nature of fat and water temperature. Oxidised lipids reduces digestibility of diets but oxidised lipids supplemented to the diets do not reduce digestibility of proteins in carp. When dietary fat is low, yearling carp secretes endogenous fats into the lumen of the intestine and this may affect estimation of apparent digestibility. In P. japonicus fatty acids are easily digested when esterified.

Differential absorption has also been observed for minerals. In common carp calcium absorption was more when its level in the diet was increased from 0.09 to 1.24% while phosphorus was held constant. Mineral absorption is also affected by the mineral base used in the diet formulation.

#### Determination of digestibility

Determination of digestibility involves measuring total quantity of nutrients ingested and the amount of corresponding nutrients egested.

$$\text{Digestibility} = \frac{\text{Amount of nutrients ingested} - \text{Amount of nutrients egested}}{\text{Amount of nutrients ingested}} \times 100$$

$$\text{True digestibility} = \frac{I - (F - F_k)}{I}$$

where I = Intake, F = Faeces egested and  $F_k$  = Endogenous metabolic faeces.

Endogenous materials such as secretion from within the intestinal tract, sloughed epithelial cells and other materials of metabolic origin also may occur in faeces. It is very difficult to measure such endogenous material and hence apparent digestibility is usually measured rather than true digestibility.

Apparent digestibility can be measured by direct faecal collection method or by indirect method using inert marker or indicator which can be external (added to the diet) or internal (integral part of the diet). Ideally, an indicator should be totally indigestible and excreted at the same time as the other gut contents. It should not reduce palatability of food and should be readily determined. Digestibility is determined indirectly by calculating the rate of indicator concentration in dry food and the same rate from the test food.

$$\text{Apparent digestibility of nutrients} = 100 - 100 \frac{\frac{\% \text{ indicator in food}}{\% \text{ indicator in faeces}}}{\frac{\% \text{ indicator in faeces}}{\% \text{ indicator in food}}} \times$$

The most popularly used indicator is chromic oxide,  $\text{Cr}_2\text{O}_3$ . Other indicators used are Polythene markers, radioactive phosphorus ( $\text{P}_{32}$ ), silica, Hydrolysis Resistant Organic Matter (HROM), Hydrolysis Resistant Ash (HRA), crude fibre, titanium (IV) oxide and metallic iron particles.

Faeces collection is very important in digestibility estimations. Faeces should represent quantitatively the ingested residue of the food consumed. Main problem in faeces collection is the leaching of nutrients in water. Faeces can be collected by sacrificing the animal, by stripping the intestine and from the rearing tank after egestion.

1. Direct faeces collection method

Materials needed:

- Specially designed aquaria for faeces collection. Troughs (closed system) can be used in its absence but faeces should be collected frequently if the latter is ned.
- Reagents and equipments required for protein, lipid and carbohydrate determination.
- Hot air oven
- Feeding trays, polythene tubes for siphoning out the faeces, aerators etc.

Procedure

The feeding trials should be conducted for 10-15 days. First the animals are to be acclimatised to the rearing conditions and to the test diet. Before the start of the experiment the animal has to be starved for one or two days for complete gut evacuation. Feeding with test diets should last at least 10 days and faeces should be collected at regular intervals, washed with little quantity of distilled water if the fish is reared in saline water, and dried in an hot air oven at 60°C. After the completion of the experiment weigh the total faeces collected and determine the nutrients in dry faeces.

Calculation

$$\text{Apparent digestibility} = \frac{I - F}{I} \times 100$$

where I = Nutrient intake and  
F = Faecal nutrient

## 2. Chromic oxide indicator method

The experimental design is same as in direct faeces collection method. The test animals should be fed with the test diet without chromic oxide for about a week before start of the experiment.

### Determination of chromic oxide content

The method involves oxidation of organic material in the sample using concentrated nitric acid followed by oxidation of insoluble (green) chromium III in chromic oxide to soluble (yellow) chromium IV, which is then determined spectrophotometrically.

### Reagents

- Conc. Nitric acid and Perchloric acid

### Apparatus

- Kjeldahl digestion tubes
- Heating mantles
- Gloves and Goggles for protection against acid fumes
- 5 ml graduated pipette with filling bulbs
- 100 ml volumetric flasks
- Spectrophotometer
- Cuvettes.

### Procedure

1. Weigh accurately 50-100 mg of sample into a piece of foil
2. Transfer sample carefully into a numbered Kjeldahl flask and reweigh the foil.
3. Add 5 ml conc.  $\text{HNO}_3$  washing the sample down from the sides of the flask.

4. Place the flask on a heating mantle and boil gently for 20 minutes. Do not allow to boil it dry, if it is add more  $\text{HNO}_3$ .
5. Turn off the mantle and allow to cool.
6. Add three ml of perchloric acid to flask with care.
7. Boil in the mantle until colour changes from green to yellow orange or brown and continue to boil for a further 10 minutes.
8. Allow the flasks to cool - if the colour reverts to green it must be reboiled.
9. Cool the flask to room temperature and then carefully wash the contents into a 100 ml volumetric flask with distilled water and make upto 100 ml.
10. Transfer an aliquote to a spectrophotometer cuvette and read optical density at 350 nm with distilled water as blank.

Calculation - 1

$$\begin{aligned} \text{Weight of sample} &= A \\ \text{Chromic oxide in the sample} &= \frac{\text{OD of sample}}{\text{OD of standard}} \times \text{Concentration of standard}^* = X \\ \% \text{ Chromic oxide in sample} &= \frac{X}{A} \times 100 \end{aligned}$$

\* Known concentration of chromic oxide should be treated in the same way as the sample for standardisation.

Calculation - 2

$$\begin{aligned} \text{Weight of sample} &= \text{Amg} \\ \text{Optical density} &= Y \\ \text{Weight of chromic oxide} &= \frac{Y-0.0032}{0.2089} = X \\ \text{in sample (mg)} & \\ \% \text{ chromic oxide in sample} &= \frac{X}{A} \times 100 \end{aligned}$$



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