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NUTRITIONAL NEEDS OF FINFISHES AND SHELLFISHES

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INTRODUCTION

Malnutrition, as it adversely affects the human being, impairs the growth, reproduction, health and well-being of the finfishes and shellfishes also. In the farming of these animals in an environment unlike their natural habitat, feeding of the stocked population with nutritionally balanced and quality diets is of critical importance not only to promote their optimal biological and physiological processes, but also to the production. In the different aquaculture systems except that practised on farming the animals feeding on the natural food available in the field, formulated feeds are provided either as supplementary feed or as whole feed. For the preparation of the formulated feed having the optimum dietary nutrient levels, the essential prerequisite is to have an understanding of the nutrient requirements of the species selected for culture.

NUTRITION REQUIREMENTS OF FINFISH

Over the past three decades considerable progress has been made in the study of the dietary nutrient requirements of a number of fishes (Halver, 1972; Cowey and Sargent, 1972, 1979; National Research Council, 1981, 1983; Millikin, 1982; Cowey and Tacon, 1983; Cho, Cowey and Watanabe, 1985).

Although fishes exhibit certain similarities with the terrestrial vertebrates in respect of basic qualitative nutrient requirements, marked difference has been noted from them in the quantitative nutrient needs at the dietary level. This difference is attributed to the carnivorous/omnivorous feeding habit of fishes, and their preference to use protein over carbohydrates as a dietary energy source. Further, as the fishes live in an ecosystem which supports them and are capable of adjusting to the temperature of the environment; they do not have to expend large amount of energy to maintain the constant body temperature and to develop an elaborate skeletal system as in the case of land-based animals. It is also observed that the fishes expend relatively low energy for reproduction. For these reasons, they are considered to be better feed converters than the other vertebrate groups. Besides, the fish have the advantage of disposing ammonia, the primary end product of nitrogen metabolism, through permeable surface unlike the land-based animals that require conversion of ammonia to urea or uric acid to dispose of the toxic ammonia building up in the tissues. This metabolic characteristic helps the fish to derive relatively more energy for the catabolism of protein than the terrestrial animals.

Fish require 40 or more essential nutrients among these the most important ones relate to protein and amino acids, lipids, essential fatty acids, vitamins and minerals.

Protein

Over 20 species of fishes have so far been studied for the dietary protein requirements principally on the basis of feeding experiments on a balanced diet containing gradual levels of quality protein and the recorded optimum growth (weight gain) of the fish. The results of these experiments have shown a high dietary requirement ranging

from 35 to 55% which is equivalent to 45-70% of the gross energy content of the diet in the form of protein. Although such high protein requirement is expected for carnivorous fishes, it is also observed in omnivorous and herbivorous fishes. The use of different dietary protein sources, non-protein energy substitutes, feeding regimes, fish age and methods employed for the determination of dietary energy content and dietary requirement are observed to result partly in the estimation of such high protein requirement. The dietary protein need is also found to be dependent on the size of the fish and environmental factors such as temperature and salinity. Small sized fishes require higher levels of protein for growth than the larger fish. Similarly, increase in dietary protein is recorded in higher environmental temperature. Recent studies and comparisons of results observed in the different feeding experiments to determine the protein requirement have shown that (1) a linear relationship exists between dietary protein requirement (g protein/kg body wt/day) and the specific growth rate, (2) the utilisation of dietary protein for new tissue growth is relatively constant within and between the individual finfishes examined and (3) the dietary protein requirements of fish when expressed relative to feed intake (g protein/kg body weight/day) and live weight gain (g protein/kg live weight gain) are not dissimilar from those of terrestrial animals. It is now recognised that the general protein requirements of fish is the requirement of essential amino acids together with some requirement of non-specific nitrogen.

Amino acids

The fish require ten essential amino acids, namely threonine, valine, methionine, isoleucine, leucine, phenylalanine, lysine, histidine, arginine and tryptophan

in the diet. Generally, quantitative amino acid requirements are determined using dose-response curves. In recent years, the methods based on plasma or serum concentration of free amino acids and carcass deposition have also been employed.

The studies carried out on the amino acid requirements have shown that significant difference in requirement exists within and between individual fish species. The following factors are found to influence the determination of amino acid requirements.

1. Formulation of amino acid test diets,
2. supply of protein in the form of free amino acid and protein bound amino acids,
3. free amino acids are more rapidly assimilated in fish than protein-bound amino acids,
4. the interaction among the essential amino acids themselves and between the essential and non-essential amino acids and between amino acids and other nutrients.

Although, the different individual essential amino acid requirements of several of the fishes have been determined, the dietary requirement of all the ten essential amino acids are established only for four species of fishes, namely, common carp, Japanese eel, channel cat fish and Chinook Salmon (Table 1).

Table 1. Essential amino acid requirements (g/kg dry weight) at stated dietary protein levels of certain fishes

	Chinook Salmon	Japanese eel	Common carp	Channel catfish
Arginine	24	17	16	10.3-17.0
Histidine	7	8	8	3.7
Isoleucine	9	15	9	6.2
Leucine	16	20	13	8.4
Lysine	20	20	22	15.0
Methionine + Cysine	16	19	12	5.6
Phenylalamine + tyrosine	21	22	25	12.0
Threonine	9	15	15	5.3
Tryptophan	2	4	3	1.2
Valine	13	15	14	7.1
Protein in diet	400	377	385	240.0

Lipids

Lipids are important as an energy source in fish diets. Excess or deficiency of lipids affects the growth as well as the body composition of the fish. If the diet is deficient of non-protein energy (Lipids and Carbohydrates), protein is used for energy requirements; if it contains excess, appetite or demand is satisfied before a sufficient quantity of protein is ingested to meet the demand for maximal rate of protein synthesis and growth. Consequently, the experiments to determine the level of dietary lipids are directed to find out the levels which could afford the maximum protein sparing effect and expressed as a function of dietary protein level. Thus, in channel catfish, smaller fishes have shown best growth with diets containing 35% crude protein and 12% lipid,

whereas larger fishes with 25% crude protein with 12% lipid. For rainbow trout, maximum protein sparing is obtained at 15-21% lipid and 35% crude protein. It has also been shown that the protein level could be reduced in marine fish diet, if the energy content is maintained at a high level. Experiments on the use of unsaturated and saturated fatty acids have indicated that the lipids in saturated form could also be used in moderation without affecting the energy requirements of the fishes.

Essential Fatty acids (EFA)

The requirements of EFA of linolenic series have been demonstrated in a number of fishes for achieving better growth rates and food conversion and to avoid certain pathological conditions. However, their requirements differ from species to species as the EFA requirement is found to be far less for channel catfish and carp than those of rainbow trout. Certain fishes such as turbot, red sea bream, black sea bream and yellow tail are found to be not capable of desaturating and chain elongating 18-carbon fatty acids. Consequently, for these fishes, it is essential to supply highly unsaturated fatty acids in the diet.

Carbohydrates

Although carbohydrates form the major source of metabolizable energy in the nutrition of mammals and birds, it is considered to be of relatively little value in fish nutrition. This low efficiency of utilisation of carbohydrates by fishes may be due to insufficient enzymatic break down in the digestive tract, insufficient absorption and inefficient metabolism of monosaccharids. Even if most of the carnivorous fishes are poorly equipped to metabolize sugars and starches, the specific and careful balance of carbohydrate sources would help to spare the protein and furnish

fibre to move other nutrients down the gastrointestinal tract for proper digestion. Recent studies have shown that atleast in certain fishes such as trout, there is no fundamental problem in the utilisation of carbohydrates and sucrose and gelatinised starch may be of practical value as components of feeds.

Vitamins

Four fat-soluble (Vitamin A, D₃, E and K) and eleven water-soluble vitamins (Thiamine, Riboflavin, pyridoxine, Pantothenic acid, Niacin, Inositol, folic acid, choline, Biotin, B₁₂ and Ascorbic acid) are required by the fish. They are required for the metabolism of other nutrients into tissue components. Many of the water-soluble vitamins function either directly or in a modified form as coenzyme. However, fat-soluble vitamins do not function as coenzymes. Specific requirements of vitamins differ from species to species and are affected by the diet composition.

Minerals

Minerals are mainly required for the maintenance of salt and water tissue balance, metabolism of other nutrients and for structural functions. The minerals required by the fish are calcium, chlorine, magnesium, phosphorous, potassium and sodium along with a number of trace elements such as cobalt, copper, iodine, iron, manganese, selenium, zinc, aluminium, chromium and vanadium. Determination of mineral requirements and trace elements in the diet is found to be extremely difficult due to the problem of limiting their concentration and their waterborne characteristics. Between the marine and fresh water fishes, the former require limited supply of minerals as some of the elements are taken from the external environment. For the latter group, mineral supplement in the diet is found to be essential. As

in the case of vitamins, the specific requirements of different minerals are found to vary from species to species.

NUTRITION REQUIREMENTS OF CRUSTACEANS

Protein

The results of various investigations carried out on the nutrition and nutritional requirements of crustaceans have been reviewed by New (1976, 1980), Castell et al. (1981), Claybrook (1983) and Dall and Moriarty (1983). Dietary protein requirements of cultivable penaeid prawns have been the subject matter of several investigations. These studies have shown that although the protein requirements for penaeid prawns vary from 15 to 80%, generally it is found around 40%. For Penaeus indicus, the optimum protein level is recorded between 35 and 40%; P. mergeriensis, 34-42%; P. monodon, 34-40%; P. japonicus, 52-57%. As in the case of fishes, factors such as protein source used in the diet, environmental factors, effect of other nutrient levels, size and age, amino acid profile of the protein source and that of the animal influence the specific requirement.

Amino acids

Amino acids in crustaceans occur both in free form and bound form as in all organisms. However, the free amino acids in most crustaceans are found to be relatively at higher levels than in vertebrate tissues. Prawns are found to require the same ten amino acids as fish. For several crustaceans such as Cancer, Homarus, Palaemon, Penaeus, Macrobrachium and Uca, these amino acids are found to be essential in the diet. In certain crustaceans, the gut symbionts are known to be capable of synthesising certain amino acids. Among the other amino acids, proline appears to enhance the growth.

Lipids

Studies on the quantitative requirements of lipids in the diet of prawns have shown that lipid levels less than 10% have given higher growth increment than that of the lipid levels at 10, 13 and 17%. Generally a lipid level between 5 and 7% in diet is suggested for prawns. The lipid levels show marked difference during the moult cycle of prawns, being low in the post-moult and premoult stages. The de novo synthesis of fatty acids of the linoleic, and linolenic series is found to be extremely limited or non-existent in crustaceans. However, these fatty acids made available in the dietary sources could be chain elongated and further desaturated. Marine crustaceans have shown to have higher levels of linolenic series of fatty acids and higher amounts of C 20 and C 22 polyunsaturated fatty acids than fresh water crustaceans, that have higher levels of linolenic type fatty acids. This indicates that EFA of linolenic series have greater value to marine crustaceans, while the fresh water crustaceans require more linoleic series or a mixture of both. A dietary requirement of 1-2% linolenic acid is indicated in the diet for prawns. Recent studies on the phospholipids requirement of larval P. japonicus have shown that phospholipids containing choline or inositol and linoleic, linolenic, eicosa pentaenoic acid and docosa hexaenoic molecules promote growth and survival.

Crustaceans also found to be incapable of synthesising sterols. Cholesterol forms the major sterol in crustaceans and it is synthesised from ergosterol, stigmasterol and -sitosterol but not from non-sterol precursors as in Astacus, Penaeus, Portunus and Panulirus. Growth is found to be better when cholesterol at 0.5% level is added as a dietary component. Cholesterol supplemented with ecdysone and cyasterone in the diet is found to increase the moulting frequency.

Carbohydrates

Carbohydrates although considered not as an essential component of the diet, penaeid prawns are found to utilise disaccharides better than the monosaccharides. In penaeid prawns including P. indicus, increased growth is recorded in the carbohydrate level upto 40%, where starch is used as the nutrient source. The inclusion of carbohydrates in the diet helps to spare more portion for growth than for energy requirement. Cellulose used as roughage and non-nutrient filler in the diet helps better utilisation of other nutrients. The amino sugar glucosamine at 0.5% level in the diet is shown to have a growth promoting effect.

Vitamins

The observations on the dietary requirements of various vitamins for crustaceans by different workers are inconsistent. Gut symbionts and bacterial contamination, it is opined, may be involved in the supply of some or all the vitamins. While the vitamins of all prosthetic groups of enzymes are found to be required by Artemia, Moina is shown to require thiamine, nicotinamide, pyridoxine, pantothenic acid, riboflavin and folic acid.

Ascorbic acid, inositol and choline are found to enhance growth and survival in Penaeus. Crustacean appears to be incapable of synthesising carotenes. Panulirus cygnus fed with a low carotene diet became pallid, indicating the requirement of vitamin A.

Minerals

Information on the mineral requirement of crustaceans is scanty. Phosphorus, potassium and trace metals are found to be required in the dietary composition of Penaeus. However, calcium, magnesium and iron are observed to be not essential in the diet. Although calcium is an obvious

requirement for crustacean for exoskeleton build up and needs to be conserved in calcium-deficient environment, utilisation of the calcium from the gastrolithes and from eating of excuviae help to meet their requirement. Further, marine crustaceans absorb directly calcium from the sea water. Since the magnesium is excreted by most crustaceans and is available to them in the sea water, and since the iron is stored in the mid gut, these minerals may not be a dietary requirement. Copper which is required for haemocyanin synthesis, is derived mainly from the food and the sea water.

Larval nutrition

There has been considerable progress in the studies on larval nutrition, particularly on the penaeid prawns and Macrobrachium rosenbergii that are cultured in several regions of the world. The recent progress made in the development of microencapsulated feed on commercial lines is a noteworthy development in this field. The various aspects of penaeid and palaemonid larval nutrition are reviewed by Rao (1983).

Although there have been several pioneering investigations on crustacean nutrition, comparison of results of studies from different laboratories have rendered difficulties in the evaluation of different diets. In order to permit direct comparison of results among different laboratories, a crab protein diet is formulated as a standard reference diet. The composition of this standard reference diet is as follows:

Crab Protein Reference Diet (%)

Crab protein	..	40
Wheat gluten	..	5
Corn starch	..	15
Dextrin	..	5
Alpha cellulose	..	17.8
Mineral mix	..	4
Vitamin mix	..	2
Dl - α -tocopherol	..	0.2
Codliver oil	..	6
Corn oil	..	3
Choline chloride	..	1
Cholesterol	..	1

Total		100
		=====
Crude protein	..	38.1% (dry wt)
Lipid	..	10.5%
Ash	..	6.5%
Gross energy (cal/g)	..	5.0

NUTRITION REQUIREMENTS OF MOLLUSCS

The larval molluscs as well as the majority of the adult bivalves procure their food by filtering plankton or suspended particles from sea water. Adult gastropods graze on algae and detritus, while the cephalopods are active predators feeding on moving live animals such as crustaceans, fishes and other molluscs. In the stomach content of oysters items such as plankton, organic waste, fungi, flagellates, larvae of various invertebrates, sand and mud, have been found. On the basis of the physiology of digestion, it is observed that the animal food material in oysters contribute only 10% or less of the total food.

As the farmed molluscs in the grow-out system derive their food from nature, nutritional aspects for the culture of molluscs assumes importance principally in the rearing of larvae and spat under controlled conditions.

It is now established that a mixed diet containing more than two types of food forms a more balanced diet for shellfishes. Experiments have shown that the shellfish larvae fed with single cell alga raised with vitamins such as B₁₂, thiamine and biotin have given better growth rates.

There have been several attempts to develop artificial diets for marine bivalves. However, these efforts have so far met with only a partial success, as clumping of food particles, leaching losses and development of bacterial population pose difficulties. Further, the food particles must also be digestible and small (2-15 μ m). The artificial diet prepared with microgel particles, microcapsules, Kaolin and trace-metal mixture has given good growth in the juveniles of Crassostrea virginica when experimented in a beaker. But the growth rate is found to be poor when the larvae are reared in a flow through system. The bacterial population developing in the flow through system appears to effect the nutrient source of the diet or aid in the breakdown and digestion of the food particles. The artificial diet had protein, soluble starch, rice starch each of 25.7% by weight, soy lecithin 14.4%, DNA and RNA, each 2.6%, phosphorus 1.3%, carboxymethyl cellulose 1.1%, trace-metal mix 1.0%, Menhaden-walled vitamin capsules, (25 capsules/ μ l, 0.82 mg lipid/ltr), lipid, 97.9%, vitamin mix 1.0%, Ascorbic acid, 1.0% and phenol red 0.1%. Menhaden-walled B₁₂ capsules (5 capsules/ μ l, 0.16 mg lipid/ltr. Egg albumen is found to be the best source of protein.

- Halver, J.E. 1972. Fish nutrition. Acad. Press. New York and London. 713 pp.
- Millikin, M.R. 1982. Qualitative and quantitative nutrient requirements of fishes. A review. Fish. Bull., 80: 655-686.
- National Research Council. 1981. Nutrient requirements of coldwater fishes. Nat. Acad. Press, Washington, D.C.
- National Research Council. 1983. Nutrient requirements of warmwater fishes and shellfishes. Ibid.
- New, M.B. 1976. A review of dietary studies with shrimps and prawns. Aquaculture, 9: 101-144.
- New, M.B. 1980. A bibliography of shrimp and prawn nutrition. Ibid., 21: 101-128.
- Rao, P. Vedavyasa 1983. A review of the studies on larval nutrition in cultivable penaeid and palaemonid prawns. Proc. Nat. Symposium on Shrimp seed Production and Hatchery management; Cochin, 21-22, January, 1983. The Marine Products Export Development Authority, 69-96.