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RECENT ADVANCES IN FINFISH AND SHELLFISH NUTRITION

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MINERAL REQUIREMENTS OF FINFISH AND SHELLFISH

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INTRODUCTION

In animal nutrition, while protein, lipid and carbohydrate are required in major quantities, vitamins and minerals are required in small quantities in the diet. Though minor in nature, the mineral nutrition is no less important. Minerals are very essential because animals are not capable of synthesising them and should be supplied through external source. Many of the minerals are vital for healthy growth of the animals and their prolonged non-availability, either through diet or through environment, may cause irrecoverable deficiency disease. On one hand mineral elements are basic units of skeletal structures of the animals on the other hand mineral ions are important co-factors of enzymes and other biological chemicals involved in life process. Minerals are of paramount importance for aquatic animals like fish and shellfish since these animals need to keep their osmotic balance through mineral ions. Mineral elements required by fish may be classified as bulk elements such as calcium, phosphorous, potassium, chlorine, sodium and magnesium (these are required comparitively in large amounts) and trace elements which are copper, cobalt, iron, iodine, manganese, selenium, zinc, aluminum, chromium and vanadium (these are required in very small quantities).

MINERAL REQUIREMENT STUDIES

Experiments to study mineral requirements are rather difficult to conduct, mainly because it is not easy to prepare diets with limiting concentrations of each mineral. Secondly feeding experiments with the animals should be of very long durations, in order to observe the manifestations of mineral requirements, deficiency and excesses. It is much more complicated in the case of aquatic animals since water is the largest contributor of a variety of mineral elements. Further there is possibility of interactions between minerals which may create complications in assessing the dietary requirements. The interactions may be either antagonism or synergism. For example large amounts of calcium in the diet may lower the availability of zinc present in the diet. Calcium deposition occurs in fish which is deficient in magnesium. Minerals have a role in many facets of metabolism such as hormones, respiratory pigments, structural elements, high-energy bonds and enzyme co-factors. Thus studies on mineral requirements, assume greater importance in fish and shellfish nutrition.

MINERAL REQUIREMENTS OF FINFISH

Among the aquatic animals, the mineral nutrition of finfish has been studied in greater detail. Fresh water fish show higher requirement of most of the minerals compared to the marine fish. The later group are capable of absorbing some of the mineral elements present in salt water. After detailed experiments, it was shown that red sea bream need only iron, potassium and phosphorous in their diet, the remainder of the elements coming from the external environment. Many of the mineral requirement studies are therefore directed on the fresh water fish species. The mineral elements calcium, phosphorous, sodium molybdenum, chlorine, magnesium, iron, selemium, iodine, manganese, copper, cobalt and zinc are recognised as essential for body functions in fish. Fluorine and chromium have also been added of late to the list of essential elements.

Calcium and phosphorous

Calcium and phosphorous are closely related in metabolism and are discussed together in fish nutrition. Major portion of calcium (99%) and phosphorous (80%) are found in bones, teeth and scales. The ratio of calcium and phosphorous in bone ash is found to be approximately 2:1.

The extra skeletal calcium is widely distributed throughout the organs and tissues and exists in diffusable and non-diffusable form. Non-diffusable form is bound to proteins and the diffusable form which is generally found as phosphate, plays a significant role in the nutrition of animal. Ionised calcium in the extracellular fluids and in the circulatory system participate in muscular activity and osmoregulation.

Extra skeletal phosphorous is present mostly in combination with proteins, lipids, sugars, nucleic acids and other organic compounds. In some species the skin also appears to be an important repository for dietary phosphorous.

As stated earlier, fish are capable of extracting calcium directly from the surrounding water through gills. On the other hand the absorption of phosphorous is negligible from the environment, and the fish mainly depend upon the dietary sources of phosphorous.

Calcium and Phosphorous are absorbed in fish in the upper gastro-intestinal tract. While calcium is rapidly deposited as calcium salts in the skeleton, phosphorous is distributed to all the major tissues. Water temperature influences the absorption of phosphorous and increases with increase in temperature. Higher content of glucose in the diet was found increase the absorption of dietary phosphorous by the fish. Absorption and retention of calcium are not influenced by any such external factors. The level of phosphorous in the diet influences the calcium retention in the body. Higher levels of dietary phosphorous enhances the retention of calcium to maintain the ratio between the two elements in the body.

The dietary phosphorous requirement of fish are as follows:-

Fish	Requirement
Atlantic salmon	0.7%
Channel catfish	0.4 - 0.47%
Common carp	0.6 - 0.7%
Red sea bream	0.68%
Rainbow trout	0.7 - 0.8%

Thus in general the phosphorous requirement in the diet of fresh water and marine finfish is almost same. Dietary calcium levels for fish are not recorded due to the absorption of this element from the environment by most of the fish species.

Many ingredients used for feed making are rich in calcium and phosphorous. Fish meal contains rich amounts of both the minerals. Calcium is generally deficient in plant ingredients and the phosphorous present in them is not available to the fish especially when the plant materials contain phytin or phytic acid. Phosphorous is readily available to the animals if potassium or sodium hydrogen phosphate or mono calcium phosphate is used in the diet. On the other hand phosphorous from tricalcium phosphate is not readily available. Fish meal is rich in tri-calcium phosphate. Due to its poor availability to fish, it is released into to the water through faeces. This can combine with ammonia in water and lead to extensive eutrophication in the ponds. Deficiency symptoms of calcium are not described in fish. Poor growth, reduced feed efficiency, low bone ash and low haematocrit levels were observed in channel catfish fed phosphorous deficient diet. Prolonged feeding of phosphorous deficient diet has resulted in lordosis and abnormal calcification of bones (brittle structure) in common carp. The symptoms were similar in red sea bream.

Magnesium

About 60% of the body magnesium is found in skeletal structure and the remaining is distributed throughout the organic and muscle tissues. It is an important enzyme co-factor and component of cell membranes.

Marine fish are capable of extracting magnesium from the environment. Since this element is very limited in fresh water, the fresh water species seem to depend upon dietary source of magnesium. The dietary requirement of magnesium for rainbow trout is 0.06-0.07% and for carp it is 0.04-0.05%. Most of the compounded feeds prepared with ingredients of animal or plant origin have adequate levels of magnesium.

Apart from the general symptoms (reduced growth and poor food conversion efficiency) magnesium deficiency in rainbow trout leads to renial calcinosis and a flaccidity of the muscle due to increase in the extra cellular fluid volume. Loss of appetite, sluggishness and convulsion followed by tetany were also observed in common carp and rainbow trout fed magnesium deficient diets.

Zinc

Zinc is an important co-factor of many enzymes like carboxy peptidase, dismutase and superoxide in the animals. Many metabolic functions are effected by its deficiency. In rain bow trout, zinc requirements are normally met by dietary levels of 15-30 mg/kg. Large amounts of calcium present in the diet appears to lower the availability of zinc to the animal due to antagonism. Calcium and zinc perhaps compete for the same binding sites of protein or have same metabolic pathway and absorption mechanism. Zinc sulphate ($ZnSO_4$) seems to be the best source of zinc in the diet.

Deficiency of zinc was found to cause 'dwarfism' and cataract of the eye in rain bow trout. However several hundred mg of Zn per kg of diet do not appear to be injurious to rainbow trout.

Copper

Different dietary levels of copper have no influence on the growth of rainbow trout. Copper when included in the diet of common carp fingerlings at 0.7 mg/kg diet, the growth was low. The growth was better with the diet containing 3.0 mg copper per kg diet. Dietary copper level for channel catfish does not appear to be more than 1.5 mg/kg dry diet. Copper concentrations of 20-30 mg/kg diet, very much reduced the growth of catfish.

Iron

Iron deficiency in the diet of red sea bream resulted in a form of microcytic, hypochromic anaemia similar to iron deficiency in land animals. Common carp fed a semi purified diet with iron grew normally but exhibited sub-clinical symptoms of hypochromic and microcytic anaemia. Iron concentration of 150 mg/kg diet was found to be required at minimum level, to prevent anaemia in red sea bream and common carp.

Manganese

Manganese deficiency in rainbow trout gives rise to abnormal curvature of the back bone and mal formation of the tail. Manganese content of 12-13 mg/kg diet, produced higher growth rates in rainbow trout and carp. Manganese sulphate ($MnSO_4$) and Manganese chloride ($MnCl_2$) are found to be good sources of manganese in fish diets.

Selenium

Selenium is a component of metallo-enzymes glutathione peroxidase. It plays an important role in the antioxidant defence mechanisms of the fish. It functions synergestically with Vitamin E. Deficiency of selenium leads to rapid onset of muscular dystrophy and exerdative diathesis. Maximum glutathione peroxidase activity was observed at dietary selenium levels of 0.15 and 0.38 mg/kg diet. At 13 mg/kg diet it was found to be toxic, causing uncoordinated spiral swimming behaviour leading to mortality in fish.

Iodine

Iodine deficiency produces goitrous condition in trout. A dietary requirement of 1.1 mg iodide/kg is recommended in the diet of chinook salmon.

The role of other trace elements in fish are not clarified. The requirements may be similar to those described for land animals.

A summary of the mineral requirements of different fish and their deficiency symptoms are given in Annexure - I.

Mineral mixtures used in the standard fish diet are given below.

I. Mineral Mixture (USP XII No. 2) (Halver 440)

Calcium phosphate (g)	13.58
Calcium lactate	32.70
Ferric citrate	2.97
Magnesium sulphate	13.20
Potassium phosphate (dibasic)	23.98
Sodium biphosphate	8.72
Sodium chloride	4.35
Aluminum chloride	0.015
Zinc sulphate	0.300
Copper chloride	0.010
Manganese sulphate	0.080
Potassium iodide	0.015
Cobalt chloride	0.100

Ogino salt Mixture for Fish

NaCl (g)	1.0
$MgSO_4$ "	15.0
NaH_2PO_4	25.0
KH_2PO_4	32.0
$Ca(H_2PO_4)_2$	20.0
Ferric citrate	2.5
Calcium lactate	3.5
Trace elements*	3.5
Trace elements	1.0
	100.0

* composition of trace elements

$ZnSO_4$	$7H_2O$ (g)	35.3
$MnSO_4$	$4H_2O$ (g)	16.2
$CuSO_4$	"	3.1
$CoCl_2$	"	0.1
KIO_3	"	0.3
Cellulose	"	45.0
		100.0

MINERAL REQUIREMENTS OF SHELLFISH

Mineral requirements of cultivable species of prawns are studied to some extent among the shellfish. Penaeid prawns were found to require calcium and phosphorous in their diets. Best growth was obtained when the diets of the prawn Penaeus japonicus diets were supplemented with 1.04% of phosphorous and 1.24% of calcium. It was indicated that the calcium and phosphorous ratio in the diet of prawn should be 1.2:1. When this ratio was increased to 2:1 growth was inhibited and pigmentation decreased. Through studies with radio activity labelled calcium (45 Ca), it was demonstrated that prawns could absorb calcium from the surrounding water. It was estimated in P. japonicus that approximately 0.83 mg of calcium is absorbed per day per gram of body weight from the environment, when the sea water contained 0.44 mg/ml of calcium. This had lead to the conclusion that the calcium requirement could be satisfied by the calcium in surrounding seawater.

Supplementation of the diet with calcium (2%), magnesium (0.3%) and iron (0.02%) did not improve the nutritive value of the diet P. japonicus. Iron in the diet rather reduced the growth. Phosphorous at 2% level in the diet effectively improved the performance of the diet.

Potassium at 1% level in the diet showed higher growth and feed efficiency.

The requirement of individual trace elements was not studied. However when a mixture of trace elements, consisting of aluminum chloride (5 mg), Zinc sulphate (90 mg), manganese sulphate (20 mg), copper chloride (5 mg), potassium iodide (5 mg) and cobalt chloride was added to the diet at 0.2% level, enhanced the efficiency of the feed; higher level of this mixture above 0.2% in diet lowered the growth.

Studies in detail, on the requirement of each individual mineral element are needed to understand thoroughly the deficiency and excess symptoms of different minerals which are not available at present. However nutritionists have been adding mineral mixtures to their diet formulations. The mineral mixture used in the diet of *P. japonicus* is given below.

Mineral mixture used in prawn diet per 100 g.

	(g)
K2HPO4	2.0
Ca3 (PO@)2	2.72
NaH2 PO4 2.H2O	0.79
Mg SO4	3.02
FeSO4. 7H2O	0.015
MnSO4. 5H2O	0.004
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REFERENCES

Deshimaru, O and Y. Yone 1978. Requirement of prawn for dietary minerals. Bull. Jap. Soc. Sci. Fish. 44(8): 907-910.

Deshimaru, O., K. Kuroki, S. Kakamoto and Y. Yone 1978. Absorption of labelled calcium - 45 Ca by Prawn from sea water. Bull. Jap. Soc. Sci. Fish., 44(9): 975-977.

Cho, C.Y., C.B. Cowey and T. Watanabe 1985. Finfish Nutrition in Asia: Methodological Approaches to Research and Development. Ottawa, Ont., IDRC, 154 p.

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Halver, J.E. (Ed:), Fish Nutrition. New York, Academic Press, 719 p.

New, M.B. 1976. A review of dietary studies with shrimp and prawns. Aquaculture, 9(2): 101-144.

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