Winter School on

'RECENT ADVANCES IN DIAGNOSIS AND MANAGEMENT OF DISEASES IN MARICULTURE'

7th to 27th November, 2002

Course Manual

Organising Committee

Prof. (Dr) Mohan Joseph Modayil
Director, CMFRI

Course Director

Dr. K. C. George
Principal Scientist,
Division of
Physiology, Nutrition and Pathology

Co-ordinators

Dr. R. Paul Raj, Head, P N P Division

Dr. P. C. Thomas, Principal Scientist

Shri. N.K. Sanil, Scientist (Sr. Scale)

Dr. (Mrs.) K.S. Sobhana, Scientist (Sr. Scale)

Indian Council of Agricultural Research
Central Marine Fisheries Research Institute
P B. No. 1603, Tatapuram P.O., Cochin 682 014

NUTRITIONAL DEFICIENCY DISEASES IN FISH

Dr.Imelda Joseph and Dr. R. Paul Raj
Central Marine Fisheries Research Institute, Kochi – 682 014

Introduction

During the past two decades, there has been tremendous development in aquaculture. As aquafarming continues to expand worldwide, production methods have shifted from traditional extensive to modified extensive, semi-intensive and intensive systems utilizing modern facilities, equipments and management practices aimed at producing higher yields per unit area. Natural food constitutes an important source of nutrients for extensive and improved extensive culture systems, whereas artificial feeds are required for semi-intensive and intensive practices. In fish and crustacean aquaculture the feed conversion ratio (FCR) falls in between 0.5 and 2.0 and hence feed represents the major expense, often accounting for more than 50% of total operating cost.

Feed-derived wastes also impact the culture environment through direct pollution, which in turn affects the culture organisms. Uneaten feeds, faeces and metabolic wastes contribute to nutrient and particulate loading of the water and substrate. These factors induce stress, depress the growth of cultured organisms and increase their vulnerability to diseases. A balanced diet and a healthy environment are vital for sustaining the health of the cultured organism. A well balanced diet results in higher production as well as provides nutrients necessary to hasten recovery from deficiency diseases. Diets can negatively influence the well being of a fish by inducing nutrient deficiencies, imbalances or toxicoses or by inducing infective agents into the fish. At any stage of development of fish, an impaired nutritional status contributes to defective host resistance. Malnourished fish may harbor latest infections, and certain physiological conditions and environmental stress may predispose them to infection.

Nutritional diseases are directly dependent on stocking density in a culture system. Nutritional problems in fish population may appear in the form of reduced fecundity, slow growth, decreased appetite, increased susceptibility to infectious diseases, morbidity with clinical signs and pathological lesions and mortality.

Nutrient Requirements

Protein

The dietary requirements of protein for cultivable fish and crustaceans range from 25 to 60%. This depends on many factors such as species, size, age, protein source, level of non-protein energy, availability of natural food and culture management. Animals, including fish must consume protein for maintenance, growth and reproduction. Fish and crustaceans require the same 10 essential amino acids, viz., arginine, histidine, isoleucine, leucine, lysine, methionine, phenylalanine, threonine, tryptophan, and valine as do terrestrial animals. Imbalance of essential amino acids and inadequate protein in the diet will result in retardation or cessation of growth, or a loss of weight due to the withdrawal of protein from less vital tissues in order to maintain the functions of more vital ones.

Lipids

Lipids are required in the diet not only as a source of energy but also as sources of essential fatty acids, sterols, phospholipids, and as carriers of fat-soluble vitamins and carotenoid pigments. The optimum dietary lipid level for shrimps and majority of fish species ranges from 6 to 10%. However carnivorous fish require higher levels of lipids (10 to 20%). Fast moving and migrating fish species require higher lipid levels (15-25%) in the diets. Fatty acids of the linoleic and linolenic series are dietary essentials for fish and crustaceans. Linolenic acid (18:3 n-3) is nutritionally superior to linoleic acid (18:2 n-6), while, eicosapentanoic acid (20:5 n-3) and docosahexanoic acid (22:6 n-3) are more effective than linolenic acid in the diets of marine species. As a source of energy, lipids have been shown to spare some protein for growth. Limited ability of crustaceans to synthesize sterols makes them susceptible to cholesterol deficiency. Cholesterol, the precursor of moulting and sex hormones in crustaceans has to be supplied through diet (0.25 to 0.4%). Phospholipids, important for biomembranes and for transport of dietary lipids in hemolymph, has also to be supplied through diet.

Carbohydrates

Carbohydrates serve as energy source, help in the synthesis of non-essential fatty acids and aid in pellet stability and binding. The utilization of carbohydrate is unclear in many warm water fishes. Disaccharides (sucrose) and polysaccharides (glycogen, dextrin and starch) are better utilized by shrimp than are monosaccharides. Shrimps tolerate 25-35% of carbohydrate in their diet. Carbohydrate levels in fish range from 20% to 45% depending on their feeding habit.

Vitamins

Vitamins are essential in fish diets for various metabolic functions. Eleven water-soluble and four fat-soluble vitamins are essential for most of the cultured aquatic organisms.

Minerals

Fish absorb minerals from water as well as from feed. Calcium and Phosphorous are required in greater levels in diets. Fish can obtain adequate calcium from water unless the water is very soft. Phosphorous is not absorbed from water, so has to be supplemented through diet. Dietary minerals essential for shrimp are calcium, phosphorous, magnesium, potassium, copper, selenium and zinc.

Deficiency diseases

Deficiencies in essential nutrients in the diet result in deficiency symptoms, which are non-specific and proved only by prolonged feeding experiments and histopathological observations. Malnourished fish harbor pathogens, which on favourable environment conditions or physiological conditions predispose them to infection. Nutritional problems can lead to reduced fecundity, poor hatchability rates, poor larval survival, decreased appetite and increased susceptibility to infectious diseases.. Most common nutritional deficiency diseases are as follows:

Starvation

Starvation refers to absolute nutrient deprivation resulting from inadequate intake or assimilation of feed. The energy needs of the organism are below maintenance levels. Typically, starved fish will have a large head and slender body and will be dark in colouration.

Protein and amino acids

In warm water fishes, specific protein and amino acid deficiency diseases are generally not recognized as a problem in pond culture systems. Feeding on experimental diets deficient in selected essential amino acids cause appetite depression and poor growth regardless of the type of amino acid in fishes. Growth suppression, skeletal deformities and exophthalmia in fish fed with maize meal deficient in tryptophan are evidenced in earlier studies. A tryptophan deficiency related scoliosis has been reported for salmonids due to improper feed formulation, hyper extended storage or excess heat in processing. Dietary methionine deficiency in salmonids leads to reduced growth rate with the development of bilateral cataracts. Fatty liver caused by feeding with poor quality protein diet with an imbalance in the amino acid profile, is also reported. With shortage of certain amino acids, the body must burn off excessive unusable amino acids as energy, and results in fat production. A feed with proper amino acid profile is the only treatment.

Lipids

In case of deficiency of non-protein energy (lipids and carbohydrate) in diets, dietary protein will be used up for energy purposes like basal metabolism and voluntary activity, rather than protein synthesis resulting in poor growth rate in fishes. Fatty acid deficiency in fish leads to poor food efficiency, and deficiency signs such as elevated muscle water content, susceptibility to caudal fin erosion, shock syndrome, decreased hemoglobin and blood cell volume, swollen pale fatty liver, degeneration of gill epithelium etc. Eicosapentanoic acid (EPA) and Docosahexanoic acid (DHA) are essential for larval and broodstock nutrition. Their deficiency in broodstock leads to poor egg quality, hatchability and poor survival of larvae (lately enriched larval feed being proved to improve immunity in larvae).

An essential fatty acid deficiency syndrome has been reported in channel catfish, carp and eel. Lipoid liver degeneration, an essential fatty acid deficiency disease reported in salmonids is not known to occur in warm water cultured fishes. Fish are capable of synthesizing most fatty acids but not the linolenic or linoleic acid series. Deficiencies of these fatty acids lead to depigmentation, fin erosion, cardiomyopathy, fatty infiltration of the liver and myxomatous degeneration of fat. The disease signs are marked if the food given is rotten or stored too long under intense cooling. Auto-oxidation of unsaturated fatty acids, which destroys vitamin E, leads to morbid changes, especially in the liver. Typically of this are deposits of ceroid, which is a fat like brownish yellow substance. The addition of vitamin E as an anti oxidant in diet prevents formation of ceroid.

Carbohydrate

Low energy diets deficient in carbohydrates generally produce growth retardation due to gluconeogenesis. Sekoke disease, described as spontaneous diabetes in carp fed with extremely high-starch diets has been reported in Japan. Eliminating the excess starch from the diets can prevent the disease.

Vitamins

Absence of a particular vitamin leads to serious metabolic disorders, which are referred to as avitaminoses, which are frequently fatal. Inadequacy of vitamins leads to deficiency symptoms, which are non-specific and termed as hypovitaminoses. In case of deficiency it is difficult to trace the cause to lack of a specific vitamin. Quantitative deficiency can lead to non-specific growth retardation and susceptibility to diseases,

which are difficult to diagnose accurately. Vitamin deficiency disease is unlikely to occur in pond culture systems, unless stocking densities are considerable. Broken back syndrome is a well-known disease of channel catfish cultured under super-intensive conditions. The disease arises if channel catfish are fed diets deficient in vitamin C for periods longer than eight weeks.

Water-soluble vitamins (C & B) are generally lost through feed processing, storage and leaching. Vitamins become unavailable due to interaction and chelation with other dietary ingredients. Use of stable form of vitamins (*eg*: L- Ascorbyl-2-monophosphate and L-Ascorbyl-2-polyphosphate for vitamin C), instead of pure forms reduces leaching loss. Major vitamins and their deficiency symptoms are given in Table 1.

Deficiencies due to presence of anti-vitamin factors in feed can be overcome by heat processing, (anti-thiamine factors present in fish and shell fish meal, rice polishing *etc*.), cooking (anti-pyridoxine factor in linseed), or heat treatments (anti-vitamin A, D, E & B 12 factors in soybean) etc.

Minerals

Mineral deficiencies arise due to dietary imbalances and interaction of dietary components. The deficiencies of fish are seen as skeletal deformities, anemia and reduced resistance to diseases. Excessive dietary calcium has an antagonistic effect on the availability of phosphorous. Major minerals and their deficiency symptoms in fish and shellfish are given in Table 2.

Conclusion

A well balanced diet fed at right time in right quantity leads to enhanced production, healthy environment and healthy animals. Proper feed management is the best key for sustainable aquaculture production.

Table 1 The essential Vitamins and their deficiency symptoms in fishes and crustaceans.

Vitamin	Deficiency Signs
Thiamine (B1)	Anorexia, poor apetite, muscle atrophy, convulsions, instability and loss of equilibrium, poor growth, congestion of fins and skin, fading of body colour, lethargy.
Riboflavin	Corneal vascularization, cloudy lens, hemorrhagic eyes, photophobia, incoordination, abnormal pigmentation of iris, striated constrictions of abdominal wall, dark colouration, poor apetite, anemia, poor growth, hemorrhage in skin and fins.
Pyridoxine	Nervous disorders, epilepticform fits, hyper-irritability, ataxia, anemia, loss of apetite, edema of peritoneal cavity, colourless serous fluid, rapid onset of rigor mortis, rapid breathing, flexing of opercles, irridiscent blue colouration, exophthalmia.

Pantothenic acid	Clubbed gills, necrosis, scarring and cellular atrophy of gills, gill exudate, prostration, loss of apetite, lethargy, poor growth, hemorrhage in skin, skin lesions and deformities.
Niacin	Poor growth, anorexia, lethargy and mortality.
Inositol	Distended stomach, increased gastric emptying time, skin lesions, de-pigmentation and poor growth.
Biotin	Loss of appetite, lesions in colon, altered colouration, muscle atrophy, spastic convulsions, fragmentation of erythrocytes, skin lesions, poor growth.
Folic acid	Lethargy, fragility of caudal fin, dark colouration, macrocytic anemia, poor growth.
Choline	Poor food conversion, hemorrhagic kidney,and intestine, poor growth, accumulation of neutral fat in hepatopancreas, enlarged liver.
Nicotinic acid	Loss of apetite, lesions in colon, jerky or difficult motion, weakness, edema of stomach and colon, muscle spasms while resting, sensitivity to sunlight, poor growth, hemorrhage in skin, tetany, lethargy, anemia.
Vitamin B_{12}	Poor apetite, low hemoglobin, fragmentation of erythrocytes, macrocytic anemia, reduced growth.
Vitamin C	Scoliosis, lordosis, impaired formation of collagen, abnormal cartilage, eye lesions, hemorrhagic skin, liver, kidney, intestine and muscle, reduced growth (Salmonids), dark colouration, loss of equilibrium, caudal fin erosion, hemorrhagic gills, short operculum, exophthalmia, fragile gill filaments (*Lates calcarifer*), Broken back syndrome (The symptoms are deformed spinal column, opercula and gill filaments, caudal fin erosion, anemia, reduced growth rate, increased susceptibility to bacterial pathogens and reduced tissue levels of vitamin C), internal and external hemorrhage, fin erosion, anorexia, erratic swimming behaviour(Channel catfsh), lower jaw erosion (eel), poor growth, high mortality, severe hemorrhages, fin necrosis, increased pigmentation and spinal flexures (*Cirrhinus mrigala*). Black death syndrome (blackening of exoskeleton, melanized

	hemoltic lesions), reduced wound repair, feed efficiency, growth and survival(Penaeid shrimps).
Vitamin A	Ascites, ceroid in liver, spleen and kidney, epicarditis, exophthalmia, microcytic anemia, pericardial anemia, fragility of red blood cells, poor growth(Salmonids), exophthalmia, oedema and kidney haemorrhages (Carps), depigmentation and soft exoskeleton (shrimps).
Vitamin D	Disturbance in calcium and phosphorous metabolism. Poor feed utilization,growth reduction and raised blood counts, slow growth rate, tetany of hite muscles and elevated T3 levels (Rainbow trout), decreased levels of ash, calcium and phosphorous (Channel catfish), poor growth, soft exoskeleton, lethargy *etc.* are reported in shrimps.
Vitamin E	Muscular dystrophy, pathological condition in male and female reproductive organs. Increased permeability of capillaries resulting in hemorrhages and oedema in various parts of the body.
Vitamin K	Anemia, prolonged coagulation time.

Table 2 **Essential minerals their metabolic functions and their deficiency signs in fishes and crustaceans.**

Mineral	Functions	Deficiency symptoms
Calcium & Phosphorous	Bone and cartilage formation, blood clotting; bone formation, high energy phosphate esters Other organophosphorous compounds	decreased growth rate and increased mortalities(channel catfish) slowed growth, scoliosis, lordosis and skull and opercular deformities.(Carps) P requirement for carp in diet is 0.8% and channel cat fish 0.45%
Magnesium	Enzyme cofactor extensively involved in the Loss of appetite, poor Metabolism of fats, carbohydrates	Lordosis, poor growth and protein growth, tetany.

Sodium	Primary monovalent cation of intracellular fluid,involved in nerve action and osmo-regulation	Not defined
Sulphur	Integral part of sulphur amino acids and collagen, Involved in detoxification of aromatic compounds	Not defined
Chlorine	Primary monovalent anion in cellular fluids,Component of digestive juice (HCl), acid-base balance	Not defined
Iron	Essential constituent of haeme in hemoglobin,Cytochromes, peroxidases *etc.*	Microcytic Homochronic anemia
Copper	Component of heme in cephalopods, crustacea Cofactor in tyrosinate and ascorbic acid oxidase	anaemia
Manganese	Cofactor for arginase and certain other metabolicEnzymes, involved inbone formation and erythrocyte regeneration	sluggish movement, loss of equilibrium, poor apetite, weight loss and mortality
Cobalt	Metal component of cynocobalamin. prevents Anemia	Not defined
Iodine	Constituent of thyroxine, regulates oxygen use	Thyroid,Hyperplasia(goitre)
Chromium	Involved in collagen formation and regulation of the rate of glucose metabolism	Not defined
Selenium	Component of metallozyme glutathione peroxidase Which contributes to the antioxidant defence mechanism in fish	Not defined

References

Comey, C.B., D. Knox, J.W. Adron, S.George and B. Pirie, 1977. The Production of Renal Calcinosis by Magnesium deficiency in Rainbow trout (*Salmogairdneri). British Journal of Nutrition*., 38: 127- 135.

Dunbar, C.E., and R.L. Herman, 1971. Viral granulona in brook trout (*Salvelines Fontinalis*) *Journal of Nutrition*, 101: 1445- 1452.

Halver. J.E, 1972. Nutritional fish diseases, In: 'Fish nutrition' (Halver J.E. Eds) 404-434p.

Harrison J.G and R.H. Richards, 1979. The pathology and histopathology of nephrocalcinosis in rainbow trout (*Salmogairdneri* Richardson) in freshwater. *Journal of Fish Diseases* , 2: 1- 12.

Masumoto.T, H.Hosokainta and S.Shirneno, 1991. Ascorbic acid role in aquaculture nutrition. In: *Proceedings of the Aquculture Feed Processing Nutriton W*orkshop. Thailand and Indonesia September 19-25, 1991.Edited by D.M.Akiyama and R.K.H.Tan.

Paul Raj. R., 1997. Aquaculture Feed. Handbook on Aquafarming. The Marine Products Export Development Authority (Ministry of Commerce ,Government of India),103p.

Richardson, N.L., D.A.Higgs, R.M. Beames and J.R. Mc Bride, 1985. Influence of ditric calcium, phosphorous, zinc and sodium phytase level on cateract incidence, growth and histopathology in chinnok salmon. *Journal of Nutrition*, 115: 553-567.