# Deficiency of protein, fat and vitamins in freshwater catfish, *Clarias batrachus*: morphological symptoms and impact on growth performance

# Priyanka Agarwal<sup>1</sup>, Farheen Jameel<sup>1</sup>, Daisy Rani<sup>2</sup> and M. Serajuddin<sup>1\*</sup>

<sup>1</sup>Fish Biology Research Lab, Department of Zoology, University of Lucknow, Lucknow-226007, U.P., India <sup>2</sup>Department of Zoology, Feroze Gandhi College, Raebareli-229001, U.P., India

\*Corresponding author: lu.fisheries@gmail.com

### Abstract

This paper briefly describes some major nutritional pathologies which have been observed in freshwater catfish, *Clarias batrachus*. These are caused by nutrient deficiencies or dietary imbalances and require attention in aquaculture. Lipids, proteins and vitamins are beneficial nutrients needed for growth and metabolic functions of fish. The nutrients not synthesized by the organism and required in the diet from outside sources are called "essential" or "indispensable" such as unsaturated fatty acids, essential amino acids and vitamins. The deficiency of indispensable nutrients (protein, fat, vitamins A, B, C and E) in the present study limited their synthesis, caused reduced body weight and showed specific morphological symptoms in *C. batrachus*. The fish recovered to a certain extent when fed with these nutrients. Reduced body weight and skin lesions were observed in protein and fat-deficient fish. Vitamin A deficiency of vitamins C and E caused scoliosis (broken back disease) and skin haemorrhage, respectively.

Keywords: Nutritional fish pathology, Nutrient deficiency

\_\_\_\_\_

## Introduction

Fishes are a rich source of nutritive and medicinal ingredients for human beings. They provide amino acids, unsaturated fatty acids and vitamins that are essential for our health. Nutrition plays a vital role in growth and condition of the farmed fish and, therefore, nutritional fish pathology, dealing with symptoms/ailments appearing in the fish due to nutrient deficiencies or dietary imbalances, should be treated without delay to support successful farming. Trichet (2010) has provided a good account of the malnutrition and vulnerability of fish stocks to diseases caused by nutrient deficiencies. The importance of scientific data on nutritional requirements of the fishes and single or multiple nutrient deficiencies has been amply highlighted by National Research Council (1993). Dietary lipids in fishes are the source of essential fatty acids (EFA) such as polyunsaturated fatty acid (PUFA) particularly n-3 and n-6 PUFAs (Sargent et al., 2002). EFA requirement are related to dietary lipid intake by the fish (Takeuchi et al., 1991, 1992; Tacon, 1992). Proteins in the form of enzyme, hormones and immunoglobulins are required for normal body functions. Vitamins on the other hand are micronutrients required in small concentrations to support specific structural and metabolic functions (Gatlin, 2002; Halver, 2002). Diets supplemented with vitamin C and E help in enhancing the macrophage function and antibody production (Blazer, 1992; Gatlin, 2002), and improving disease resistance and stress tolerance in fish (Koshio, 2007; Lim et al., 2008). Vitamins C and E rich diets improve immune response and disease resistance to fish (Ortuno et al., 2000; Clerton et al., 2001; Cuesta et al., 2001; Sahoo and Mukherjee, 2002; Lin and Shiau, 2005, Wang et al., 2006). Vitamin E also reduces susceptibility to stress under crowding conditions (Montero et al., 2001; Trenzado et al., 2007). Fat soluble vitamins viz. vitamin A, D, E and K are deposited in the body along with body lipids, hence fish may not show deficiency symptoms for a long period of time. Water soluble vitamins, however, are not present in substantial amounts in the body; hence they show early remarkable symptoms. The present work was planned to study the specific morphological symptoms resulting from deficiency of particular nutrients in *C. batrachus* and recovery of the fish by providing the deficient nutrient in the diet.

\_\_\_\_\_

# **Materials and Methods**

Sixty individuals of *C. batrachus* (length=30 ± 5 cm; weight=  $180 \pm 10$ gm) were purchased from commercial fishermen, station road, Raebareli (U.P.), during the month of November 2017 and acclimatized to captive conditions for 2 weeks. The water was changed every day and the fish were fed with control diet twice (morning: 7-8 hrs; evening: 17-18 hrs) a day @ 5% of their body weight. The 7 dietary treatments were given to the 7 groups of the fish, each comprising 5 specimens. Diet 1 was offered to the control group which comprised complete diet with no nutritional deficiency; diets 2-7 were made deficient in protein, fat, vitamins A, B, C and E, respectively. The details are given in Table-1. The ingredients of each diet were mixed thoroughly in a mixer grinder and pellets of 1-1.5 cm diameter were made after mixing with 30% de-ionized water, air-dried at room temperature and stored in the freezer until fed.

Volume: 03 (1) | July 2019, 9 - 12

Diet	1 (control)	2 (protein deficient)	3 (fat deficient)	4 (vit. A deficient)	5 (vit. B deficient)	6 (vit. C deficient)	7 (vit. E deficient)
Fish meal (protein rich)	35%	-	35%	35%	35%	35%	35%
Nutrela (protein rich)	35%	-	35%	35%	35%	35%	35%
Sunflower oil (fat rich)	6%	6%	-	6%	6%	6%	6%
Cod liver oil (fat rich)	2%	2%	-	2%	2%	2%	2%
Vitamin A tablets/kg	5000 IU	5000 IU	5000 IU	-	5000 IU	5000 IU	5000 IU
Vitamin B tablets/kg	400 mg	400 mg	400 mg	400 mg	-	400 mg	400 mg
Vitamin C tablets/kg	150 mg	150 mg	150 mg	150 mg	150 mg	-	150 mg
Vitamin E tablets /kg	50 mg	50 mg	50 mg	50 mg	50 mg	50 mg	-
Multivitamin tablets /kg	10 tablets	10 tablets	10 tablets	-	-	-	-
Multiminerals tablets/kg	25 tablets	25 tablets	25 tablets	25 tablets	25 tablets	25 tablets	25 tablets

Table 1. Proximate composition of the experimental diets

The experimental procedure was divided into 3 parts: (1) fish was made deficient of a particular nutrient, (2) morphological symptoms of nutrient deficiency were observed in the fish and (3) recovery in the nutrient-deficient fish was studied by providing the deficient nutrient in the diet. Seven different batches of fish were observed for different periods of time interval until they showed symptoms of deficiency. After the appearance of deficiency signs, each deficient batch was provided with control diet.

# **Results and Discussion**

Different deficiency signs and morphological symptoms in *C. batrachus* were noticed in the absence of a specific nutrient

as compared to control. These symptoms were confirmed by observing the affected fish regaining normal health after the deficient nutrient was provided in the diet. The signs and morphological symptoms of nutrient deficiency and recovery of the symptoms of different batches of the fish are summarized in Table 2.

Different batches of the fish showed different deficiency symptoms at different time intervals which are given in Table 3. The different batches of the fish recovered to an appreciable extent when fed with the nutritionally balanced diet.

Deficient nutrient	Deficiency symptoms	Recovery of the fish		
Control	No deficiency symptoms	No symptoms		
Proteins	Dorsal/caudal fin erosion	Dorsal/caudal fins were seen to be recovered		
	Skin lesions on body	Skin regained the normal form		
	Reduction in body weight	Body weight began to increase to an appreciable extent		
Fats	Depigmentation of skin	Pigmentation of skin was regained		
	Skin lesions on body	Skin lesions were healed		
	Reduced body weight	Body weight began to increase		
	Reduced appetite and growth	Appetite and growth were regained		
	Haemorrhage on fins	Haemorrhage on fins was reduced		
Vitamin A	Skin lesions were seen	Broken skin was healed		
	Eroded barbles	Eroded barbles begin to appear		
	Exophthalmia was prominent	Exophthalmia was seen to be reduced		
	Edema on ventral side	Reduced edema		
	Loss of appetite	Appetite was regained		
Vitamin B	Eroded barbles	Eroded barbles begin to appear		
	Haemorrhage of skin and fins	Haemorrhage on skin and fins healed		
	Sluggishness/abnormal swimming	Normal swimming behavior was observed		
	behavior			
	Loss of appetite	Appetite was regained		
	Reduced body weight	Body weight began to increase		
Vitamin C	Scoliosis (broken back disease)	Scoliosis recovered to some extent		
	Caudal fin erosion	Eroded fins began to recover		
	External hemorrhage	External hemorrhage became normal		
	Reduced growth	Growth was regained		
Vitamin E	Eroded barbles	Eroded barbles begin to appear		
	Eroded Skin	Eroded skin repaired		
	Caudal fin erosion	Eroded fins recovered		
	Reduced weight	Body weight increased		

Table 2. Deficiency signs and recovery of morphological symptoms of different deficient nutrients in *C. batrachus* 

# **Table 3.** Time interval of appearance and recovery of Deficiency symptoms and growth parameters in different nutrient deficient batch in *C. batrachus*

Deficient nutrient	Time interval of deficiency	Recovery time interval*	Initial weight (g)	Final weight (g)	Weight gain/loss (%)	Specific growth rate (% day <sup>.1</sup> )	Survival rate (%)
Control	-	-	180±10.2	187±11.1	+3.9	+16.67	100 %
Protein	4 weeks	5 weeks	177±10.2	166± 8.01	-6.2	-39.28	100 %
Fat	6 weeks	4 weeks	174±10.2	168±9.03	-3.45	-14.28	100 %
Vitamin A	5 weeks	4 weeks	183±10.2	179±10.02	-2.18	-11.43	100 %
Vitamin B	3 weeks	3-4 weeks	175±10.2	170±10.02	-2.86	-23.81	100 %
Vitamin C	2 weeks	3-4 weeks	186±10.2	184±10.02	-1.07	-14.28	100 %
Vitamin E	4 weeks	4 weeks	182±10.2	179±10.02	-1.65	-10.71	100 %

\*recovery time interval is noted from the time the fish has been provided the deficient nutrient in the diet.

Volume: 03 (1) | July 2019, 9 - 12

Protein deficiency caused degeneration of skeletal muscle in C. batrachus, resulting in reduced body weight and growth. Fat deficiency led to depigmentation of skin in C.batrachus. Nicolaids and Woodall (1962) also reported a similar depigmentation in the skin of Chinook salmon when fed with fat-free diet. Pigmentation was restored when fatty acid was added to the diet. Vitamin A deficiency in the diet caused exophthalmia. This may be due to inadequate amount of this nutrient leading to impairment in the formation of visual pigment rhodopsin. Edema on ventral side of the fish was also observed as it was reported in channel catfish, Ictalurus punctatus (Dupree, 1966). The appetite and growth of the catfish also suffered due to Vitamin B deficiency. These symptoms may be due to the deficiency of vitamin B1 as it is involved in oxidation of glucose in cells and is essential for good appetite and normal growth. Eroded barbles were seen which may be due to the deficiency of Pantothenic acid. Similar observations were reported by Murai and Andrews (1978, 1979) in fingerlings of channel catfish. Vitamin C deficiency led to reduced collagen synthesis in the fish resulting into scoliosis (broken back disease) and external haemorrhage. Butthep et al. (1985) also noticed these symptoms. Vitamin E is necessary for muscle cell respiration and lack of this nutrient in the diet caused muscular dystrophy and skin lesions. Earlier, these symptoms were documented by Murai and Andrews (1974) in Ictalurus punctatus.

#### Conclusions

The deficiency of protein, fat, vitamins A, B, C and E were found to be detrimental for fish growth and development. The investigated deficiency symptoms of these nutrients will be useful for taking care of farmed fish that can help to reduce the effect of dietary imbalance on farmed fish health and in turn increase their productivity.

#### Acknowledgements

The authors thank the Head, Department of Zoology, University of Lucknow, for providing lab facilities and administrative support. Priyanka Agarwal is also thankful to University Grant Commission (UGC) for the award of research fellowship.

#### References

Blazer, V.S. (1992). Nutrition and disease resistance in fish. **Annual Review of Fish Diseases** 2, 309–323.

Butthep, C., Sitasit, P. & Boonyaratpalin, M. (1985). Water soluble vitamins essential for the growth of *Clarias*. In: **Finfish Nutrition in Asia: Methodological Approaches to Research and Development,** pp 118-129. IDRC, Ottawa, ON, CA.

Clerton, P., Troutaud, D., Verlhac, V., Gabaudan, J. & Deschaux, P. (2001). Dietary vitamin E and rainbow trout (*Oncorhynchus mykiss*) phagocyte functions: effect on gut and on head kidney leucocytes. **Fish and Shellfish Immunology** 11, 1–13.

Cuesta, A., Esteban, M.A., Ortuno, J. & Meseguer J. (2001). Vitamin E increases natural cytotoxic activity in seabream (*Sparus aurata* L.). **Fish and Shellfish Immunology** 11, 293–302.

Dupree, H.K. (1966). Vitamins essential for growth of channel catfish, *Ictalurus punctatus*. **Technical paper No.7, Bureau of Sport Fisheries and Wildlife**, Washington, D.C., 12 pp. Gatlin, D.M. (2002). Nutrition and fish health. In: **Fish Nutrition, Third edition** (J.E. Halver, R.W. Hardy, eds.), pp 672–703. Academic Press, San Diego, CA.

Halver, J.E. (2002). The vitamins. In: **Fish Nutrition, third edition** (J.E. Halver, R.W. Hardy, eds.), pp 61–141. Academic Press, San Diego, CA.

Koshio, S. (2007). Vitamins. In: **Dietary Supplements for the Health and Quality of Cultured Fish** (H. Nakagawa, M. Sato, D.M. Gatlin, eds.), pp 35–46. CAB International, Oxon, UK.

Lim, C., Yildirim-Aksoy, M. & Klesius, P.H. (2008). Nutrition and disease resistance in fish. In: **Feeding and Digestive Functions in Fish** (J.E.P. Cyrino, D.P. Bureau, R.G. Kapoor, eds.), pp. 479–545. Science Publishers, Plymouth, UK.

Lin, Y.H. & Shiau, S.Y. (2005). Dietary vitamin E requirement of grouper, *Epinephelus malabaricus*, at two lipid levels, and their effects on immune responses. **Aquaculture** 248, 235–244.

Montero, D., Tort, L., Robaina, L., Vergara, J.M. & Izquierdo, M.S. (2001). Low vitamin E in diet reduces stress resistance of gilthead seabream (*Sparus aurata*) juveniles. **Fish and Shellfish Immunology** 11, 473–490.

Murai, T. & Andrews, J.W., (1974). Interactions of dietary alphatocopherol, oxidized menhaden oil and ethoxyquin on channel catfish (*lctalurus punctatus*). **The Journal of Nutrition**, 104, 1416-1431.

Murai, T. & Andrews, J.W. (1978). Thiamine requirement of channel catfish fingerlings. **The Journal of Nutrition**, 108, 176-180.

Murai, T. & Andrews, J.W. (1979). Pantothenic acid requirements of channel catfish fingerlings. **The Journal of Nutrition**, 109, 1140-1142.

National Research Council (1993). Nutrient Requirements of Fish. National Academy Press, 114 pp. National Academy Press, Washington, DC.

Nicolaides, N. & Woodall, A.H. (1962). Impaired pigmentation in chinook salmon fed diets deficient in essential fatty acids. **The Journal of Nutrition** 78, 431-437.

Ortuno, J., Esteban, M.A. & Meseguer, J. (2000). High dietary intake of alpha-tocopherol acetate enhances the non-specific immune response of gilthead seabream (*Sparus aurata L.*). **Fish and Shellfish Immunology** 10, 293–307.

Sahoo, P.K. & Mukherjee, S.C. (2002). Influence of high dietary alphatocopherol intakes on specific immune response, nonspecific resistance factors and disease resistance of healthy and aflatoxin B-1-induced immunocompromised Indian major carp, *Labeo rohita* (Hamilton). **Aquaculture Nutrition** 8, 159–167.

Sargent, J.R., Tocher, D.R. & Bell, J.G. (2002). The lipids. In: **Fish Nutrition, third edition** (J.E. Halver, R.W. Hardy, eds.), pp 181–257. Academic Press, San Diego, CA.

Tacon, A.G. (1992). Nutritional fish pathology. Morphological signs of nutrient deficiency and toxicity in farmed fish. **FAO Fisheries Technical Paper** 330, 75.

Takeuchi, T., Shiina, Y. & Watanabe, T. (1991). Suitable protein and lipid levels in diet for fingerlings of red sea bream *Pagrus major*. **Nippon Suisan Gakkaishi** 57, 293–299.

Takeuchi, T., Shiina, Y. & Watanabe, T. (1992). Suitable levels of n-3 highly unsaturated fatty acids in diet for fingerlings of red sea bream. **Nippon Suisan Gakkaishi** 58, 509–514.

Trenzado, C.E., de la Higuera, M. & Morales, A.E. (2007). Influence of dietary vitamins E and C and HUFA on rainbow trout (*Oncorhynchus mykiss*) performance under crowding conditions. **Aquaculture** 263, 249–258.

Trichet, V.V. (2010). Nutrition and immunity: an update. **Aquaculture Research** 41, 356–372.

Wang, L., Mai, K.S., Liufu, Z.G., Ma, H.M., Xu, W., Ai, Q.H., Zhang, W.B., Tan, B.P. & Wang, X.J. (2006). Effect of high dietary intakes of vitamin E and n-3 HUFA on immune responses and resistance to *Edwardsiella tarda* challenge in Japanese flounder (*Paralichthys olivaceus*, Temminck and Schlegel). Aquaculture Research 37, 681–692.