A SEMINAR PAPER ON

DIETARY REQUIREMENT OF TRACE ELEMENTS IN FISH

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ON
DIETARY REQUIREMENT OF TRACE ELEMENTS IN FISH[1]
By
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ABSTRACT

The study was carried out to know the dietary trace element requirements in fish. An element is considered as dietary essential when its absence or insufficiency in diet causes deficiency syndrome and supplementation in normal level brings back normal health. An organism can neither grow nor remain healthy without the element in question. All forms of aquatic inhabitants require some inorganic elements in little or trace amounts for their normal growth and metabolism. Trace minerals do not exist only by themselves but in combination with others. Therefore, too much of one element may lead to imbalances in others resulting in disease or adverse effect in metabolism. In comparison to the farmed animals, the knowledge on dietary essentiality of minerals in fish is scarce being mainly restricted to iron, copper, manganese, zinc, iodine, selenium, cobalt and chromium as components of body fluids, co-factors in enzymatic reactions, structural units of non-enzymatic macromolecules, *etc.* The trace minerals requirement found by different researchers in fish are; Iron 30-170, Copper 3-5, Manganese 2-20, Zinc 15-70 and Selenium 0.15 to 2 (mg/kg diet). The deficiency and excess amount of these trace elements both have the negative effects in fish. The importance of trace minerals as essential ingredients in diets, although in small quantities, is also evident in fish. More research should be done in case of freshwater fishes.

Keywords: Fish nutrition, Trace minerals, Requirement, Deficiency, Iron, Zinc, Manganese, Copper, Selenium.

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CHAPTER I

INTRODUCTION

Trace elements that are essential for the nutrition of animals are usually required in very low amounts in dietary dry matter. These elements include Fe, Zn, Cu, Mn, Se, I, Co, and Mo. In fish nutrition, mineral elements are very important (Hossain and Yoshimatsu, 2014). They are integral parts of a large number of enzymes and they may participate in the energy metabolism of cells through red–ox reactions of the electron transport chain. All terrestrial and aquatic organisms require minerals for their normal life processes. Fish may absorb elements from the surrounding water, but the diet is the main source of essential elements (Lall and Bishop, 1977). Minerals serve a variety of functions as both intra– and extracellular components. They serve as structural components of hard–tissue matrices (e.g., bone, fin rays, scales, teeth, and exoskeleton) and components of soft tissues (e.g., sulfur in proteins, phosphorus in phospholipids, and nucleic acids). They also are components of metalloproteins (e.g., iron in hemoglobin, copper in hemocyanin, and zinc in carboxypeptidase), and serve as cofactors and/or activators of a variety of enzymes (e.g., zinc activation of alkaline phosphatase). The physiological functions of minerals are well defined for humans and some terrestrial animals, but this information for fish has not been well established. Freiden, (1984) described that an element is considered essential when its deficient intake produces an impairment of function and when restoration of physiological levels of the element prevents or relieves the deficiency. Organisms can neither grow nor complete its life cycle without those inorganic elements. They should have a direct influence on the organisms and involved in the metabolism. The principle minerals present in micro quantities (or trace levels) are iron, manganese, zinc, copper, cobalt, selenium, chromium and iodine (Lall, 1979). There are approx. fifteen trace elements considered to be essential in animals. Among these the physiological role of a deficiency of chromium, cobalt, copper, fluorine, iodine, iron, manganese, molybdenum, selenium and zinc is well recognized (Lall, 1989). However, not all of the minerals essential for warm–blooded animals have been found to be essential in the diet for fish. The concentration of minerals in the body of an aquatic organism depends on the food source, environment, species and stages of development and physiological status of the animal. In marine food chains, a unique transport of trace minerals has been reported (Bernhard and Andreae, 1984). It has been shown that whole–body element concentrations are reduced owing to insufficient dietary intake of trace elements (Ogino and Yang, 1980; Maage and Julshamn, 1993). Many mineral bioavailability crises are being increasingly recognized in humans and animal nutrition. Several factors influencing bioavailability include the level and form of the nutrient, particle size and digestibility of the diet, physiological and pathological conditions of the fish, waterborne mineral concentration and the species under consideration. Among these factors, those related to the chemical state are important because the element may assume different molecular forms, valence state and ligands when ingested from different diets (Forbes and Erdman, 1983). Some biologically important compounds contain minerals as an inherent part of their structure, e.g., hemoglobin and vitamin B12. Information on nutritional requirements of freshwater fish for trace elements is scarce particularly because many are needed in extremely small amounts and this pose difficulty in analysis. Sub lethal effects of several metals on aquatic organisms have been well demonstrated by Bryan and Darracott, (1979). Most of the sub lethal toxicity appears to be of a biochemical origin and causes morphological, physiological (growth, swimming performance, respiration and reproduction) and behavioral changes (Bryan, 1976).

Although the trace minerals are required by the animal in very small quantities (usually less than 100 mg/kg dry diet), they are absolutely required for normal growth. If excess amounts of the elements are ingested and assimilated, toxicity may develop. Early research on mineral requirements of fish was primarily aimed at determining the optimum dietary levels necessary good growth and prevention of deficiency for signs. However, In the past few decades the understanding of the interrelationships between nutrition, immunity, and disease resistance in terrestrial animals has progressed rapidly (Davis and Gatlin, 1996). Dietary requirements are established for macro minerals such as Ca, K, Mg, Na and P and micro minerals such as Cu, Fe, I, Mn, Se and Zn for one or more fish species (NRC, 2011). Available data on the requirements of different minerals are limited and poorly defined when compared to data for other nutrients in fish or to data on minerals in terrestrial animals. It is recognized that nutrient requirement of an animal should be determined in terms of a specific response criterion at a given age, sex, weight gain and body composition (Baker, 1986). However, it is much more complicated in fish due to the close interaction with the aquatic environment unlike in terrestrial animals (Lall, 2002). The present study was therefore undertaken based on the above premise and the following were its main objectives:

- To review the requirements of trace elements in fish, and
- To highlight the effects of trace elements in fish.

CHAPTER II

MATERIALS AND METHODS

This seminar paper is exclusively a review paper so all of the information has been collected from the secondary sources. During the preparation of the review paper, I went through various relevant books, journals, proceedings, reports, publications, internet etc. Findings related to my topic have been reviewed with the help of the library facilities of Bangabandhu Sheikh Mujibur Rahman Agricultural University. I got suggestion and valuable information from my major professor and my course instructors. After collecting all the available information, I myself complied the collected information and prepared this seminar paper.

CHAPTER III

REVIEW OF MAJOR FINDINGS AND DISCUSSION

As all the trace elements are not necessary for fishes, the requirements and effects of some most important trace elements are reviewed here.

3.1 Iron (Fe)

Iron is a trace element that is essential for the production and normal functioning of hemoglobin, myoglobin, cytochromes, and many other enzyme systems. It helps to increase growth, diet oxidation and anaemia prevention in fish (Chanda *et al*., 2015). Fe is absorbed and transported in the body in a protein–bound form (Lovell, 1989). It is a trace mineral of fundamental importance for most higher animals, including fish, because of its functions in oxidation–reduction activity and oxygen transport. In biological systems, Fe can exist in the ferrous (Fe2+) or ferric (Fe3+) state, and this permits Fe to donate or accept electrons and thus participate in the oxidation–reduction reactions, including those involved in oxygen transport. It occurs in the animal body as a component of the respiratory pigment (heme compounds), such as hemoglobin in red blood cells and myoglobin in muscle. Fish can absorb soluble Fe from water across the gill membrane (Roeder and Roeder, 1966); however, intestinal mucosa is considered the major site of iron absorption (Lall, 1989). Iron also is an important micronutrient that has been shown to affect immune system function and host defense against infection. Either a deficiency or an excess of iron can compromise the immune system (Bhaskaram, 1988).

3.1.1 Availability

The iron content of blood plasma and of the water medium has been found to be positively correlated in salmonids (Merk, 1987). Feeds of animal origin such as fish meal and meat meal are rich sources of iron, containing about 400–800 mg/kg, Oil seeds contain 100–200 mg Fe/kg, while cereals contain 30–60 mg/Fe kg. The iron from animal sources may occur as porphyrin, myoglobin and haemoglobin. It may be in a complexed form with phytin in cereals. The availability of iron in the various feedstuffs for fish depends on its form (Watanabe *et al*., 1988).

3.1.2 Dietary Requirement

The total dietary iron requirements have been determined for certain species of fish (Table 1). Total dietary levels of iron required to maintain optimum hematological values and iron status were 30 mg iron/kg diet for channel catfish (Gatlin and Wilson, 1986a), 60 to 100 mg/kg of diet for Atlantic salmon (Maage and Julshamn, 1993); 150 mg/kg diet for red sea bream (Sakamoto and Yone, 1978b); and 170 mg/kg diet for eel (Nose and Arai, 1976), In Tilapia 150–160 mg/kg (Shiau and Su, 2003). Ling *et al*. (2010) found that in case of common carp, iron requirement is 146.1 mg/kg diet.

Table 1. Iron Requirement of Certain Species of Fish

Fish Species	Requirements (mg/kg diet)
Atlantic salmon (*Salmo salar)*	60
Channel catfish (*Ictalurus punctatus)*	30
Eel (*Anguilla japonica)*	170
Rainbow trout (*Oncorhynchus mykiss)*	60
Red sea bream (*Pagrus major)*	150
Common Carp (*Cyprinus carpio)*	146.1
Grass carp (*Ctenopharyngodon idella)*	101
Tilapia (*Oreochromis nilotica)*	150–160

(Source: Lim *et al.*, 2001 and Ling *et al*., 2010)

Per cent weight gain, SGR, FE and FI of juvenile Common carp fed the diets containing varying levels of iron are given in Table 2. PWG, SGR and FE were significantly improved with increasing dietary iron levels. Ling *et al*. (2010) found that all these factors were positively affected with increasing levels of iron and highest found in 146.1 mg/kg.

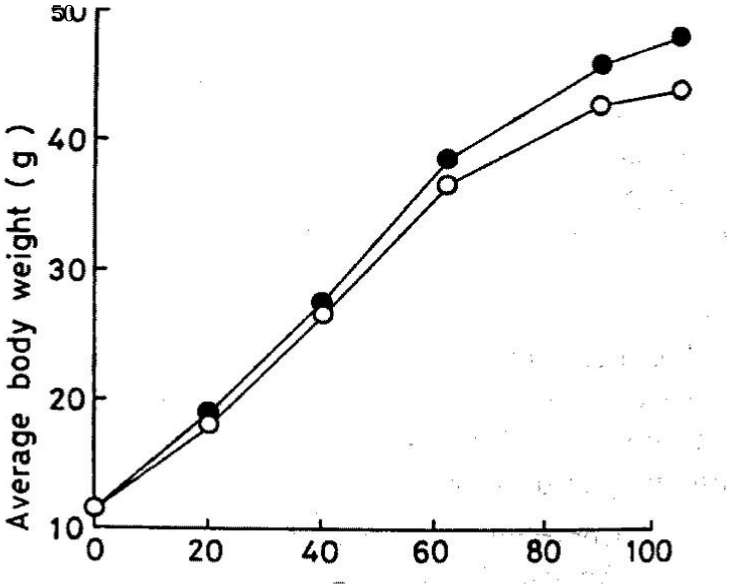
Table 2. The effects on various parameters of Common carp fed diets containing graded levels of iron (mg/kg)

	Dietary iron levels				
	53.9	90.0	115.6	146.1	176.0
IBW	11.3 ± 0.0	11.4 ± 0.0	11.3 ± 0.0	11.4 ± 0.0	11.3 ± 0.0
FBW	54.7 ± 0.7	60.5 ± 1.9	61.0 ± 3.5	64.2 ± 3.6	63.5 ± 2.2
PWG	382 ± 8	433 ± 19	438 ± 30	465 ± 31	460 ± 19
FI	55.2 ± 0.7	57.0 ± 0.1	58.0 ± 0.5	59.7 ± 0.9	59.3 ± 0.5
FE	78.5 ± 2.3	86.2 ± 3.3	85.6 ± 5.4	88.5 ± 4.7	88.0 ± 4.2
SGR	2.62 ± 0.03	2.79 ± 0.06	2.80 ± 0.09	2.88 ± 0.09	2.87 ± 0.06

IBW= Initial Body Weight, FBW=Final Body Weight, PWG= Percentage Weight Gain
FI= Feed Intake, FE= Feed Efficiency, SGR= Specific Growth Rate

(Source: Ling *et al.*, 2010)

As shown in Fig. 1, the control group fed the diet containing ferric chloride exhibited a slightly higher growth rate than the group fed the diet without the iron supplement. (Sakamoto and Yone, 1978a)



Days

Figure 1. Growth of carp fed the diets with and without iron supplementation.
(○, Without; ●, With). (Source: Sakamoto and Yone, 1978a)

3.1.3 Deficiency and Toxicity Signs

Iron deficiency is not a common problem in fish culture because commercial diets normally contain certain amounts of fish meal and/or animal proteins that are rich sources of iron. However, iron deficiency has been produced in several fish species cultured under laboratory conditions and fed low–iron diets (Table 3). Decreased appetite, feeding activity, and feed efficiency, along with reduced growth and reduced serum iron and transferrin saturation in the blood have also been observed in channel catfish fed iron–deficient diets (Gatlin and Wilson 1986a). Excessive levels of dietary iron can be toxic to fish. Toxicity of dietary iron was observed in rainbow trout, *Oncorhynchus mykiss*, fed greater than 1,380 mg iron (as ferrous sulfate)/kg diet (Desjardins, Hicks, and Hilton 1987).

Table 3. Iron Deficiency Signs in Certain Species of Fish

Species	Deficiency Symptoms
Atlantic salmon *(Salmo salar)*	Reduced hemoglobin, hematocrit, mean corpuscular hemoglobin, and mean corpuscular volume.
Channel catfish *(Ictalurus punctatus)*	Reduced appetite, growth, feed efficiency, red blood cell count, hemoglobin, hematocrit, plasma iron, and transferrin saturation
Common carp *(Cyprinus carpio)*	Reduced hemoglobin, hematocrit, mean corpuscular hemoglobin, mean corpuscular volume, and mean corpuscular hemoglobin concentration.
Eel *(Anguilla japonica)*	Decreased red blood cell count, hemoglobin, hematocrit, mean corpuscular hemoglobin, mean corpuscular volume, and mean corpuscular hemoglobin concentration.

(Source: Lim *et al.,* 2001)

3.2 Copper (Cu)

Copper (Cu) is an essential trace element for all animals including fish (Mertz, 1986; Lall, 1989). It plays a fundamental role in the activity of enzymes such as cytochrome oxidase, superoxide dismutase, lysyl oxidase, dopamine hydroxylase and tyrosinase. In addition, copper–proteins and chelates also have metabolic roles (Watnabe *et al.,* 1997). Copper metabolism revealed similarities to mammals in the distribution of copper and copper dependent enzymes (Syed and Coombs, 1982). Marine invertebrates, especially mollusks, possess a blue colored Cu containing complex in the haemolymph called the haemocyanin. Copper also serves as an oxygen carrier in haemolymph of these organisms (Lall, 1989).

3.2.1 Availability

Most feeds and the aquatic medium generally contain copper in amounts adequate for fish. Plant and animal protein feed ingredients contain 5–30 mg/kg of copper, when products and fish solubles are relatively rich sources of copper. Copper levels are high in eyes (iris and chloroide), liver, brain and heart. A copper– protein complex ceruloplasmin exhibiting oxidative activity occurs in blood plasma (Watnabe *et al.*, 1997). Copper and zinc may be antagonistic because of the similar nature of the valence shell hybrids (Hill and Matrone, 1970). Moreover, during processing of feed copper content can be increased by using protein enriched sources. Due to metal contamination in processed feed ingredients wide variation can occurs in copper content (Watnabe *et al.*, 1997).

3.2.2 Dietary Requirement

The copper requirement in different fish ranges from 3–5 mg/ kg dry diet as reported (Watnabe *et al.*, 1997). The total dietary copper requirements have been determined for certain species of fishes given bellow (Table 4). For Proper enzymatic activity, metabolism, proper growth some optimum values of copper were determined. Ogino and Yang, (1980) found in case of Rainbow trout and Carp dietary copper requirement was 3 mg/kg, In Channel catfish 5 mg/kg (Gatlin and Wilson, 1986b), In Atlantic salmon 5 mg/kg (Berntssen *et al.*, 1999). Tang *et al*, (2013) found 3.75mg/kg for Grass carp.

Table 4. Copper requirement studies reported in some fish

Fish Species	**Requirements (mg/kg diet)**
Rainbow trout	3
Carp	3
Channel catfish	5
Atlantic salmon	5
Common carp	3
Grass carp	3.75
Sea bass	4

(Source: Antony *et al*., 2016)

As shown in Table 5, FBW, PWG, FI, FE, and SGR were significantly increased with increasing dietary Cu level up to 3.75 mg/kg diet and declined thereafter (Tang *et al*., 2013).

Table 5. The effects of dietary Cu on the growth and survival rate of Grass carp

Parameters	Dietary Cu levels (mg/kg diet)				
	0.74	2.26	3.75	5.25	6.70
IBW	280.45±0.46	283.12±3.49	280.19±3.33	285.79±1.85	282.32±2.12
FBW	452.40±5.41	569.34±4.68	688.12±6.41	634.84±8.81	598.10±4.88
PWG	61.31±1.72	101.10±0.99	145.61±2.88	122.13±2.22	111.85±0.88
FI	308.98±2.99	476.14±1.51	627.13±0.50	553.97±1.66	529.28±1.13
FE	55.66±2.16	60.11±0.36	65.05±0.88	63.01±1.27	59.66±0.68
SGR	0.85±0.019	1.25±0.009	1.60±0.021	1.43±0.018	1.34±0.007
Survival rate	88.22 ± 6.77	90.22 ± 7.69	95.2 ± 3.77*	86.4 ± 6.38	82.47 ± 9.54

(Source: Tang *et al*., 2013)

Analysis by quadratic regression, the requirement of dietary Cu for grass carp (282–688 g) based on PWG was estimated to be 4.78 mg/kg diet (Figure 2) (Tang *et al*., 2013).

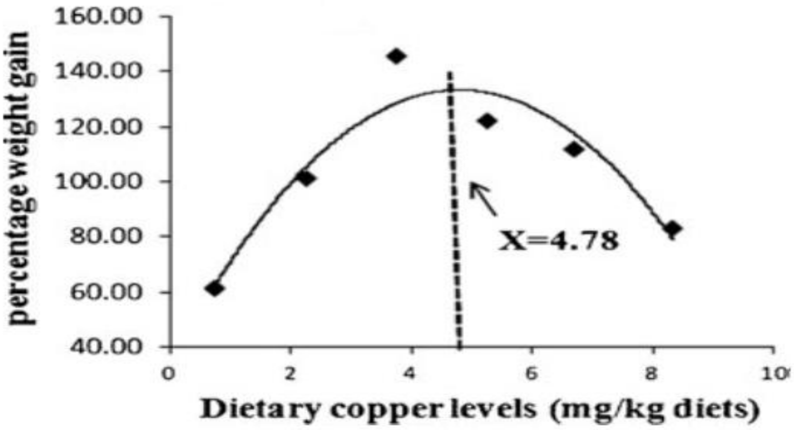


Figure 2. Percentage weight gain for Grass carp fed diets containing graded levels of copper for 8 weeks. (Source: Tang *et al*., 2013)

Figure 3. represents the protease activity (PA) in the digestive tract of red sea bream fed test diets for 60 days. PA showed the highest value in fish fed the diet supplemented with 2 mg kg−1 Cu when compared with the other amounts. However, no significant differences were reported between fish fed 2, 3 and 4 mg kg−1 Cu. (Basuini *et al*., 2016)

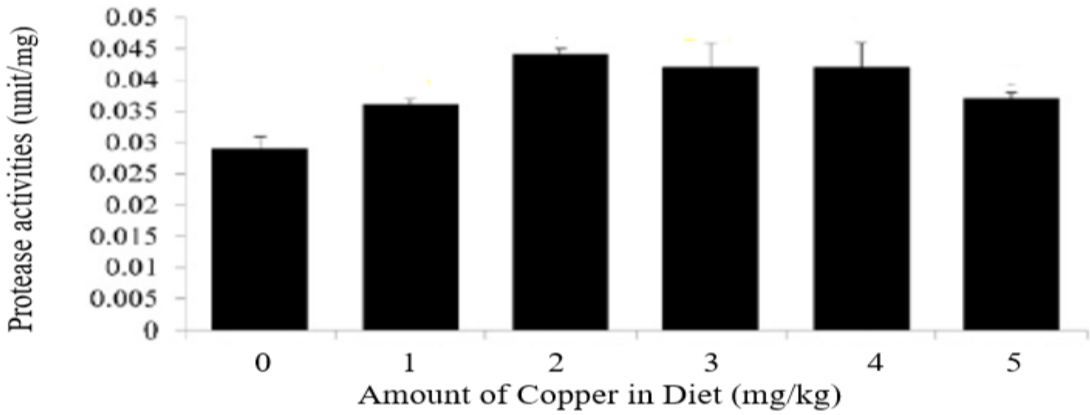


Figure 3. Protease activities in the digestive tract of red sea bream fed test diets for 60 days. (Source: Basuini *et al*., 2016)

3.2.3 Deficiency and Toxicity Signs

When the diets of carp and rainbow Trout had low levels of copper, there was a decline in the tissue copper content (Table 6) (Ogino and Yang, 1980). Copper toxicity may cause damage to gills and necrosis in liver and kidney. High levels of copper depressed growth and impaired feed conversion in channel catfish (Murai *et al*., 1981). In the same fish, Gatlin and Wilson, (1986b) showed that a deficiency of copper affected the activities of cytochrome c oxidase in heart and copper–zinc superoxide dismutase in liver. The toxicity and tissue retention of copper contained in water was affected by dietary ascorbic acid in carp and rainbow trout (Yamamoto *et al.*, 1977, 1981).

Table.6 Copper Deficiency Signs in Certain Species of Fish

Species	Deficiency Signs
Channel catfish	Reduced heart cytochrome c Oxidase, Reduced liver Cu–Zn superoxide dismutase
Common Carp	Reduced growth and cataract formation
Atlantic salmon	Hitra disease (Coldwater bacterial disease caused by *Vibrio salmonicida*)
Rainbow trout	Reduce tissue copper content, Damage to gills, Necrosis in liver and kidney

(Source: Chanda *et al*., 2015)

3.3 Zinc (Zn)

In Fish Nutrition, Zinc is an important trace element as it is involved in various metabolic pathways and serves as a specific cofactor of several enzymes. (Chesters, 1991). Zinc is required for normal growth, increase feed efficiency and reduce cataract problems (Chanda *et al*., 2015). The primary functions of zinc are based on its roles as a cofactor in several enzyme systems and as a component of a large number of metalloenzymes, including carbonic anhydrase, alkaline phosphatase, carboxypeptidase, alcohol dehydrogenase, glutamic dehydrogenase, lactate dehydrogenase, ribonuclease, and DNA polymerase (NRC 1980). Fish can absorb zinc from both water and dietary sources. However, dietary zinc is more efficiently absorbed than waterborne zinc (NRC, 1993).

3.3.1 Availability

Fish can derive zinc from dietary sources as well as from water. The bioavailability of zinc in various fish meals has been found to be inversely related to the tricalcium phosphate content of the meal. Thus, it is generally lowest in white fish meals, which contain the highest level of tricalcium phosphate, and slightly higher in brown fish meals (Watanabe *et al*., 1988). Reduced bioavailability of zinc in response to calcium phosphate supplementation also has been observed in rainbow trout (Satoh *et al*., 1987a). Practical diets often contain feedstuffs that are relatively high in phytate, which also may reduce the bioavailability of zinc. The effect of phytate on zinc availability has been documented in a variety of fish (Gatlin and Phillips, 1989). The bioavailability of dietary zinc is affected by dietary levels of calcium, phosphorus and phytic acid, protein source, and form of zinc (NRC 1993). Satoh, Takeuchi, and Watanabe, (1987a) showed that the availability of zinc to rainbow trout was highest in zinc sulfate, lowest in zinc chloride, and intermediate in zinc nitrate or carbonate.

3.3.2 Dietary Requirement

The dietary zinc requirement has been determined for several fish species (Table 7). The requirement values reported are 15 to 30 mg zinc/kg diet for rainbow trout and common carp, (Ogino and Yang, 1978), 20 mg zinc/kg diet for channel catfish (Gatlin and Wilson, 1983). Maage and Julshamn, (1993) reported 37–67 mg/kg for Atlantic salmon, Liang *et al*. (2012) found 55 mg/kg for grass carp and for Magur 30 mg/kg (Sapkale and Singh, 2011).

Table 7. Zinc requirements studies reported in some fish

Fish Species	Requirements (mg/kg diet)
Channel catfish	20
Common carp	15–30
Tilapia	30
Atlantic Salmon	37–67
Rainbow trout	15–30
Grass carp	55
Magur	30

(Source: Davis and Gatlin, (1996) and Antony *et al*., 2016)

Weight gain and feed efficiency data (Table 8) for tilapia fed the basal diet were significantly lower than those for tilapia fed diets containing supplemental zinc. There was a significant difference in weight gain and feed efficiency between fish fed the diet supplemented with 30 mg Zn/kg and all other experimental groups. Growth increased rapidly in all experimental fish, but the zinc–deficient fish (diet 1) gained weight at less than half the rate of the fish fed the diet supplemented with 30 mg Zn/kg. The mortality rate (Table 8) of Nile tilapia fed the basal diet was significantly higher than that of fish fed the other experimental diets. Fish fed the other dietary zinc levels had either no or very low mortality, which did not appear to be related to dietary treatment. (Eid *et al*., 1994).

Table 8. Performance of fingerling Nile tilapia fed diets with various zinc concentrations

Supplemental zinc (mg/kg)	Initial wt. (g)	Final wt. (g)	Weight gain(%)	Feed efficiency	Mortalit y (%)
0	8.4	16.0	90.47	0.69	10.0
5	8.3	16.5	98.79	o. 75	8.0
10	8.4	17.2	104.76	0.83	3.0
20	8.4	18.0	104.28	0.87	2.0
30	8.5	23.1	171.76	1 .20	0.0
40	8.5	19.1	124.70	0.93	0.0
50	8.5	21.1	148.23	0.97	0.0
60	8.4	18.3	117.85	0.90	1.0
80	8.5	18.7	120.00	0.90	2.0

(Source: Eid *et al*., 1994)

Requirement of Rainbow Trout for Dietary Zinc

Ogino and Yang, (1978) reported that the fish given the diet containing 1 ppm zinc showed very low growth rate, high mortality and the deficiency symptoms in the eyes and in the fins (Figure 4). Whereas, there were no mortality as well as no deficiency signs of zinc in the fish on diets containing more than 5 ppm zinc. However, the growth rate of the fish received the diet containing 5 ppm zinc was somewhat lower than those obtained with diets containing 15 and 30 ppm zinc. From these results, the requirement of rainbow trout for dietary zinc is estimated to be 15 to 30 ppm.

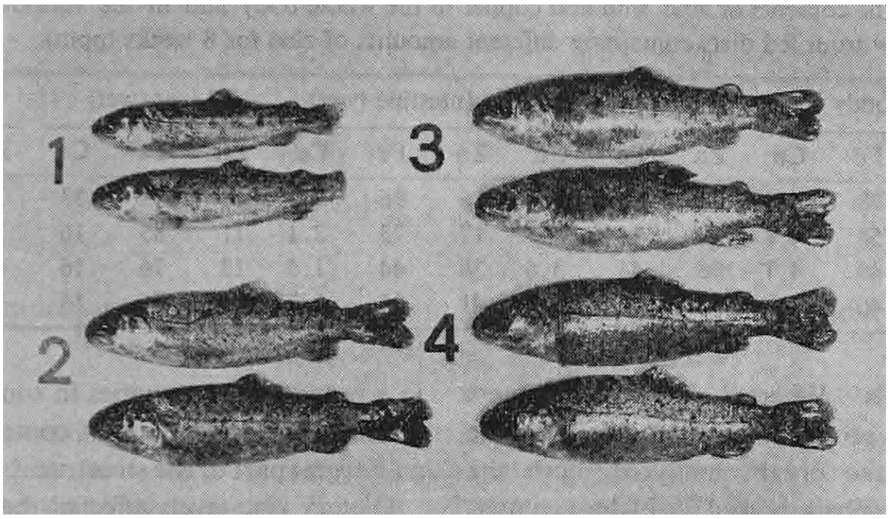


Figure 4. Rainbow trout reared with diets containing different amounts of zinc. Number in the photograph indicates the zinc content; 1) 1 ppm; 2) 5 ppm; 3) 15 ppm and 4) 30 ppm.

Source: Ogino and Yang, (1978)

Broken–line analysis showed that, based on the Zn content in whole body, the requirements of dietary Zn for maintaining maximum Zn storages in juvenile grass carp were 53.8 mg/ kg (Figure 5) (Liang *et al*., 2012).

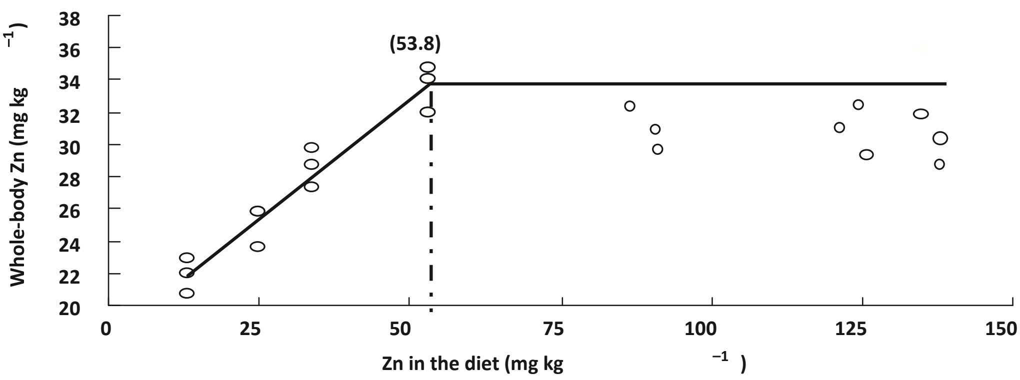


Figure 5. Effect of dietary Zn on the whole–body Zn content of juvenile grass carp–fed experimental diets for 8 weeks. The breakpoint of the broken–line is 53.8 mg/kg dietary Zn.

(Source: Liang *et al*., 2012)

3.3.3 Deficiency and Toxicity Signs

Ogino and Yang, (1978) found in rainbow trout that zinc deficiency caused growth depression, lens cataracts, and short body dwarfism (Poor growth, loss of appetite, high mortality and erosion of skin and fins were reported in zinc–deficient common carp (Ogino and Yang, 1979). Deficiency signs are given in Table 9.

Table 9. Zinc Deficiency Signs in Certain Species of Fish

Species	**Deficiency Signs**
Channel catfish	Dwarfism, low appetite, Zn –Ca levels, and serum Zn concentrations
Common Carp	Poor growth, loss of appetite, high mortality and erosion of skin and fins, Reduce hatchability
Red drum	slow growth, poor feed efficiency and survival, and reduced bone and scale
Rainbow trout	Cataracts, Reduced growth, high mortality, eroded fins and skins, Short body dwarfism

(Source: (Watnabe *et al*., 1997 and Chanda *et al*., 2015)

Zinc deficiency signs observed in red drum were slow growth, poor feed efficiency and survival, and reduced bone and scale zinc concentrations (Gatlin, O'Connell and Scarpa, 1991). Channel catfish fed zinc deficient diets had reductions in weight gain, appetite, survival, serum zinc content, serum alkaline phosphatase activity, and bone zinc and calcium levels (Gatlin and Wilson, 1983).

3.4 Manganese (Mn)

Manganese is important for fish and is widely distributed in fish and animal tissue. Manganese is necessary for the normal functioning of brain, proper lipid and carbohydrate metabolism, improve growth and fat synthesis in the liver of fish (Chanda *et al*., 2015). The mineral has two roles: first as a cofactor for enzymes which form metal–enzyme complexes; and second as an integral part of metalloenzymes (Clark *et al*., 1987). Srivastava and Agrawal, (1983) reported the mechanism of water dissolved manganese uptake, but it is better absorbed through diet intake. Manganese has very important role in brood stock nutrition. The manganese content of the diet influences its level and that of other trace elements in gonads (Satoh *et al.*, 1987b).

3.4.1 Availability

Manganese availability differs in various inorganic salt supplements. Utilization of manganese in the form of manganese oxide is poor in Atlantic salmon (Lall, 1989). Manganese sulphate and manganese chloride are ranked as superior manganese sources for carp over manganese carbonate (Satoh *et al*., 1987b). Good growth was recorded in carp provided manganese sulphate in the diet and it was found that manganese supplies increased protein synthesis and prevented fat synthesis in the liver (Romanenko, 1984). Depending on the fish, the content of manganese in fish meal ranges from 4 to 38 mg/kg, Capelin meal and herring meal contain only 4–12 mg Mn/kg. Among the plant sources, cereals contain 8–50 mg Mn/kg and corn 4–11 mg Mn/kg. Rice bran, wheat middlings and corn distillers' dried soluble are good sources of manganese. The availability of manganese in different fish meals to carp was very high and not affected by tricalcium phosphate present in the diet (Satoh *et al*., 1989). As the carp lacks a stomach, it cannot dissolve tricalcium phosphate and the chances of interaction between manganese and calcium or phosphate are few. Moreover, Supplementation of tricalcium phosphate to semi purified diets caused a reduction in Mn absorption leading to low levels of the mineral in vertebrae of carp (Satoh *et al.*, 1992). The content of manganese in fish meal ranges from 4 to 38 mg/kg, depending on the fish. Capelin meal and herring meal contain only 4–12 mg Mn/kg. Among the plant sources, cereals contain 8–50 mg Mn/kg and corn 4–11 mg Mn/kg. Rice bran, wheat middlings and corn distillers' dried soluble are good sources of manganese.

3.4.2 Dietary Requirement

Dietary requirements of different fish species are determined by different researchers (Table 10). Chanda *et al*. (2015) reported Mn requirements in case of Channel catfish 2.4 mg/kg, Rainbow trout and Carp 12 – 13mg/kg, Atlantic salmon 7.5 – 10.5 mg/kg and Juvenile stage fish 2–15 mg/kg. In case of Tilapia and Grass carp mn requirements were 13–15 mg/kg and 15 mg/kg, respectively (Antony *et al*., 2016).

Table 10. Dietary Mn requirement of certain fish species

Fish Species	Requirements (mg/kg diet)
Channel catfish	2.4
Common carp	12–13
Tilapia	13–15
Atlantic Salmon	7.5–10.5
Rainbow trout	7.5–10.5
Grass carp	15
Juvenile stage fish	2 – 15

Source: (Chanda *et al*., 2015 and Antony *et al*., 2016)

Growth and feed Efficiency were poor in fish receiving various diets without supplementary manganese, but when the diet groups received additional manganese there was an effective improvement. It also was determined that more than 15 mg Mn/kg diet was necessary to sustain normal growth in rainbow trout (Table 11) (Satoh *et al*., 1991).

Table 11. Effect of various supplementary manganese levels on growth, feed efficiency and appearance of cataract in rainbow trout after 26 weeks of feeding

Supplementary Mn (mg/kg)	Total Mn in diet (mg/kg)	Growth rate (%)	Feed efficiency	Cataract
0	5.1	3817	1.05	7
5	9.7	4021	1.07	7
10	14.4	4104	1.11	3
15	19.1	4296	1.05	0
20	23.9	4136	1.06	0

Source: (Satoh *et al*., 1991)

Zhang *et al*. (2016) found that Mn increases the Specific Growth rate (SGR) of Large Yellow croaker. Broken–line analysis of SGR showed that the requirement of dietary Mn for large yellow croaker was 16.44 mg/kg (Figure 6).

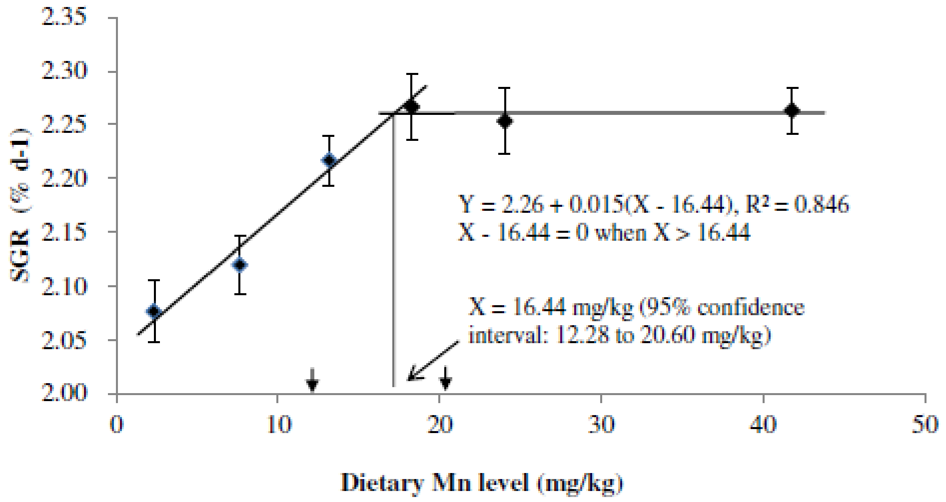


Figure 6. Relationship between specific growth rate (SGR) and dietary Mn levels based on a broken–line regression analysis, where X represents dietary Mn requirement of large yellow croaker.

4.3.3 Deficiency and Toxicity Signs

An inadequate supply of manganese usually results in retardation of growth (Table 12). Ogino and Yang, (1980) obtained poor growth in rainbow trout and carp when the diets were low in manganese. When young tilapia was deprived of manganese, Ishac and Dollar, (1968) observed reduced intake of food, loss of equilibrium, depressed growth and increased mortality. Other deficiency signs such as dwarfism linked to disturbances in bone formation and cataract of eye lens have been observed in rainbow trout and carp. A reduction in skeletal manganese content has been noted corresponding to insufficient dietary supply of that mineral (Satoh *et al*., 1983a, b, c). The importance of manganese has been recognized in broodstock nutrition. The manganese content of the diet influences its level and that of other trace elements in gonads (Satoh *et al*., 1987b). The absence of manganese in a fish meal diet significantly influenced the mineral composition of common carp gonads. The eggs produced by broodstock of brook trout and rainbow trout fed fish meal diets lacking manganese contained only low levels of this trace metal and subsequently hatchability was poor (Lall, 1989).

Table 12. Deficiency Signs of manganese in some fish

Species	**Deficiency Signs**
Common Carp, Tilapia and Rainbow trout	Reduced growth, Skeletal abnormalities, Cataracts, Dwarfism
Brook trout	Poor hatchability and low Mn level of eggs
Rainbow trout	Disturbance in bone metabolism, Low activity of Cu–Zn superoxide, dismutase and Mn superoxide dismutase in cardiac muscle and liver,
Tilapia	Reduced intake of food, loss of equilibrium, depressed growth and increased mortality

Source: (Watnabe *et al*., 1997 and Chanda *et al*., 2015)

3.4 Selenium (Se)

Selenium has been found to be an essential trace element for all animals studied, including fish. Selenium is a component of the enzyme glutathione peroxidase (Rotruck *et al*., 1973). This enzyme catalyzes reactions necessary for the conversion of hydrogen peroxide and fatty acid hydroperoxides into water and fatty acid alcohols by using reduced glutathione, thereby protecting cell membranes against oxidative damage (NRC, 1993). Selenium compounds are also capable of protecting heavy metal toxicity like cadmium and mercury (Watnabe *et al.*, 1997).

3.4.1 Availability

Fish derive selenium from both water and diet. Fishmeal and marine by–products are the rich sources of Selenium to fish. Plant materials vary widely in their Selenium content. Selenium is available from various feed components and other selenium–containing compounds. Selenite and selenate are inorganic selenium compounds, whereas selenomethionine, selenium–methylselenomethionine, selenocystine and selenocysteine are organic complexes. Fish meal and marine by–products employed as feed ingredients provide adequate selenium to fish. Plant materials vary widely in their selenium content. Generally, the availability of selenium from fish meals has been considerably lower than from plant derived products (Hodson and Hilton, 1983).

3.4.2 Dietary Requirement

Fish derive selenium from both diet and water. High levels of Selenium (40 – 130 µg/l) in water are toxic to fish. Han *et al.,* (2011) reported that the dietary selenium requirement is 1.18 mg/kg in case of Gibel carp (*Carassius auratus gibelio*) (Watnabe *et al*., 1997). Selenium requirements are given in Table 13.

Table 13. Dietary requirement of selenium in some fishes

Fish Species	Requirements (mg/kg diet)
Channel catfish	0.25
Gibel carp	1.18
Atlantic Salmon	0.14
Rainbow trout	0.15–0.38

Source: Antony *et al*., 2016

The Se concentration in cobia fresh liver progressively increased with increasing concentration of dietary Se within the range of 1.33–2.23mg/kg. The Se concentration of the whole body increased significantly with increasing levels of dietary Se. Based on linear regression of wholebody Se concentration, a minimum dietary requirement for Se was estimated to be 0.811mg/kg diet (Figure 7) (Liu *et al*., 2010).

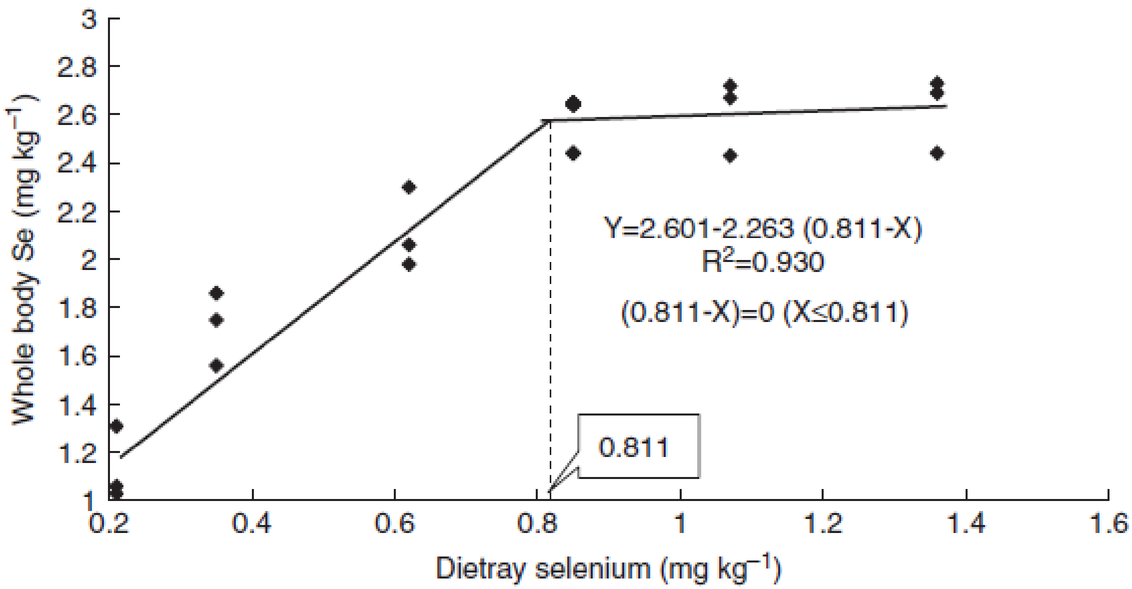


Figure 7. Relationship between dietary selenium level and whole–body selenium of cobia fed the six diets for10 weeks.

(Source: Liu *et al*., 2010)

Broken–line linear regression analysis of WG (Figure 8) indicated the requirement level of Se level for the maximal growth of juvenile Nile tilapia could be 1.06 mg/kg Se diet (Lee *et al*., 2016).

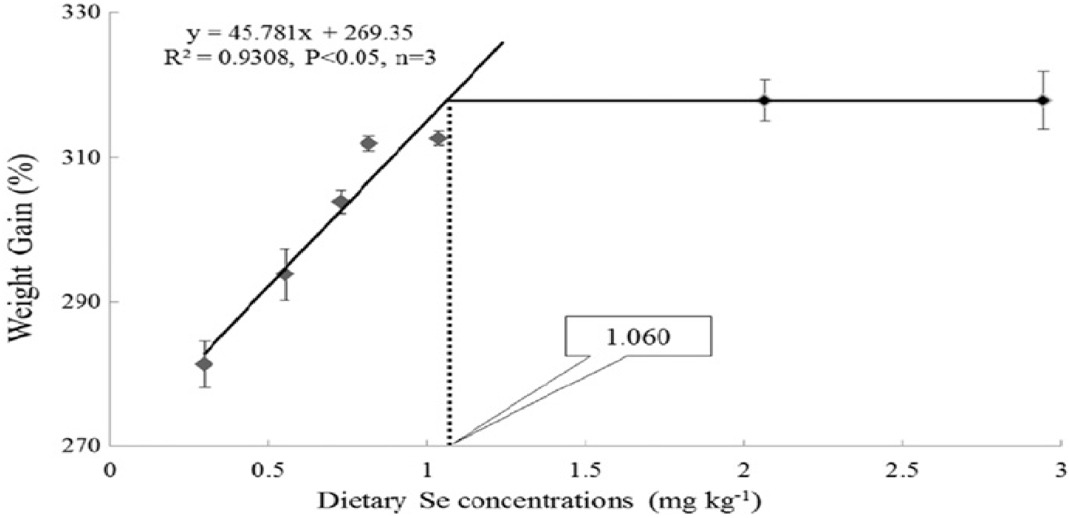


Figure 8. Broken–line regression analysis of weight gains juvenile nile tilapia, *Oreochromis niloticus*, fed different levels of organic Se for 10 weeks.

(Source: Lee *et al*., 2016)

Deficiency signs

Selenium deficiency results growth depression, loss of appetite, lethargy, reduced muscle tone and mortality in Atlantic salmon (Poston and Combs, 1979). Deficiency of selenium and vitamin E are the probable factors responsible for Hitra disease in farmed salmon (Poppe *et al.,* 1986). Apart from the deficiency symptoms, it is observed that a reduction in tissue concentration of vitamin E and selenium, low haematocrit values and increased haemolytic rates (Watnabe *et al.,* 1997) (Table 14).

Table 14. Deficiency signs of selenium in some fish

Species	**Deficiency Signs**
Channel catfish	Growth depression
Common Carp	Growth depression
Rainbow trout	Growth depression, Exudative diathesis
Atlantic Salmon	Muscular dystrophy

Source: (Watnabe *et al*., 1997 and Chanda *et al*., 2015)

CHAPTER IV
CONCLUSION

- Trace minerals are essential for fish and is involved in the normal metabolism and life processes. The minerals are required in extremely small quantity in the diet but excess supplementation through a continuous process can cause severe toxicity. The trace minerals requirement in fish are; Iron 30-170, Copper 3-5, Manganese 2-20, Zinc 15-70, and Selenium 0.15 to 2 (mg/kg diet).
- In deficient condition, growth is retarded with abnormal metabolism in particular. It is also a matter of concern about the potential causes of conditioned trace element deficiencies such as food processing methods, dietary interactions, disease conditions and genetic disorders. Fish can absorb part of the required minerals directly from the water through gills or even through their entire body surface. The minerals absorbed from water do not meet the total requirement and a certain supplementation through the diet is required whether in the natural food or supplementary feed. It should be taken care of so that specific diet could be composed with supplemented trace minerals to meet up the need. These approaches should highlight future research and potential strategies to maximize the activity on the requirement of trace mineral nutrition in fish. Once this requirement is met, the normal growth, survival and essential cellular metabolism may be ensured.

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