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Dietary Mineral Requirements of Fish and Marine Crustaceans*

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ABSTRACT: Information concerning mineral nutrition of fish and marine crustaceans is rather limited compared to that available for terrestrial animals; however, significant advances have been made in recent years. This article summarizes pertinent information about mineral nutrition of fish and marine crustaceans, including classification and general functions of the minerals; dietary essentiality and/or quantitative requirements; bioavailability and dietary interactions; and general recommendations for dietary supplementation. Dietary requirements for approximately ten minerals (calcium, copper, iodine, iron, magnesium, manganese, phosphorus, potassium, selenium, and zinc) have been identified for fish, and seven minerals (calcium, copper, phosphorus, potassium, magnesium, selenium, and zinc) have been recommended for inclusion in penaeid shrimp and lobster feeds. Based on current literature, it is clear that when evaluating the dietary essentiality of a mineral, not only growth must be evaluated, but also tissue stores, biochemical indicators (e.g., enzyme reaction rates), and effects on the general development of the animal should be assessed. Due to limited studies concerning dietary mineral requirements of marine fish and crustaceans, methodologies and requirement estimates for other species are recommended as guides for research with marine fish and crustaceans.

KEY WORDS: minerals, diet, fish, shrimp.

I. INTRODUCTION TO MINERAL NUTRITION

A dietary source of 23 minerals has been demonstrated as essential in 1 or more animal species. These elements are divided into two groups: the 7 macrominerals (calcium, chlorine, magnesium, phosphorus, potassium, sodium, and sulfur) and 16 trace minerals (aluminum, arsenic, cobalt, chromium, copper, fluorine, iodine, iron, manganese, molybdenum, nickel, selenium, silicon, tin, vanadium, and zinc). For the

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chicken, 13 minerals have been identified as essential in the diet and 6 have been identified as producing beneficial effects (Scott et al., 1982). Thirteen minerals are required in the diet of most terrestrial animals and may be essential for aquatic animals. Among these minerals, eight are cations: calcium (Ca^{2+}), copper (Cu^{2+}), iron (Fe^{2+}), magnesium (Mg^{2+}), manganese (Mn^{2+}), potassium (K^{+}), sodium (Na^{+}), and zinc (Zn^{2+}); and five are anions or are usually found in anionic groupings: chloride (Cl^{-}), iodide (I^{-}), molybdate (MoO_4^{2-}), phosphate (PO_4^{3-}), and selenite (SeO_3^{2-}) (Scott et al., 1982).

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 more soluble minerals (calcium, phosphorus, sodium, potassium, and chloride) function in osmoregulation, acid-base balance, and in the production of membrane potentials.

With the exception of osmoregulation, the maintenance of osmotic balance between body fluids and the water in which the animal lives, the biochemical functions of minerals in aquatic species appear to be similar to those in terrestrial animals (Lovell, 1989). Freshwater species lose ions to the hypotonic environment and therefore suffer from hydration, whereas the reverse is true for marine species. Unlike terrestrial animals, which are primarily limited to a dietary source of minerals, aquatic animals may be able to utilize, to some extent, minerals dissolved in the water to meet physiological requirements. Calcium, copper, iron, magnesium, potassium, sodium, selenium, and zinc are generally derived from the water to satisfy part of the physiological requirements of fish (National Research Council, 1993). However, phosphates and sulfates are more effectively obtained from feed sources (Lall, 1989). Independent of the mode of intake, a deficiency will occur if the animal's physiological requirements are not met.

II. DIETARY ESSENTIALITY OF AND REQUIREMENTS FOR MINERALS BY FISH AND MARINE CRUSTACEANS

Ten minerals (calcium, phosphorus, magnesium, potassium, copper, iron, zinc, manganese, selenium, and iodine) have been identified as essential in the diet of fish (Tables 1 and 2), and seven minerals (calcium, copper, magnesium, phosphorus, potassium, selenium, and zinc) have been recommended for inclusion in penaeid shrimp and lobster feeds (Tables 3 and 4). The large discrepancy in knowledge of mineral nutrition between terrestrial and aquatic species illustrates the relative infancy of aquatic animal nutrition and the related difficulty of conducting research on mineral nutrition of aquatic species. Problems associated with the quantification of mineral requirements include identification of the potential contribution of minerals from the water; leaching of mineral from the feeds prior to consumption; availability of suitable test diets; limited data on mineral bioavailability and the

TABLE 1
Macromineral Requirements of Fish

Mineral	Species	Recommended supplement (g/100 g diet)	Rearing condition[a]	Ref.
Calcium	*Ictalurus punctatus*	1.5	FW Practical diet	Andrews et al., 1973
		Dispensable	FW (14 mg Ca/l)[b]	Lovell, 1978
		0.45	FW (Ca free)	Robinson et al., 1986
	Tilapia aureus	0.17–0.65	FW (Ca Free)	Robinson et al., 1984
		0.7	FW (Ca Free)	Robinson et al., 1987
	Chrysophrys major	Dispensable	SW	Sakamoto and Yone, 1976a
	Cyprinus carpio	Dispensable	FW (20 mg Ca/l)	Ogino and Takeda, 1976
	Oncorhynchus mykiss	Dispensable	FW (20–23 mg Ca/l)	Ogino and Takeda, 1978
	O. keta	Dispensable	FW (20 mg Ca/l)	Watanabe et al., 1980
	Poecilia reticulata	Dispensable	FW (40 mg Ca/l; 0.6 mg P/l)	Shim and Ho, 1989
Phosphorus	*I. punctatus*	0.80	FW Practical diet	Andrews et al., 1973
		0.45	FW (0.03 mg P/l)	Lovell, 1978
		0.33[c]	FW (0.04 mg P/l)	Wilson et al., 1982
	C. carpio	0.6–0.7	FW (0.002 mg P/l)	Ogino and Takeda, 1976
	T. aureus	0.50	FW (Ca-free)	Robinson et al., 1987
	O. mykiss	0.7–0.8	FW (0.002 mg P/l)	Ogino and Takeda, 1978
	O. keta	0.5–0.6	FW (0.002 mg P/l)	Watanabe et al., 1980
	Salmo salar	0.6	FW (<0.5 mg P/l) 0.7% dietary P from plant sources	Ketola, 1975
	Poecilia reticulata	0.53–1.23	FW (40 mg Ca/l; 0.6 mg P/l)	Shim and Ho, 1989
	Sunshine bass (*Morone chrysops* × *M. saxatilis*)	0.5	FW (150 mg $CaCO_3$)	Brown et al., 1993
	C. major	0.68	SW	Sakamoto and Yone, 1978a
	Sciaenops ocellatus	0.86	BW (5–6 ppt)	Davis and Robinson, 1987

TABLE 1 (continued)
Macromineral Requirements of Fish

Mineral	Species	Recommended supplement (g/100 g diet)	Rearing condition[a]	Ref.
Ca:P ratio	*I. punctatus*	1.5:0.8	FW Practical diet	Andrews et al., 1973
		No relationship	FW (14 mg Ca/l)	Lovell, 1978
	C. major	0.34:0.68	SW	Sakamoto and Yone, 1973
		No relationship	SW	Sakamoto and Yone, 1976a
	C. carpio	No relationship	FW (20 mg Ca/l)	Ogino and Takeda, 1976
	O. mykiss	No relationship	FW (20–23 mg Ca/l)	Ogino and Takeda, 1978
	O. keta	No relationship	FW (20 mg Ca/l)	Watanabe et al., 1980
Potassium	*O. Ishawytscha*	0.6–1.2	FW No replication	Shearer, 1988
	I. punctatus	0.26	FW (4 mg K/l)	Wilson and El Naggar, 1992
	C. major	Dispensable	SW	Sakamoto and Yone, 1978b
Sodium/ chloride	*I. punctatus*	Dispensable	FW Practical diet	Murray and Andrews, 1979
	O. mykiss	Dispensable	FW Levels up to 11.6% produced no adverse effects	Salmon and Eddy, 1988
	S. ocellatus	2.0	FW and BW (6 ppt)	Gatlin et al., 1992
		Dispensable	35 ppt	
Sodium	*C. major*	Dispensable	SW	Sakamoto and Yone, 1978b
Magnesium	*I. punctatus*	0.04	FW (1.6 mg Mg/l)	Gatlin et al., 1982
	P. reticulata	0.054	FW (2.07 mg Mg/l)	Shim and Ng, 1988
	O. mykiss	0.06–0.07	FW (3.1 ppm Mg)	Ogino et al., 1978
		0.05	FW (0.05 mmol Mg/l	Knox et al., 1981
			1.2 mg Mg/l	
		0.06	FW (1.3 mg Mg/l)	Shearer, 1989
	T. niloticus	0.059–0.077	FW (1.0 mg Mg/l)	DaBrowska et al., 1989
	T. mossambicus	Dispensable	FW	Velden et al., 1991
	T. aureus	0.023	FW	Reigh et al., 1991
	C. carpio	0.06	FW (9.4 mg Mg/l)	Dabrowska et al., 1991
	C. major	Dispensable	SW (0.012% basal diet)	Sakamoto and Yone, 1979a

[a] *Freshwater (FW); saltwater (SW); brackish water (BW).*
[b] *( ) Indicate concentration in rearing water.*
[c] *Apparent available phosphorus.*

TABLE 2
Trace Mineral Requirements of Fish

Mineral	Species	Recommended supplement (mg/kg diet)	Rearing condition[a]	Ref.
Copper	*Ictalurus punctatus*	1.5	FW	Murai et al., 1981
		5	FW	Gatlin and Wilson, 1986b
	Cyprinus carpio	3	FW	Ogino and Yang, 1980
	Oncorhynchus mykiss	3	FW	Ogino and Yang, 1980
		3.5	FW	Julshamn et al., 1988
Iron	*I. punctatus*	30	FW (0.43 mg Fe/l)[b]	Gatlin and Wilson, 1986a
	C. carpio	199[c]	FW	Sakamoto and Yone, 1978d
	Chrysophrys major	199[c]	SW	Sakamoto and Yone, 1976b
		150	SW	Sakamoto and Yone, 1978c
Zinc	*I. punctatus*	20	FW (25 μg Zn/l)	Gatlin and Wilson, 1983
		150	FW with 1.1% phytate	Gatlin and Wilson, 1984c
	C. carpio	15–30	FW (10 μg Zn/l)	Ogino and Yang, 1979
	O. mykiss	15–30	FW (11 μg Zn/l)	Ogino and Yang, 1978
		20–40	FW	Satoh et al., 1987
		40	4% Tricalcium phosphate	
		80	7% Tricalcium phosphate	
	Tilapia aureus	20	FW (4 μg Zn/l)	McClain and Gatlin, 1988
	T. niloticus	30	FW	Eid and Ghonim, 1994
	Sciaenops ocellatus	20	BW (6 ppt)	Gatlin et al., 1991
Manganese	*I. punctalus*	2.4	FW (2 μg Mn/l)	Gatlin and Wilson, 1984b
	C. carpio	12–13	FW	Ogino and Yang, 1980
	O. mykiss	12–13	FW	Ogino and Yang, 1980
	T. mossambica	1.7 mg Mn/kg	FW	Ishac and Dollar, 1968
Selenium	*I. punctatus*	0.25	FW Adequate Vitamin E	Gatlin and Wilson, 1984a

TABLE 2 (continued)

Trace Mineral Requirements of Fish

Mineral	Species	Recommended supplement (mg/kg diet)	Rearing condition[a]	Ref.
	O. mykiss	0.15–0.38	FW (0.4 µg Se/l) 0.4 IU E/g diet	Hilton et al., 1980
		0.07	Prevented frank deficiency	Poston et al., 1976
		3	May produce toxicity	
		13	Toxic	
Iodine	*O. tshawytscha*	0.6–1.1	FW (0.2 µg I/l)	Woodall and LaRoche, 1964

[a] *Freshwater (FW); saltwater (SW); brackish water (BW).*

[b] *( ) Indicate concentration in rearing water.*

[c] *1 and 199 mg Fe/kg diet tested.*

TABLE 3
Macromineral Requirements of Penaeid Shrimp and Lobster

Mineral	Species	Recommended supplement (g/100 g diet)	Ref.
Calcium	*Penaeus japonicus*	Dispensable	Deshimaru and Yone, 1978
		1.2	Kitabayashi et al., 1971
		1.0–2.0	Kanazawa et al., 1984
	P. vannamei	Dispensable	Davis et al., 1993c
Phosphorus	*P. japonicus*	2.0	Deshimaru and Yone, 1978
		1.0–2.0	Kanazawa et al., 1984
		1.0	Kitabayashi et al., 1971
		1% Ca, 0.75	Civera, 1989
	P. vannamei	0.03% Ca, ≤0.34[a]	Davis et al., 1993c
		1% Ca, 0.5–1.0	
		2% Ca, 1.0–2.0	
Ca:P Ratio	*Homarus americanus*		
	Juvenile	0.56:1.10	Gallagher et al., 1978
	Adult	1:1	Gallagher et al., 1982
	P. japonicus	1:1	Kanazawa et al., 1984
		1.24:1.04	Kitabayashi et al., 1971
	P. californiensis	2.06:1	Huner and Colvin, 1977
		≤2.42:1	
	P. vannamei	Poor correlation	Davis et al., 1993c
Potassium	*P. japonicus*	1.0	Deshimaru and Yone, 1978
		0.9	Kanazawa et al., 1984
Magnesium	*P. japonicus*	Dispensable	Deshimaru and Yone, 1978
		0.3	Kanazawa et al., 1984

[a] *Level in the basal diet.*

nutritional requirements of many species. Despite these problems, mineral requirement data for a variety of species originating from diverse environments are becoming established.

A. MACROMINERALS

1. Calcium and Phosphorus

Calcium and phosphorus are two of the major constituents of the inorganic portion of feeds. Quantitatively, calcium and phosphorus function primarily as structural components of hard tissues (e.g., bone, exoskeleton, scales, and teeth). In addition to its structural functions, calcium is essential for blood clotting (vertebrates), muscle function, proper nerve impulse transmission, osmoregulation, and as a cofactor for enzymatic processes (National Research Council, 1993). Phosphorus is a component of a variety of organic phosphates, such as nucleotides, phospholipids, coenzymes, deoxyribonucleic acid (DNA), and ribonucleic acid. Inorganic phosphates also serve as important buffers to maintain normal pH of intra- and extracellular fluids (Zubay, 1983).

TABLE 4
Trace Mineral Requirements of Penaeid Shrimp and Lobster

Mineral	Species	Recommended supplement (mg/kg diet)	Ref.
Copper	*Penaeus japonicus*	Dispensable	Kanazawa et al., 1984
	P. orientalis	53	Liu et al., 1990
	P. vannamei	16–32	Davis et al., 1993a
Iron	*P. japonicus*	Dispensable	Deshimaru and Yone, 1978
		Dispensable	Kanazawa et al., 1984
	P. vannamei	Dispensable	Davis et al., 1992b
Manganese	*P. japonicus*	Dispensable	Kanazawa et al., 1984
Zinc	*P. vannamei*	15 (32 total)[a]	Davis et al., 1993b
		200[b] (218 total)[a]	
Selenium	*P. vannamei*	0.2–0.4	Davis, 1990

[a] *Total level in the diet.*
[b] *With 1.5% phytate supplemented to the diet.*

There are many factors influencing the absorption, distribution, and excretion of calcium and phosphorus. Dietary calcium is absorbed primarily from the intestine by active transport. In vertebrates, the vitamin 1,25-dihydroxycholecalciferol functions in the maintenance of serum calcium and phosphorus levels by altering the rate of intestinal absorption (via Ca^{2+}-binding protein), renal resorption, and bone mobilization (Zubay, 1983). There is some evidence that vitamin D and its metabolites may affect calcium homeostasis in teleosts. However, in a recent study (O'Connell and Gatlin, 1994), dietary supplementation of cholecalciferol (vitamin D_3) was not required for blue tilapia, *Tilapia aureus*, to utilize dietary calcium for growth and tissue mineralization in low-calcium water. The role of vitamin D_3 in intestinal calcium uptake of fish, therefore, is not well established as it is in terrestrial vertebrates.

Regardless of the form in which calcium and phosphorus are ingested, their absorption is dependent on solubility at the point of contact with the absorbing membranes; hence, mineral sources that are soluble at luminal pH are potentially more available. The National Research Council (1983) summarized the relative availability or apparent absorption of various sources of phosphorus for four species of fish. In general, bioavailability of phosphorus (and minerals in general) has been found to be positively correlated with the solubility of the mineral in water. Monobasic phosphates of sodium and potassium are highly available (90 to 95%) sources of phosphorus for channel catfish, *Ictalurus punctatus* (Lovell, 1978); common carp, *Cyprinus carpio* (Ogino et al., 1979); red sea bream, *Chrysophrys major* (Sakamoto and Yone, 1979b); and rainbow trout, *Oncorhynchus mykiss* (Ogino et al., 1979). Di- and tribasic phosphates are highly available to red sea bream (Sakamoto and Yone, 1979b). Dibasic calcium phosphate has an availability of 65% for the channel catfish (Lovell, 1989), and di- and tribasic calcium phosphates have availability values of 71 and 64%, respectively, for rainbow trout (Ogino et al., 1979). For common carp, the bioavailability of phosphorus from different sources is more variable; tribasic calcium phosphate has an availability of only 13%, dibasic calcium

phosphate has an availability of 46%, and monobasic calcium phosphate is 94% available (Ogino et al., 1979). In the presence of adequate phosphorus, Nakamuara and Yamada (1980) determined the availability of calcium for common carp to be 58, 37, and 27% from calcium lactate, tribasic calcium phosphate, and calcium carbonate, respectively. The differences in calcium availability were influenced by the lack of acidic digestion in common carp.

The bioavailability of phosphorus to marine shrimp appears to be similar to that of the carp. Apparent phosphorus availability values for *Penaeus vannamei* include calcium phosphate monobasic, 46%; calcium phosphate dibasic, 19%; calcium phosphate tribasic, 10%; potassium phosphate monobasic, 68%; sodium phosphate monobasic, 68% (Davis and Arnold, 1994).

In addition to chemical form (solubility) affecting mineral availability, other nutrients affect the availability of certain minerals. For example, lactose may interact with absorptive cells to increase their permeability to calcium ions; large intakes of iron, aluminum, and magnesium may interfere with the absorption of phosphorus by forming insoluble phosphates, and fats may interact with calcium to form insoluble soaps (Maynard et al., 1979). Hence, availability of calcium and phosphorus may be dependent upon the mineral form (solubility at intestinal pH), and dietary levels of calcium, phosphorus, vitamin D, iron, aluminum, manganese, potassium, magnesium, and fat.

Phosphorus in fish meal exists mainly in the form of insoluble hydroxyapatite originating from hard tissues such as bones. It has been shown (Watanabe et al., 1988) that the availability of phosphorus contained in fish meal is fairly low for carp (10 to 33%) compared with rainbow trout (60 to 81%). Based on unpublished data, Akiyama and Dominy (1989) reported the apparent absorption of phosphorus from fish meat to be 46.5% for *P. vannamei*. Phytate phosphorus, which constitutes approximately 67% of the phosphorus in grains, also is poorly available to fish (Lovell, 1989) and shrimp (Civera et al., 1990; Davis et al., 1993b). Civera et al. (1990) found that the presence of phytate inhibited the availability of dietary calcium and phosphorus to *P. vannamei* and *P. japonicus*, presumably due to the formation of insoluble complexes in the digestive system. The apparent availability of phytate phosphorus was determined to be 47.3% for *P. japonicus* and 8.4% for *P. vannamei*. Similarly, Davis et al. (1993b) reported that the presence of 1.5% phytate inhibited the availability of dietary phosphorus and zinc to *P. vannamei*. It should be noted that phytate phosphorus can account for a considerable portion of the phosphorus in practical diet formulations. The treatment of plant proteins with phytase has been demonstrated to increase the availability of this form of phosphorus to trout (Ketola, 1994) and consequently may be utilized to increase the availability of phosphorus from this component of the diet.

Calcium requirements of crustacea have been reported (Kitabayashi et al., 1971; Gallagher et al., 1978; Kanazawa et al., 1984); however, recent work indicates that *P. vannamei* raised in seawater does not require supplemental calcium (Davis et al., 1993c). Similarly, calcium deficiency in fish has been produced only in low-calcium water (Robinson et al., 1984, 1986, 1987). Although the diet plays an important role, it appears that both shrimp and fish can absorb some minerals from the water via drinking, and by direct absorption via the gills, skin, or both (Deshimaru et al., 1978; National Research Council, 1993). The calcium requirement of channel catfish may be totally or partially satisfied through absorption of calcium from the water;

however, the dietary calcium source and level can influence growth and bone mineralization of channel catfish, even in water containing adequate levels of calcium (Gatlin and Scarpa, 1991). Due to the low concentration of phosphorus in natural waters (Boyd, 1981), absorption of significant amounts of phosphorus from fresh- and saltwater is unlikely (Lall, 1991), making a dietary source of phosphorus potentially more critical for both fish and shrimp.

Phosphorus requirements of 1% (Kitabayashi et al., 1971), 1 to 2% (Kanazawa et al., 1984), and 2% (Deshimaru and Yone, 1978) of the diet have been recommended for shrimp. Davis et al. (1993c) demonstrated that the dietary phosphorus requirement of *P. vannamei* was dependent upon the calcium content of the diet and that in the absence of calcium supplements, the basal diet (0.03% Ca, 0.34% P) contained adequate phosphorus for normal growth. The dietary phosphorus requirements of fish species range from 0.3 to 0.9% of the diet (Table 1). The large disparity between dietary requirements of fish and shrimp may indicate that dietary phosphorus requirements of shrimp have been overestimated.

Gallagher et al. (1978) examined the effects of varying the dietary calcium-to-phosphorus ratio (Ca:P) on juvenile lobsters (*Homarus americanus*). Based on growth and histological studies of the endocuticle, a Ca:P of 0.51 (0.56:1.10) was found to be best for lobster juveniles, whereas a Ca:P of 1.55 and greater resulted in abnormalities of the endocuticle. In *P. japonicus*, a dietary Ca:P of 1:1 has been recommended (Kitabayashi et al., 1971; Kanazawa et al., 1984). Davis et al. (1993c) reported that the supplementation of calcium to the basal diet appeared to inhibit phosphorus bioavailability and the Ca:P did not totally explain the inhibitory effects of calcium. Based on these studies, it appears that calcium may affect phosphorus availability and that calcium levels in excess of 2.5% should be avoided. Although there does not appear to be a fixed Ca:P ratio that will produce optimal results, it appears that a ratio < 2:1 (Ca:P) provides good results in commercial formulations.

Considering that there is a great deal of interest in the deleterious effects of phosphorus in the effluent waters of aquaculture facilities, care should be taken to minimize over supplementation of dietary phosphorus. The phosphorus released from uneaten food and excreta must be minimized because it stimulates algal growth. In order to achieve optimum growth and lower phosphorus output, nutritionists must select feed ingredients with high phosphorus bioavailability while supplying the least possible amount of phosphorus in the diet (Lall, 1991).

2. Sodium, Chloride, and Potassium

Sodium, chloride, and potassium are recognized as being essential for a number of physiological processes. Dietary deficiencies of sodium and chloride have not been demonstrated in fish (National Research Council, 1993). The supplementation of high levels (4.5 to 11.6% of the diet) of sodium chloride (NaCl) to the diet has been reported to inhibit feed efficiency of rainbow trout raised in freshwater, presumably due to nutrient dilution (Salman and Eddy, 1988). Additionally, there were no positive or negative effects on channel catfish raised in freshwater (Murray and Andrews, 1979) or Atlantic salmon (*Salmo salar*) raised in fresh- or seawater (Shaw et al., 1975) due to NaCl supplementation.

Recent work with the euryhaline red drum (*Sciaenops ocellatus*) has demonstrated that, at low salinities, the supplementation of NaCl to the diet resulted in

increased growth (Gatlin et al., 1992). One possible explanation for this positive response is an increase in amino acid absorption. Boge et al. (1983) demonstrated that, in addition to being energized by an Na^+-gradient, amino acid transport by brush-border membrane vesicles prepared from enterocytes of the sea bass (*Dicentrarchus labrax*) is dependent on the presence of Cl^- ions. The addition of dietary NaCl at low salinities may increase the absorption of amino acids and/or satisfy other metabolic requirements, thus resulting in a physiological advantage for some species. The supplementation of NaCl (7 to 10%) to practical diet formulations has also been found to increase survival of fish being transferred from fresh- to saltwater (Al-Amoudi, 1987; Zaugg et al., 1983), presumably through the stimulation of osmoregulatory function and gill sodium and potassium ATPase activity (Al-Amoudi, 1987).

A dietary potassium requirement has been identified for channel catfish (Wilson and El Naggar, 1992) and chinook salmon (*O. tshawytscha*) in freshwater (Shearer, 1988), but not for red sea bream in saltwater (Sakamoto and Yone, 1978b), possibly indicating that marine fish can obtain adequate levels of potassium from the water. Results with marine shrimp are less clear and limited by the relative infancy of this area of research in crustaceans. Kanazawa et al. (1984) reported that diets containing 0.9% potassium improved growth of *P. japonicus* compared with diets containing 1.8% potassium. Deshimaru and Yone (1978) recommended dietary supplementation of 1% potassium based on the comparative growth of shrimp fed a diet without magnesium and potassium supplementation; however, potential interactions between potassium and magnesium were not evaluated. Davis et al. (1992a) found that the individual deletion of potassium from a semipurified diet did not result in a significant depression in tissue potassium or growth; however, tissue levels of magnesium were affected, indicating a potential interaction. Consequently, additional research should be conducted with marine species to clearly define the dietary essentiality of potassium and its specific physiological effects.

Most freshwater and all seawater probably contain sufficient amounts of sodium, potassium, and chloride ions to satisfy the physiological needs of fish (Lovell, 1989). Those ions are found in substantial amounts in most feedstuffs, making the necessity of dietary supplementation unlikely.

3. Magnesium

In vertebrates, approximately 60% of total body magnesium is located in bone, in which about one third of it is combined with phosphate and the remainder is adsorbed loosely on the surface of the mineral structure (Pike and Brown, 1975). In soft tissues, magnesium occurs both intra- and extracellularly. Magnesium is essential for maintenance of intra- and extracellular homeostasis in fish (Moyle and Cech, 1982) and crustaceans (Mantel and Farmer, 1983). Magnesium is essential for cellular respiration and phosphate transfer reactions involving adenosine tri-, di-, and monophosphate. It is an activator for all thiamine pyrophosphate reactions and is involved in the metabolism of fats, carbohydrates, and proteins.

Dietary magnesium deficiencies have been documented for a variety of freshwater fishes (Table 1). Deficiency signs include poor growth, anorexia, lethargy, muscle flaccidity, convulsions, vertebral curvature, high mortality, and depressed magnesium levels in the whole-body, blood serum, and bone (Lovell, 1989). Fish

in freshwater, which contains 1 to 3 mg Mg/l, require 0.025 to 0.07% magnesium in the diet (Lovell, 1989). Seawater typically contains high levels of magnesium (1350 mg/l) and magnesium is excreted by marine crustacea and fish, resulting in blood levels lower than that of the external medium. Thus, marine species may not require a dietary source of magnesium (Dall and Moriarty, 1983).

Red sea bream reared in seawater showed no signs of deficiency when fed diets containing as little as 0.012% magnesium (Sakamoto and Yone, 1979a). Early research with *P. japonicus* indicated that supplementation of 0.3% magnesium did not improve the nutritive value of a semipurified diet (Deshimaru and Yone, 1978). Kanazawa et al. (1984) reevaluated the magnesium requirement of *P. japonicus* and reported that dietary supplementation of 0.1 to 0.5% magnesium improved the nutritive value of the diet. However, in this series of studies weight gain was very low (<100%) and a dietary essentiality was not established. Davis et al. (1992a) reported a depression in hepatopancreas magnesium levels of *P. vannamei* in response to the deletion of magnesium from a semipurified diet; however, weight gain (>1471%) and magnesium levels of the carapace were unaffected. Based on the high levels of magnesium in seawater and the magnesium requirements of freshwater fish, a dietary magnesium requirement for marine species would not be expected. However, due to the scarcity of studies concerning marine species, further investigation into the dietary requirements of marine fish and shrimp is required to establish the need for dietary magnesium supplementation. Because most feed ingredients, especially those of plant origin, are high in magnesium, supplementation of magnesium to practical diets is generally not necessary. Due to the low bioavailability of magnesium from white fish meals (Watanabe et al., 1988), some diets designed for freshwater species may be marginally deficient and may require magnesium supplementation.

B. MICROMINERALS

1. Copper

Copper functions in hematopoiesis and in numerous copper-dependent enzymes (O'Dell, 1976), including lysyl oxidase, cytochrome *c* oxidase (CCO), ferroxidase, tyrosinase, and superoxide dismutase (SOD). Lysyl oxidase functions in the formation of cross-links during the synthesis of collagen and elastin. The failure of collagen maturation (cross-linking) in the organic matrix of bone accounts for increased fragility of bones and the associated abnormalities of copper deficiencies (O'Dell, 1976). Failure of collagen and elastin cross-linking and an undefined muscular defect result in enlargement of the heart and cardiac failure in copper-deficient animals.

Copper is also involved in the absorption and metabolism of iron and functions in the formation of hemoglobin in vertebrates. Crustaceans utilize hemocyanin as the oxygen-carrying pigment. This copper-containing pigment has an analogous role to hemoglobin in red-blooded animals (Lovell, 1989). Depledge (1989) estimated that, on a fresh-weight basis, 40% of the whole-body copper load in shrimp is found in hemocyanin. This suggests a considerable increase in the physiological demand for copper by crustaceans above that required by vertebrates.

In addition to the physiological functions of copper, high levels of environmental copper have been found to be toxic to a variety of marine species (Bryan, 1976).

Copper contamination in the marine environment is generally due to an increase in anthropogenic input and occurs primarily in coastal and estuarine areas, where copper concentrations (up to 0.6 mg Cu/l) far exceed the background copper level of (0.5 µg Cu/l) in seawater (Bjerregaard and Vislie, 1986). Due to toxic effects of dissolved copper, shrimp hatcheries routinely use water that has been treated with ethylenediaminetetraacetic acid to chelate free copper (Lawrence et al., 1981). Although a great deal of research has been conducted on the toxicity of dissolved copper, the dietary essentiality of this nutrient for marine species has received little attention.

Dietary deficiencies of copper have been documented for several freshwater fish (Ogino and Yang, 1980; Murai et al., 1981; Gatlin and Wilson, 1986b; Julshamn et al., 1988), but the dietary essentiality of copper has not been evaluated in marine fish. Dietary requirements for copper range from 1.5 to 5 mg Cu/kg diet. In addition to growth and feed efficiency, copper-dependent enzymes, such as ceruloplasmin, copper- and zinc-dependent SOD, and CCO, have been shown to be excellent indicators of copper nutrition (Gatlin and Wilson, 1986b).

Kanazawa et al. (1984) found that the dual deletion of iron and copper had no significant effect on growth and survival of *P. japonicus*. However, in this series of experiments, percent weight gain was low (40%) and survival was poor (57%); hence, the nutritional stress or the quality of the diet may not have been adequate to induce a dietary deficiency. Davis et al. (1993a) demonstrated a dietary copper deficiency in *P. vannamei* fed semipurified diets containing <34 mg Cu/kg diet. Deficiency signs included poor growth; reduced copper levels in the carapace, hepatopancreas, and hemolymph; and enlargement of the heart. Similarly, based on growth, survival, CCO activity, and tissue mineralization, Liu et al. (1990) reported a dietary copper requirement of 53 mg Cu/kg diet for *P. orientalis*. These results indicate that shrimp cannot meet their physiological needs for copper from seawater and that a dietary source is required for maximum growth and tissue mineralization. This also indicates that species utilizing copper as a component of their respiratory pigment have an increased copper requirement over species utilizing iron-based respiratory pigments.

2. Iron

Iron is a trace element that is essential for the production and normal functioning of hemoglobin, myoglobin, cytochromes, and many other enzyme systems. In vertebrates, the principal role of iron is as a component of hemoglobin. Red blood cells are regenerated periodically and most of the iron is recycled. That which is not recycled is excreted via the bile into the intestine. Like other elements of low solubility, such as zinc and copper, iron is absorbed and transported in the body in a protein-bound form (Lovell, 1989). In vertebrates, mucosal transferrin binds Fe^{2+} in the intestinal lumen and transports it across the mucosal brush border. Within the cell, Fe^{3+} is bound to apoferritin forming ferritin. The amount of apoferritin in the mucosa is regulated by physiological needs for iron. The iron-bound transferrin is then transported in the blood, where the iron is again released at target tissues (liver and hematopoietic tissue). Iron and other minerals of low solubility are not easily excreted; thus, mineral

excesses are deposited in cells of the digestive system, which are then sloughed into the digestive tract for elimination.

In crustaceans, the hepatopancreas has been found to be the organ richest in iron. Storage cells containing iron have been reported in crayfish, *Procambarus clarkii* (Ogura, 1959; Miyawaki et al., 1961), and the crab, *Cancer irroratus* (Martin, 1973). Iron-transporting proteins have been found in the hemolymph of two species of crabs (Ghidalia et al., 1972; Depledge et al., 1986). These observations indicate the presence of a regulatory mechanism similar to that of vertebrates. In addition to the digestive system, gills appear to play an active role in iron metabolism. In *C. irroratus*, iron accumulates by forming a coating around the branchial lamellae during the intermolt cycle, which is then rejected at ecdysis along with the integument (Martin, 1973). Absorption of iron from the water through the gills could provide an additional source of iron.

Iron deficiencies have been documented for several species of fish (Table 2); however, dietary deficiencies for shrimp have not been observed (Deshimaru and Yone, 1978; Kanazawa et al., 1984; Davis et al., 1992b). Iron deficiency causing hypochromic microcytic anemia has been reported for freshwater fish, such as the brook trout, *Salvelinus fontinalis* (Kawatsu, 1972) and common carp (Sakamoto and Yone, 1978d), and in marine fish, such as the red sea bream (Sakamoto and Yone, 1976b) and the yellowtail, *Seriola quinqueradiata* (Ikeda et al., 1973). However, growth depression was not observed in these iron-deficient fish. Gatlin and Wilson (1986a) characterized the iron-deficiency signs of channel catfish and found that fish fed the basal diet (9.6 mg Fe/kg) exhibited suppressed growth and feed efficiency, as well as reduced hemoglobin, hematocrit, plasma iron, transferrin saturation, and erythrocyte-count values. The authors concluded that a minimum of 20 mg supplemental Fe/kg diet (30 mg total Fe/kg) was required by channel catfish for best growth and hematological values.

In addition to physiological problems caused by iron deficiency, excessive levels of iron can be toxic as well. Excessive iron supplementation appears to have potentially adverse effects on growth of *P. japonicus* (Deshimaru and Yone, 1978; Kanazawa et al., 1984). Additionally, iron-catalyzed lipid oxidation increases with iron supplementation, which in turn adversely affects feed stability (Desjardins et al., 1987). Iron is one of the primary metals involved in lipid oxidation and ferrous iron is a more potent catalyst of lipid peroxidation than ferric iron (Chvapil et al., 1974; Lee et al., 1981). Ferrous iron catalyzes the formation of hydroperoxides and free radical peroxides by providing a free radical initiator in the presence of unsaturated fatty acids and oxygen. Marine fish and crustacean diets contain primarily polyunsaturated fats; therefore, supplementation of ferrous iron to the diet could be expected to affect the stability of the diet through increased lipid oxidation (rancidity) and reduced stability of ascorbic acid (Hilton, 1989). Consequently, excessive supplements and restrictions of ingredients with high levels of iron are recommended in practical diet formulations. Although many practical diets may contain considerable levels of endogenous iron, little is known about its form and availability (Lall, 1989). Hence, a low level of supplementation (ca. 10% of dietary requirement) of an available source is often recommended to ensure adequacy of the diet.

3. Iodine

Iodine is an essential element for a variety of animals. Nearly every cell in the body contains iodine; however, in vertebrates the thyroid gland is the main location of iodine reserves. Thyroid hormones, which contain iodine, are known to have roles in thermoregulation, intermediary metabolism, reproduction, growth and development, hematopoiesis and circulation, as well as neuromuscular functioning (National Research Council, 1980). The minimum dietary iodine requirement of fish has not been defined; however, 1 to 5 mg I/kg diet has been found to be adequate (National Research Council, 1993). The physiological essentiality of iodine has not been evaluated in shrimp.

4. Manganese

Manganese functions as a cofactor in several enzyme systems, including those involved in urea synthesis from ammonia, amino acid metabolism, fatty acid metabolism, and glucose oxidation (National Research Council, 1980). Principal signs of manganese deficiency in terrestrial species include reduced growth rate, skeletal abnormalities, convulsions, reduced righting ability, abnormal reproductive function in males and females, and ataxia in the newborn (National Research Council, 1980).

Dietary deficiencies in fish have resulted in poor growth, skeletal abnormalities, high embryo mortalities, and poor hatch rates (National Research Council, 1993). A total dietary manganese content of 12 to 13 mg/kg has been recommended for the common carp and rainbow trout (Ogino and Yang, 1980); however, Gatlin and Wilson (1984b) found that 2.4 mg Mn/kg diet was sufficient for normal growth and health of channel catfish. Kanazawa et al. (1984) found that supplementation of 10 and 100 mg Mn/kg diet did not improve the growth of *P. japonicus*. However, it should be noted that percent weight gain during the study was <70% and the nutritional stress placed on the shrimp would not be considered adequate to reduce body stores enough to induce a deficiency. Because the manganese content of seawater is very low (0.01 mg/l), significant absorption from the water is unlikely. Thus, a dietary source of manganese could be necessary for marine shrimp and fish.

5. Selenium

Selenium is a trace element that functions as a component of the enzyme glutathione peroxidase, which converts hydrogen peroxide and lipid hydroperoxides into water and lipid alcohols, respectively. Thus, this enzyme functions in protecting the cell from deleterious effects of peroxides (Little et al., 1970). Glutathione peroxidase acts along with vitamin E to function as a biological antioxidant to protect polyunsaturated phospholipids in cellular and subcellular membranes from peroxidative damage (Lovell, 1989). In addition to its enzymatic functions, selenium helps protect against mercury toxicosis by forming a mercuric-selenium complex. This protein-bound complex is diverted from the kidney (where inorganic mercury devoid of selenium is deposited) to the liver and spleen, where its toxicity is considerably reduced (National Research Council, 1980).

Complementary functions of selenium and vitamin E may allow these nutrients to interact physiologically (Gatlin et al., 1986). Selenium and vitamin E interrelationships have been investigated in several animal species, and a variety of common and unique deficiency signs have been described (National Research Council, 1993). Differing responses, especially with respect to gross deficiency signs, were observed when Atlantic salmon (Poston et al., 1976), rainbow trout (Bell et al., 1985), and channel catfish (Gatlin et al., 1986) were fed diets without supplemental selenium, vitamin E, or both nutrients.

Levels of 0.15 to 0.38 mg Se/kg diet (Hilton et al., 1980) and 0.25 mg Se/kg diet (Gatlin and Wilson, 1984a) were required to provide maximum growth and glutathione peroxidase activity in rainbow trout and channel catfish, respectively. In zooplanktonic daphnids, a selenium deficiency in the medium resulted in cuticle deformation and a depression in reproduction. In the presence of replete zinc, 1 ppb Se was adequate (Keating and Dagbusan, 1984); however, in the absence of detectable zinc, 5 ppb Se in the medium was required to eliminate deficiencies characteristic of selenium deprivation (Keating and Caffrey, 1989). Davis (1990) found that juvenile *P. vannamei* grew best when fed semipurified diets supplemented with 0.2 to 0.4 mg Se/kg diet. Although a specific level has not been quantified, it appears that shrimp have a dietary requirement for selenium.

Practical diets containing >15% fish meal should contain adequate selenium and should not require supplementation, although selenium content and availability have been shown to vary considerably among different feedstuffs. Diets formulated with predominantly plant ingredients may require a selenium supplement. Due to the very toxic nature of sodium selenite or sodium selenate, they should not be handled in the pure form. A selenium premix, which usually contains 0.06 g Se/100 g, can be purchased for inclusion in the feed. Selenium supplementation is regulated by the United States Food and Drug Administration and supplemental selenium at 0.1 mg/kg is approved for minor-use animals such as fish and crustaceans.

6. Zinc

Zinc is required for normal growth, development, and function in all animal species that have been studied (National Research Council, 1980). Zinc functions as a cofactor in several enzyme systems and is a component of a large number of metalloenzymes, including carbonic anhydrase, carboxypeptidases A and B, alcohol dehydrogenase, glutamic dehydrogenase, D-glyceraldehyde-3-phosphate dehydrogenase, lactate dehydrogenase, malic dehydrogenase, alkaline phosphatase, aldolase, SOD, ribonuclease, and DNA polymerase (National Research Council, 1980).

A dietary requirement for zinc has been quantified for a variety of freshwater fishes fed semipurified diets: 20 mg Zn/kg diet for channel catfish (Gatlin and Wilson, 1983) and blue tilapia, *T. aureus* (McClain and Gatlin, 1988); 15 to 30 mg Zn/kg diet for carp (Ogino and Yang, 1979); and 15 to 30 mg Zn/kg diet for rainbow trout (Ogino and Yang, 1978). In daphnids reared under controlled trace-element exposure, utilizing a controlled media system (Keating, 1985), the absence of detectable zinc resulted in decreased lifespan and increased demand on the organism's pool of available selenium (Keating and Caffrey, 1989). The marine shrimp,

P. vannamei, has been found to require 33 mg Zn/kg diet to maintain normal tissue mineralization (Davis et al., 1993b). The zinc requirement of red drum also has been determined to be 20 mg Zn/kg diet (Gatlin et al., 1991).

Practical diets contain feedstuffs that are relatively rich sources of zinc (e.g., fish meal); however, for fish the bioavailability of zinc in feedstuffs is generally low, making supplementation essential (Watanabe et al., 1988). 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 (Ketola, 1979; Hardy and Shearer, 1985; Satoh et al., 1987).

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 terrestrial animals (Oberleas et al., 1962; O'Dell et al., 1964; Savage et al., 1964; Lo et al., 1981), fish (Gatlin and Wilson, 1984c; Richardson et al., 1985; McClain and Gatlin, 1988; Gatlin and Phillips, 1989; Satoh et al., 1989), and shrimp (Davis et al., 1993b). Consequently, practical diets are often supplemented at levels in excess (100 to 150 mg Zn/kg diet) of the dietary requirement to overcome the effects of inhibitory agents, such as phytate.

III. FACTORS AFFECTING BIOAVAILABILITY OF MINERALS

In order to meet an animal's physiological requirements for various minerals, dietary sources must be available. Numerous factors affect the availability of minerals. The most soluble, and consequently the most readily absorbed, forms are the simple state of the element or ionic group of atoms (e.g., Ca^{2+}, Mg^{2+}, Mn^{2+}, PO_4^{3-}). However, in nature, compounds differing in electric charge bind with the minerals, forming stable compounds that are less soluble in water. Although such compounds have low solubility in water, the acidic condition of the gastric stomach allows dissociation of the compounds into salts that can be easily absorbed in the intestine. Consequently, in animals without acidic digestive systems, the bioavailability of minerals is generally reduced.

Although the gastric stomach generally increases the availability of minerals, some minerals may interact after being released into the alkaline intestine by forming insoluble precipitates. Excessive levels of calcium and phosphorus react with magnesium and zinc to form insoluble precipitates. Additionally, colloids, such as particles of clay and insoluble salts of aluminum, magnesium, iron, and other elements, strongly absorb cations. This absorption occurs both through chemical union with highly electronegative areas of the colloidal surface and through attraction of the cation by physical forces (Scott et al., 1982). Consequently, the bioavailability of a given mineral will be dependent on the dissociation of the mineral as well as interactions with other dietary components.

With regard to the bioavailability of minerals, there has been a great deal of interest in chelated minerals. If an element is chelated by a compound that will

release it in ionic form at the site of absorption or will be readily absorbed as the intact chelate, this form may greatly enhance the absorption of the element by preventing its conversion to insoluble chemical compounds in the intestine or by preventing its strong adsorption in insoluble colloids (Scott et al., 1982). Chelated minerals may have potential as dietary supplements, yet research concerning those compounds with respect to aquatic species has been very limited. Compared with inorganic sources, chelated minerals are generally less sensitive to the inhibitory action of other compounds (i.e., phytate and fiber) and may have a higher bioavailability in practical diets. Consequently, chelated minerals are widely used in the livestock industry, and to a lesser extent in aquatic feeds, to satisfy a portion of dietary mineral requirements. As prices of chelated minerals decrease and the benefits of these compounds are better defined, their use will likely increase.

Of the feed ingredients used in practical animal diets, fish meal is the richest source of endogenous minerals. Research on the bioavailability of minerals contained in fish meals has demonstrated that there is considerable variation among species (perhaps due to luminal pH), and that the bioavailability of minerals is affected by meal type and ash content. Fish meals are relatively rich in minerals but low mineral availability, and inhibitory interactions in fish meals require supplementation of available sources of copper, phosphorus, magnesium, manganese, and zinc to prevent dietary deficiencies and maximize fish growth (Watanabe et al., 1988). As the aquatic animal feed industry increases its use of less expensive plant feedstuffs, which are generally poor sources of minerals and may contain factors that reduce the bioavailability of minerals, the need for mineral supplements should increase.

IV. GENERAL RECOMMENDATION

Information on the mineral requirements of aquatic organisms has expanded considerably but is still limiting for most species. Undoubtedly, as our understanding of mineral requirements expands along with establishment of species and environmental differences, our ability to tailor mineral delivery systems will become more precise. Currently, it is recommended that semipurified or purified research diets should contain replete mineral premixes and that environmental conditions under which these diets are used should be reported. A mineral premix that has produced good growth and survival of *P. vannamei* and a variety of fish species fed semipurified diets is presented in Table 5. Because practical diets will generally contain a substantial amount of endogenous minerals, a complete mineral premix may not be required. With the exception of phosphorus and magnesium (which may be marginally deficient), there is no evidence that the other macrominerals require supplementation in commercial diets. Indeed, some macrominerals should be minimized to prevent interactions with other nutrients (e.g., calcium) and excessive levels of pollutants (e.g., phosphorus). Due to low mineral bioavailability, deficient levels, and the presence of inhibiting agents, most practical diets will require supplementation with a highly available form of phosphorus and trace minerals. Although required supplementation will vary with diet formulation and environmental conditions, a generalized trace mineral premix for practical diets designed for fish and shrimp is presented in Table 6.

TABLE 5
Complete Mineral Premix for *P. vannamei* and Finfish Fed Semipurified Diets

	g/1000 g premix	
Ingredient	***P. vannamei*[a]**	**Finfish[b]**
Calcium phosphate monobasic	500.00	136.00
Calcium lactate	100.00	348.49
Magnesium sulfate heptahydrate	80.00	132.00
Potassium citrate	100.00	
Potassium phosphate monobasic	100.00	240.00
Sodium chloride	74.00	45.00
Aluminum chloride		0.15
Chromic potassium sulfate	0.55	
Cobalt chloride		1.00
Cupric sulfate pentahydrate	1.40	0.50
Ferrous sulfate heptahydrate	2.00	5.00
Manganous sulfate monohydrate	2.00	0.70
Potassium iodide	0.045	0.15
Sodium selenate	0.018	0.011
Zinc sulfate heptahydrate	22.00	3.00
Filler	17.987	87.999

[a] *Mineral premix at 4% inclusion will provide (as g/100 g diet): Ca, 0.42; Cl, 0.18; K, 0.33; Na, 0.11; P, 0.60; (as mg/kg diet) Cr, 4.0; Cu, 22.3; Fe, 16.0; Mg, 315.5; Mn, 26.0; I, 1.4; Zn, 200.1; Se, 0.3.*

[b] *Mineral premix at 4% inclusion will provide (as g/100 g diet): Ca, 0.27; Cl, 0.11; K, 0.43; Mg, 0.052; Na, 0.13; P, 0.38; (as mg/kg diet) Al, 1.2; Cu, 5.1; Fe, 40.2; Mn, 9.1; I, 4.6; Zn, 27; Se, 0.2.*

TABLE 6
Generalized Trace Mineral Premix for Finfish and Shrimp[a]

Ingredient[b]	**g/100 g premix**
Cobalt chloride	0.004
Cupric sulfate pentahydrate	0.250
Ferrous sulfate heptahydrate	4.000
Magnesium sulfate heptahydrate	28.398
Manganous sulfate monohydrate	0.650
Potassium iodide	0.067
Sodium selenite	0.010
Zinc sulfate heptahydrate	13.193
Filler	55.128

[a] *For shrimp, cupric sulfate pentahydrate = 0.550 g/100 g premix; ferrous sulfate = 2.00 g/100 g premix. Thus providing (as mg/kg diet) Cu, 10.95; Fe, 20.10.*

[b] *Trace mineral premix at 5% inclusion will provide (as mg/kg diet): Co, 0.050; Cu, 4.98; Fe, 40.18; Mg, 140; Mn, 10.57; Se, 0.211; I, 2.58; Zn, 150.*

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