5.16 Nutritional Requirements of Grouper (*Epinephelus* spp.)

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Abstract

This paper covered information on the nutritional requirements for various grouper species. It also provides information on the requirements of vitamin, mineral and also the essential fatty acid. Feed and feeding are also being described.

Introduction

Grouper is the most popular maricultured fish in Southeast Asia and live grouper command a high price in Hong Kong, China. The red grouper, *Epinephelus akaara* is a preferred species as it symbolizes good fortune [in Hong Kong] and is often served at wedding dinners (Tseng & Ho, 1988). *E. akaara* is a slow growing grouper in comparison to *E. tauvina*, which is widely cultivated in Asian countries.

Groupers are sluggish fish. In their natural habitat they normally rest in rocky areas or at the bottom of cages when they are cultured. In a culture environment, suspended material such as used automobile tyres increase the resting surface area. The sluggish nature of the grouper helps reduce feeding frequency.

Groupers can withstand salinity ranging from 15 to 45 ppt. They can also withstand washing with freshwater for longer than fifteen minutes. The optimum temperature range is 22-28°C. When the temperature is below 15°C, the fish do not eat. The grouper is euryphagous, and prefers crustaceans and live food (Randall, 1965).

Longly and Hildebrand (1941) reported that *E. mario* feed indifferently day or night. *E. akaara* favours feeding just before sunset. Cultured red groupers exhibit special feeding behaviour. The fish can be trained to know when they will be fed. When they sense the sounds of chopping of trash fish or knocking of a wood plank, they gather at the cage edge. As the fish have a suspicious nature, they keep a look out for food but do not move. However, if one of them approaches the food, all will immediately snatch at it, sometimes injuring themselves in the process. Groupers usually eat one to three pieces of minced trash fish and then swim away. They do not eat food which falls to the net bottom. Due to this special feeding behaviour, groupers are generally mix-cultured with sea breams who act as scavengers and stimulate groupers to feed.

Ingredient	Percent	
Fish meal	34	
Meat and bone meal	10	
Soybean meal	15	
Groundnut meal, expellar	10	
Ricebran, solvent extracted	10	
Leaf meal	5	
Таріоса	8	
Vitamin and mineral mixture	1	
Soybean or corn oil	4	
Squid or pollack liver oil	3	
BHT	0.02	
Ethoxyquin	0.02	

 Table 1: Composition of formulated diet for grouper E. tauvina and seabass Lates calcarifer (Kanazawa 1984)

E. tauvina is a hermaphrodite (gynandrous) fish. Males are generally absent in the smaller length-classes. The sexually mature males are more than 740 mm in standard length at the age of nine years or more. Chen et al. (1977) reported that three year old females could be transformed into males by an oral application of methyltestosterone.

Groupers are potentially important aquaculture species since they are fast growing, accept dry pellet feed, have high feed efficiency and have a very high economic value. A success of larval rearing and a success of spawning in captivity constitute crucial factors. Trash fish is the most common feed used in the cage culture of grouper. A high price, shortage of supply, variable quality, and poor feed conversion rate indicate that trash fish is neither nutritionally adequate nor economically suitable for grouper. It is important to develop a balanced and cheap diet for this fish.

Nutrient Requirements

As groupers are carnivorous, their dietary protein requirements are high. Teng et al. (1977) reported that a maximum weight gain by *E tauvina* was achieved at a dietary protein concentration of 50%. These researchers suggested that a dietary protein concentration of 40% was the most economical.

Sukhawongs et al. (1978) conducted experiments on two sizes of *E. tauvina* (20 g to 30 g, and 60 g to 70 g). They were fed diets with 30%, 40%, 45%, or 50% levels of protein. The best weight gain was observed at 50% dietary protein level. An optimum protein level of 45% for *E. tauvina* was also reported by E1-Dakour and George (1982). Teng (1979) found that 40% dietary protein was optimum for *E. salmonoides* (formerly cited as *E. tauvina*).

Wongsomnuk et al. (1978) observed a comparable trend for a higher protein requirement by smaller sized fish. Teng et al. (1978) determined the optimum dietary protein level for 65 g to170 g estuarine grouper (*E. salmonoides*) fed to satiation with moist diets having sun-dried tuna muscle as the protein source to be 40% on a dry matter basis. The dietary energy of the 40% protein diet was 3,302 kcal/kg on a dry weight basis calculated on an energy value of 3.9, 8.0 and 1.6 kcal/g for protein, lipid and carbohydrate respectively giving a P:E ratio of 121 mg protein/kcal. The optimum diet had a lipid content of 13.5% on a dry weight basis. Chen and Tsai (1994) reported that the dietary protein level that yielded maximum growth in *E. malabaricus* was 47.8% based on a broken-line model estimation of the weight gain results. Shiau and Lan (1996) fed diets with two dietary protein levels (50% and 44%) and four energy levels at each protein level (305, 340, 375, 410 kcal of GE per 100 g diet). The results show that when the energy level of a diet is maintained at 340-375 kcal per 100 g, the dietary protein level for juvenile grouper can be lowered from 50% to 44%.

Tucker (1991) reported that the optimum protein energy ratio for *E. tauvina* weighing 60-130 g was about 94 mg/kcal GE, and 142 and 162 mg/kcal DE for *E. malabaricus* and *E. striatus*, respectively. The data from Shiau and Lan (1996) suggested that the optimum P:E ratio for *E. malabaricus* was 117 mg to 129 mg/kcal GE. New (1987) reported that the optimum concentration of lipid in the grouper diet was about 14%.

Vitamin and Mineral Requirements

The ascorbic acid requirement has been studied in *E. tauvina* by Boonyaratpalin et al. (1993). A study was conducted to determine the effects of vitamin C in the form of Lascorbyl 2-phosphate-Mg on feeding rate, growth, feed efficiency, % hydroxyproline of protein, survival, and deficiency signs in juvenile E. malabaricus. A practical diet was supplemented with 0 mg, 30 mg, 60 mg and 100 mg of L-ascorbyl 2-phosphate-Mg per kg dry diet. The experiment was carried out for sixteen weeks in aquaria. The feeding rate, growth, feed efficiency, % hydroxyproline of protein and survival of fish fed with the diet without supplementary L-ascorbyl 2-phosphate-Mg were significantly lower (P<0.05) compared with other treatments. Fish fed a diet without supplementary L-ascorbyl 2-phosphate-Mg showed deficiency signs. The signs of vitamin C deficiency included loss of appetite, short snout, erosion of the opercula and fins, haemorrhaging eyes and fins, exophthalmia, swollen abdomen, abnormal skull, falling pharyngobranchials, severe emaciation, scoliosis and lordosis. The minimum level of Lascorbyl 2-phosphate-Mg required for normal growth was 30 mg/kg dry diet. Histological observation of the gills of scorbutic fish showed hyperplasia of epithelial cells of primary and secondary lamellae, fusion of secondary lamellae, gill cartilage distortion and detachment of the epithelium from the basal membrane of the secondary lamellae. By electron microscopy, degeneration of chloride cells, mucous cells and epithelium cells as well as hyperplasia and edema of epithelium cells were observed. (Phromkunthong et al, 1993)

E. malabaricus spawners were reared for nine months using three feed formulae. Formula 1(control) was fresh *Caranx*, Formula 2 was fresh *Caranx* supplemented with vitamin E at 200 mg/kg feed and Formula 3 was fresh *Caranx* supplemented with 400 mg vitamin E per kilogram of feed. After the study period, Formula 3 feed resulted in the highest percentage of eggs collected and was, therefore, considered to be the optimum feed for grouper propagation (Chulawittayanukul, 1986).

Essential Fatty acid

Ruangpanich and Boonliptanond (1993) determined the essential fatty acid requirement of grouper. Larvae between one and fifteen days of age fed essential fatty acids (n3 HUFA)-enriched rotifers showed no significant difference in survival compared to rotifers maintained in *Chlorella* as food source. This gives an indication that larvae were not deficient in essential fatty acids when given *Chlorella* rotifers. Therefore, rotifers should be maintained in *Chlorella* media for six hours before feeding fish larvae. This can increase the levels of essential fatty acids in rotifers up to 12%, which is considered optimum for the feeding of fish larvae (Watanabe, 1983, Ruangpanich and Boonliptanond, 1993).

When fifteen day-old larvae were fed brine shrimp without fatty acid supplementation, the fish began to show general body weakness and mortality starting at twenty-one days of age resulting in total mortality by thirty days. When larvae were fed brine shrimp enriched with fish oil at 25 or 50 ml/m³ of the rearing water, they showed normal growth. Some developed into juveniles at the age of thirty-seven days and all become juveniles by age fifty days. The survival rate for these two treatments was 50.5% and 56% respectively.

Pechmanee and Assavaaree (1993) reported that n-3 HUFA content of rotifers fed fish oil emulsified with raw egg yolk and a commercial emulsified oil was higher than that of rotifer fed *Chlorella* sp. After three to six hours of feeding, the n-3 HUFA content of rotifers fed fish oil, emulsified with raw egg yolk was 17.9% and 13.8% of the total lipid respectively, whereas the n-3 HUFA content in rotifers fed a commercial emulsified oil was 14.6% and 19.3% of total lipid respectively. It is suggested that rotifer should be enriched with n-3 HUFA before feeding grouper larvae after six days of age.

Feed and Feeding

The traditional grouper feed in floating net cages is trash fish, similar to that for seabass. Commercial, slow-sinking, extruded feed is available in some Asian countries. This commercial feed is believed to contain not less than 43% crude protein, not less than 6% fat, not more than 16% ash, 3% fiber and 12% moisture. A practical feed formulation based on availability of local feed ingredients as recommended by Kanazawa (1984) and Tacon et al. (1989) is presented in Tables 1 and 2, respectively.

Chua and Teng, (1978) conducted a three month study on the effect of feeding frequency on the growth of young *E. tauvina* being cultured in floating net cages. Seven feedings [to satiation] frequencies were studied: once every day, every two days, every three days, every four days, every five days, twice a day, and three times a day.

Optimal growth, good feed conversion ratio, and higher survival rates were observed in feeding to satiation once every two days. Weight gain was substantially lower in treatments involving once every three, four and five days. No enhancement in growth was observed when the frequency was increased to two or three feedings daily. The fact that feed conversion ratios were similar in fish fed to satiation with one feeding in five, four and two days suggests that feed intake is an important growth factor. Total feed intake was significant in fish fed once every two days. The intake of food was found to be closely related to the amount of food remaining in the stomach, with the intake being maximal when the stomach was empty. The food deprivation time of grouper is about thirty-six hours, at which time over 95% of the food is digested at a water temperature of 30 ± 1 °C. Hence, feeding the fish once every two days greatly enhanced intake and use of feed. Dawes (1930), Moore (1941) and Hickling (1962) found that if a second meal was taken too soon after the first, or if fish accept too much feed, digestion was less efficient.

Ingredient	Formulation	
	1	2
Brown fish meal	75	66
Shrimp head meal	-	5
Squid liver powder	-	5
Suehiro UGF	-	5
Wheat middlings	8.6	3
Wheat flour	10	10
Zeolite	0.75	0.75
Fish oil	4.2	3.8
Soy lecithin	0.75	0.75
Choline chloride (50%)	0.40	0.40
Vitamin premix AGJT/F1*	0.33	0.33
Mineral premix AGJT/F1**	0.037	0.037
Total	100.067	100.067

Table 2: Composition of formulated test diets for grouper (Tacon et al. 1989)

*Vitamin premix AGJT/F1 supplies per kilogram of dry diet: Vitamin A 4000 IU, Vitamin D3 2000 IU, Vitamin E 200 mg, Vitamin K_3 8 mg, Thiamine 32 mg, Riboflavin 40 mg, Pyridoxine 32 mg, Pantothenic acid 120 mg. Nicotinic acid 160 mg. Biotin 0.4 mg, Folic acid 8 mg, Vitamin B_{12} 0.04 mg, Inositol 300 mg, Vitamin C 800 mg.

**Mineral premix AGJT/F1 supplies per kilogram of dry diet: Iron 30 mg, Zinc 50 mg, Manganese 25 mg, Copper 3 mg, Cobalt 0.5 mg, Iodine 3 mg, Trivalent chromium 0.25 mg, Selenium 0.10 mg.

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The effect of food ratio on the growth and yield of the grouper raised in floating netcages was investigated. A ration provided at 5% of body weight yielded the best body weight, a uniform size, a relatively better survival rate, and the highest feed efficiency. Rations provided at 1.41% and 5.75% of body weight were considered suitable for maintenance, and best weight gain respectively whereas ration provided at 9% was considered to be representing maximum feed intake. The fish were more uniform in size when fed at 5% to 8% of body weight. Although the yield increased with an increase in ration rate, the percentage increase between net yield of fish on 5% ration over 2% ration was 59.6% while the difference between net yield on rations above 5% was 26.8% to 33.6%. For economic production, the ration should be approximately 5% of body weight supplied every two days (Chua and Teng, 1982).

It appears that the nutrient requirement of grouper is similar to that of seabass and that practical seabass feed is expected to work equally well for grouper. Frequency is a major difference between these two species of fish.

References

- Boonyaratpalin, M. J. Wannagowat. and C. Borisut. 1993. L-ascorbyl 1-2 phosphate-Mg as a dietary vitamin C source for grouper. Presented at the seminar on fisheries 1993, Department of Fisheries, 16-17 September 1993.
- Chen, F. Y. M. Chow. T. M. Chao. and R. Lim. 1977. Artificial spawning and larval rearing of the grouper, *Epinephelus tauvina* (Forsskal) in Singapore. Singapore J. Pri. Ind. 5 (1) 1.21.
- Chen, H. Y. and J.C. Tsai. 1994. Optimal dietary protein level for the growth of juvenile grouper, *Epinephelus malabaricus*, fed semipurified diets. Aquaculture, 119: 265-271.
- Chua, T.E. and S.K. Teng. 1978. Efffects of feeding frequency on the growth of young estuary grouper, *Epinephelus tauvina* (Forsskal), cultured in floating cages. Aquaculture, 14:31-47.
- Chua, T.E. and S.K. Teng. 1982. Efffects of food ration on the growth, condition factor, food conversion efficiency and net yield of estuary grouper, *Epinephelus salmonoides* (Maxwell), cultured in floating net cages. Aquaculture, 27:273-283.
- Chulawittayanukul, P. 1986. Effect of vitamin E on maturation and reproduction of grouper. Paper presented at the 3rd Brackish Water Fisheries Seminar, Brackish Water Division, Department of Fisheries. Bangkok, Thailand. 6 p.
- Dawes, B. 1930. The absorption of fats and lipoids in plaice. J. Mar. Biol. Ass. U.K., 17:75 102.
- El Dakour, S and K.A. George. 1982. Growth of (*Epinephelus tauvina*) fed on different protein : energy ratios. Kuwait Inst. Sci. Res. Ann Res. Rep. pp 75-77.
- Hickling, C.F. 1962. Fish culture. Faber and Faber, London. 295 pp.
- Kanazawa, A. 1984. Feed formulation for penaeid shrimp, seabass, grouper and rabbitfish culture in Malaysia. pp 61-78. *In*: FAO, Malaysia Coastal Aquaculture Development Project. FI: DP MAL/77/008, Field Document 2.
- Longly, W.H. and S.F. Hildebrand. 1941. Systematic Catalogue.
- Moore, W.G. 1941. Studies on the habits of fishes. Ecol. 22: 91-96.
- New, M.B. 1987. Feed and feeding of fish and shrimp: A manual on the preparation and presentation of compound feeds for shrimp and fish in aquaculture. United Nations Development Program, Food and Agriculture Organization of the United Nations, Rome, pp 111.
- Pechamanee, T. and M. Assavaaree. 1993. Nutritional value of rotifer, *Branchionus plicatilis*, fed with emulsified oils rich in n3HUFA. The Proceedings of Grouper Culture, Songkhla, Thailand p. 63-67.
- Phromkunthong, W.M. Boonyaratpalin and W. Verakunpiriya. 1993. Histopathology of the gills of ascorbic acid deficient grouper *Epinephelus malabaricus*. Fish Pathol, 28(4): 151-159.

Randall, J.E. 1965. Food habits of reef fishes of the West Indies. Stud. Trop. Oceanog., 5: 665 -847.

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- Ruangpanich N. and P. Boonliptanond. 1993. Some factors affecting the growth and survival of grouper larvae (*Epinephelus malabaricus*). National Institute of Coastal Aquaculture, Department of Fisheries, Thailand Technical Paper No. 20.
- Shiau, S.Y. and C.W. Lan. 1996. Optimum dietary protein level and protein to energy ratio for growth of grouper (*Epinephelus malabaricus*) Aguaculture 145: 259-266.
- Sukhawongs, S. N. Tanakumchup. and S. Chungyampin. 1978. Feeding experiment on artificial diet for greasy grouper, Epinephelus tauvina in nylon cages. Annual Report of Songkhla Fisheries Station, Department of Fisheries. pp 103-117.
- **Tacon, A.G.J. et al. 1989.** The food and feeding of seabass (Lates calcarifer), grouper (Epineephelus tauvina) and rabbitfish (Siganus canaliculatus) in floating net cages at the National Sea Farming Development Center, Lampung, Indonesia. Sea Farming Development Project, FAO. INS/81/008/Technical Paper/13.
- **Teng, S.K. 1979.** Studies on the culture of the estuary grouper, *Epinephelus salmoides*, Maxwell (Pisces: Serranidae) in floating net cages. P 61. *In*: Grouper Abstracts, Brackish water Aquaculture Information System, Compiled by A. Divina and V. Zamora et al. SEAFDEC, Philippines, 1987.
- Teng, S. K. T.B. Chua and P.E. Lim. 1977. Preliminary observation on the dietary protein requirement of estuary grouper, *Epinephelus tauvina* (Forsskal) cultured in floating net cages. P 60. *In*: Grouper Abstracts, Brackish water Aquaculture Information System. Compiled by M.A.V. Divina and V. Zamora et al. SEAFDEC, Philippines, 1987.
- Teng, S. K. T.B. Chua and P.E. Lim. 1978. Preliminary observation on the dietary protein requirement of estuary grouper, *Epinephelus tauvina* (Forsskal) cultured in floating net cages, Aquaculture 15 (3) 257-272.
- Tseng, W.Y and S.K. Ho. 1988. Grouper culture, a practical manual. Chien Cheng Publisher, Kaohsiung. R.O.C. 134 pp.
- Tucker, J.W. Jr. 1991. Marine fish nutrition. In: G.L. Allan and W. Dall (Eds.) Proc. Aquaculture Nutrition workshop, Salamander Bay, 15-17 April 1991, NSW fisheries, brackish water Fish Culture Research Station, Salamander Bay, Australia.
- Watanabe, T. T. Arakawa, C. Kitajima and S. Fujita. 1983. Improvement of dietary value of live foods for fish larvae by feeding them on n3 highly unsaturated fatty acid and fat soluble vitamin. Bulletin of the Japanese Society of Scientific Fisheries, 49(3): 471-479.
- Wongsomnuk, S. P. Parnichsuka and Y. Danayadol. 1978. Experiment on nursing of grouper, *Epinephelus tauvina* (Forsskal) with various mixed feeds. Pp. 79-102. *In*: Annual Report of Songkhla Fisheries Station, Department of Fisheries.