



Optimizing the growth performance of brown-marbled grouper, *Epinephelus fuscoguttatus* (Forsk.) , by varying the proportion of dietary protein and lipid levels

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ABSTRACT

Nine practical diets containing different protein (450, 500, or 550 g/kg) and lipid (80, 120, or 160 g/kg) levels in a 3×3 factorial experimental design were fed to triplicate groups of brown-marbled grouper, *Epinephelus fuscoguttatus* (initial weight, 8.8 ± 1.0 g) for eight weeks. Fish were cultured in a flow-through seawater system with a stocking density of 15 fish per tank. Fish fed Diet 50/16 (500 g/kg protein and 160 g/kg lipid) had the highest final body weight and specific growth rate at the end of feeding trial. In general, diets with 450 g/kg protein, irrespective of lipid levels, produced fish with lower growth performance compared to other fish groups. Meanwhile, increasing dietary protein content to 550 g/kg did not improve growth rates of the cultured fish. The feed conversion ratio (FCR) mirrored the growth rate trend with better value of FCR observed in fish fed Diet 50/16. The results indicated that a diet formulated with 500 g/kg protein and 160 g/kg lipid was optimal for rearing brown-marbled grouper fingerlings <40 g body weight. Increasing lipid level in the diets from 80 to 160 g/kg appeared to have a protein sparing effect which should help reduce feeding costs.

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1. Introduction

Groupers have been identified as the most desired fish in the live reef fish trade in the Southeast Asia region with retail prices ranging from US\$5 to \$180 per kilogram (Sadovy et al., 2003). Grouper species such as *Epinephelus* spp., *Cromileptes altivelis* and *Plectropomus* spp. are the dominant species in this live fish trade (Pierre et al., 2008). However, brown-marbled grouper (*E. fuscoguttatus*) have many advantages as an aquaculture species due to its tolerance to captive conditions, good market price and demand in the live fish trade market, and higher growth rates compared to other grouper species such as the red grouper (*E. akaara*), humpback grouper (*C. altivelis*) and coral trouts (*Plectropomus* spp.) (Boonyaratpalin, 1997; Rachmansyah et al., 2009). Unfortunately, one of the limiting factors to the expansion of this industry are high production costs associated with fish feeds and often the unsustainable approach of using trash fish as a main source of feed (Tuan and Williams, 2007). Designing species-specific feeds can improve the performance of the cultured fish, and determining the optimal protein and lipid ratios are important in achieving this goal.

Abbreviations: CF, condition factor; DL, dietary lipid; DP, dietary protein; FCR, feed conversion ratio; FI, feed intake; HSI, hepatosomatic index; NPU, net protein utilization; PER, protein efficiency ratio; SGR, specific growth rate; VSI, viscerosomatic index; WG, weight gain.

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Table 1
Ingredient, proximate composition and energy content of experimental diets (DM basis).

Ingredients (g/kg)	Protein/lipid 45/8	45/12	45/16	50/8	50/12	50/16	55/8	55/12	55/16
Fish meal ^a	639.0	639.0	639.0	710.0	710.0	710.0	781.0	781.0	781.0
Tapioca starch ^b	60.0	60.0	60.0	60.0	60.0	60.0	60.0	60.0	60.0
α-Cellulose	209.0	169.0	129.0	145.0	105.0	65.0	81.0	41.0	1.0
CMC ^c	15.0	15.0	15.0	15.0	15.0	15.0	15.0	15.0	15.0
Vitamin Premix ^d	30.0	30.0	30.0	30.0	30.0	30.0	30.0	30.0	30.0
Mineral Premix ^e	20.0	20.0	20.0	20.0	20.0	20.0	20.0	20.0	20.0
Dicalcium phosphate	10.0	10.0	10.0	10.0	10.0	10.0	10.0	10.0	10.0
Fish oil ^f	17.0	57.0	97.0	10.0	50.0	90.0	3.0	43.0	83.0
Proximate composition									
Moisture	118.9	119.0	118.0	116.7	119.1	12.3	115.8	112.6	111.0
Crude protein	455.1	456.1	452.2	495.6	495.1	498.7	552.1	548.2	548.6
Lipid	76.8	122.2	154.1	78.4	121.6	161.8	80.4	117.9	158.0
Ash	103.2	101.7	106.3	118.3	123.8	111.3	127.5	119.3	136.8
GE ^g (MJ/kg)	20.1	21.1	21.7	20.7	21.0	22.1	20.3	21.2	21.9
P/E ^h (g/MJ)	22.7	21.6	20.8	24.7	23.6	22.6	27.2	25.9	25.1

^a Danish fish meal.

^b Tapioca AAA brand. Bake with Me Sdn. Bhd.

^c Carboxymethyl cellulose (CMC), Sigma.

^d Vitamin mixture (g/kg mixture): ascorbic acid, 45.0; inositol, 5.0; choline chloride, 75.0; niacin, 4.5; riboflavin, 1.0; pyridoxine HCl, 1.0; thiamine HCl, 0.92; D-calcium pantothenate, 3.0; retinyl acetate, 0.60; vitamin D3, 0.083; Menadione, 1.67; DL alpha tocopherol acetate, 8.0; D-biotin, 0.02; folic acid, 0.09; vitamin B12, 0.00135. All ingredients were diluted with alpha cellulose to 1 kg.

^e Mineral mixture (g/kg mixture): Calcium phosphate monobasic, 270.98; Calcium lactate, 327.0; Ferrous sulphate, 25.0; Magnesium sulphate, 132.0; Potassium chloride, 50.0; Sodium chloride, 60.0; Potassium iodide, 0.15; Copper sulphate, 0.785; Manganese oxide, 0.8; Cobalt carbonate, 1.0; Zinc oxide, 3.0; Sodium salenite, 0.011; Calcium carbonate, 129.274.

^f Cod liver oil, Seven Seas Brand.

^g Calculated gross energy (GE) = based on combustion values of 23.6 MJ/kg for protein, 39.5 MJ/kg for lipid and 17.2 MJ/kg for carbohydrate.

^h P/E = Protein to energy ratio.

Protein is often the most expensive ingredient in fish feeds, and sufficient levels are necessary to support high growth rates. On the other hand, if provided in excess, protein can be catabolized into energy which should be avoided as this will increase feeding costs as well as contributing to higher nitrogenous waste (NRC, 2011). To prevent this from occurring, lipid can be utilized as a concentrated energy source, thus sparing protein for growth, while also providing essential fatty acids (Watanabe, 1982; Lee et al., 2002). Therefore, when formulating fish diets, careful considerations must be made regarding protein to lipid ratios since appropriate combinations may lead to improved feeding efficiencies, higher growth and/or better survival rates of fish (Shiau and Lan, 1996; Tuan and Williams, 2007).

In general, studies showed that groupers require high dietary protein levels to achieve optimum growth. In a study using casein-based diets, Luo et al. (2004) reported that the protein requirement of *E. coioides* juveniles, was close to 480 g/kg diet. Usman et al. (2005) reported that humpback grouper (*C. altivelis*) growth was optimal when fed diets containing 530 g/kg dietary protein (DP). Tuan and Williams (2007) observed that the growth of Malabar grouper (*E. malabaricus*) cultured in indoor tanks was best when fed 550 g/kg DP and 120 g/kg dietary lipid (DL) level with fishmeal, krill hydrolysate, wheat gluten and casein as the protein source. Chen and Tsai (1994) reported a lower dietary protein requirement of about 480 g/kg for Malabar grouper with casein as protein source. A factorial approach study indicated that 2–250 g white grouper (*Epinephelus aeneus*) required higher DP (450–550 g/kg) and moderate DL of 100–120 g/kg while larger fish 250–750 g require lower DP (400–420 g/kg) and higher DL (130–140 g/kg) (Lupatsch and Kissil, 2005). Surprisingly, despite the brown-marbled grouper being the most widely cultured grouper species in the Asia Pacific region, very few studies have been carried out on the nutritional requirement of this commercially important species. To address this issue, the present study was conducted to investigate the optimum protein to lipid ratios in the formulated diets for brown-marbled grouper fingerlings.

2. Materials and methods

2.1. Experimental diets

Nine experimental diets were formulated to provide a 3 × 3 factorial arrangement of three levels of crude protein (450, 500, or 550 g/kg) and three levels of lipid (80, 120, or 160 g/kg) which yielded Diet 45/8, 45/12, 45/16, 50/8, 50/12, 50/16, 55/8, 55/12, and 55/15, respectively. Danish fish meal and cod liver oil (Seven Seas®) were used as the source of protein and lipid, respectively. All dry ingredients were mixed together in a mixer before adding the fish oil and water. The moist dough was then screw-pressed through a 3 mm die and the strands of feeds were oven-dried at 40 °C for 3 h. Diets were refrigerated at 4 °C until used. Table 1 shows the formulation and proximate composition of the experimental diets. Gross energy (GE) was calculated using combustion values for protein 23.6 MJ/kg, lipid 39.5 MJ/kg and carbohydrate 17.2 MJ/kg, respectively (NRC, 2011). The amino acid and fatty acid composition of experimental diets are listed in Tables 2 and 3, respectively.

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