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ORIGINAL ARTICLE

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Effect of dietary lipid level on growth performance and feed utilization of juvenile kelp grouper *Epinephelus bruneus*

Kakuta Yoshii · Fumiaki Takakuwa · Hung Phuc Nguyen · Toshiro Masumoto · Haruhisa Fukada

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Abstract A growth trial was conducted to evaluate the effect of dietary lipid level on juvenile kelp grouper *Epinephelus bruneus*. Juvenile kelp grouper were fed isonitrogenous diets (470 g/kg crude protein) with four levels of crude lipid at 60 g/kg (CL60), 130 g/kg (CL130), 210 g/kg (CL210), and 270 g/kg (CL270) for 56 days. The highest growth performance and feed utilization were found in the CL130 diet group. A high dietary lipid level (CL270 diet) significantly decreased growth performance and feed utilization. A significant difference in apparent digestibility was only observed in protein, which was highest in the CL130 diet groups. The highest retention for protein, energy, and lipid was found in the CL130 diet group. The dietary lipid levels significantly changed whole-body and liver compositions, the highest being the CL60 diet group for crude protein level and the CL210 diet group for crude lipid level. Based on a second-order polynomial regression analysis of crude lipid level against specific growth rate and protein efficiency ratio, the optimum dietary lipid level for kelp grouper was estimated to be 152 and 154 g/kg diet, respectively.

Keywords Dietary lipid level · Digestibility · Feed utilization · Growth · Kelp grouper

K. Yoshii · F. Takakuwa · H. P. Nguyen · T. Masumoto · H. Fukada (✉)
Laboratory of Fish Nutrition, Faculty of Agriculture, Kochi University, B200 Monobe, Nankoku, Kochi 783-8502, Japan
e-mail: fukaharu@kochi-u.ac.jp

Introduction

The demand for aquatic products is rapidly increasing with the growing human population and a change in preferences towards tasty and healthier foods [1]. Groupers are an important species in Japan, Taiwan, Hong Kong, and Southeast Asia. Major Asian grouper species are cultured in these areas. Groupers are good candidates for aquaculture because of their high value, good adaptability to a crowded environment, and rapid growth [2].

The kelp grouper *Epinephelus bruneus* is a warm seawater fish distributed from south Japan to the Philippines. Juvenile and adult kelp groupers live in clean water of offshore reefs. During the juvenile stage, groupers live in shallow water, where they can find hiding places. As they grow, they move to deeper water. Groupers reach market size within 2–3 years. The price of the fish is US $40 per kg in Japan, higher than for other fish. Kelp grouper aquaculture is a new industry in Japan, and little is known about the nutrient utilization and dietary requirements of the fish [3].

Information on the optimum dietary protein and lipid levels can be used to improve efficiency of cultivation and reduce feed costs [4–6]. Optimum dietary protein and lipid levels minimize undesirable nitrogenous waste output and improve the water quality of fish-farm effluents. To date fish feed developed for other species or trash fish supplemented with minerals and vitamins have mainly been used as feed in kelp grouper culture. For fish feed, the use of dietary protein as an energy source is undesirable because it is more expensive than nonprotein energy sources [6]. Fish oil is used as an energy and essential fatty acid source. An adequate dietary lipid level decreased the use of protein as an energy source in rainbow trout *Oncorhynchus mykiss* (formerly *Salmo gairdneri*) [7] and also reduced nitrogenous output to the environment [8, 9]. A diet containing up

to 300 g/kg crude lipid for Atlantic salmon *Salmo salar* significantly reduced use of protein as an energy source and improved feed efficiency [10]. However, excessive dietary lipid negatively affected growth performance and reduced feed consumptions and utilization of other nutrients, which led to fat deposition on viscera or the carcass [11]. Excessive lipid in the diet not only suppresses fatty acid synthesis, but also reduces the ability of fish to digest and assimilate lipids [12]. Therefore, a proper ratio of digestible protein (DP) to digestible energy (DE) (DP/DE ratio) is necessary to achieve high growth rate and feed conversion efficiency [13].

Groupers store dietary lipid in the liver, which can lead to "fatty liver condition" in aquaculture. This is undesirable because the fish utilize dietary energy ineffectively; furthermore, excessive accumulation reduces physiological ability [14, 15]. Recent studies have shown that the requirement for dietary protein and lipid differs among grouper species. Juvenile orange-spotted grouper *E. coioides* showed lower growth performance with a higher dietary lipid level of 160 g/kg, and the optimum crude protein and lipid levels were 480 and 110 g/kg, respectively [16]. The optimum crude protein and lipid levels for humpback grouper *Cromileptes altivelis* are 530 and 120 g/kg, respectively [17]. Furthermore the optimum DP/DE ratio for white grouper *E. aeneus* decreased with fish growth [18]. In juvenile Malabar grouper *E. malabaricus* the optimum crude protein and lipid levels were 550 and 120 g/kg, respectively [19]. When juvenile Malabar grouper were fed with a high-lipid diet, weight gain and specific growth rate decreased with the lipid level. These data suggest that the determination of optimum crude lipid level is needed to develop optimum diet for groupers.

This study aimed to investigate the effects of dietary lipid levels for future determination of optimum dietary DP/DE ratio in juvenile kelp grouper. Growth performance, apparent digestibility, and nutrient retention responses of the fish were used to derive an optimum dietary lipid level.

Materials and methods

Experimental diets

Four experimental diets (Table 1) were formulated to provide 470 g/kg protein and crude lipid of 60 g/kg (CL60), 130 g/kg (CL130), 210 g/kg (CL210), or 270 g/kg (CL270). Fish meal and fish oil were used as main protein and lipid sources, respectively. All dry ingredients were mixed before the addition of fish oil. Feed ingredients were mixed with 45% fresh water. The mixture was pelleted to 2.2 mm diameter (Hiraga Kosakusyo Co. Ltd., Kobe, Japan). The gross energy levels in the diets were in the range 19.3–22.6 MJ/kg. The DP/DE ratios of the diets were in the range 20.7–29.8 g/MJ. Diets were stored at −20°C until use.

Rearing system and experimental design

Kelp grouper *E. bruneus* juveniles hatched and reared at Yoshikawa Aquaculture Farm (Kagawa, Japan) were used in this study. The growth trial was performed at the Kochi Prefectural Fisheries Experimental Station (Kochi, Japan). The fish were acclimatized in cylindrical fiberglass tanks (1000 l capacity; 250 fish per tank) for 15 days. During the acclimation period, the fish were fed a commercial feed (Marubeni Nisshin Feed, Tokyo, Japan) twice daily (09:00 and 17:00). The experiment was conducted as a factorial design (four lipids levels × single protein level) in triplicates. A total of 240 fish were distributed into 12 square

Table 1 Composition of experimental diets for kelp grouper

Diet	CL60	CL130	CL210	CL270
Crude protein (g/kg)[g]	470	470	470	470
Crude lipid (g/kg)[g]	60	130	210	270
Ingredients (g/kg)				
Fish meal[a]	521	521	521	521
Krill meal[b]	100	100	100	100
Fish oil[c]	3	73	143	213
Vitamin mixture[d]	55	55	55	55
Mineral mixture[e]	55	55	55	55
Guar gum	20	20	20	20
CMC Na[f]	35	35	35	35
Cellulose	211	141	71	1
Proximate compositions (g/kg)[h]				
Crude protein	484	482	471	474
Crude fat	64	132	214	272
Crude ash	94	87	86	84
Gross energy (MJ/kg)[h]	19.3	20.4	21.7	22.6
DP/DE (g/MJ)[h]	29.8	25.8	22.6	20.7

[a] Dry matter, 88%; crude protein, 80%; crude fat, 9%

[b] Dry matter, 82%; crude protein, 66%; crude fat, 9%

[c] Riken feed oil Ω (Eiken Shoji Co. Ltd, Tokyo, Japan)

[d] Vitamins (mg/100 g dry diet): thiamine HCl, 1.6; pyridoxine HCl, 1.6; nicotinic acid, 4.8; inositol, 113; folic acid, 1.6; choline chloride, 779; calcium ascorbate, 119; menadione-$NaHSO_4$, 5.13; riboflavin, 2.93; calcium pantothenate, 9.33; biotin, 4.67; cyanocobalamin, 1.07; vitamin A palmitate, 2.67; α-tocopherol, 117; α-cellulose, 837

[e] Minerals (mg/100 g dry diet): KH_2PO_4, 209; $Ca(H_2PO_4)_2 \cdot H_2O$, 247; calcium lactate, 113; iron citrate 66; $ZnSO_4 \cdot H_2O$, 8; $CuSO_4 \cdot 4H_2O$, 5.04; $CoCl_2 \cdot 6H_2O$, 0.04; KIO_3, 0.12; α-cellulose, 443.2; dextrin, 360

[f] Carboxymethyl cellulose sodium salt

[g] By calculation

[h] By analysis

fiberglass tanks (200 l capacity; 20 fish per tank; initial biomass density, 0.56 kg/m^3). Mean initial body weight was 6.38 ± 0.06 g. Sand-filtered seawater was supplied to each tank (flow rate, 4 l/min), which was continuously aerated. Dissolved oxygen was maintained above 4.2 mg/l, and water temperature ranged from 24.4°C to 30.5°C during the experimental period. During the 56-day experiment, the fish were hand-fed experimental diets (dry weight) at 3.6% body weight per day (on weekdays at 09:00 and 17:00 and on Saturday at 09:00). The amount of feed was adjusted every 2 weeks by batch weight.

Fish from each tank were batch-weighted and counted on days 0, 12, 24, 36, 48, and 56. These fish were starved for 24 h before analysis. At the beginning of the trial, 15 fish were randomly selected and killed with an overdose of 2-phenoxyethanol. Five fish were used for analysis of whole-body composition. Ten fish were used for analysis of liver composition and hepatosomatic index. At the end of the trial, 13 fish from each experimental tank were killed as described above. Three fish were used for the analysis of whole-body composition. Whole-body and liver weights were measured for 10 fish to estimate the hepatosomatic index. The liver was used for analysis of liver composition. These samples were stored at −20°C until analysis.

Digestibility measurement

Nutrient digestibility was determined immediately after the growth trial with eight square tanks and 96 fish (mean initial body weight, 15.2 ± 0.6 g). After random distribution to each tank, fish were acclimatized on experimental diets for 1 week. Experimental diets were the same as those used in the growth trial, with the addition of chromium oxide (Cr_2O_3, 5 g/kg). Feces were collected using a vacuum collector. Apparent digestibility was calculated by the following formula:

$$\begin{aligned}\text{Apparent digestibility} = 100 - 100 \\ \times (\%\ Cr_2O_3 \text{ in diet}/\%Cr_2O_3 \text{ in feces}) \\ \times (\%\text{nutrition in feces}/\%\ \text{nutrition in diet}).\end{aligned}$$

Analyses

The whole body and liver were finely ground and stored at −30°C until analysis. Crude protein, crude lipid, crude ash, and moisture were determined by AOAC methods [20]. The chromic oxide contents of the experimental diets and feces were measured by photometry according to the methods of Furukawa and Tsukahara [21]. The gross energy of the experimental diets and feces was measured using a bomb calorimeter (Ogawa Sampling, Saitama, Japan). These experiments were conducted in duplicate.

Statistical analyses

Normality for all data was confirmed using a Kolmogorov–Smirnov test ($P < 0.05$) with Ekuseru–Toukei 2008 (Social Survey Research Information Co., Ltd., Tokyo, Japan). After the normality test, all data were analyzed by one-way analysis of variance followed by Tukey–Kramer tests. The statistical significance of differences between treatment groups were assessed at a 5% level with Statcel 2 (OMS Publishing, Saitama, Japan). Second-order polynomial regression analysis of the relationship between specific growth rate or protein efficiency ratio and dietary lipid level was used to estimate the optimal dietary lipid level in the diets for juvenile kelp grouper.

Results

Growth, feed efficiency, hepatosomatic index, and survival

Fish in one of the tanks for the CL130 and one for the CL210 diet groups were infected by viral nervous necrosis and more than 50% of the fish died. These two tanks were removed from the analyses. Juvenile kelp grouper readily accepted the experimental diets. Growth performance and feed efficiency are shown in Table 2. The CL130 diet group showed the highest values in weight gain, specific growth rate, feed efficiency ratio, and protein efficiency ratio among the treatments. The CL130 diet group showed significantly higher growth performance compared with the CL60 and CL270 diet groups. The daily feeding ratio was significantly higher in the CL270 diet group compared with the other dietary groups. With the exception of the CL60 diet group, there were no significant differences in hepatosomatic index. The survival rate was not significantly different among the dietary groups.

Whole-body and liver compositions

Proximate compositions of whole body and liver are shown in Table 3. The moisture content was significantly lower in whole body of the CL130 and CL270 diet groups compared with the CL60 diet group and in the liver of the CL210 diet group compared with the other dietary group. The protein content of whole body and liver in the CL60 and CL130 diet groups was significantly higher than that of the other dietary groups. The lipid content of whole body in the CL210 and CL270 diet groups was almost the same; however, the liver lipid content of the CL210 group was significantly higher than those of other dietary groups. There were no significant differences in ash contents among the treatments.

Table 2 Growth performance and feed utilization efficiency of kelp grouper fed experimental diets with different lipid levels for 56 days

	Dietary group			
	CL60	CL130	CL210	CL270
Initial body weight (g)	6.37 ± 0.01	6.39 ± 0.00	6.37 ± 0.02	6.38 ± 0.00
Final body weight (g)	14.8 ± 0.21^{a}	18.0 ± 0.12^{b}	16.2 ± 0.09^{ab}	13.9 ± 0.54^{a}
Weight gain (%)[A]	132 ± 2.89^{ab}	182 ± 1.50^{c}	154 ± 1.00^{bc}	118 ± 8.39^{a}
Specific growth rate (%/day)[B]	0.65 ± 0.01^{ab}	0.80 ± 0.00^{c}	0.72 ± 0.00^{b}	0.60 ± 0.03^{a}
Feed efficiency (%)[C]	75.6 ± 1.05^{ab}	92.4 ± 0.60^{c}	83.2 ± 2.55^{bc}	66.2 ± 3.82^{a}
Daily feeding ratio (%/day)[D]	2.74 ± 0.03^{a}	2.64 ± 0.01^{a}	2.68 ± 0.01^{a}	2.96 ± 0.04^{b}
Protein efficiency ratio[E]	1.55 ± 0.03^{ab}	1.91 ± 0.02^{c}	1.83 ± 0.01^{bc}	1.39 ± 0.08^{a}
Hepatosomatic index (%)[F]	2.27 ± 0.04^{a}	2.82 ± 0.06^{b}	2.89 ± 0.14^{b}	2.73 ± 0.17^{b}
Survival rate (%)[G]	90.0 ± 2.89	100 ± 0.00	92.5 ± 2.50	91.7 ± 3.33

Values are mean ± standard error (SE) of duplicate or triplicate tanks except hepatosomatic index ($n = 10$)

Values in the same line with different lower-case superscripts are significantly different ($P < 0.05$)

[A] Weight gain = (final total body weight − initial total body weight)/initial body weight × 100

[B] Specific growth rate = 100 × (ln final mean weight − ln initial mean weight)/trial days

[C] Feed efficiency = (final total body weight − initial total body weight)/total dry feed × 100

[D] Daily feeding ratio = 100 × g dry feed intake/[(initial fish number + final fish number)/2 × (g initial average body weight + g final average body weight)/2]/feeding days

[E] Protein efficiency ratio = weight gain/total protein intake

[F] Hepatosomatic index = individual liver weight/wet body weight × 100

[G] Survival rate = (final fish number − initial fish number)/initial fish number × 100

Table 3 Whole-body and liver composition (wet weight basis) of kelp grouper fed experimental diets with different lipid levels for 56 days

	Dietary group				
	Initial	CL60	CL130	CL210	CL270
Whole-body composition (%)					
Moisture	71.9 ± 0.30	69.5 ± 0.09^{b}	67.6 ± 0.13^{a}	68.7 ± 0.71^{ab}	67.7 ± 0.34^{a}
Protein	17.7 ± 0.09	20.7 ± 0.16^{b}	20.7 ± 0.31^{b}	18.8 ± 0.02^{a}	19.2 ± 0.39^{a}
Lipid	4.44 ± 0.12	4.24 ± 0.04^{a}	6.81 ± 0.14^{b}	8.25 ± 0.10^{c}	8.16 ± 0.27^{c}
Ash	5.25 ± 0.08	5.37 ± 0.07	5.19 ± 0.26	5.06 ± 0.05	5.42 ± 0.01
Liver composition (%)					
Moisture	85.1 ± 0.9	79.9 ± 0.18^{c}	76.9 ± 0.83^{b}	74.0 ± 0.19^{a}	78.6 ± 0.53^{bc}
Protein	7.70 ± 0.17	11.1 ± 0.28^{b}	9.89 ± 0.87^{ab}	8.85 ± 0.31^{a}	8.96 ± 0.07^{a}
Lipid	4.04 ± 0.02	6.77 ± 0.12^{a}	11.6 ± 0.33^{b}	14.6 ± 0.53^{c}	11.8 ± 0.65^{b}
Ash	1.69 ± 0.01	2.33 ± 0.06	2.19 ± 0.06	2.18 ± 0.22	2.12 ± 0.12

Values are mean ± SE of duplicate or triplicate tanks except initial ($n = 5$ for whole-body composition; $n = 10$ for liver composition). Statistical analysis was performed without initial group. Values in the same line with different lower-case superscripts are significantly different ($P < 0.05$)

Nutrient retention

Protein and lipid retention were not significant different except in the CL270 diet group (Table 4). The CL270 diet group showed significantly lower protein and lipid retention. The highest values for protein and lipid retention were found in the CL130 diet group. The highest value for energy retention was in the CL210 diet group. A significant difference in energy retention was observed only between the CL60 and CL210 diet groups.

Apparent digestibilities of protein, lipid, and energy

Apparent digestibility of protein in the CL130 diet group was significantly higher than that in the CL210 and CL270 diet groups (Table 5). Apparent digestibility of protein

Table 4 Protein, lipid, and energy retentions of kelp grouper fed experimental diets with different lipid levels for 56 days

	Dietary group			
	CL60	CL130	CL210	CL270
Protein retention (%)[A]	35.5 ± 0.89[b]	42.5 ± 1.18[b]	35.6 ± 0.05[b]	27.4 ± 2.00[a]
Lipid retention (%)[B]	48.7 ± 1.94[b]	52.5 ± 1.13[b]	42.6 ± 0.60[b]	27.5 ± 2.09[a]
Energy retention (%)[C]	21.4 ± 1.56[a]	28.1 ± 0.76[ab]	29.5 ± 1.33[b]	23.3 ± 1.58[ab]

Values are mean ± SE of duplicate or triplicate tanks. Values in the same column with different lower-case superscripts are significantly different ($P < 0.05$)

[A] Protein retention = 100 × (final body protein − initial body protein)/total protein intake

[B] Lipid retention = 100 × (final body lipid − initial body lipid)/total lipid intake

[C] Energy retention = 100 × (final body energy − initial body energy)/total energy intake

Table 5 Protein, lipid, and energy digestibilities of kelp grouper fed experimental diets with different lipid levels

	Dietary group			
	CL60	CL130	CL210	CL270
Protein (%)	85.5 ± 0.14[bc]	86.6 ± 1.16[c]	79.4 ± 1.47[b]	69.8 ± 1.16[a]
Lipid (%)	94.8 ± 0.33	94.8 ± 0.55	94.6 ± 0.88	92.6 ± 0.62
Energy (%)	71.2 ± 1.59	79.3 ± 2.21	75.8 ± 3.24	70.6 ± 3.10

Values are mean ± SE of duplicate tanks. Values in the same column with different lower-case superscripts are significantly different ($P < 0.05$)

decreased with an increase in crude lipid from 130 to 270 g/kg. No significant differences were found in apparent digestibility values of lipid and energy.

Estimation of requirements

Second-order polynomial regression analysis was used to estimate the optimal dietary lipid level for juvenile kelp grouper (Fig. 1). The second-order polynomial regression curves $[Y = (-0.0015 \times 10^{-2})X^2 + 0.0046X + 0.4373$ $(R^2 = 0.862)$; $Y = (-0.0043 \times 10^{-2})X^2 + 0.0133X + 0.9036$ $(R^2 = 0.903)]$ were used to determine the optimal point for the maximum specific growth rate or protein efficiency ratio. The results showed that the maxima were at 152 g/kg for specific growth rate and 154 g/kg for protein efficiency ratio.

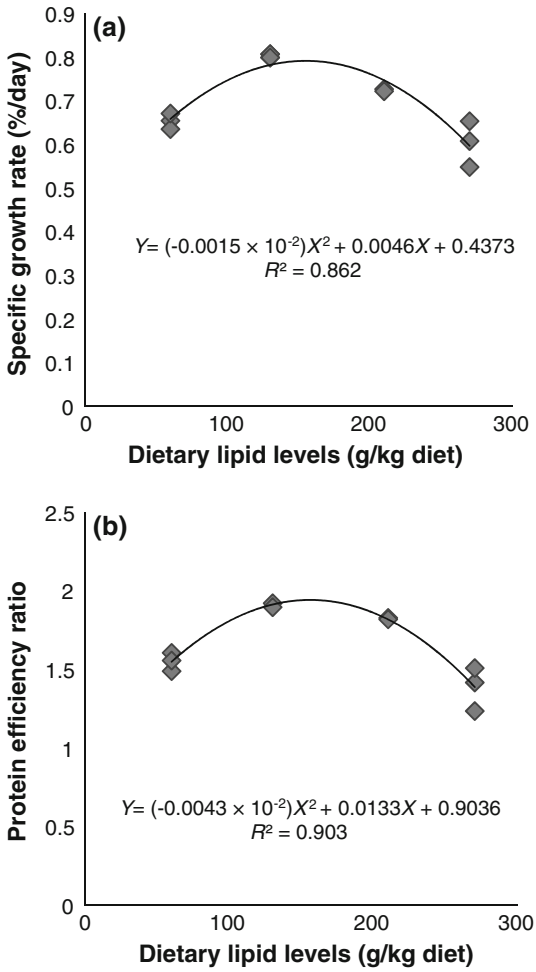

Fig. 1 **a** Relationship between dietary lipid level and specific growth rate of juvenile kelp grouper as fitted by second-order polynomial regression analysis. **b** Relationship between dietary lipid level and protein efficiency ratio of juvenile kelp grouper as fitted by second-order polynomial regression analysis

Discussion

In our study, juvenile kelp grouper fed with the CL130 diet showed the best growth performance, while the CL270 diet group had the lowest growth performance. High protein digestibility and lipid utilization are possible factors in the good growth performance for juvenile kelp grouper fed the CL130 diet. Protein digestibility and lipid utilization were reduced when the CL270 diet was fed. Furthermore, the CL270 diet group showed low protein and lipid retention, which might relate to the lower growth performance. It

appears that juvenile kelp grouper cannot utilize diets containing high levels of lipid, as reported for juvenile Malabar grouper [19]. However, juvenile kelp grouper fed on the low-lipid diet (CL60 diet) also showed low growth performance, despite relatively high protein and energy digestibilities. As supported by the lower protein efficiency ratio, the dietary protein in the CL60 diet might be used as an energy source due to the lack of dietary energy. The protein efficiency ratio in fish is regulated by the nonprotein energy source and is a good parameter for evaluating the "protein-sparing effect" [15, 22]. Nonprotein energy sources, such as lipids and carbohydrates, can be effectively used to reduce the requirement for protein [5, 6, 23–25]. In our study, juvenile kelp grouper effectively utilized lipid and showed a higher protein efficiency ratio, when fed the CL130 diet. Growth performance, feed efficiency, and nutrient retention decreased linearly with increasing dietary lipid levels from CL130 to CL270. Similar results were reported in juvenile orange-spotted grouper [16], humpback grouper [17], and juvenile Malabar grouper [19]. These observations suggest that groupers show similar trends in lipid utilization.

A strong correlation between specific growth rate and protein retention was observed in this study ($P < 0.05$, $R^2 = 0.830$, $n = 10$; Pearson correlation coefficient test). This observation was consistent with the finding of a positive correlation between protein retention and growth performance in juvenile Malabar grouper [19]. Such a positive correlation was also reported in cuneate drum *Nibea miichthioides* [26] and silver barb *Puntius goniontus* [27]. However, when the diet containing a high crude lipid level (i.e., the CL270 diet) was fed to juvenile kelp grouper, lipid retention significantly decreased. Differences in capacity to store lipid and ability to generate lipid-storing cells and tissues have been reported in several species [28, 29]. Thus, our results suggest that the kelp grouper's ability to store lipid is not high. There was no significant difference in energy retention between the CL130 and the CL210 diet groups. However, the specific growth rate was significantly higher in the CL130 diet group compared with the CL210 diet group. This indicated that the CL130 diet supplied enough energy for growth and extra lipids were not used as energy in juvenile kelp grouper.

Apparent digestibility of lipid by the kelp grouper did not differ significantly among the dietary groups. However, apparent digestibility of protein decreased significantly with an increase in crude lipid level from 210 to 270 g/kg, which is probably one of the causes of the lowered growth performance described above. It has been reported that sharp snout seabream *Diplodus puntazzo* showed no decrease in apparent digestibility of protein and an increase of apparent digestibility of lipid with an increase in dietary lipid level [30]. The digestibilities of protein and lipid for a diet with high lipid might differ among fish species.

When juvenile kelp grouper were fed diets containing high crude lipid levels, the crude protein levels of whole body and liver significantly decreased. In the present study, the crude protein levels of whole body and liver were negatively affected by an increase in lipid level, as reported in cuneate drum fed a similar diet [26]. Juvenile Malabar grouper showed a significant increase in whole-body protein level with an increase in crude lipid in feed [19]. In this study, whole-body and liver lipid contents increased with increasing crude lipid level from 60 to 210 g/kg, and were highest in the CL210 diet group. Similar results have been reported in juvenile Malabar grouper [19, 31], orange-spotted grouper [16], and humpback grouper [17]. When the high-lipid diet (CL270 diet) was fed, liver lipid content decreased. The difference in liver lipid content according to dietary lipid levels was reflected in the hepatosomatic index. Senegalese sole *Solea senegalensis* [32] showed decreased enzymatic activity for lipid synthesis when a high-lipid diet was fed. This is thought to be due to suppression of lipid retention by decreasing lipid synthesis when excessive lipid is consumed. The CL270 diet group might have decreased lipid synthesis, resulting in low lipid retention.

In this study, second-order polynomial regression analysis was used to estimate the optimal dietary lipid level for juvenile kelp grouper. Specific growth rate and protein efficiency ratio were used as markers for growth and nutrient utilization in the analysis. The optimal crude lipid levels were 152 g/kg for specific growth rate and 154 g/kg for protein efficiency ratio. The optimal crude lipid level was similar to that of humpback grouper [17] and higher than that of juvenile Malabar grouper [19, 31] when dietary crude protein levels were similar.

In the present study, maximum growth performance, protein, energy, and lipid retention, whole-body protein content, and apparent protein digestibility were found in the CL130 diet group. A high-lipid diet (CL270 diet) negatively affected growth performance and digestibility of protein in juvenile kelp grouper. From the second-order polynomial regression analysis, the optimum dietary lipid level for kelp grouper was estimated to be 152–154 g/kg diet.

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