Utilization of Waste porphyra Products as Eco-friendly Feed Ingredients

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Tsuyoshi SUGITA[*1], Takeshi YAMAMOTO[*1], and Takao YASHIMATSU[*2]

Abstract We investigated the growth and hepatopancreatic enzyme activities in red sea bream, *Pagrus major*, fed diets containing waste nori, *Porphyra* spp. (red algae, purple laver) with different protein to fat ratios. Fingerlings of 10.1 g average body weight were fed for 10 weeks on the optimum fat and protein (11 and 55 %) control diet (CTD) and two higher fat and lower protein (19 and 42 % and 25 and 36 %, respectively) experimental diets (MFD and HFD). All diets were supplemented with 5 % waste nori. Weight gain, specific growth rate and feed efficiency in fish fed the CTD and MFD were higher than those in fish fed the HFD. Plasma total cholesterol, phospholipid, and free fatty acid concentrations and whole body crude fat content were increased as dietary fat level increased, whereas hepatopancreatic lipogenic enzyme activities (glucose-6-phosphate dehydrogenase, 6-phosphogluconate dehydrogenase, NADP-isocitrate dehydrogenase and NADP-malate dehydrogense) were decreased as the dietary fat level increased. Aspartate aminotransferase activity of the MFD group was significantly lower than the activity of the CTD group. These findings suggest that dietary fat content can be increased from 11% to 19% by supplementing 5% waste nori in fingerling red sea bream diets.

Key words: *Pagrus major*, *Porphyra*, eco-friendly fish feed, high fat low protein diet, hepatopancreatic enzyme activity

Introduction

In recent years, environmental pollution by nitrogen and phosphorus loading from fish farming has become a serious social problem. These pollutants are considered to originate mainly from fish meal, which is a main feed ingredient in fish feeds. Furthermore, the drastic decrease of feed-grade fish landings worldwide and increasing demand of formula feed production for aquaculture has made the market price of fish meal to increase. Therefore, from the viewpoints of environment and economics, dietary fish meal content should be reduced as much as possible.

The protein-sparing effects dietary of fat have been demonstrated with rainbow trout, *Oncorhynchus mykiss* (Takeuchi et al. 1978; Beamish and Medland 1986), yellowtail, *Seriola quinqueradiata* (Shimeno et al. 1980), Atlantic salmon, *Salmo salar* (Hillestad and Johnsen 1994), and Atlantic halibut, *Hippoglossus hippoglossus* (Aksnes *et al*. 1996), whereas no such effects were observed with tilapia, *Oreochromis niloticus* (Shimeno *et al*. 1993) or Japanese flounder, *Paralichthys olivaceus* (Kikuchi *et al*. 2000). In our previous report ofnfingerling red sea bream, *Pagrus major* (Sugita *et al*. 2007a), a protein-sparing effect by fat was not also found.

Nori, *Porphyra* spp. (red algae, purple laver) are commercially and culturally important species used widely as traditional Japanese food. Nori products contain high amounts of taurine, an important amino acid for the growth of marine fish, as well as minerals and vitamins. In recent years, occurrence of discolored nori with no commercial value

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[*1] Feed Research Group, Aquaculture Systems Division, Inland Station, National Research Institute of Aquaculture, Fisheries Research Agency, Tamaki, Mie 519-0423, Japan
[*2] Feed Research Group, Aquaculture Systems Division, National Research Institute of Aquaculture, Fisheries Research Agency, Minamiise, Mie 516-0193, Japan

(i.e., waste nori) has become a serious problem. Therefore, effective uses of the waste nori have been sought. There have been several studies on the use of various kinds of algae meal in fish feed (Stanley and Jones 1976, Tsai 1979, Appler 1985). It has been confirmed that a small amount of algae added to fish feed significantly improves growth, lipid metabolism, body composition, and disease resistance (Nakagawa and Kasahara 1986, Satoh *et al*. 1987, Xu *et al*. 1993, Mustafa *et al*. 1995). However, the supplemental effects of the waste nori on the growth and lipid metabolism have not been studied in red sea bream. We compared the growth and hepatopancreatic enzyme activities in red sea bream fed diets containing waste nori with different protein to fat ratios.

Materials and Methods

Experimental Diets

The formulation and proximate composition of the experimental diets are shown in Table 1. The control diet (CTD) contained 69% brown fish meal (BFM), 4.7% pollock liver oil (Riken Vitamin, Tokyo, Japan) and 10.5% gelatinized potato starch (Tokai Denpun, Shizuoka, Japan) as the main protein, fat, and carbohydrate sources, respectively. Medium fat and medium protein diet (MFD) and the highest fat and the lowest protein diet (HFD) contained 13.7 and 19.3% pollock liver oil. On the other hand, the BFM levels of MFD and HFD were reduced to 53 and 45%. The gelatinized potato starch content was reduced to 9% (MFD) and 5.1% (HFD). All diets were supplemented with 5% waste nori, which was supplied from Saga Prefectural Ariake Fisheries Research and Development Center, Koshiro, Saga, Japan. Krill meal was supplemented to all diets as a feeding stimulant. Chromium oxide was included as a marker for nutrient digestibility measurement. Cellulose was used as a filler. All the ingredients were thoroughly mixed, moistened by the addition of 35% water (w/w), and pelleted using a meat chopper (Hanaki Mfg Co., Ltd., Tokyo, Japan). Then the pellets were dried at 50℃ for 6 h and stored at -20℃ until fed to the fish.

Analytical nutrient and energy contents of the test diets are given in Table 1. The content of crude protein, crude fat and crude sugar in the CTD were 55, 11, and 11%, respectively, which are considered optimum (Furuichi *et al*. 1971, Yone *et al*. 1971, Yone 1976, Takeuchi *et al*. 1991). Crude protein contents were 42% in the MFD and 36% in the HFD, and crude sugar contents were 9% and 7%. On the other hand, crude fat contents were 19% in the MFD and 25% in the HFD. The phosphorus content in the CTD was 2.8%, and the content decreased as the dietary BFM level decreased (i.e., the dietary fat level increased), and the value was 2.4% (MFD) and 2.1% (HFD). Gross energy content in all diets ranged from 20.1-22.9MJ/kg.

Fish and Feeding Procedures

Fingerlings of red sea bream were obtained from the Kinki University Fish Nursery Center, Uragami, Wakayama, Japan and transferred to the Nansei Station of the National Research Institute of Aquaculture, Minamiise, Mie, Japan. The fish were initially stocked in two 500 L fiber reinforced plastic circular tanks, supplied with sand filtered and aerated sea water. The fish were fed a commercial feed (Kyowa Hakko Kogyo Co., Ltd., Tokyo, Japan) until they reached nearly 8.5 g. Then 20 fish of a similar size were selected and stocked in each of six 100 L plastic circular tanks with duplication for each dietary treatment. Seawater was supplied at a flow rate of 3 L/min, and the commercial feed was fed to the fish for another two weeks to acclimate them to the experimental conditions. The water temperature during the experiment was 19.0±1.7℃.

Just before the start of the feeding trial, the fish were weighed individually, and eight fish were randomly sampled from a surplus tank after being anesthetized in 0.01% 3-ethyl aminobenzoate methanesulfonic acid (initial fish). In the eight fish sampled, four were used for proximate whole body composition analysis. After taking blood from the other four fish, the hepatopancreas was taken for analyses of proximate composition and metabolic enzyme activity. After measuring the hematocrit, the blood was centrifuged at 4,000 x g for five minutes to separate the plasma. All samples were stored in a freezer at -80℃ until analyzed. The

Table 1. Formulation and proximate analysis of the experimental diets

Diets	CTD[1]	MFD[1]	HFD[1]
P/F[2]	55/11	42/19	36/25
Ingredient (% wet weight)			
Brown fish meal	69.0	53.0	45.0
Waste nori	5.0	5.0	5.0
Pollock liver oil	4.7	13.7	19.3
Gelatinized potato starch	10.5	9.0	5.1
Cellulose	–	8.5	14.8
Constant components[3]	10.8	10.8	10.8
Analysis (dry weight basis)			
Crude protein (N x 6.25 %)	54.5	41.6	35.7
Crude fat (%)	11.4	19.4	24.7
Crude sugar (%)	10.5	9.1	6.9
Phosphorus (%)	2.76	2.35	2.09
Gross energy (MJ/kg diet)	20.1	21.9	22.9

[1] CTD: control diet, MFD: medium fat diet, HFD: high fat diet.

[2] Crude protein : crude fat.

[3] 3.0% krill meal, 5.0% mineral mixture (Ogino et al. 1979), 1.5% vitamin mixture (Sugita et al. 2007a), 1.0% chromic oxide (chromic oxide : cellulose=1 : 1), 0.3% guar gum.

experimental diets were fed to the fish with an initial mean weight of 10.1 g to satiation, three times daily (0900, 1300, and 1700), six days per week for 10 weeks. At the end of the feeding trial, the fish were weighed individually, and seven fish per tank were sampled after one day starvation. In the seven fish sampled, four were used for whole body proximate composition analysis and the other three fish were used for analyses of plasma components and hepatopancreatic proximate composition and enzyme activity.

Another 38 fish (11.0±0.3 g) were selected from the surplus tank and stocked in each of six 50 L tanks for fecal collection (Sugita *et al.* 2007b). Seawater was supplied at a flow rate of 4.5 L/min, and the commercial feed was fed to the fish for another seven days to acclimate them to the experimental conditions. Each experimental diet was fed to the fish to satiation twice in the morning. For the initial six days, the fish were acclimated to the diets and fecal samples were not collected. Feces excreted from 1730 on one day to 0830 the next day, were collected immediately prior to the next satiation feeding each day for six days, pooled, and freeze-dried. The water temperature during fecal collection was 19.0±1.7℃.

Blood Component Analysis

Blood was taken using a heparinized syringe. Hematocrit was measured using microhematocrit tubes. After sealing, the tubes were centrifuged at 10,000 x g for 5 min. The contents of plasma glucose, triglyceride, total cholesterol and total protein were determined using an automatic analyzer (SPOTCHEM SP-4410, Arkray, Kyoto, Japan). Plasma free fatty acid (FFA) and phospholipid were measured using commercial kits (NEFA C Test Wako and Phospholipids C Test Wako, Wako Pure Chemical Industries, Ltd., Osaka, Japan) and free amino acid (FAA) was assayed using dinitrofluorobenzene (Goodwin 1968).

Chemical Component Analysis

Determination of moisture, crude protein, crude fat, and ash in the test diets; feces; whole fish; and hepatopancreas were assayed by 10 h drying at 110℃, semi-micro Kjeldahl method (N x 6.25), diethyl ether extraction, and 5 h heating at 600℃, respectively (Shimeno 1974). Crude sugar content in the diets was determined by measuring simple sugar liberated in boiling 1N sulfuric acid for 4 h, based on the phenol-sulfuric acid method (Shimeno

1974). Phosphorus contents of the diet, feces, and whole fish were determined using ammonium molybdate and ascorbic acid (Lowry and Lopez 1946). The gross energy content of the diets was measured using a CA-4PJ bomb calorimeter (Shimadzu, Kyoto, Japan). The chromium oxide contents of the diets and feces were measured using a spectrophotometer (UVmini 1240, Shimadzu) after the samples were hydrolyzed in nitric acid and perchloric acid (Furukawa and Tsukahara 1966).

Enzyme Activity Analysis

Hepatopancreas samples were homogenized with nine volumes of 3-morpholinopropansulfonic acid buffer (MOPS-buffer; 30 mM, pH7.0) using a Polytron (Kinematica AG, Switzerland, PTA 10S). The homogenate was centrifuged at 12,000 x g for 30 min at 4℃. The resulting supernatant was used to analyze the activities of the following enzymes: phosphofructokinase (PFK, EC 2.7.1.11), glucose-6-phosphatase (G6Pase, EC 3.1.3.9), fructose-1,6-bisphosphatase (FBPase, EC 3.1.3.11), glucose-6-phosphate dehydrogenase (G6PDH, EC 1.1.1.49), 6-phosphogluconate dehydrogenase (6PGDH, EC 1.1.1.44), NADP-isocitrate dehydrogenase (ICDH, EC 1.1.1.42), aspartate aminotransferase (GOT, EC 2.6.1.1) and alanine aminotransferase (GPT, EC 2.6.1.2). These enzyme activities were assayed by the methods described previously (Sugita et al. 2001, 2007a). Pyruvate kinase (PK, EC 2.7.1.40) activity was measured as the rate of NADH reduction (Moon and Johnston 1980). NADP-malate dehydrogenase (MDH, EC 1.1.1.40) activity was determined as the rate of NADP reduction (Ochoa 1995). The absorbance of samples during the measurements of enzyme activities was measured using UVmini 1240 spectrophotometer (Shimadzu) equipped with a thermo-controlled cell positioner CPS-240A (Shimadzu). All the enzyme activities were expressed as μmol of substrate or coenzyme converted per min per 100 g body weight.

Statistical Analysis

The effects of diet on growth, feed utilization, apparent digestibility and biochemical parameters were evaluated by one-way ANOVA and Fisher`s Protected Least Significant Difference test. A probability level of less than 0.05 was considered significant. The statistical analyses were carried out using the Stat View program (SAS Institute, Cary, North Carolina, USA).

Results

Apparent Digestibility

Table 2 shows the apparent digestibility of nutrients in the experimental diets in fingerling red sea bream. The apparent digestibility of crude protein was not significantly different among the dietary groups. The apparent digestibility of crude fat in fish fed the CTD and MFD was higher than the digestibility of the HFD. The apparent phosphorus absorption increased as the dietary fat level increased (i.e., the dietary BFM level decreased).

Growth and Feed Performances of Fish in the Feeding Trial

Growth and feed performance are presented in Table 3. Weight gain (WG), specific growth rate (SGR), and feed efficiency (FE) in fish fed the CTD and MFD were higher than those in the HFD

Table 2. Apparent digestibility of nutrients of the experimental diets in fingerling red sea bream (%)[1]

Diets	CTD[2]	MFD[2]	HFD[2]
P/F[2]	55/11	42/19	36/25
Crude protein	91.8±0.1	91.2±0.1	91.0±0.5
Crude fat	99.1±0.1[b]	99.1±0.1[b]	98.6±0.0[a]
Phosphorus	48.3±0.2[a]	56.2±1.5[b]	59.8±0.1[c]

[1] Values are mean ± SD of two samples. Values in the same row followed by the same letter are not significantly different (P >0.05).

[2] See the footnote of Table 1.

group. On the other hand, feed consumption rate was highest in the HFD group. Protein efficiency ratio (PER) and protein retention in the fat-rich diet (MFD and HFD) groups were higher compared with those of the CTD group. Phosphorus retention was not significantly different among the dietary groups.

Hematological Characteristics

Table 4 shows the results of hematological parameter analyses of fingerling red sea bream. Plasma FFA, total cholesterol and phospholipid concentrations increased as the dietary fat level increased. Plasma glucose, triglyceride, FAA and total protein concentrations were not different among the dietary groups. Hematocrit level was decreased as the dietary P/F decreased; however, it was not significantly influenced by dietary treatment.

Whole body and Hepatopancreas Composition

The whole body composition of the initial and the final fish for each diet are shown in Table 5. Moisture and crude protein contents decreased as dietary fat level increased, whereas crude fat content increased as dietary fat level increased. Ash and phosphorus contents were not significantly different among the dietary groups. Table 5 also shows the hepatopancreas composition of fingerling red sea bream. Hepatopancreatic crude protein and crude fat contents, and hepatosomatic index (HSI) was not significantly influenced by the dietary treatment.

Hepatopancreatic Enzyme Activity

The hepatopancreatic enzyme activities of the initial and final fish are shown in Table 6. The activities of G6Pase, FBPase, G6PDH, 6PGDH, ICDH, and MDH were the highest in the CTD groups, and those activities decreased as the dietary fat level increased. GOT activity of the MFD group was significantly lower than that of the CTD group, whereas GOT activity of the HFD group was not significantly different from that of

Table 3. Growth and feed performances of fingerling red sea bream fed the experimental diets [1]

Diets	CTD[2]	MFD[2]	HFD[2]
P/F[2]	55/11	42/19	36/25
Initial BW (g/fish)	10.10±0.02	10.10±0.01	10.12±0.01
Final BW (g/fish)	45.93±0.48	46.05±1.18	39.62±0.24
WG (%)[3]	355±5.7^{b}	356±11.4^{b}	292±1.9^{a}
SGR (%)[4]	2.16±0.02^{b}	2.17±0.04^{b}	1.95±0.01^{a}
Feed consumption rate (% BW/day)[5]	1.55±0.03^{a}	1.57±0.00^{ab}	1.62±0.02^{b}
FE (%)[6]	112.8±2.5^{b}	109.8±2.4^{b}	97.4±1.8^{a}
PER[7]	2.07±0.05^{a}	2.64±0.06^{b}	2.73±0.05^{b}
Retention (%)			
Protein[8]	38.6±0.3^{a}	47.1±1.0^{b}	45.3±2.4^{b}
Phosphorus[9]	38.7±4.8	41.8±0.1	44.4±4.0

[1] Values are mean ± SD of duplicate tanks. Values in the same row followed by the same letter are not significantly different (P >0.05).

[2] See the footnote of Table 1.

[3] Weight gain (WG) =100 x (final BW-initial BW)/initial BW.

[4] Specific growth rate (SGR) =100 x {ln(final BW) - ln(initial BW)}/rearing period (days).

[5] Expressed as 100 x feed intake/[{(initial BW + final BW)/2} x rearing period (days)].

[6] Feed efficiency (FE) =100 x (final BW - initial BW)/dry feed intake.

[7] Protein efficiency ratio (PER) =(final BW - initial BW)/dry feed protein intake.

[8] Expressed as 100 x {(final BW x final body protein) - (initial BW x initial body protein)} /(total feed intake x feed protein).

[9] Expressed as 100 x {(final BW x final body phosphorus) - (initial BW x initial body phosphorus)} /(total feed intake x feed phosphorus).

Table 4. Hematological characteristics of fingerling red sea bream fed the experimental diets [1]

Diets		CTD[2]	MFD[2]	HFD[2]
P/F[2]		55/11	42/19	36/25
	Initial[3]	Final		
Glucose (mg/100mL)	69	44±3	51±11	49±6
Triglyceride (mg/100mL)	77	86±15	76±3	125±34
FFA (mEq/L)[4]	0.30	0.22±0.02[a]	0.28±0.02[ab]	0.37±0.07[b]
Total cholesterol (mg/100mL)	166	163±16[a]	266±35[b]	255±39[ab]
Phospholipid (mg/100mL)	458	460±36[a]	674±47[b]	721±14[b]
FAA (mg/100mL)[5]	45.4	54.2±6.9	46.6±0.8	45.3±1.5
Total protein (g/100mL)	2.3	2.9±0.2	2.9±0.2	2.7±0.3
Hematocrit (%)	24.8	25.5±1.9	23.8±3.7	19.9±0.8

[1] Values are mean ± SD of duplicate tanks (3 fish/tank). Values in the same row followed by the same letter are not significantly different (P >0.05).

[2] See the footnote of Table 1.

[3] Average values of 4 fish.

[4] FFA, Free fatty acid.

[5] FAA, Free amino acid.

Table 5. Proximate composition of whole body and hepatopancreas of fingerling red sea bream fed the experimental diets

Diets		CTD[1]	MFD[1]	HFD[1]
P/F[1]		55/11	42/19	36/25
	Initial[2]	Final		
Whole body[3]				
Moisture (%)	75.9	71.6±0.4[b]	69.7±0.4[a]	69.2±0.0[a]
Crude protein (N x 6.25 %)	17.1	17.4±0.3[b]	16.9±0.1[ab]	16.2±0.2[a]
Crude fat (%)	2.4	5.7±0.6[a]	9.0±0.1[b]	9.6±0.1[b]
Ash (%)	4.9	5.0±0.0	4.8±0.2	5.0±0.1
Phosphorus (%)	0.97	0.92±0.07	0.88±0.01	0.94±0.04
Hepatopancreas[4]				
Crude protein (N x 6.25 %)	10.7	14.8±0.2	15.5±0.4	15.3±2.0
Crude fat (%)	4.3	5.6±1.3	7.5±2.1	6.2±1.1
HSI (%)[5]	1.30	1.37±0.02	1.31±0.19	1.25±0.04

[1] See the footnote of Table 1.

[2] See the footnote of Table 4.

[3] Values are mean ± SD of duplicate tanks (pooled samples, 4 fish/sample). Values in the same row followed by the same letter are not significantly different (P >0.05).

[4] Values are mean ± SD of duplicate tanks (pooled samples, 3 fish/sample). Values in the same row followed by the same letter are not significantly different (P >0.05).

[5] Hepatosomatic index (HSI) = 100 x hepatopancreas weight/BW. Values are mean ± SD of duplicate tanks (3 fish/tank).

Table 6. Hepatopancreatic metabolic enzyme activities of fingerling red sea bream fed the experimental diets[1]

Diets		CTD[2]	MFD[2]	HFD[2]
P/F[2]		55/11	42/19	36/25
		(μmol/min/100g body weight)		
	Initial[3]	Final		
PFK[4]	2.42	3.20±0.40	2.95±0.44	2.40±0.50
PK[4]	13.0	14.9±0.4	15.0±1.4	14.1±1.4
G6Pase[4]	5.69	8.54±0.36^{b}	6.92±0.38^{ab}	5.46±0.96^{a}
FBPase[4]	5.54	5.42±0.04^{b}	3.52±0.29^{a}	3.34±0.36^{a}
G6PDH[4]	23.1	41.1±0.2^{c}	12.9±1.1^{b}	8.4±1.0^{a}
6PGDH[4]	9.4	19.6±0.1^{c}	11.9±1.2^{b}	6.9±0.4^{a}
ICDH[4]	41.0	44.0±1.6^{b}	35.7±2.6^{a}	37.0±1.1^{a}
MDH[4]	1.54	4.66±0.41^{b}	1.65±0.06^{a}	1.25±0.18^{a}
GOT[4]	42.2	78.2±2.6^{b}	58.0±0.5^{a}	64.9±7.4^{ab}
GPT[4]	20.3	22.1±0.7	19.9±6.0	24.1±0.2

[1] Values are mean ± SD of duplicate tanks (3 fish/tank). Values in the same row followed by the same letter are not significantly different (P >0.05).

[2] See the footnote of Table 1.

[3] See the footnote of Table 4.

[4] PFK: Phosphofructokinase, PK: Pyruvate kinase, G6Pase: Glucose-6-phosphatase, FBPase: Fructose-1,6-bisphosphatase, G6PDH: Glucose-6-phosphate dehydrogenase, 6PGDH: 6-Phosphogluconate dehydrogenase, ICDH: NADP-Isocitrate dehydrogenase, MDH: NADP-Malate dehydrogenase, GOT: Aspartate aminotransferase, GPT: Alanine aminotransferase.

the CTD group. The activities of PFK, PK, and GPT were not significantly different among the dietary groups.

Discussion

In our previous report on fingerling red sea bream (Sugita *et al*. 2007a), growth and feed performances decreased as the dietary fat level increased (i.e., dietary protein level decreased). Therefore, the authors concluded that fingerling red sea bream did not effectively utilize the high fat diet. In the present experiment, crude protein and crude fat contents of the experimental diets were similar to those in the previous experiment. However, growth and feed performances of the MFD group were equal to those of the CTD group. The difference between the present and previous experiments was just the waste nori diet supplementation. The present experiment clearly showed that supplemental waste nori enhanced the utilization of the higher fat and lower protein diet. Since dietary protein contents of MFD and HFD were lower than CTD, PER and protein retention of the MFD and HFD groups were higher than the CTD group. WG, SGR, FE and apparent crude fat digestibility were not different between the CTD group and the MFD groups, whereas those were lowest in the HFD group. These results suggest that waste nori supplementation can improve utilization of high fat and low protein diets to some degree in fingerling red sea bream. The supplemental effect was not observed in fish fed the high fat and low protein diet (HFD), because digestible energy intake for fish fed the HFD might have been reduced due to the decrease of apparent fat digestibility.

Lipogenic enzymes (G6PDH, 6PGDH, ICDH and MDH) play an important role in the generation of reduced form nicotinamide adenine dinucleotide phosphate (NADPH) for lipid synthesis in the liver (Kheyyali et al. 1989). In the present experiment, the activities of these enzymes significantly decreased in the MFD and HFD groups. On the

other hand, the concentrations of total cholesterol, phospholipid and FFA in the plasma, and the content of crude fat in the whole body increased as the dietary fat level increased. These results indicate that the accumulated body fat components did not originate mainly from dietary protein and carbohydrate but from dietary fat. Similar depression in hepatic lipogenic enzyme activity and increase in fat deposition by a high fat and low protein diet was reported in yellowtail (Shimeno *et al.* 1980; Shimeno *et al.* 1981), carp *Cyprinus carpio* (Kheyyali *et al.* 1989), tilapia (Shimeno *et al.* 1993) and Japanese flounder (Kikuchi *et al.* 2000).

Although the GOT activity of the HFD group was not different from the CTD group, the activity of the MFD group was significantly lower than the CTD group. The response of GOT activity of the MFD group meant the depression of breakdown from protein to amino acid. These results suggest that protein-sparing effect by fat was demonstrated with fish fed the MFD but the effect was limited compared to the case in rainbow trout, yellowtail and so on as mentioned in the introduction. Judging from the growth performance and hepatopancreatic enzyme activities of the present experiment and our previous report (Sugita et al. 2007a), the dietary fat content can be increased from 11% to 19%; i.e., dietary protein content can be effectively reduced from 55% to 42%, by supplementing 5% waste nori in fingerling red sea bream diets.

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