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Effects of Dietary Nutrient Composition on Compensatory Growth of Grower Olive Flounder *Paralichthys olivaceus* under Different Feeding Regimes at Suboptimal Temperature

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The effects of dietary nutrient composition on compensatory growth of grower olive flounder (*Paralichthys olivaceus*) under different feeding regimes at suboptimal temperature were determined. Four hundred five fish weighing 271.2 g were distributed into 27 300 L flow-through tanks (15 fish per tank). Nine treatments were prepared in triplicate: fish were hand-fed with control (C) diet for 12 weeks (12WF-C); four groups of fish were starved for 1 week and then fed C, high-protein (HP), high-lipid (HL), or combined high-protein and high-lipid (HPL) diets for 11 weeks; these groups are referred to as 11WF-C, 11WF-HP, 11WF-HL, and 11WF-HPL, respectively. Four other groups of fish were starved for 2 weeks and then fed C, HP, HL, and HPL diets for 10 weeks; these groups are referred to as 10WF-C, 10WF-HP, 10WF-HL, and 10WF-HPL, respectively. Weight gain and specific growth rate of fish from 12WF-C group were greater than those of fish from 11WF-C, 11WF-HP, 11WF-HL, 10WF-C, 10WF-HP, and 10WF-HL groups, but not different from those of fish from 11WF-HPL and 10WF-HPL groups. Feed-efficiency ratio of fish from 12WF-C, 11WF-HP, 11WF-HPL, 10WF-HL, and 10WF-HPL groups was higher than that of fish in 11WF-C, 11WF-HL, 10WF-C, and 10WF-HP groups. The results of this study demonstrated that grower olive flounder subjected to 1- or 2-week feed deprivation were able to achieve full compensatory growth at suboptimal temperature only when fed HPL diet.

Key words: Olive flounder (*Paralichthys olivaceus*), Compensatory growth, Dietary nutrient composition, Grower, Suboptimal temperature

Introduction

Application of compensatory growth is an effective, practical fish-culture method in terms of improving feeding activity and accelerating the growth rate after refeeding; it allows savings on feed and labor costs while the fish are starved. Compensatory growth of fish varies depending on species, fish size, feed allowance, dietary nutrient content, duration of the feeding trial, feeding regime, physiological/nutritional state, and social factors (Jobling et al., 1994; Rueda et al., 1998; Gaylord and Gatlin, 2000; Hayward et al., 2000; Gaylord and Gatlin, 2001; Gaylord et al., 2001; Ali et al., 2003; Huang et al., 2008; Cho and Heo, 2011).

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The optimum temperature condition for growth of olive flounder (*Paralichthys olivaceus*), one of the most commercially important marine finfish for aquaculture in Eastern Asia, has been reported to be 20-25°C (Iwata et al., 1994). However, the temperature frequently rises above 30°C in summer and falls below 10°C in winter in this area. Olive flounder overwinters to grow to marketable size and grows slowly at low or suboptimal temperature due to its slow metabolism. Under this condition, compensatory growth can be one of the most effective fish culture techniques adopted by fish farmers to regain accelerated growth of the fish.

We reported that juvenile olive flounder subjected to 2 weeks of feed deprivation were able to achieve full compensatory growth in 8-week trials (Cho, 2005; Cho et al., 2006a). In addition, juvenile olive flounder subjected to temperatures as low as 8.5°C for 10 days achieved full compensatory growth when grown at 22.0°C for the following 30 days (Huang et al., 2008).

Lipid in the fish body was primarily utilized as the energy source for maintenance of basal metabolism and survival while fasting (Rueda et al., 1998; Gaylord and Gatlin, 2000), and the body weight of the fish decreased in proportion to feed deprivation (Cho, 2005; Cho et al., 2006a). In addition, Gaylord and Gatlin (2001) showed that increasing dietary protein levels from 32% to 37% achieved a desirable effect on cumulative weight gain of channel catfish (*Ictalurus punctatus*). Therefore, there is a body of evidence indicating that manipulation of dietary nutrient composition can affect the compensatory growth of fish.

A variety of feeding trials with juvenile olive flounder have been reported (Kikuchi, 1999; Lee et al., 2000; Alam et al., 2002; Kim et al., 2005; Cho et al., 2006b; Kim et al., 2006; Lee et al., 2006); on the other hand, only a few feeding trials (Kim et al., 2009, 2010) at suboptimal temperatures involving grower fish have been performed to date. Kim et al. (2010) reported that the optimum dietary protein and lipid levels for grower olive flounder averaging 255 g were 45% and 14%, respectively, at 15.5°C and were equivalent to an estimated energy level of 17.4 kJ/g diet. No study on compensatory growth of grower olive flounder has been performed yet. In this study, therefore, effects of dietary nutrient composition on compensatory growth of grower olive flounder with different feeding regimes at suboptimal temperature were determined.

Materials and Methods

Fish and experimental conditions

Grower olive flounder were purchased from a private farm and acclimated to the experimental conditions for 4 weeks. During the acclimation period, the fish were fed commercial extruded pellets containing 50% crude protein and 10% crude lipid once a day at a ratio of 0.5% biomass. Four hundred five fish (initial body weight: 271.2 ± 0.14 g) were randomly distributed into 27 300 L flow-through tanks (water volume: 250 L; 15 fish per tank). The water source was sand-filtered natural seawater, and aeration was supplied to each tank. The flow rate of water into each tank was 8.6 L/min. Water temperature ranged from 9.8°C to 16.9°C (mean ± SD: 12.9 ± 1.96°C), and the photoperiod followed the natural condition.

Preparation of the experimental diets

The ingredients and nutrient composition of the experimental diets are given in Table 1. Four experimental diets were prepared: control (C), high protein (HP), high lipid (HL), and combined high protein and high lipid (HPL). The crude protein and lipid content of the C diet was 48.5% and 9.1%, respectively. The crude protein content of the HP diet and the crude lipid content of the HL diet were increased to 53.4% and 14.1% by supplementation with fish meal and soybean oil at the expense of wheat flour, respectively. The combined crude protein and crude lipid content of the HPL diet was increased to 53.1% and 14.1%, respectively. Ingredients of the experimental diets were well mixed with water at the ratio of 7:3 and pelletized with a pellet extruder. The experimental diets were dried overnight at room temperature and stored at -20°C until use.

Table 1. Ingredients and nutrient composition (%, dry matter basis) of the experimental diets

	Experimental diets			
	C	HP	HL	HPL
Ingredients (%)				
Fishmeal	30	38	30	38
Casein	10	10	10	10
Dehulled soybean	13	13	13	13
Corn gluten meal	4	4	4	4
Wheat flour	34	26.6	28.8	21.5
Soybean oil	4.9	4.3	10.1	9.4
CMC	2	2	2	2
Vitamin premix[1]	1	1	1	1
Mineral premix[2]	1	1	1	1
Choline	0.1	0.1	0.1	0.1
Nutrient (%, DM basis)				
Crude protein	48.5	53.4	48.3	53.1
Crude lipid	9.1	9.2	14.1	14.1
Ash	5.9	6.8	5.8	6.8
Fiber	3.0	2.9	3.0	2.9
NFE[3]	33.5	27.7	28.8	23.1
EE (kJ/g diet)[4]	17.6	17.5	18.7	18.5

[1]Vitamin premix and [2]Mineral premix were same as Cho et al. (2007)'s study.
[3]NFE calculated by differences [100 - (crude protein + crude lipid + ash + fiber)].
[4]Estimated energy (EE) calculated based on 16.7 kJ/g for protein and carbohydrate, and 37.7 kJ/g for lipid (Garling and Wilson, 1976).

Design of the feeding trial

Nine treatments were prepared in triplicate: fish in the control group were hand-fed the C diet to satiation twice a day, 7 days a week, for 12 weeks (12WF-C); four groups of fish were starved for 1 week and then fed C, HP, HL, and HPL diets to satiation twice a day, 7 days a week, for 11 weeks; these groups are referred to as 11WF-C, 11WF-HP, 11WF-HL, and 11WF-HPL, respectively. Four other groups of fish were starved for 2 weeks and then fed C, HP, HL, and HPL diets to satiation twice a day, 7 days a week, for 10 weeks; these groups are referred to as 10WF-C, 10WF-HP, 10WF-HL, and 10WF-HPL, respectively.

Analysis of chemical composition of the experimental diets and fish

Proximate analysis of the experimental diets was performed. At the beginning and end of the trial, five fish were sampled and sacrificed for proximate analysis. Crude protein was determined by the Kjeldahl method (Kjeltec 2100 Distillation Unit, Foss Tecator, Hoganas, Sweden), crude lipid was deter-mined using an ether-extraction method, moisture was determined by oven drying at 105°C for 24 h, fiber was determined using an automatic analyzer (Fibertec, Tecator, Sweden), and ash was determined using a muffle furnace at 550°C for 4 h; all analyses were carried out according to standard AOAC methods (1990).

Statistical analysis

One-way ANOVA and Duncan`s multiple-range test (Duncan, 1955) were used to analyze the significance ($P<0.05$) of the difference among the means of treatments by using SAS Program Version 9.1 (SAS Institute, Cary, NC, USA).

Results and Discussion

Survival of fish ranged from 95.6% to 100% and was not significantly ($P>0.8$) different among groups (Table 2). However, weight gain and specific growth rate (SGR) of grower olive flounder in the 12WF-C group were significantly ($P<0.008$ and $P<0.006$, respectively) higher than those of fish in the 11WF-C, 11WF-HP, and 11WF-HL groups, which were deprived of feed for 1 week, and those of fish in the 10WF-C, 10WF-HP, and 10WF-HL groups, which were deprived of feed for 2 weeks, but not significantly ($P>0.05$) different from those of fish in the 11WF-HPL and 10WF-HPL groups. This indicated that full compensatory growth could be achieved in grower olive flounder fed the HPL diet after 1- or 2-week feed deprivation at suboptimal temperature, but not in fish fed the C, HP, or HL diets.

The dietary optimum protein and lipid levels for growth of grower olive flounder were 45% and 14%, respectively, which were equivalent to an estimated energy level of 17.4 kJ/g diet when fish averaging 255 g were fed 40, 45, and 50% protein diets with two lipid levels (7 and 14%) to satiation twice daily at a mean temperature of 15.5°C for 14 weeks (Kim et al., 2010). Supplementation of both protein and lipid levels in the diet (HPL diet) achieved compensatory growth of grower fish in this study, partially agreeing with Cho and Heo’s (2011) study reporting that a diet containing a combined protein and lipid level effectively improved compensatory growth of juvenile olive flounder at optimum temperature, but a diet containing the combined protein and lipid level with 5% amino acid supplementation did not. Increased dietary protein levels from 32% to 37% might have had a desirable effect on cumulative weight gain of channel catfish, but increased dietary lipid levels did not, although the authors concluded that dietary manipulation had not augmented the growth rate of the fish in a 6-week feeding trial (Gaylord and Gatlin, 2001).

Poorer weight gain and specific growth rate (SGR) of grower olive flounder in the 11WF-C and 10WF-C groups than in the 12WF-C group in this study contradicted earlier studies (Cho, 2005; Cho et al., 2006a) showing that juvenile olive flounder subjected to 2-week feed deprivation were able to achieve full compensatory growth in 8-week trials. This probably resulted from differences in fish size, water temperature, and/or the duration of feeding trials among the studies.

SGR (0.22%/day) of grower olive flounder in the 12WF-C treatment in this study was comparable to that (0.20%/day) of grower fish averaging 287 g fed a commercial diet to satiation once a day at a mean temperature of 12.1°C for 15 weeks (Kim et al., 2009) and within values (0.18-0.56%/day) obtained from grower fish averaging 255 g fed diets containing various protein and lipid levels daily at 15.5°C for 14 weeks (Kim et al., 2010). Therefore, growth of grower olive flounder in the 12WF-C treatment in this study seemed to be relatively well achieved.

Feeding rate (FR) ($P>0.3$), condition factor (CF) ($P>0.5$), and hepatosomatic index (HSI) ($P>0.3$) of fish were not significantly different among treatments (Table 3). However, given that 13.6% lower FR in the 10WF-HPL treatment produced 6.6% less weight gain compared to that of fish in the 12WF-C treatment in this study, these two variables must be evaluated to determine the feasibility of compensatory growth of grower olive flounder under these experimental conditions. However, unlike in this study, HSI was a good index to indicate com-

Table 2. Survival (%), weight gain (g/fish) and specific growth rate (SGR) of grower olive flounder fed the diets containing various nutrient contents with different feeding regime for 12 weeks

Treatments	Initial weight of fish (g/fish)	Final weight of fish (g/fish)	Survival (%)	Weight gain (g/fish)	SGR[1]
12WF-C	271.1 ± 0.17	326.8 ± 17.00	95.6 ± 4.44	55.6 ± 16.98^{a}	0.22 ± 0.061^{a}
11WF-C	271.1 ± 0.11	281.6 ± 1.66	100.0 ± 0.00	10.5 ± 1.77^{c}	0.05 ± 0.007^{c}
11WF-HP	271.3 ± 0.11	299.9 ± 3.08	100.0 ± 0.00	28.6 ± 3.19^{bc}	0.12 ± 0.013^{bc}
11WF-HL	271.3 ± 0.18	287.2 ± 3.58	95.6 ± 4.44	15.9 ± 3. 69^{bc}	0.07 ± 0.015^{bc}
11WF-HPL	271.1 ± 0.20	310.4 ± 7.91	100.0 ± 0.00	39.3 ± 7.78^{ab}	0.16 ± 0.030^{ab}
10WF-C	271.3 ± 0.16	284.8 ± 6.75	95.6 ± 2.22	13.5 ± 6.74^{c}	0.06 ± 0.028^{c}
10WF-HP	271.1 ± 0.23	283.7 ± 2.33	97.8 ± 2.22	12.6 ± 2.30^{c}	0.05 ± 0.010^{c}
10WF-HL	270.9 ± 0.04	300.0 ± 4.37	97.8 ± 2.22	29.1 ± 4.40^{bc}	0.12 ± 0.018^{bc}
10WF-HPL	271.4 ± 0.06	305.3 ± 2.36	97.8 ± 2.22	33.9 ± 2.41^{abc}	0.14 ± 0.009^{abc}
P-value			$P > 0.8$	$P < 0.008$	$P < 0.006$

Values (mean of triplicate ± SE) in the same column sharing a common superscript are not significantly different (P>0.05).
[1]Specific growth rate (SGR, %/day) = (Ln final weight of fish - Ln initial weight of fish) × 100/days of feeding trial.

Table 3. Feeding rate (FR), feed efficiency ratio (FER), protein efficiency ratio (PER), condition factor (CF) and hepatosomatic index (HSI) of grower olive flounder fed the diets containing various nutrient contents with different feeding regime for 12 weeks

Treatments	FR[1]	FER[2]	PER[3]	CF[4]	HSI[5]
12WF-C	0.22 ± 0.042	0.89 ± 0.070^{a}	1.94 ± 0.200^{a}	0.94 ± 0.040	1.36 ± 0.098
11WF-C	0.17 ± 0.001	0.26 ± 0.042^{b}	0.54 ± 0.087^{c}	0.95 ± 0.063	1.30 ± 0.175
11WF-HP	0.17 ± 0.002	0.70 ± 0.066^{a}	1.31 ± 0.124^{ab}	0.92 ± 0.034	1.17 ± 0.199
11WF-HL	0.17 ± 0.008	0.36 ± 0.081^{b}	0.82 ± 0.165^{bc}	0.85 ± 0.020	0.97 ± 0.181
11WF-HPL	0.18 ± 0.006	0.87 ± 0.141^{a}	1.64 ± 0.265^{a}	0.87 ± 0.040	1.26 ± 0.165
10WF-C	0.17 ± 0.003	0.35 ± 0.148^{b}	0.69 ± 0.319^{c}	0.88 ± 0.027	1.00 ± 0.161
10WF-HP	0.17 ± 0.001	0.34 ± 0.054^{b}	0.62 ± 0.113^{c}	0.88 ± 0.037	1.18 ± 0.180
10WF-HL	0.18 ± 0.003	0.65 ± 0.103^{a}	1.39 ± 0.180^{ab}	0.91 ± 0.023	1.53 ± 0.079
10WF-HPL	0.19 ± 0.002	0.74 ± 0.060^{a}	1.41 ± 0.103^{ab}	0.92 ± 0.008	1.37 ± 0.079
P-value	$P > 0.3$	$P < 0.0007$	$P < 0.0007$	$P > 0.5$	$P > 0.3$

Values (mean of triplicate ± SE) in the same column sharing a common superscript are not significantly different (P>0.05).
[1]Feeding rate (FR, %) = (100 × feed consumption of fish)/[(initial weight of fish + final weight of fish)/2]/feeding day.
[2]Feed efficiency ratio (FER) = Wight gain of fish/feed consumed.
[3]Protein efficiency ratio (PER) = Weight gain of fish/protein consumed.
[4]Condition factor (CF) = Body weight (g) × 100/total length (cm)^3.
[5]Hepatosomatic index (HSI) = Liver weight × 100/body weight.

pensatory growth of fish in previous studies (Gaylord and Gatlin, 2000; Cho, 2005).

Feed efficiency ratio (FER) of grower olive flounder from 12WF-C, 11WF-HP, 11WF-HPL, 10WF-HL, and 10WF-HPL groups was significantly (P<0.0007) higher than those of fish from 11WF-C, 11WF-HL, 10WF-C, and 10WF-HP groups in this study. Protein efficiency ratio (PER) of fish from 12WF-C and 11WF-HPL groups was significantly (P<0.0007) higher than those of fish from 11WF-C, 11WF-HL, 10WF-C, and 10WF-HP groups, but not significantly (P>0.05) different from those of fish from 11WF-HP, 10WF-HL, and 10WF-HPL groups. Similarly, improved FER and/or PER of fish achieving full compensatory growth was observed in the previous studies (Gaylord and Gatlin, 2001; Cho, 2005; Oh et al., 2007; Huang et al., 2008; Cho and Heo, 2011).

No difference in FR, but significantly higher weight gain, SGR, FER, and PER of fish from 12WF-C group compared to those of fish from 11WF-C and 10WF-C groups in this study indicated that the poor performance of fish from 11WF-C and 10WF-C groups did not directly result from less feed consumption. Nevertheless, hyperphagia was a common phenomenon to explain compensatory growth of fish in the several studies (Rueda et al., 1998; Ali et al., 2003; Cho et al., 2006a; Oh et al., 2007; Huang et al., 2008).

Table 4. Chemical composition (%, wet weight) of the whole body excluding the liver and the liver of grower olive flounder at the end of the 12 week trial

Treatments	Whole body excluding liver			
	Moisture	Crude protein	Crude lipid	Ash
12WF-C	72.3 ± 0.03	18.5 ± 0.07[abc]	3.3 ± 0.10	3.1 ± 0.03
11WF-C	73.0 ± 1.05	17.7 ± 0.32[c]	3.1 ± 0.20	3.4 ± 0.21
11WF-HP	72.7 ± 0.30	19.1 ± 0.45[ab]	3.5 ± 0.18	3.3 ± 0.14
11WF-HL	72.4 ± 0.23	19.5 ± 0.21[a]	4.1 ± 0.23	3.6 ± 0.22
11WF-HPL	72.4 ± 0.56	18.4 ± 0.38[abc]	3.5 ± 0.20	3.3 ± 0.23
10WF-C	74.4 ± 1.49	18.4 ± 0.42[abc]	3.5 ± 0.20	3.3 ± 0.03
10WF-HP	71.9 ± 0.45	19.5 ± 0.67[a]	3.4 ± 0.29	3.5 ± 0.15
10WF-HL	73.1 ± 0.38	17.9 ± 0.19[bc]	3.5 ± 0.07	3.2 ± 0.06
10WF-HPL	72.3 ± 0.78	18.2 ± 0.41[abc]	3.5 ± 0.18	3.2 ± 0.12
P-value	$P > 0.4$	$P < 0.03$	$P > 0.2$	$P > 0.6$

Treatments	Liver		
	Moisture	Crude protein	Crude lipid
12WF-C	66.7 ± 2.61	9.3 ± 0.09	16.3 ± 0.44
11WF-C	63.2 ± 1.58	10.0 ± 0.20	15.6 ± 0.72
11WF-HP	65.8 ± 0.92	10.0 ± 0.39	15.6 ± 0.68
11WF-HL	69.4 ± 1.85	10.1 ± 0.27	15.5 ± 0.44
11WF-HPL	68.4 ± 2.45	9.1 ± 0.23	16.6 ± 0.57
10WF-C	70.0 ± 1.78	9.4 ± 0.06	15.7 ± 0.72
10WF-HP	66.6 ± 1.28	9.7 ± 0.44	16.7 ± 0.49
10WF-HL	68.8 ± 2.36	9.8 ± 0.55	16.7 ± 0.60
10WF-HPL	68.8 ± 1.69	10.1 ± 0.32	16.6 ± 0.32
P-value	$P > 0.4$	$P > 0.3$	$P > 0.7$

Values (mean of triplicate ± SE) in the same column sharing a common superscript are not significantly different ($P>0.05$).

Moisture ($P>0.4$), crude lipid ($P>0.2$), and ash ($P>0.6$) content of whole body excluding liver and moisture ($P>0.4$), crude protein ($P>0.3$), and crude lipid ($P>0.7$) content of liver in grower olive flounder were not significantly different among groups (Table 4). However, crude protein content of whole body of fish excluding liver of fish from 11WF-HL and 10WF-HP groups was significantly ($P<0.03$) higher than those of fish from 11WF-C and 10WF-HL groups, but was not significantly different from those of fish from 12WF-C, 11WF-HP, 11WF-HPL, 10WF-C, and 10WF-HPL groups. Chemical composition of whole body was not affected by either feeding regime (Gaylord and Gatlin, 2000; Kim et al., 2005; Cho et al., 2006a) or dietary nutrient content (Gaylord and Gatlin, 2001; Kim et al., 2006) in the previous studies.

The results of this study demonstrate that grower olive flounder subjected to 1- or 2-week feed deprivation were able to achieve full compensatory growth at suboptimal temperature only when they were fed a combined high-protein and high-lipid (HPL) diet.

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