# Could a Manipulation of Dietary Nutrient Contents Including Phosphorous Affect Compensatory Growth of Juvenile Olive Flounder *Paralichthys olivaceus*?

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## Abstract

I hypothesized that the manipulation of dietary nutrient contents including phosphorous could affect compensatory growth of juvenile olive flounder, *Paralichthys olivaceus*. Thirty fish averaging 34.8 g per tank were randomly chosen and distributed into 15 flow-through 180-L tanks. Three experimental diets were prepared: the control (C) diet, high protein and lipid (HPL) diet, and HPL diet with supplementation of calcium phosphate-monobasic (HPLP). Five treatments were prepared in triplicate: fish were hand-fed daily with the C diet twice a day for 8 weeks (C-8W); fish were starved for 1 week, and then fed with the HPL or HPLP diets twice a day for 7 weeks, and referred to as HPL-7W and HPLP-7W, respectively; and fish were starved for 2 weeks, and then fed with the HPL or HPLP diets twice a day for 6 weeks, and referred to as HPL-6W and HPLP-6W, respectively. The body weight of fish with C-8W, HPL-7W and HPLP-7W treatments was higher than fish with HPL-6W and HPLP-6W treatments on week 2, 4 and 6 after an initiation of the trial. At the end of the 8-week trial, fish with HPLP-7W and HPLP-7W treatments overcompensated, as compared to fish with C-8W treatment. Full compensation was not achieved in fish subjected to the 2-week feed deprivation (HPL-6W and HPLP-6W treatments). Overall feed intake by fish was proportional to weeks of feeding. Feed conversion ratio of fish with HPLP-7W, HPL-6W and HPLP-6W treatments was higher than fish with C-8W treatment. The study showed that dietary supplementation of protein and lipid resulted in overcompensation of juvenile olive flounder subjected to a 1-week feed deprivation, but not a 2-week feed deprivation. Additionally, dietary supplementation of phosphorous did not further improve compensatory growth of fish.

Key words: Paralichthys olivaceus, Olive flounder, Compensatory growth, Dietary nutrient content, Phosphorous

# Introduction

Compensatory growth of fish is rapid or faster than normal growth rate of fish resulting from refeeding after fasting or undernutrition. Compensatory growth of fish varies depending on fish species, fish size, water temperature, feed allowance, dietary nutrient content, duration of feeding trial and feeding regimes (Bilton and Robins, 1973; Rueda et al., 1998; Wang et al., 2000; Gaylord and Gatlin, 2001; Zhu et al., 2001; Cho, 2005; Cho et al., 2006; Oh et al., 2007, 2008; Bavčevič et al., 2010).

In our earlier studies (Cho, 2005; Cho et al., 2006), juvenile olive flounder *Paralichthys olivaceus* could reach full compensatory growth after being subjected to a 2-week feed deprivation in the 8-week trials. A later study showed an increased dietary crude protein and lipid content from 50% to 55% and 10% to 14%, respectively, led to overcompensation and full compensation of juvenile olive flounder fed for 7 and 6 weeks

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\*Corresponding Author E-mail: chosunh@hhu.ac.kr after 1-week and 2-week feed deprivation, respectively, as compared to fish fed on the 50% crude protein diet with 10% lipid daily for 8 weeks (Cho and Heo, 2011). Manipulation of dietary nutrient composition effectively improved compensatory growth of channel catfish *Ictalurus punctatus* fed for 11 days after experiencing short-term (3-day) feed deprivation in three cycles (Gaylord and Gatlin, 2001).

Ibáňez et al. (2012) recently reported that food availability and type between different grow-out facilities resulted in compensatory growth of juvenile cyprinid roach *Rutilus rutilus* (L.) causing scale shape differences. Additionally, Cho and Heo (2011) reported that fish achieving compensatory growth became fatter. Lovell (1988) explained that approximately 85% to 90% of the phosphorous in fish is in bone and scales. Therefore, dietary supplementation of phosphorous content may affect the process of fish achieving compensatory growth.

I hypothesized that the manipulation of dietary nutrient contents including phosphorous could affect compensatory growth of juvenile olive flounder.

### **Materials and Methods**

#### Fish and experimental conditions

Juvenile olive flounder were purchased from a private hatchery and acclimated to the experimental conditions for 1 week. During the acclimation period, fish were hand-fed with the commercial feed twice a day. Thirty fish (initial body weight of fish 34.8 g) per tank were randomly chosen and distributed into 15 flow-through 180-L tanks (water volume 150 L). The water source was the sand-filtered natural seawater and aeration supplied to each tank. The flow rate of water into each tank was 10 L/min. Water temperature ranged from  $18.5^{\circ}$ C to  $23.6^{\circ}$ C (mean  $\pm$  SD,  $22.4 \pm 1.05^{\circ}$ C) and photoperiod was left in a natural condition.

#### Preparation of the experimental diets

Three experimental diets were prepared (Table 1): the control (C) diet, high protein and lipid (HPL) diet, and HPL diet with supplementation of calcium phosphate-monobasic (HPLP). The C diet was prepared to satisfy dietary nutrient requirements of olive flounder (Lee et al., 2000, 2002). Additionally, the HPL diet was prepared by an increase of fishmeal and soybean oil instead of wheat flour and cellulose. The HPLP diet was prepared by an inclusion of 0.5% calcium phosphate-monobasic into the HPL diet instead of cellulose.

Fishmeal, soybean meal and corn gluten meal, and soybean oil were used as the protein and lipid sources, respectively. Ingredients of the experimental diets were well mixed with water at a 7:3 ratio and pelletized by a pellet-extruder. The experimental diets were dried overnight at room temperature and stored at  $-20^{\circ}$ C until use.

#### Design of the feeding trial

Five treatments were prepared in triplicate: fish were handfed daily with the C diet twice a day (09:00 and 17:00), 7 days a week, for 8 weeks (C-8W) were designated as a control group; fish starved for 1 week, and then fed with the HPL or HPLP diets twice a day, 7 days a week, for 7 weeks were referred to as HPL-7W and HPLP-7W, respectively; and fish starved for 2 weeks, and then fed with the HPL or HPLP diets twice a day, 7 days a week, for 6 weeks were referred to as HPL-6W and HPLP-6W, respectively. Fish were weighed collectively from each tank in bi-weekly intervals throughout the 8-week trial.

#### **Statistical analysis**

One-way ANOVA and Duncan's multiple range test (Duncan, 1955) were used to analyze the significance of the differences among the means of treatments by using SAS program version 9.3 (SAS Institute, Cary, NC, USA).

## **Results and Discussion**

Survival of juvenile olive flounder ranged from 91.1% to 97.8% and was not significantly (P > 0.05) different among treatments (Table 2).

No difference in body weight of juvenile olive flounder was

Table 1. Chemical composition of the experimental diets (%, DM basis)

	<b>Experimental diets</b>			
Composition (%)	Control (C) diet	HPL diet	HPLP diet	
Fishmeal	48.5	57.2	57.2	
Dehulled soybean meal	9.8	9.8	9.8	
Corn gluten meal	18.3	18.3	18.3	
Wheat flour	12.4	7.9	7.9	
Soybean oil	3.9	7.9	7.9	
Calcium phosphate-monobasic*			0.5	
Cellulose	9	1	0.5	
$CMC^{\dagger}$	3	3	3	
Vitamin premix <sup>‡</sup>	1	1	1	
Mineral premix <sup>§</sup>	1	1	1	
Choline	0.1	0.1	0.1	
Nutrients (%, DM basis)				
Crude protein	50.3	55.4	55.3	
Crude lipid	9.6	14.3	14.3	

HPL, high protein and lipid; HPLP, HPL diet with supplementation of calcium phosphate-monobasic.

<sup>\*</sup>Calcium phosphate-monobasic was purchased from Duksan Pure Chemical Co. Ltd. (Ansan, Korea), <sup>†</sup>Carboxymethyl cellulose (CMC) was purchased from Sigma Chemical Co. (MO, USA), <sup>†</sup>Vitamin premix and <sup>§</sup>Mineral premix were same as Cho and Heo (2011).



**Fig. 1.** Change in body weight (g/fish) of olive flounder *Paralichthys olivaceus* fed the experimental diets with different feeding regime (means of triplicate  $\pm$  SE). C-8W, fish were hand-fed daily with the C diet twice a day for 8 weeks; HPL-7W and HPLP-7W, fish were starved for 1 week, and then fed with the HPL or HPLP diets twice a day for 7 weeks; HPL-6W and HPLP-6W, fish were starved for 2 week, and then fed with the HPL or HPLP diets twice a day for 6 weeks. \*Body weight of fish in C-8W, HPL-7W and HPLP-7W treatments was significantly (P < 0.05) higher than that of fish in HPL-6W and HPLP-6W treatments. \*\* Body weight of fish in HPLP-7W, HPL-7W, HPL-7W and C-8W treatments was significantly (P < 0.05) higher than that of fish in HPL-6W and HPLP-6W treatments, but no significant (P > 0.05) difference was found between HPL-7W and C-8W or HPLP-7W and HPL-7W treatments.

found at the beginning of the trial: however, fish body weight with C-8W, HPL-7W and HPLP-7W treatments was significantly (P < 0.05) higher than fish with HPL-6W and HPLP-6W treatments on week 2, 4 and 6 after the trial initiation (Fig. 1). At the end of the 8-week trial, fish with HPLP-7W and HPL-7W treatments overcompensated as compared to fish with C-8W treatment, agreeing with Cho and Heo's (2011) study showing that fish fed a 55% protein diet with 14% lipid for 7 weeks after 1-week feed deprivation overcompensated as compared to fish fed a 50% protein diet with 10% lipid daily for 8 weeks.

However, in this study, a slight, but not significant improvement in compensatory growth of fish with HPLP-7W treatment as compared to fish with HPL-7W treatment was observed (Fig. 1), indicating a dietary inclusion of phosphorous had no distinctive effect on compensatory growth of fish. In addition, since dietary supplementation of phosphorous can result in a deterioration of water quality, phosphorus supplementation must be carefully considered a practical application on a fish farm. Turano et al. (2008) also reported that soluble reactive phosphorous was 41% and 24% lower in ponds with a cycled 3-week feeding after a 3-week feed deprivation and 3-week feeding after a 6-week feed deprivation, respectively, than in ponds with daily satiation feeding for 18 weeks.

Overcompensation of fish, as compared to fish in a control group (no feed deprivation) was observed in only a few studies. Hayward et al. (1997) reported that hybrid sunfish (*Lepomis cyanellus*  $\times$  *L. macrochirus*) subjected to repeating cycles (2 or 14 days) of no feeding and refeeding overcompensated only when the fish experienced the fixed no-feed periods of either 2, 4, 6, 10, or 14 days followed by periods of refeeding, as compared to fish fed meal worms *Tenebrio molitor* to satiation daily for 105 days. Additionally, channel catfish with 3-day feed deprivation only overcompensated, as compared to fish with daily satiation feeding when fish were fed a 36% protein commercial catfish feed to satiation daily or received repeating cycles of fixed feed deprivation for either 1, 2, or 3 days followed by periods of refeeding for 10 weeks (Chatakondi and Yant, 2001).

Full compensation was not achieved in fish subjected to a 2-week feed deprivation (HPL-6W and HPLP-6W treatments) in this study. This result contradicts Cho and Heo's (2011) study showing that juvenile olive flounder fed a 55% protein diet with 14% lipid for 6 weeks after a 2-week feed deprivation could reach full compensatory growth in the 8-week trial or Cho (2005) and Cho et al.'s (2006) studies showing that juvenile olive flounder were able to reach full compensatory growth up to the 2-week feed deprivation in the 8-week trials. Gaylord and Gatlin (2001) also reported that cumulative weight gain was increased in catfish fed the diets containing 37% crude protein, as compared to fish fed 32% protein when fish were fed to satiation for 6 weeks or 11 days of feeding after a 3-day feed deprivation in three cycles.

Hyperphagia is a common phenomenon explaining com-

Treatments	Initial weight (g/fish)	Survival (%)	Feed intake (g/fish)	FCR <sup>*</sup>	$\mathbf{CF}^{\dagger}$	HSI <sup>‡</sup>
C-8W	$34.7\pm0.06$	$91.1 \pm 1.11$	$59.1\pm0.17^{\rm a}$	$0.86\pm0.025^{\text{b}}$	$0.6\pm0.05^{\rm b}$	$4.5\pm1.45^{\rm a}$
HPL-7W	$34.9\pm0.03$	$95.6 \pm 1.11$	$56.1 \pm 0.53^{b}$	$0.93\pm0.012^{ab}$	$0.5\pm0.02^{\rm c}$	$2.7\pm0.08^{ab}$
HPLP-7W	$34.8\pm0.02$	$96.7 \pm 3.33$	$52.9\pm0.57^{\rm c}$	$1.02\pm0.007^{\rm a}$	$0.4\pm0.01^{\circ}$	$3.0\pm0.08^{\rm ab}$
HPL-6W	$34.9\pm0.03$	$97.8 \pm 1.11$	$40.9\pm1.57^{\rm d}$	$1.02\pm0.037^{\rm a}$	$1.1\pm0.06^{\rm a}$	$2.1\pm0.05^{\rm b}$
HPLP-6W	$34.9\pm0.03$	$96.7 \pm 3.33$	$41.3\pm0.28^{\rm d}$	$0.98\pm0.064^{\rm a}$	$1.1\pm0.04^{\rm a}$	$1.9\pm0.03^{\rm b}$

Table 2. Performance and biological index of juvenile olive flounder Paralichthys olivaceus fed the experimental diets with different feeding regime

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

C-8W, fish were hand-fed daily with the C diet twice a day for 8 weeks; HPL-7W and HPLP-7W, fish were starved for 1 week, and then fed with the HPL or HPLP diets twice a day for 7 weeks; HPL-6W and HPLP-6W, fish were starved for 2 week, and then fed with the HPL or HPLP diets twice a day for 6 weeks. <sup>\*</sup>Feed conversion ratio (FCR) = total feed consumption/[(final weight of fish+ dead weight of fish) – initial weight of fish], <sup>†</sup>Condition factor (CF) = fish weight (g) × 100/total length (cm)<sup>3</sup>, <sup>†</sup>Hepatosomatic index (HSI) = liver weight (g) × 100/fish weight (g). pensation of fish. Fish exhibited hyperphagia in many studies (Rueda et al., 1998; Wang et al., 2000; Zhu et al., 2004; Cho, 2005; Cho et al., 2006; Cho and Heo, 2011) showing full compensation of fish subjected to feed deprivation for a certain period of time. However, overall feed intake by fish was proportional to the weeks of feeding in this study. This could explain why fish in the 2-week feed deprivation groups (HPL-6W and HPLP-6W treatments) did not achieve full compensation, as compared to fish with C-8W treatment.

Feed conversion ratio (FCR) of olive flounder with HPLP-7W, HPL-6W and HPLP-6W treatments was significantly (P < 0.05) higher than that of fish with C-8W treatment, but not significantly (P > 0.05) different from that of fish with HPL-7W treatment. Similarly, improved FCR of fish achieving full compensatory growth was observed in other studies (Gaylord and Gatlin, 2001; Cho, 2005; Oh et al., 2007; Kim et al., 2010; Cho and Heo, 2011).

Condition factor (CF) of olive flounder with HPL-6W and HPLP-6W treatments was significantly (P < 0.05) higher than that of fish with C-8W, HPL-7W and HPLP-7W treatments. Additionally, CF of fish with C-8W treatment was significantly (P < 0.05) higher than fish with HPL-7W and HPLP-7W treatments. Unlike this study, however, a previous study observed a high CF in fish reaching full compensatory growth (Cho and Heo, 2011).

Hepatosomatic index (HSI) is a good index to indicate compensatory growth of fish (Gaylord and Gatlin, 2000; Cho, 2005). Bavčevič et al. (2010) showed that gilthead sea bream *Sparus aurata* did not compensate in length, but increased the fish's condition and proposed that length should always be analyzed (or another measure incorporates length, such as condition) when characterizing compensatory growth. In the present study, the HSI is olive flounder with C-8W treatment was significantly (P < 0.05) higher than that of fish with HPL-6W and HPLP-6W treatments, but not significantly (P > 0.05) different from that of fish with HPL-7W and HPLP-7W treatments.

When considering the present results, dietary supplementation of protein and lipid resulted in overcompensation of juvenile olive flounder subjected to a 1-week feed deprivation, but not a 2-week feed deprivation. In addition, dietary supplementation of phosphorous did not further improve compensatory growth of fish.

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