Effects of Water Temperature and Feeding Rate on Growth and Body Composition of Grower Olive Flounder *Paralichthys olivaceus*

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사육 수온 및 사료 공급율이 넙치의 성장 및 체조성에 미치는 영향

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Abstract

A 3×2 factorial experiment was conducted to investigate effects of water temperature and feeding rate on growth and body composition of olive flounder. Triplicate groups of fish (initial body weight of 118 g) fed a extruded pellet (55% protein and 4708 cal/g) to satiation and at restricted feeding rates of 0.25 and 0.4% body weight per day (BW/d) at the different water temperatures (13 and 18°) for 9 weeks. Weight gain increased significantly with increase in feeding rates at each temperature. Weight gain of fish fed to satiation was significantly higher at 18 than 13°C, whereas, that of fish fed at 0.25 and 0.4% BW/d were significantly or slightly lower at 18 than 13°C. Feed efficiency and protein efficiency ratio of fish fed to satiation were not significantly different between 13 and 18°C, but those of fish fed at 0.25 and 0.4% BW/d were significantly higher at 13 than 18°C. The major finding of this study is that satiation feeding is efficient for optimal growth and feed efficiency of grower oliver flounder (116-164 g) in suboptimal water temperatures. The maintenance feeding ration which is zero growth performance, were 0.30 and 0.41% BW/day at 13 and 18°C, respectively. In the restricted feeding regime, compromised growth of fish were worsen in higher water temperature (18°C vs. 13°C). It might be related to high metabolic rate of fish that spend more energy for maintenance metabolism. Based on these results, we suggest that a satiation feeding regime is recommended for a productive growth of grower olive flounder in the suboptimal temperature.

Key words : Water temperature, Feeding rate, Olive flounder

I. Introduction

Water temperature, feeding rate, and fish size are the three most important factors on the growth of fish (Brett, 1979). Particularly, water temperature greatly influences the metabolic rate, growth and feed intake of fish.

Fish feeding is one of the most important factors in commercial fish farming because feed costs account for major portion of the production cost in

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fish culture. Olive flounder (*Paralichthys olivaceus*) is the most commercially important aquaculture species in Korea (Statistics Korea, 2014). It has been reported that the optimum temperature for growth of olive flounder is 20-25 °C, but this fish consumes less feed gradually as temperatures decreases from 20 to 10 °C (Iwata et al., 1994).

Olive flounder are exposed to suboptimal water temperature during year round culture in Korea. Thus, knowledge of adequate feed regime at all relevant environmental temperatures as well as different fish sizes are needed for efficient feeding of olive flounder throughout the year.

Some studies about effects of feeding rate on flounder in suboptimal water temperature showed considerable decrease in growth and feed efficiency of flounder at restricted feeding groups (Kim et al., 2009; Kim et al., 2010).

However, limited information is available regarding effects of satiation or restricted feeding rate on grower flounder at different water temperatures.

The objective of this study was to investigate effects of feeding rate on growth, feed utilization and body composition of grower olive flounder at different suboptimal water temperatures.

II. Materials and methods

1. Fish and Rearing Conditions

Olive flounder were obtained from a local fish farm (Uljin, Korea). They were acclimated to laboratory conditions for 2 weeks before the beginning of the feeding trial. Fish (118±5.1 g) were randomly allocated in two sets of 400 L cylindrical plastic tanks (180 L water volume) with 20 fish to each tank of a flow-through aquarium

system. After initial weighing, water temperature in heated water tanks was elevated to 5° C in 0.5° C per 1 hour. Unheated natural seawater and heated seawater were supplied at a flow rate of 5 L/min in each set of nine tanks, and the difference in water temperatures was maintained about 5° C. The temperatures of natural and heated seawater ranged from 11.3 to 15.1° C and from 16.2 to 20° C (Mean±SD; 13.2 ± 1.6 and $18.0\pm1.6^{\circ}$ C), respectively, throughout the feeding trial. Photoperiod was left at natural conditions (ca. 11-h light/13-h dark). All fish in each tank were weighed collectively at the beginning and the end of feeding trial after being fasted for 24 h.

2. Experimental design

A 3 (feeding rate) \times 2 (water temperature) factorial design with triplicate per treatment was applied in this study. Three feeding rates (satiation, 0.4 and 0.25% body weight per day) were tested on randomly assigned triplicate groups of fish at different mean water temperatures (13 and 18°C). During an acclimation period for 2 weeks in experimental condition, a satiation feeding rate for olive flounder was measured at 0.60% of body weight based on daily *ad libitum* feeding data (control). Two feeding ration was also set up at 0.40 and 0.25% of body weight by deduct 30 and 60% from satiation feeding rate (0.60% of body weight), respectively.

Fish were hand-fed a commercial extruded pellet (Suhup feed Co. Ltd., Uiryeong, Korea; 55% protein, 12% lipid and 4708 cal/g) two times a day at 09:00 and 17:00 h with equal amount of meal according to feeding rates for 9 weeks. Satiation feeding was determined by the point of cessation of voluntary feeding activity by fish. Uneaten food Effects of water temperature and feeding rate on growth and body composition of grower olive flounder *Paralichthys olivaceus*

was removed and weighed after the fish stopped eating, and deducted from feed consumption. Records were kept for daily feed consumption and mortalities. To assess growth and feed efficiency, fish were collectively weighed in each tank after 4 weeks from the beginning of feeding trial. The amount of diet was adjusted accordingly for the subsequent 5 weeks. Growth as measured by percent body weight increase (% BWI) was calculated as $100 \times (BWf-BWi)/(BWi)$, where BWi and BWf were the average initial and final body weights, respectively. Feed efficiency was calculated as (BWf-BWi)/AFC, where AFC was the total amount of diet fed to a tank divided by the number of fish in that tank.

3. Sample Collections and Chemical Analysis

At the end of the feeding trial, five fish in each tank were collected and stored at -75 °C for chemical analysis. Whole body samples were analyzed for proximate composition using a ground and homogenized mixture. Crude protein was determined by the Kjeldahl method using Auto Kjeldahl System (Gerhardt VAP500T/TT125, KG, Germany). Crude lipid was determined with ether extraction using Soxhlet extractor (Velp SER 148, Usmate, Italy). Moisture was determined by drying at 105°C for 6 h. Ash was determined by muffle furnace at 550°C for 6 h. Gross energy of diet was analyzed using and adiabatic bomb calorimeter (Parr-6200, Moline, IL, USA).

4. Statistical Analysis

The data were subjected to a factorial ANOVA to test the effects of feeding rate and water temperature on fish performance, and Duncan's multiple range test (Duncan 1955) was used for comparisons between means at an $\alpha = 0.05$ significance level. All statistical analyses were carried out by using the SPSS program Version 11.5 for Windows (SPSS Inc., Chicago, Illinois, USA).

III. Results

1. Growth Performance

Growth performances of fish fed at different feeding rates at 13 and 18°C for 9 weeks are shown in <Table 1>. Survival was significantly affected by water temperature (P < 0.05), but not by feeding rate. Survival of fish fed the diet at 0.25% body weight per day (BW/d) at 18°C was lowest and significant different from that of fish at 13°C (P<0.05). However, there was no significant difference in survival among treatment except for fish fed the diet at 0.25% BW/d at 18°C. Final mean weight and weight gain were significantly affected by feeding rate (P<0.001), but not by water temperature. Specific growth rate was significantly affected by feeding rate (P<0.001) and water temperature (P<0.05). Fish showed significant increases in final mean weight, weight gain and specific growth rate at both 13 and 18°C when feeding rates were increased. Final mean weight, weight gain and specific growth rate of fish fed to satiation were significantly (P<0.05) higher at 18°C than 13°C. On the contrary, final mean weight and weight gain of fish fed at 0.25 were significantly (P < 0.05) lower at 18 °C than 13 °C.

Feed efficiency and protein efficiency ratio were significantly affected by both feeding rate (P<0.001) and water temperature (P<0.001). Feed efficiency and protein efficiency ratio increased significantly (P<0.05) with increase in feeding rates at same

(Table 1> Growth performances of olive flounder fed at different rates in 13 and 18°C for 9 w

Temperature		13°C			18°C	
Feeding rate (%/BW/d)	0.25	0.4	Satiation	0.25	0.4	Satiation
Initial mean weight (g/fish)	117±5.6	116±2.0	117±3.8	118±1.3	119±2.3	121±3.5
Survival (%)	$100{\pm}0.0^{a}$	$100{\pm}0.0^{a}$	100±0.0 ^a	93±1.8 ^b	98±1.9 ^a	96±1.9 ^{ab}
Final mean weight (g/fish)	114±4.8°	123±3.6°	146±3.9 ^b	100 ± 1.6^{d}	118±1.6 ^c	164±6.7 ^a
Weight gain (%/fish) ¹	-3±1.4 ^d	6±2.8°	25±1.2 ^b	-15±1.0 ^e	-1±1.3 ^{cd}	36±3.9 ^a
Specific growth rate (%/day) ²	-0.05 ± 0.03^{d}	$0.10 \pm 0.05^{\circ}$	$0.39{\pm}0.02^{b}$	-0.28±0.02 ^e	-0.02 ± 0.02^{d}	$0.54{\pm}0.05^{a}$
Feed efficiency $(\%)^3$	-20±10.4 ^c	23±10.7 ^b	68 ± 2.0^{a}	-145±11.4 ^d	-19±8.3°	62±4.9 ^a
Protein efficiency ratio ⁴	-0.36±0.19°	$0.42{\pm}0.20^{b}$	$1.25{\pm}0.04^{a}$	-2.80 ± 0.22^{d}	-0.38±0.16 ^c	1.20±0.09 ^a
Daily feed intake (%) ⁵	$0.25{\pm}0.00^{d}$	$0.41 \pm 0.00^{\circ}$	$0.57{\pm}0.02^{b}$	$0.26{\pm}0.01^{d}$	0.41 ± 0.00^{c}	$0.71{\pm}0.04^a$
Two-way ANOVA						
	Feeding rate	Temperature	Interaction			
Survival (%)	P<0.2	P<0.01	<i>P</i> <0.2			
Final mean weight (g/fish)	P<0.001	P<0.9	P<0.01			
Weight gain (%/fish)	P<0.001	P<0.2	P<0.001			
Specific growth rate	P<0.001	P<0.05	P<0.001			
Feed efficiency (%)	P<0.001	P<0.001	P<0.001			
Protein efficiency ratio	P<0.001	P<0.001	P<0.001			
Daily feed intake	P<0.001	P<0.05	P<0.05			

BW = body weight. Values (mean \pm SEM of three replications) in the same row not sharing a common superscript are significantly different (P<0.05).

¹ (Final body weight - initial body weight) \times 100/initial body weight.

² 100×[(ln(final body weight) - ln(initial body weight)]/day.

³ Fish wet weight gain \times 100/feed intake (dry matter).

⁴ Fish wet weight gain/protein intake.

⁵ Feed intake (dry matter) × 100/[(initial fish wt.+ final fish wt.+ dead fish wt.) × days fed/2].

water temperature. Feed efficiency and protein efficiency ratio of fish fed to satiation were not significantly different between 13° C and 18° C, but those of fish fed at 0.25 and 0.4% BW/d were significantly (*P*<0.05) higher at 13° C than 18° C. Significant interaction of feeding rate and water temperature on weight gain, specific growth rated, feed efficiency and protein efficiency ratio of fish were observed (*P*<0.001). Daily feed intake was similar as the originally designed feed ingrate.

2. Body Composition

Proximate compositions of whole body in fish fed at different feeding rates at 13 and 18°C for 9

weeks are shown in <Table 2>. The contents of moisture and crude lipid were significantly affected by water temperature (P<0.01) and feeding rate (P<0.05). Moisture content of fish tended to decrease at both 13 and 18°C when feeding rate was increased, but the opposite trends were found for crude lipid content. An increasing trend in moisture content, and a decreasing trend in crude lipid content were observed with an increase in water temperature from 13 to 18°C when fish fed at 0.25 and 0.4% BW/d. No significant differences were observed in contents of crude protein and ash in the whole body.

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Temperature	13°C			18°C		
Feeding rate (%/BW/d)	0.25	0.4	Satiation	0.25	0.4	Satiation
Moisture (%)	76.3±0.67 ^{ab}	75.2±0.79 ^b	74.5±0.60 ^a	77.9±0.23ª	75.7±0.15 ^b	75.5±0.64 ^b
Crude protein (%)	16.3±0.26	17.3±0.13	16.9±0.38	16.0±0.72	17.2±0.58	17.4±0.23
Crude lipid (%)	2.0±0.49 ^{bc}	$3.3{\pm}0.31^{ab}$	3.4±0.57 ^a	1.0±0.21°	2.1 ± 0.34^{abc}	$3.4{\pm}0.38^{a}$
Ash (%)	3.8±0.19	3.4±0.26	2.8±0.65	4.1±0.30	2.9±0.40	3.5±0.86
Two-way ANOVA						
	Feeding rate	Temperature	Interaction			
Moisture (%)	P<0.01	P<0.05	<i>P</i> <0.6			
Crude protein (%)	<i>P</i> <0.1	<i>P</i> <0.9	<i>P</i> <0.7			
Crude lipid (%)	P<0.01	P<0.05	<i>P</i> <0.4			
Ash (%)	<i>P</i> <0.3	P<0.8	P<0.5			

<Table 2> Proximate compositions of whole body in olive flounder fed at different rates in 13 and 18°C for 9 weeks

BW = body weight. Values (mean \pm SEM of three replications) in the same row not sharing a common superscript are significantly different (P < 0.05).

IV. Discussion

In this study, the increase of water temperature from 13 to 18°C improved feed intake and growth of flounder fed to satiation. The increased feed intake at the higher water temperature may be the primary response of fish to an increase in temperature (Bureau et al., 2002), and these results agree with a previous study (Iwata et al., 1994). Similar results have been reported for other fish species (Goolish and Adelman, 1984; Peres and Oliva-Teles, 1999).

However, fish fed the diet at 0.25 and 0.4% BW/d showed lower growth and feed efficiency with increase in water temperature from 13 to 1 8°C. Similarly, feed efficiency of juvenile turbot at the restricted feeding group (35% and 60% of satiation) was significantly higher at 16°C than 2 2°C (Van Ham et al., 2003). Water temperature is the major determining th metabolic rate and energy expenditure of poikilothermic animal. Within fish species and certain temperature ranges, increasing water temperature results in a curvilinear (almont linear) increase of basal metabolism (NRC, 2011).

This may be because that increasing temperature has a negative effect on growth due to higher energy cost for maintenance metabolism (Xiao-Jun and Ruyung, 1992).

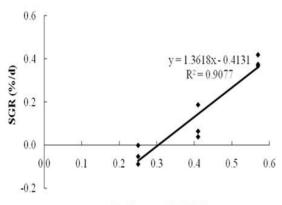
Flounder at the restricted feeding group (0.25 and 0.4% BW/d) showed a considerable lower growth and feed efficiency than those of fish at the satiated feeding group in the same water temperature in this study. This may be because that a large proportion of nutrient in the diet was used to maintain life and only a small proportion of surplus nutrient after basic maintenance was available for growth (Hung and Lutes, 1987). Similar results have been reported for other fish species (Hung et al., 1989; Mihelakakis et al., 2002). It has been suggested that optimum feed efficiency is achieved at feeding levels below that required for maximum growth in salmonids (Elliot, 1976; Kolsater, 1995). However, this study showed a higher feed efficiency for fish at satiated feeding group than at restricted feeding group. Similarly, higher or similar feed efficiency of fish at the satiated feeding group compared with restricted feeding group in previous flounder study (Cho et

al., 2006; Kim et al., 2007). Therefore, satiation feeding is efficient for optimal growth and feed efficiency of grower olive flounder in suboptimal water temperatures.

Specific growth rate (SGR) was plotted against feeding rate (FR), and the data were found to be best fit a linear curve as follows:

[Fig 1] (1) SGR (13°C) = 136.18 FR-0.413 (r^{2} =0.91) [Fig 2] (2) SGR (18°C) = 178.26 FR-0.735 (r^{2} =0.98)

The feeding rates for maintenance calculated by substituting 0 for SGR in equations (1) and (2) were 0.30% and 0.41% BW/d at 13° C and 18° C, respectively, when fish fed a diet with 55% protein and 4708 cal/g.

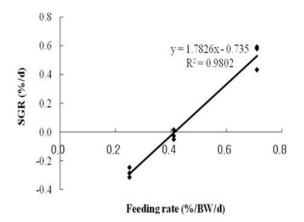


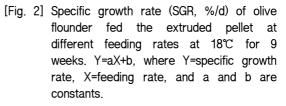
Feeding rate (%/BW/d)

[Fig. 1] Specific growth rate (SGR, %/d) of olive flounder fed the extruded pellet at different feeding rates at 13°C for 9 weeks. Y=aX+b, where Y=specific growth rate, X=feeding rate, and a and b are constants.

Some studies have shown that with increasing feeding rate, the growth increases at higher feeding rate and decreases at lower feeding rate (Arzel et al., 1998; Van Ham et al., 2003). In this study, weight gain showed a linear increase with increased feeding rate from 0.25 to 0.7% (satiation) BW/day.

Similar results have been founded in the previous flounder study (Cho et al., 2006; Kim et al., 2007).





For commercial fish farming, restricted feeding without growth suppression is preferable for economical and environmental reason. In this study, the weight gain of fish at restricted groups such as 37-72% satiation (0.25 and 0.4% of daily feeding rate) was lower than that of fish at satiated groups. Similarly, Seo et al. (2005a; 2005b) reported that the restricted feeding groups (80 and 85% of satiation) showed lower weight gain than that of satiation feeding group in flounder studies. Cho et al. (2006) reported that flounder (initial weight of 17 g) at 70-90% of satiation achieved lower growth compared to satiation group.

Reduced lipid and slightly increased moisture contents of fish at the restricted feeding group in this study agreed with previous flounder study showing that the body lipid content of fish decreased with a decrease in feed consumption (Lee et al., 2000; Kim et al., 2007). Generally, the body Effects of water temperature and feeding rate on growth and body composition of grower olive flounder *Paralichthys olivaceus*

lipid content increased clearly with increasing feeding rates, while body moisture content tended to decrease. And, the whole body moisture content is inversely related to body lipid content (Shearer, 1994). Similar trend have been also reported in other fish species (Hung et al., 1993; Mihelakakis et al., 2002). Lower lipid content was observed in fish fed at 0.25 and 0.4% BW/d at 18° C than 1 3° C. Similarly, Van Ham et al. (2003) reported that juvenile turbot fed to 35% of satiation showed reduced body lipid reared at 22° C compared to 1 6° C.

The major finding of this study is that satiation feeding is efficient for optimal growth and feed efficiency of grower oliver flounder (116-164 g) in suboptimal water temperatures. The maintenance feeding ration which is zero growth performance, were 0.30 and 0.41% BW/day at 13 and 18° C, respectively ([Fig. 1 and 2]). In the restricted feeding regime, compromised growth of fish were worsen in higher water temperature (18° C vs. 1 3° C). It might be related to high metabolic rate of fish that spend more energy for maintenance metabolism. Based on these results, we suggest that a satiation feeding regime is recommended for a productive growth of grower olive flounder in the suboptimal temperature.

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