

Effects of Photic Conditions on Growth Performance in Juveniles of the Goldlined Spinefoot, *Siganus guttatus* (Bloch, 1787)

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Abstract

Fish growth is influenced by photic environments, including light intensity, cycle (photoperiod), and wavelength. Here, we investigated the effects of photic conditions on somatic growth of the juvenile goldlined spinefoot *Siganus guttatus*. Rearing 3- and 11-month-old juveniles under three different light intensities (10%, 0.5–1.0%, and < 0.1% of natural sunlight at culmination) for 8 weeks resulted in little difference in the growth parameters of individuals (specific growth rate [SGR] and condition factor [CF]). High SGR was observed in 4-month-old juveniles reared under constant light (LL) and long-day (LD=14:10), but not under short-day (LD=10:14), for 8 weeks, and in 1- and 9-month-old juveniles exposed to long-day of green LED lights for 4 and 8 weeks, respectively. Real-time quantitative PCR revealed that mRNA abundance of growth hormone in the pituitary and insulin-like growth factor-I in the liver increased in the juveniles reared under long-day, suggesting that the endocrine axis is stimulated under this condition. Thus, somatic growth of the juveniles can be manipulated by alternating the photoperiod and light wavelength. Finally, growth manipulation is more effective in younger fish.

Introduction

Light is a potent external factor that strongly influences growth performance in fish. Steady and reliable fish growth is influenced by the sensitivity of photoreceptors in sensory organs to photic stimuli and from there, transduction into internal stimuli and conveyance to peripheral organs. Somatic growth of fish at both the larval and juvenile stages is thus expected to improve under optimal photic environments for each species. However, compared to other environmental factors, such as temperature, light is more complex because of periodic, simultaneous changes in the light cycle (photoperiod), light intensity, and light wavelength (Boeuf & Le Bail, 1999; Sánchez-Vázquez & López-Olmeda, 2018). Therefore, understanding optimal light

stimuli to maximize somatic growth of fish at larval and juvenile stages is essential for ideal aquaculture outcomes.

Numerous studies have shown that long-day conditions promote somatic growth of juveniles (Biswas, Seoka, Tanaka, Takii, & Kumai, 2006; Biswas, Takaoka, Kumai, & Takii, 2016; Rad, Bozaoğlu, Ergene-Gözükara, Karahan, & Kurt, 2006; Shahkar *et al.*, 2015). Constant light (LL) conditions maximize somatic growth (Biswas *et al.*, 2016; 2006; Shahkar *et al.*, 2015; Worrall, Carter, Wilkinson, & Porter, 2011). Growth-promotion under long-day conditions is attributed to improved appetite, larger food portions, and higher food conversion efficiency (Folkvord & Otterå, 1993).

Further, there is an optimal light intensity range that enhances somatic growth of juveniles under culture

conditions (Biswas & Takii, 2016; Tian, Zhang, Xu, Wang, & Liu, 2015; Wang, Cheng, Liu, Yan, & Long, 2013; Yoseda *et al.*, 2008). In visual feeders, insufficient light intensity is responsible for low visual recognition of food (Copeland & Watanabe, 2006), while high light intensity leads to a decrease in stress responses (Tian *et al.*, 2015). Inadequate light intensities are thus likely to cause poor growth performance and high mortality.

In addition, particular light wavelength(s) stimulates somatic growth in fish (Takahashi *et al.*, 2016; Yamanome, Mizusawa, Hasegawa, & Takahashi, 2009). However, perception of light wavelength may be closely related to the development of visual opsins in photoreceptors and habitat depth selection in respective species. Variations of several endocrine systems at the hypothalamus and pituitary level have also been proposed in light-induced growth (Boeuf & Le Bail, 1999; Porter, Duncan, Handeland, Stafansson, & Bromage, 2001; Takahashi *et al.*, 2016). Most of these findings have been obtained using juveniles or larvae of temperate species. To date, knowledge on the relationship between light stimuli and somatic growth in juveniles of tropical species is limited, although photic environments in low latitude waters differ markedly from that of higher latitude waters.

The goldlined spinefoot *Siganus guttatus* is widely distributed in the tropical and subtropical waters of the Eastern Indian Ocean and the Western Pacific. This species is reported to be highly active at night (Ayson, Reyer, & de Jesus-Ayson, 2014), suggesting high sensitivity to low light intensity. The goldlined spinefoot is an important for human food and actively farmed commercial species in countries of Southeast Asia. Increasing knowledge to promote growth based on light manipulation will be invaluable for this aquaculture species; however, the only attempt at growth promotion involved the construction of biologically active recombinant growth hormones (Funkenstein *et al.*, 2005).

The aim of this study was to investigate the effect of light cycle, light intensity, and light wavelength on somatic growth of the goldlined spinefoot during juvenile stages. The effect of light wavelength was evaluated using light-emitting diode (LED) lights because the use of these lights is applicable in developing countries due to their low energy consumption and long operating life.

Materials and Methods

Animals

Fry (approximately 2000 individuals) ranging from 0.08 to 0.15 g body weight (BW) were collected from Tei-ma river (26°33'28"N-128°04'16"E) and Minato river (26°40'41"N-127°53'13"E), northern Okinawa, Japan, using landing nets deployed during the daytime low tide around the new moon periods in July and August. Fry

were held in indoor concrete tanks (10 metric ton capacity) with running seawater and aeration under a natural photoperiod (ranging from 10.31 h to 13.37 h) and water temperature (ranging 19.1±0.22 in February to 29.5±0.13°C in July) at Sesoko Station, Tropical Biosphere Research Center, University of the Ryukyus, Japan, until use. Commercial pellets (Pure Gold EP1 and EP2; Nisshin-Marubeni, Tokyo, Japan) were given to fish daily at 10.00 h until they were satiated (2% of body weight). All of the experiments were conducted in compliance with the Animal Care and Use Committee guidelines of the University of the Ryukyus and regulations for the care and use of laboratory animals in Japan.

Experimental Procedures and Sampling

Experiment 1

The effects of light intensity on somatic growth were studied using juveniles (approximately 3-month-old) of 3.27±0.10 g BW and 5.7±0.07 cm total length (TL). Three outdoor polyethylene tanks (500 L capacity) with running seawater and aeration were covered with black and white mesh nets to adjust light intensities to 10% (8800 lx), 0.5–1.0% (640 lx), and <0.1% (34.2 lx) natural sunlight at culmination (89,200–96,600 lx) at the water surface of each tank. A total of 74 individuals were housed in tanks and satiated daily with pellets (5% of body weight). Fish were acclimated for 1 week. Following this week, the BW and TL of individual fish in each tank were measured every 2 weeks for 2 months. Fish were taken from each tank and anesthetized using 0.01% 2-phenoxyethanol (Kanto Chemical, Tokyo). After measuring BW and TL, fish were returned to tanks to recover. The same experiment was carried out using 11-month-old juveniles of approximately 54.60±0.93 g BW and 15.0±0.09 cm TL. In this case, fish (n=31) were housed in tanks and satiated daily with pellets. Growth parameters were calculated as follows:

$$\text{Specific growth rate (SGR, \%day}^{-1}\text{);} \\ [(\ln W_f - \ln W_i)/t] \times 100$$

$$\text{Condition factor (CF, \%);} \\ (\text{body weight} / \text{total length}^3) \times 100$$

Where W_f , W_i , and t are the final weight, initial weight, and time period (day), respectively.

Experiment 2

The effects of day length on somatic growth were studied using 4-month-old juveniles of approximately 8.72±0.47 g BW and 7.7±0.14 cm TL. Six indoor glass aquaria (60 L capacity) with running seawater and aeration were set under conditions of LL, long-day (LD=14:10, light on at 06.00 h and off at 20.00 h), and

short-day (LD=10:14, light on at 08.00 h and off at 18.00 h). Two aquaria were used for each light condition. Light intensity at the water's surface was adjusted to 1000 lx using fluorescent lights set at 20W. Fish (n=12) were housed in each aquarium and fed daily with pellets (5% of body weight). After being anesthetized, BW and TL of fish in each aquarium were measured every 2 weeks, and the SGR was calculated. This experiment was carried out for 2 months.

Experiment 3

The effects of light wavelengths on somatic growth were studied using juveniles (9-month-old) of approximately 14.82±0.95 g BW and 9.0±0.19 cm TL. We used light-emitting diode (LED) lights (OPTILED; Optiled Lighting International Ltd., Kwun Tong, Hong Kong) of red (peak at 625 nm), green (520 nm), blue (450 nm), and white (5000 K). LED lights were set on each aquarium with running seawater and aeration. Photon flux density at the water's surface of each aquarium was adjusted to 0.850 to 0.960 W/m². Fish (n=12) were housed in each aquarium, and held for 2 months with daily feeding, under long-day (LD=14:10) conditions. After being anesthetized, BW and TL of fish in each aquarium were measured every 2 weeks for 4 weeks, and the GR was calculated. The same experiment was carried out using juveniles (approximately 1-month-old) of 1.65±0.03 g BW and 4.6±0.03 cm TL. A total of 22 juveniles were housed in each aquarium. This experiment was conducted for 1 month.

Experiment 4

The effects of photoperiod (LD=14:10, LD=10:14, and LL) on the mRNA expressions of growth hormone (GH) in the pituitary and insulin-like growth factor (IGF)-I in the liver were studied using 11-month-old fish, respectively. Aquaria were set according to the experiments 2. After fish were acclimatized under these conditions for 1 week, they were anesthetized with 2 phenoxyethanol at 00.00 h and 12.00 h. The pituitary and liver were taken from each fish and immersed in TriPure Isolation Reagent (Roch Molecular Systems, Pleasanton, CA, USA) and held at -80°C.

Total RNA was extracted from the frozen liver using TriPure Isolation Reagent according to the manufacture's protocol and treated with recombinant DNase I (RNase-free, Takara Bio, Otsu, Japan). To avoid genomic DNA contamination. First-strand cDNA was synthesized from 500 ng of total RNA using the Prime-Script RT reagent kit (Perfect Real Time; Takara Bio) according to the manufacture's protocol. The nucleotide sequences of GH, IGF-I, and β -actin (GenBank accession nos. AB031298, AY198184, and AB643460, respectively) were previously determined (Ayson *et al.*, 2000). The gene-specific primers of GH, IGF-I, and β -actin for real-time quantitative polymerase chain reaction (qPCR)

were as follows: GH (5'-TCC ACC AGG TTG CTC AGA GA-3' for GH-F1 and 5'-CCG GAT GTA ATC GGA GTT GC-3' for GH-R1), IGF-I (5'-GCT GCA GTT TGT GTG TGG AG-3' for IGF-I-F and 5'-CTT TGG AAG CAG CAC TCG TC-3' for IGF-I-R), and β -actin (5'-TAC CAC CAT GTA CCC TGG CAT C-3' for β -actin-F and 5'-TAC GCT CAG GTG GAG CAA TGA-3' for β -actin-R) (Ayson & Takemura, 2006). Each PCR was conducted in a final volume of 10 μ l containing 5 μ l of SYBR *Premix Ex Taq II* (Takara Bio), 0.2 μ l each of forward and reverse primers, 3.8 μ l of nuclease-free water, and 0.8 μ l of cDNA template. The qPCR cycling conditions were 95°C for 3 min, 35 cycles of 95°C for 30 s, 55°C for 30 s, and 75°C for 30 s, followed by 95°C for 15 s, 60°C for 15 s, and 95°C for 5 s. The relative mRNA expression levels of IGF-I and β -actin genes were calculated using the $\Delta\Delta$ Ct method, and β -actin expression was used to normalize transcript levels. Measurement of each gene was performed in duplicate.

Statistical Analysis

All data were expressed as mean±standard error of the mean. Changes in initial weight, final weight, SGR, and CF in three experiments were compared using one-way analysis of variance (one-way ANOVA) followed by Tukey-Kramer post hoc test. The effects of photoperiod (LD=14:10, LD=10:14, and LL) on the mRNA expressions of GH in the pituitary and IGF-I in the liver were compared two-way ANOVA with day-length (LL, long-day, and short-day) and time (00.00 h and 12.00 h) as independent variables. Significant differences between 00.00 h and 12.00 h were compared using Student-*t* test.

Results

Juveniles of the goldlined spinefoot were reared under conditions of 10% (8800 lx), 0.5–1.0% (640 lx), and <0.1% (34.2 lx) natural sunlight at culmination (Table 1). Changes in BW of 3-month-old and 11-month-old juveniles were monitored for 8 weeks. There was no statistical difference ($P>0.05$) in BW of juveniles among the three light intensities, although a rapid increase in BW was observed in the 3-month-old juveniles (3.33–3.42 times), but not in the 11-month-old ones (1.4–1.5 times), of the three groups. No statistical difference ($P>0.05$) in SGR was observed among the three groups, whereas CF was significantly higher ($P<0.05$) in the juveniles reared under high light intensity.

Four-month-old juveniles were reared under different photoperiod conditions (Table 2). As a result, there was little increase (1.72 times) in BW of juveniles under short-day conditions during the experimental period. On the other hand, constant and steady growth was observed in juveniles under long-day (3.37 times) and LL conditions (4.85 times). The highest growth was observed in juveniles held under LL conditions. SGR of the juveniles under long-day and LL conditions was significantly higher ($P<0.05$) than that of the juvenile

Table 1. Effect of light intensity on growth performance of juvenile goldlined spinefoot. *

Parameters	Light intensity (%)**		
	1	10	100
<i>3-month-old juveniles</i>			
Final weight (g)	10.89±0.96 ^a	11.18±0.69 ^a	11.15±0.62 ^a
Mean weight gain	3.33	3.42	3.41
SGR (% day-1)	2.01±0.15 ^a	2.02±0.11 ^a	2.02±0.11 ^a
CF	1.72±0.03 ^a	1.69±0.02 ^{ab}	1.65±0.01 ^b
<i>11-month-old juveniles</i>			
Final weight (g)	78.18±3.80 ^a	80.50±3.45 ^a	79.93±3.31 ^a
Mean weight gain	1.43	1.47	1.46
SGR (% day-1)	0.60±0.09 ^a	0.66±0.08 ^a	0.65±0.07 ^a
CF	1.61±0.04 ^a	1.61±0.04 ^a	1.48±0.01 ^b

*Different letters indicate statistically significant differences (P<0.05).

**Each group had light intensity of 1, 10, and 100% of natural light.

Table 2. Effect of day-length on growth performance of juvenile goldlined spinefoot. *

Parameters	Day-length**		
	Short-day	Long-day	Constant light
<i>4-month-old juveniles</i>			
Final weight (g)	15.63±1.73 ^a	29.39±2.36 ^b	42.33±2.57 ^c
Mean weight gain	1.72	3.37	4.85
SGR (% day-1)	0.77±0.19 ^a	1.99±0.15 ^b	2.69±0.11 ^c
CF	1.88±0.13 ^a	1.71±0.02 ^a	1.81±0.02 ^a

*Different letters indicate statistically significant differences (P<0.05).

**Conditions of short-day, long-day, and constant light had light phase of 10, 14, and 24 hours, respectively.

under short day conditions. On the other hand, CF did not show significant difference among photoperiod conditions (P>0.05).

The effects of light wavelengths on the somatic growth of 1-month-old juveniles reared under long-day conditions (LD=14:10) for 4 weeks. Increases in BW (3.96 – 5.17 times) were observed in juveniles, which were all exposed to LED light sources. Significantly higher gain of BW (P<0.05) was seen in juveniles held under green and white LED lights. The highest SGR was recorded in juveniles held under green LED. A similar experiment carried out using 9-month-old juveniles showed relatively slow growth under four LED lights (1.18 – 2.09 times). Under green LED light, significantly higher SRG (P<0.05) was observed under green LED light (Table 3).

The mRNA expressions of GH in the pituitary and IGF-I in the liver were compared among photoperiod (11-month-old fish) (Figure 1). There was a day-low and night-high variation in mRNA expression of GH in the pituitary under all conditions. Its expression under short-day was significantly higher (P<0.05) than under long-day (00.00 and 12.00 h) and LL (00.00 and 12.00 h) (Figure 1A). When the juveniles were reared under LL conditions, significantly higher IGF-I mRNA abundance (P<0.05) was notified at 12.00 h than at 00.00 h. There was no statistical difference (P>0.05) in the juveniles reared under long-day and short-day conditions,

although a day-high and night low variation of its gene abundance was observed in these groups (Figure 1B).

Discussion

Here, we report the effects of different photic conditions (light intensity, photoperiod, and light wavelength) on somatic growth of tropical goldlined spinefoot juveniles. Somatic growth of this species was not influenced by light intensity. When natural light intensity was manipulated, robust growth was promoted even under conditions of low light intensity. Physiological perception of low light intensity may be a natural characteristic of this species because it is capable of recognizing brightness at night during the full and new moon periods and, likely, of using this light change for entrainment of the lunar-spawning cycle (Takemura, Sri Susilo, Rahman, & Morita, 2004; Takemura, Ueda, Hiyakawa, & Nikaido, 2006). This idea is supported by a recent study showing that rhodopsin is highly expressed in the eyes of this species, as in those of nocturnal species (Takeuchi *et al.*, unpublished data). Unlike our results, Atlantic cod *Gadus morhua* larvae reared at higher light intensities (1200 and 2400 lx) had better growth compared to those reared at lower light intensities (300 and 600 lx) (Puvanendran & Brown, 2002). Similarly, a range of light intensity suitable for

Table 3. Effect of LED lights on growth performance of juvenile goldlined spinefoot. *

Parameters	Light color (%)**			
	Blue	Green	Red	White
<i>1-month-old juveniles</i>				
Final weight (g)	6.46±0.39 ^a	8.25±0.37 ^b	6.70±0.31 ^a	8.37±0.60 ^b
Mean weight gain	4.03	4.94	3.96	5.17
SGR (% day ⁻¹)	4.53±0.20 ^a	10.51±0.32 ^b	4.63±0.15 ^a	5.32±0.25 ^a
CF	1.83±0.03 ^a	1.76±0.02 ^a	1.75±0.02 ^a	1.84±0.02 ^a
<i>9-month-old juveniles</i>				
Final weight (g)	17.52±4.57 ^a	30.99±3.15 ^b	23.86±3.90 ^{ab}	25.32±3.39 ^{ab}
Mean weight gain	1.18	2.09	1.61	1.71
SGR (% day ⁻¹)	0.13±0.43 ^a	1.24±0.20 ^b	0.80±0.31 ^{ab}	0.91±0.23 ^{ab}
CF	1.70±0.04 ^a	1.74±0.04 ^a	1.86±0.06 ^a	1.82±0.02 ^a

*Different letters indicate statistically significant differences ($P < 0.05$).

**LED lights of blue, green, and red had peak wavelengths of 450 nm, 520 nm, and 625 nm.

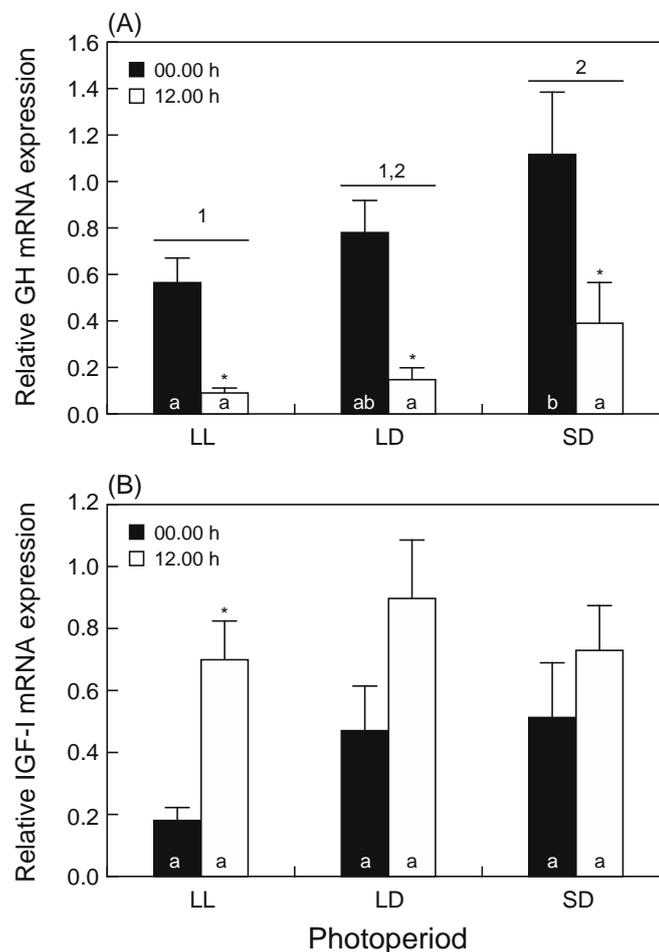


Figure 1. Effect of different photoperiodic conditions on mRNA abundance of (A) growth hormone (GH) in the pituitary and (B) insulin-like growth factor-I (IGF-I) in the liver of juvenile goldlined spinefoot. Eleven-month-old fish were reared under conditions of constant light (LL), long-day (14-hour light and 10-hour darkness, LD), and short-day (10-hour light and 14-hour darkness, SD) for 1 weeks. The pituitary and liver were taken from each individual at 12.00 h and 00.00 h, and total RNA was extracted and reverse-transcribed. Expression levels of GH mRNA in the pituitary and IGF-I mRNA in the liver were measured using real-time quantitative PCR. The data were normalized by determining the amount of β -actin, and expressed as means±standard error of the mean (SEM). The numerals ("1" and "2") denote significant day-length effect, and values with different numbers are statistically significant ($P < 0.05$, two-way ANOVA); the letters ("a", "ab", and "b") denote significant day-length effects within each group, and bars with different letters are significantly different ($P < 0.05$, two-way ANOVA). Asterisks ("*") indicate statistical difference ($P < 0.05$) between 00.00 h and 12.00 h.

growth promotion was noted in juveniles of the orange-spotted grouper *Epinephelus coioides* (Wang *et al.*, 2013), the blunt snout bream *Megalobrama amblycephala* (Tian *et al.*, 2015), and the Chinese longsnout catfish *Leiocassis longirostris* (Han, Xie, Lei, Zhu, & Yang, 2005). It appears that high light intensity precedes stress responses, including the elevation of liver oxidation rates and depression of immunity (Tian *et al.*, 2015), although thresholds for these effects differ among species. Therefore, the effect of light intensity of growth promotion appears to be species specific (Puvanendran & Brown, 2002). Larvae of the leopard coral grouper *Plectropomus leopardus* require high light intensity (3000 lx) for better feeding, growth, and survival under rearing conditions (Yoseda *et al.*, 2008). Here, we showed steady growth in juveniles of the goldlined spinefoot even at high light intensity (8800 lx). A wide range of optimal light intensity for somatic growth may be attributed to adaptive strategies of this species, which inhabits turbid inshore reefs among mangroves and is found on the drop-offs of inshore fringing reefs down to 6 m depth (Ayson *et al.*, 2014).

The somatic growth of fish larvae or juveniles is elicited by rearing under long-day conditions (Biswas *et al.*, 2016; 2006; Rad *et al.*, 2006; Shahkar *et al.*, 2015). The effectiveness of LL conditions for growth promotion is apparent in larvae of the Atlantic cod (Puvanendran & Brown, 2002), and in juveniles of the red sea bream *Pagrus major* (Biswas *et al.*, 2006), the Nile tilapia *Oreochromis niloticus* (Rad *et al.*, 2006), barramundi *Lates calcarifer* (Worrall *et al.*, 2011), striped knifejaw *Oplegnathus fasciatus* (Biswas *et al.*, 2016). Similar results were obtained in the present study, in which somatic growth of the juvenile goldlined spinefoot was promoted under conditions of long-day (LD=14:10) and LL.

Under LL conditions, food intake by juveniles of diurnal species has been shown to be increased due to stimulation of swimming and foraging activity (Biswas *et al.*, 2016), while an increase in growth hormone (GH) stimulated appetite (Johnsson & Björnsson, 1994). In the case of the goldlined spinefoot, however, it is unlikely that GH plays a key role in growth promotion under LL conditions, because a day-low and night-high pattern of GH mRNA expression disappeared in the pituitary of this species when fish were reared under LL or constant darkness (Ayson & Takemura, 2006). In support of the previous report, similar daily patterns of GH mRNA expression were observed in the pituitary. The present study also showed that its variation was higher under short-day than long-day and LL, suggesting its expression during the dark period. On the other hand, the present study showed that abundance of IGF-I mRNA increased in the liver during the photophase, suggesting that IGF-I is involved in stimulating somatic growth in the juveniles. Thus, there is likely to be little influence of the GH-IGF axis in promoting somatic growth of juveniles under conditions of

photoperiodicity. By contrast, thyroid hormones likely participate in growth promotion due to the alternation of photoperiod environments, because mRNA expression of type II iodothyronine deiodinase (D2) in the hypothalamus of the goldlined spinefoot changes according to photoperiod conditions, i.e., day-night difference in its expression was notable under long-day conditions (Wambiji *et al.*, 2011). Because this enzyme plays a role in converting 3,5,3',5'-tetraiodothyronine (T4) to 3,5,3'-triiodothyronine (T3) and is involved in regulating thyroid hormone-dependent processes in various tissues (Köhrle, 1999), and, subsequently, T3 stimulates hepatic IGF-I mRNA expression in the Mozambique tilapia *Oreochromis mossambicus* (Schmid, Lutz, Kloas, & Reinecke, 2003), the hypothalamo-pituitary-thyroid axis may be activated in this species under long-day and LL conditions. A similar idea was proposed in growth promotion of juveniles of the red drum, *Sciaenops ocellatus* (Leiner & MacKenzie, 2001).

The effect of light wavelength on somatic growth of the juvenile goldlined spinefoot was studied using LED lights because the use of these lights is applicable in developing countries due to their low energy consumption and long operating life. This light source recently adopted in similar researches (Shin, Lee, & Choi, 2012; Takahashi *et al.*, 2016). Improved growth promotion was obtained when fish were reared under long-day conditions (LD=14:10) using green LED light. Compared with light intensity and photoperiod, the effect seemed to be quick and powerful, suggesting that there is a particular range of light wavelengths that stimulates somatic growth of juveniles. A similar effect on somatic growth was reported in the barfin flounder *Verasper moseri* reared under short-day conditions (LD=9:15) and green light (Yamanome *et al.*, 2009). Red light was observed to have an inhibitory effect on somatic growth in the barfin flounder and the gilthead seabream under conditions of LD=9:15 and LD=12:12, respectively (Karakatsouli *et al.*, 2007; Yamanome *et al.*, 2009), while red light (LD=12:12) had a stimulatory effect on TL of the rainbow trout, *Oncorhynchus mykiss* (Karakatsouli *et al.*, 2008). Rearing fish under unsuitable light wavelength conditions induces stress responses, including decreases in liver total lipids and plasma glucose, as well as increases in brain serotonergic and dopaminergic activities (Karakatsouli *et al.*, 2007).

When the barfin flounder was reared at 6.6°C under short-day conditions (LD=10.5:13.5) using three light wavelengths (blue at 464 nm, green at 518 nm, and red at 635 nm), mRNA abundance of melanin-concentrating hormone-2 in the brain was lower under green than under red light, and that of proopiomelanocortin-C in the brain was higher under blue and green light compared with red light. Although there was no difference in the mRNA abundance of GH in the pituitary, increased plasma levels of insulin and IGF-I were observed in fish under green light. It has been

suggested that the endocrine system for food intake is modulated by exposure of fish to a specific range of light wavelengths (Takahashi *et al.*, 2016). The present study did not evaluate the influence of light wavelengths on the expression profiles of these genes in the hypothalamus and pituitary of the goldlined spinefoot. However, it is likely that, in addition to photoperiod, green light stimulates the endocrine axis in relation to IGF-I synthesis.

In conclusion, somatic growth in juveniles of the goldlined spinefoot was accelerated by alternation of the photoperiod and the selection of suitable light wavelengths. There is a stage-dependent effect of light manipulation on somatic growth in the Nile tilapia; high growth performance was observed at the fingerling stage (5.0–30.0 g), when fish were exposed to LL at 800 lx (Rad *et al.*, 2006). By contrast, growth performance at the fry stage (0.02–1.24 g) was higher than that at the fingerling stage (2.33–51.35 g), when reared under LL at 2500 lx (El-Sayed & Kawanna, 2004). Because somatic growth of 3-month-old juveniles was higher compared to that of 11-month-old juveniles in the goldlined spinefoot, the photoperiod may be manipulated using younger fish with a higher sensitivity to light. It may be proposed that usage of long-day conditions with green LED has advantages of aquaculture promotion in certain species including the goldlined spinefoot.

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