Effects of intermittent feeding on compensatory growth, feed intake and body composition in Asian sea bass (*Lates calcarifer*)

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Abstract

This experiment was conducted to investigate the effects of various starvation and refeeding periods on growth, feed utilization and body composition in Asian sea bass (*Lates calcarifer*) with an average initial weight of 30.26±1.4 g (mean±SE). The fish were exposed to three different regimes: the control group fed twice daily to apparent satiation throughout the experiment (C), the first group starved for 4 days and refed for 16 days, this cycle was repeated two times (T_1) and the second group starved for 8 days and refed for 32 days (T_2). At the end of experiment, there were not any significant differences in growth and feeding performance among different treatments (p>0.05). Daily feed intake was significantly higher in the deprived fish than in the control fish (p<0.05). There were no differences in moisture, lipid, ash and nitrogen free extract (NFE) content of carcass at the end of different starvation and refeeding periods between the deprived and control fish (p>0.05). Starvation had a significant effect on protein content on one sampling date during the experimental period; protein content in T_2 on day 8 was significantly lower compared to the control (p<0.05). Sea bass showed complete compensation indicating a high ability of the deprived fish to grow sufficiently to fully compensate for weight loss during starvation. The results suggested that the feeding schedule involving starvation-refeeding cycles could be a promising feed management option for the culture of this species.

Keywords: Compensatory growth, Feed deprivation, Body composition, Sea bass

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Introduction

In nature, a lot of environmental factors control growth such as temperature, food, and predators. Distribution of food can vary in different times of the day or year, so fish can experience short or long periods of starvation until they reach food (Jobling, 1994). Starvation can produce a phase of rapid growth that is greater than normal growth rates; this phenomenon is known as compensatory growth (Dobson and Holmes, 1984). Therefore, it can be illustrated that the ability to compensate starvation periods is an important adaptation in a fluctuating and unpredictable environment (Maclean and Metcalfe, 2001). In aquaculture, fish may experience food deprivation in response to several factors such as temperature fluctuation, pre-harvesting, and prior to handling (Davis and Gaylord, 2011). Compensatory growth results have been inconsistent in different fish species. There are several examples where full compensation has been observed in rainbow trout, *Oncorhynchus mykiss* (Quinton and Blake, 1990; Dobson and Holmes, 1984), barramundi, *Lates calcarifer* (Tian and Qin, 2003, 2004), Arctic charr (Miglavs and Jobling, 1989a), gibel carp, *Carassius auratus gibelio* (Xie *et al*., 2001), Atlantic salmon (Skilbrei, 1990); partial compensation has been observed in hybrid tilapia (Wang *et al*., 2000), Persian sturgeon, *Acipenser persicus* (Yarmohammadi *et al*., 2013) Arctic charr (Miglavs and Jobling, 1989b); no compensation has been reported in common carp, *Cyprinus carpio* L. (Schwarz *et al.*, 1985) and great sturgeon, *Huso huso* L. (Falahatkar, 2012). overcompensation was observed in channel catfish (Chatakondi and Yant 2001) and hybrid sunfish (Hayward *et al.*, 1997).

The Asian sea bass, also known as Barramundi, is an important aquaculture species in the Indo-Pacific region, found through the eastern edge of the Persian Gulf to China, Taiwan and Southern Japan and Australia. These fish are carnivorous, euryhaline and produced commercially in ponds, cages and recirculating tanks in both fresh and seawater. The annual production of Asian sea bass has increased during the last years, and the world grand total production of this species went up to 69,116 (t) in 2011 (FAO, 2012). Compensatory growth has been reported in various fish including both warm water and coldwater species. However, there is little information on the short-term effects of starvation and refeeding on Asian sea bass. For example, Tian and Qin (2003, 2004) studied long-term effects of starvation (1 to 3 weeks) and refeeding on growth and feeding performance in Asian sea bass. Thus, this study was conducted to investigate a compensatory growth response, growth performance and feed utilization and body composition in Asian sea bass.

Materials and methods

250 juvenile sea bass (*Lates calcarifer*) with an initial average weight of 25.4 g were transported from the Marine Fish

Cultivation and Propagation Commercial Center (Delvar, Iran) to the research station of Persian Gulf Research and Study Centre (Bushehr, Iran). After two weeks of acclimation, fish were distributed randomly into nine 300-L circular fiberglass tanks. Three treatments with three replicates were established which were stocked with twenty fish. The control group (C) was fed to satiation level twice daily (0800 h and 1600 h) with 3 mm commercial formulated feed especially for carnivorous fish such as trout (manufactured by Kimiagaran Taghzie, Shahrekord, Iran) containing 40% crude protein, 16% crude lipid, 2.5% crude fiber, 9% ash. Treatment 1 (T_1) was starved for 4 days and refed for 16 days, this cycle was repeated two times. Treatment 2 (T_2) was starved for 8 days and refed for 32 days, this cycle was repeated one time. The average water temperature (WTW, Germany), oxygen concentration (HACH-sension1) and salinity (WTW, Germany) were 27±1°C, 90–100% saturation and 42.5±0.2‰, respectively, which were monitored weekly. Photoperiod was 12h Dark and 12 h Light.

Fish were weighed (to the nearest 0.01g) (Sartorius, Germany), and total length was measured (to the nearest 0.1 cm), at the start of the experiment and every 10 days thereafter. All indices were calculated as follows: specific growth rate (SGR % /day)=100[(lnWt-lnW0)/t]; weight gain (WG)=100[(Wt-W0)/W0], where Wt and W0 are final and initial weight (g) and t is the feeding duration (day); condition factor (CF)=100[W/L3], where L=length (cm); feed conversion ratio (FCR)=intake (g, dry weight)/wet weight gain (g); total food (TF)=g feed/60 day; protein efficiency ratio (PER)=wet weight gain/protein consumed (dry matter); daily feed intake (DFI)=g feed/day (according to Nafisi and Soltani, 2008).

For analysis of initial body composition, 8 fish were frozen at the start of the experiment, during the experiment, and at the end of each starvation and refeeding period. Fish were randomly sampled and sacrificed, pooled and dried to constant weight at 105°C for determination of moisture content. The dried samples were homogenized for determining the following: Crude protein was determined by micro Kjeldahl method (N×6.25) after acid digestion, lipid by ether-extraction method using a Soxtec system, fiber by acid and alkaline digestion then combustion in a muffle oven at 550°C for 5 h and moisture content by drying at an oven temperature of 120°C for 5 h (AOAC, 1995).

Statistical analyses were performed using SPSS, version 15.0 for windows. At the beginning of the experiment, possible differences in initial weight, length and body composition of fish were tested by one-way analysis of variance (ANOVA). Analysis of covariance (ANCOVA) was used to test for differences between treatments in body composition at the start of the experiment, after each starvation and refeeding and at the end of the experiment, using sampling days as covariates. The differences in the variables among the treatments were tested using one-way ANOVA at regular sampling dates. *Post hoc* comparisons between sample means were tested by Tukey and LSD test. Data were expressed as means±standard error (SE) and differences were considered statistically significant at $p<0.05$ level.

Results

During the 40 day experiment, mortality was low and ranged from 0-1 fish per tank for all treatments. At the end of the experiment, no significant difference ($p>0.05$) in final mean weight, specific growth rate (SGR), condition factor (CF) and weight gain (WG) were found between the starved fish and the control fish.

Feeding performances of different treatments are shown in Table 1. At the end of the experiment, there were not any significant differences in FCR, PER and total food consumption between the control group and the deprived groups ($p>0.05$). However, the control fish had the highest FCR when compared to the other treatments. At the end of the experiment, daily feed intake in the deprived groups was significantly higher than the control group. These values were 134.42, 124.6% in the control fish in T_1 and T_2 fish, respectively (Table 2).

In the present study, analysis of whole body composition at the start, the end and the remaining sampling dates of experiment indicated that any significant difference did not show in lipid, ash, moisture and NFE between the deprived fish and the control fish. Protein content was not significantly different during the whole period of the experiment except on day 8 that after the starvation cycle, fish in the deprived group had significantly lower protein content (18.18±0.05) than that in the control (18.72±0.02). Results of ANCOVA for the effects of sampling days and interaction effects of feeding regime (treatments) and sampling days on the variables are presented in Table 3. Significant differences in moisture, lipid, ash and protein contents between the control and deprived groups were observed with respect to sampling days.

Discussion

Compensatory growth of fish has many advantages in aquaculture, including efficient feed utilization and /or enhanced growth rate, minimized food wastage and more flexible feeding regimes (Tian and Qin, 2004), but results have been inconsistent in different species.

Table 1: Growth performance values of *Lates calcarifer* in three feeding regimes (mean ± SE). No significant differences were observed among the three groups.

Parameters	Treatments		
	C	T_1	T_2
Initial weight(g)	30.53±1.87	30.76±1.41	31.61±1.75
Final weight(g)	45.24±1.07	46.03±0.57	46.74±1.72
Weight gain (%)	50.16±5.51	50.19±6.43	47.85±2.55
Specific growth rate (%day)	0.95±0.25	0.96±1.20	0.90±0.057
Condition factor	1.31±0.09	1.42±0.02	1.43±0.01

Table 2: Feeding performance values of *Lates calcarifer* in three feeding regimes (mean ± SE). Different superscript letters denote significant differences between the experimental groups.

Parameters	Treatments		
	C	T1	T2
FCR	1.73±0.43	1.55±0.05	1.64±0.04
PER	1.52±9.81	1.47±5.69	1.55±7.32
DFI	$8.52±0.54^{b}$	$11.09±0.39^{a}$	$10.28±0.39^{ab}$
TF	341.20±21.90	329.27±12.71	347.33±16.35

FCR: feed conversion ratio; PER: protein efficiency ratio; DFI: daily feed intake ;TF: total food.

Table 3: Results of analysis of covariance (ANCOVA) in changes in body composition during the experiment. Sampling day was used as covariate.

Dependent variable	Day effect		Treatment*Day effect	
	F	P	F	P
Moisture	26.83	0.00	0.38	0.68
Protein	66.79	0.00	0.27	0.76
Ash	9.44	0.05	1.25	0.30
Lipid	127.43	0.00	0.1	0.90
Nitrogen free extract	0.43	0.51	0.58	0.56

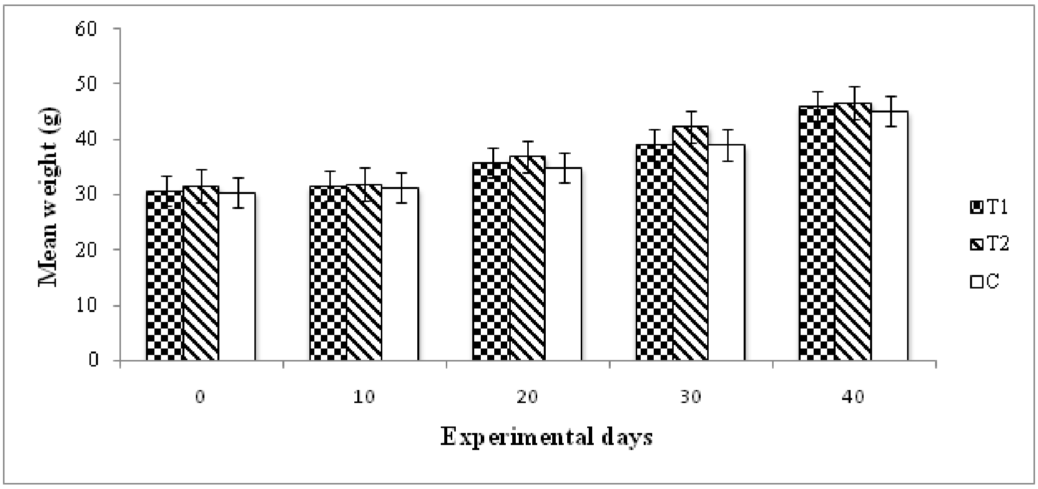

Figure 1: Mean weight of sea bass (*Lates calcarifer*) subjected to different cycles of starvation and refeeding for 40 days. No significant differences observed in three groups.

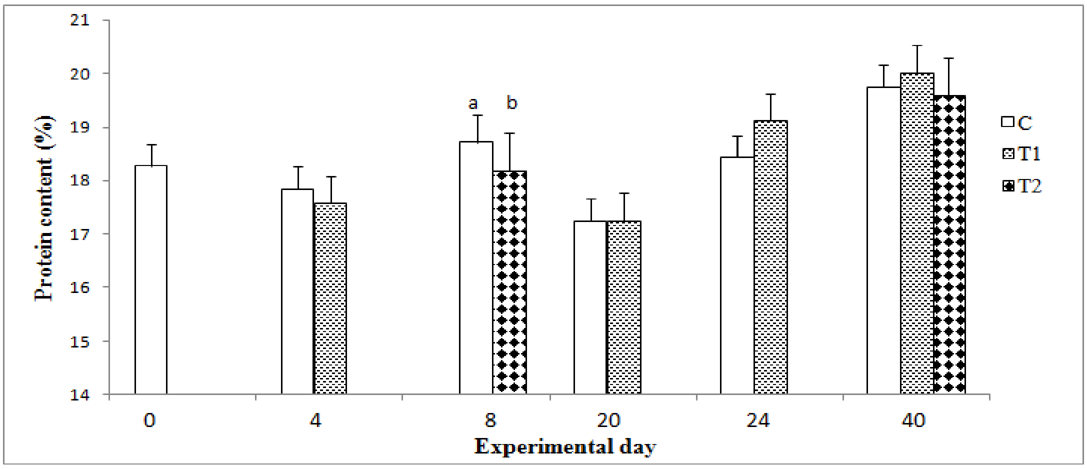

Figure 2: Protein content of sea bass (*Lates calcarifer*) reared under three feeding regimes (mean±SE). Different superscript letters denote significant differences between the experimental groups ($p<0.05$).

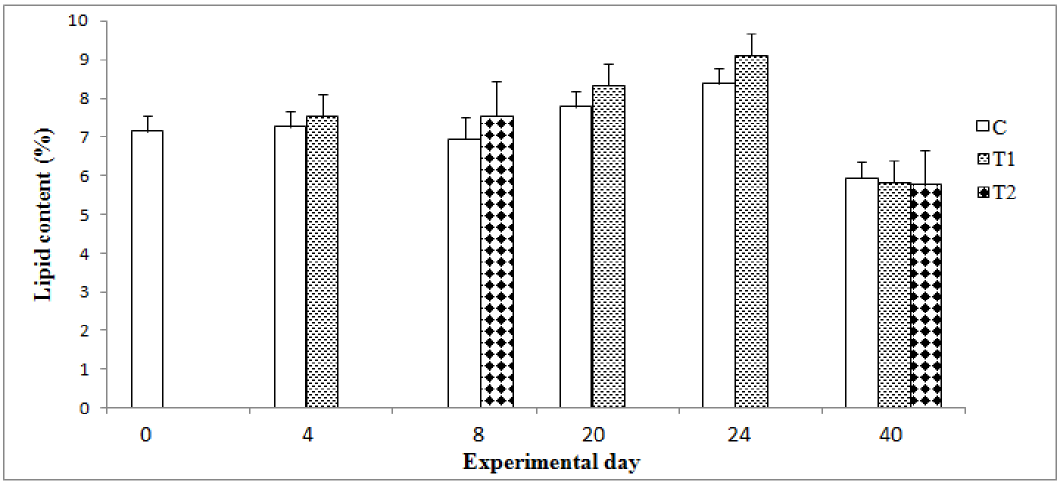

Figure 3: Lipid content of sea bass (*Lates calcarifer*) reared under three feeding regimes (mean±SE). No significant differences observed in three groups.

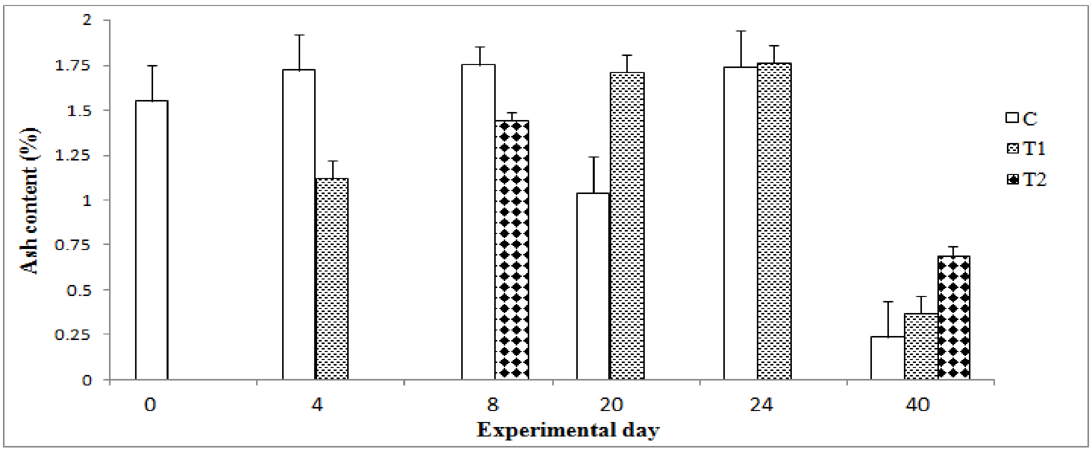

Figure 4: Ash content ofs sea bass (*Lates calcarifer*) reared under three feeding regimes (mean±SE). No significant differences observed in three groups.

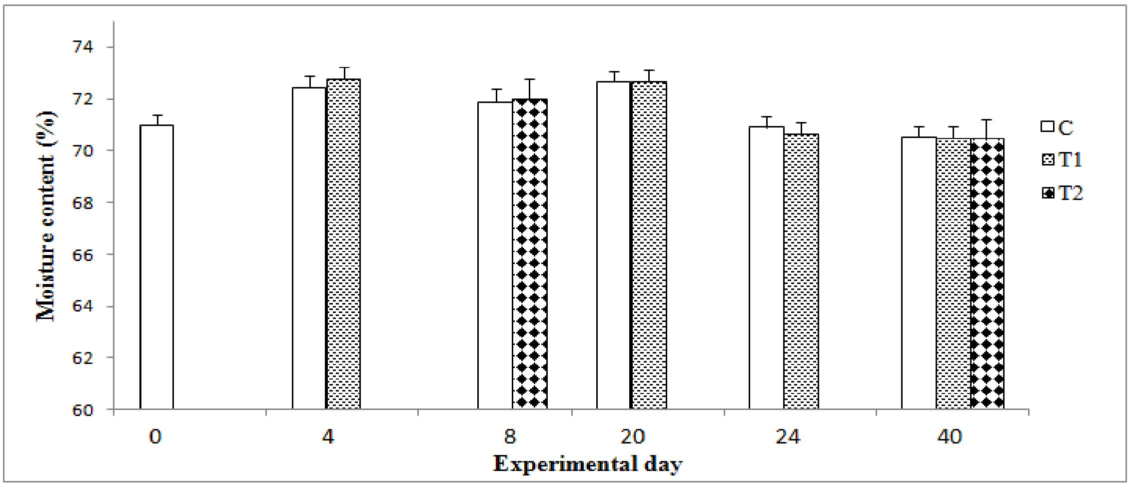

Figure 5: Moisture content of sea bass (*Lates calcarifer*) reared under three feeding regimes (mean±SE). No significant differences observed in three groups.

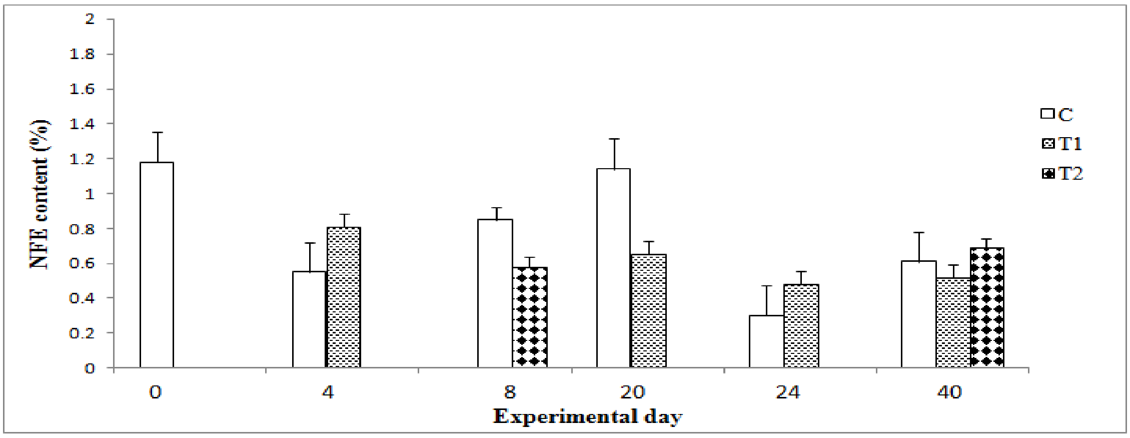

Figure 6: NFE content of sea bass (*Lates calcarifer*) reared under three feeding regimes (mean±SE). No significant differences observed in three groups.

Different fish species including coldwater and warm water fish have shown different compensatory growth responses to different feeding and starvation regimes; complete compensation in European minnows (Russel and Wootton, 1992), rainbow trout (Weatherly and Gill, 1981; Dobson and Holmes, 1984; Quinton and Blake, 1990; Jobling and Koskela, 1996; Nikki *et al.*, 2004 (fasted for 2 or 4 days), Azodi *et al*., 2013), pikeperch (Mattila *et al*., 2009 (fasted for 1 day)), olive flounder (Cho and Cho, 2009), Chinese sturgeon (Liu *et al*., 2011), gibel carp (Qian *et al*., 2000; Xie *et al.*, 2001), roach (Abolfathi *et al*., 2012) sailfin molly (Morshedi *et al*., 2013) and Siberian sturgeon (Morshedi *et al.*, 2013). In addition, there are several examples for partial compensation where fish have not fully compensated for the lost growth (Miglavs and Jobling, 1989a; Jobling *et al*., 1993; Kankanen and Pirhonen, 2009; Ribeiro and Tsuzuki, 2010). These results indicate an ability of various fish species to exhibit compensatory growth. In the present study, the deprived groups showed a clear compensatory growth response at the end of the experiment, because they reached the weight of restricted groups, and SGR value in deprived groups were not significantly different compared to that in the control group.

The results of Xie *et al*., (2001) on gibel carp and Eroldogan *et al*., (2008) on sea bream are in agreement with the results of the current study; they did not observe any significant difference in SGR between different treatments at the end of the experiment indicating a high ability of the sea bass to grow sufficiently to fully compensate for weight loss during starvation. This may be due to reduced metabolic rate during feed deprivation as a result of decreased activity (Love, 1970; Jobling, 1980; Eroldogan *et al.,* 2006) and increased daily feed intake or a combination of both (Heide *et al.,* 2006). Condition factor (CF) did not vary significantly between the treatments indicating that compensatory mechanisms had occurred (Kankanen and Pirhonen, 2009). The present differences among these experiment could be due to different experimental protocols or condition, temporal differences, physiological condition and severity of feed deprivation (Jobling, 1987; Jobling and Koskela, 1996).

At the end of the experiment, no significant difference were found in FCR and PER between the control group and the deprived groups, but there were significant difference in daily feed intake between these groups. It is possible that hyperphagia isthe main mechanism of compensatory growth. Our results are in agreement with Tian and Qin (2003) and Ranta and Pirhonen (2006), but in conflict with Foss *et al*., (2002) and Jiwyam (2010).

The total body composition (moisture, ash, protein, lipid, nitrogen free extract) of the fish subjected to starvation at the end of the experiment was similar to that of the control fish. This is in accordance with results on rainbow trout (Quinton and Blake, 1990), barramundi (Tian and Qin, 2003; 2004) and sea bream (Eroldoĝan *et al*., 2008). In the present study, fish fasted for 8 days showed significant fall in protein content, whereas such a trend was not observed in fish starved for 4 days. It could be assumed that in the fish deprived food for 8 days, protein reserves were early mobilized for supply of energy. Fauconneau *et al*., (1985) reported a rapid fall in protein deposition and protein turnover during starvation in fish. Also, Grigorakis and Alexis (2005) reported a rapid fall in protein deposition and protein turnover during starvation in gilthead sea bream (*Sparus aurata*, L.). The common response of fish undergoing a period of starvation is to satisfy the energy requirement by utilization of body stores. Overall, two principal groups of fish have been identified on the basis of their metabolic response to starvation: those that primarily use muscle protein as the principal fuel and those that primarily use lipids (Storer, 1967; Niimi, 1972; Jezierska *et al.*, 1982;

Mendez and Wieser, 1993). The effect of starvation on utilization of reserve protein and lipid seems to be species-specific (Ince and Thorpe, 1976; Mehner and Wiese, 1994), which may have caused the difference in the results. The present study indicated that sea bass adapted to short-term periods of starvation and could defend body composition in these periods.

Our aim was to test whether dividing these 8 days of starvation into different cycles of food deprivation and subsequent feeding would evoke any compensatory growth response.

In conclusion, starvation periods and refeeding resulted in complete compensatory response in juvenile sea bass. The results indicated that sea bass could be well adapted to starvation periods (especially 4 days of starvation) without significantly affecting growth. Growth and feeding performances of the starved fish was comparable to that of fish fed to satiation. However, further research on physiological and metabolic responses is needed to completely understand the mechanism in this species. The results suggested that the feeding schedule involving starvation–refeeding cycles could be a promising feed management option for the culture of this species.

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