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Abstract An experiment was conducted to determine the effects of optimal feeding frequency on growth performance, feed efficiency, nutrient utilization and body composition in juvenile flounder. Four feeding frequencies of one (F1), two (F2), three (F3) and four (F4) times a day were evaluated as treatments in triplicate for a period of 60 days. Forty-seven hatchery produced juveniles (2.5 ± 0.7 g) were stocked into 160-L rectangular fiberglass tanks and fed with a commercial diet containing 54 % protein to satiation. At the end of the experiment, the final weight of F1, F2, F3 and F4 were 5.06 ± 0.29, 5.91 ± 0.42, 6.24 ± 0.42 and 6.16 ± 0.46 g, respectively. Growth rates and feed consumption were highest in F3 and lowest in F1. Feed conversion ratios ranged between 0.83 ± 0.04 and 0.90 ± 0.03. There were no significant differences in either moisture or ash content of the fish groups. Protein content decreased with increasing feeding frequency, while lipid content partly increased with increasing feeding frequency. Nitrogen intake and lipid intake were significantly higher in fish fed under F3 and F4 treatment, whereas the nitrogen gain decreased and lipid gain increased with increasing feeding frequency. It is concluded that the flounder juveniles can achieve maximum growth performance and better nutrient utilization when they are fed a given ration two times a day. The findings have practical significance toward establishing an appropriate flounder nursery rearing and will directly benefit the nursery operations.

Keywords *Platichthys flesus luscus* · Optimum feeding · Feed intake · Nutrient

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Introduction

Growth of fish is largely influenced by a number of factors, such as feeding frequency, feed intake, feed type and ration. Of the feeding practices, feeding frequency is one of the most important variables influencing growth performance and feed conversion ratio due to it is one of the highest time-consuming activities in the husbandry practices (Kim et al. 2007; Biswas et al. 2010; Lee and Pham 2010; Aydın et al. 2011a). Thus, researchers always stress an aquatic species should be receive an optimal feeding frequency its successful culture. The optimal feeding frequency improves fish growth, survival and feed conversion ratio, reduces size variation and lower the waste load that must be captured before it leaves the farm and pollutes the environment (Goddard 1996; Kubitza and Lovshin 1999; Xie et al. 2011).

Several studies have been executed on feeding frequency in a numerous of flatfish species including plaice-*Pleuronectes platessa* (Jobling 1982), yellowtail flounder-*Limanda ferruginea* (Dwyer et al. 2002), Black Sea turbot-*Psetta maxima* (Türker 2006; Aydın et al. 2011b), olive flounder-*Paralichthys olivaceu* (Lee et al. 2000a; Lee and Pham 2010), Atlantic halibut-*Hippoglossus hippoglossus* (Schnaittacher et al. 2005) and Japanese flounder (Kim et al. 2007). In general, optimum feeding frequency in flatfish seems to be dependent on fish size, water temperature and diet composition (Table 1). Among these flatfish reported in Table 1, flounder (*Platichthys flesus*) is an important commercial fish in European, Mediterranean and Black Sea and has potential as an aquaculture species. Although seed production (Şahin 2000), reproductive performance (Şahin et al. 2008; Aydın et al. 2011c), optimum rearing temperature (Aydın et al. 2011a) and juvenile abnormality (Aydın 2012) of the flounder were studied in Turkey, to the best of the authors' knowledge, very little information is yet available on feeding frequency in flounder. Therefore, the objective of this study was to find out the effects of feeding frequency on growth, body composition and nutrient utilization of juvenile flounder.

Materials and methods

The study was carried out at the Central Fisheries Research Institute, in Trabzon, Turkey. Hatchery-reared juveniles (2.5 ± 0.7 g) were randomly stocked into twelve indoor rectangular fiberglass tanks (160-L), each stocked with 47 fish, for 60 days. Sea water with a salinity of 17.7 ± 0.01 ‰ was pumped from 40 m depth, and water flow was 5 l/min for all experimental tanks. The seawater used in the hatchery was pre-treated using pressurized sand filters and a UV sterilization system and was given to all tanks from one reservoir. The water was aerated with two air stones at a moderate rate in each tank. Temperature was measured twice in a day, and dissolved oxygen (DO), pH and salinity values were measured once a week. During the experiment, water temperature was at 18.4–23.4 °C, pH was 7.4–8.1, salinity was 17.6–17.9 and DO was higher than 80 % (DO > 5.0 mg/L).

A total of four feeding frequency groups were set up in three replicates as follows: one meal (F1) in a day at 0800, two meals a day (F2) at 0800 and 1700, and three meals a day (F3) at 0800, 1230 and 1700, and four meals a day (F4) at 0800, 1100, 1400 and 1700. Fish were fed on commercial 2 mm floating-extruded sea bass diet (Çamlı Feed Corporation, Turkey), containing 54 % crude protein, 14 % crude lipid, 1 % crude fiber and 13 % crude ash while having a gross energy of 19.8 kJ/g. Fish were fed manually to apparent satiation (fullness) according to feeding frequency. The food weight consumed by the fish in each

Table 1 Compilation of studies reporting feeding trials in which feeding frequency tested in flounder species

Species	Scientific name	Initial weight (g)	Feeding frequency	Days	Temperature	Diet (protein/lipid)	Major remarks	References
Plaice	*Pleuronectes plutessu*	29.5	1 Meal/day 1 Meal/2 days	70	Ambient	Moist (49/7.5)	1 meal/day higher growth	Jobling (1982)
Yellowtail flounder	*Limanda ferruginea*	6.8	2 Meals/2 days 1 Meal/day 2 Meals/day 4 Meals/day	70	7 °C	Extruded (55/10)	2 meals/day higher growth and lower FCR	Dwyer et al. (2002)
Turbot	*Psetta maxima*	15.2	2 Meals/day 2 Meals/2 days 2 Meals/4 days	60	5–7.5 °C	Extruded (42/18)	2 meals/day and 2 meals/2 days higher growth	Türker (2006)
Japanese flounder	*Paralichthys olivaceus*	6.3	2 Meals/day 3 Meal/day	60	12 °C 17 °C	Extruded (na) Moist pellet (na)	2 meals/day optimum at 12 °C Growth at 22 > 12 °C	Kim et al. (2007)
Japanese flounder	*Paralichthys olivaceus*	11.1	1 Meal/2 day 1 meal/day 2 Meals/day 3 Meals/day	49	20.3 °C	Sinking moist pellet (52/9) Sinking moist pellet (52/9) Floating extruded (52/8.5)	Extruded feed higher growth at 3 meals/day	Lee and Pham (2010)
Turbot	*Psetta maxima*	23.8	1 Meal every other 2 days, 1 meal/day 2 Meals/day	97	13 °C	Extruded (45/12)	1 meal/day higher growth and feed efficiency	Aydın et al. (2011b)
Flounder	*Platichthys flesus luscus*	2.5	1 Meal/day 2 Meals/day 3 Meals/day 4 Meals/day	60	20 °C	Extruded (57/14)	2 meals/day higher growth	Present study

na not available

tank was measured at each feeding time on these days (Wang et al. 1998). Uneaten pellets were collected 45 min after each feeding and counted to calculate actual feed intake.

All biometric data were taken only after feeding had been ceased for 24 h. Fish in the tanks was weighed individually. The coefficient of variation was used to examine the inter-individual weight variation among the fish in each tank.

Sample collection and chemical analysis

At the beginning of the experiment, an initial sample of 20 fish from the common pool of fish was killed for whole-body proximate analysis. Three fish from each tank were captured and killed by lowering the body temperature in a freezer and stored at −80 °C for subsequent chemical analysis. All assays for proximate composition analysis were performed using standard methods (AOAC 1995). The moisture content was determined by drying samples in an oven at 105 °C for 24 h to reach constant weight. Ash was determined by incineration of sample in a muffle furnace at 550 °C for 4 h. Crude protein was determined by the Kjeldahl method. For the analysis, an automated distillation unit (Kjeltec System 1002) was used. The lipid content was determined by extraction with diethyl ether in a Soxhlet system extractor.

Data calculations and statistical analysis

The effects of feeding frequency on the growth performance of fish were assessed by the following:

1. Weight Gain; $\text{WG} = [(\text{final body weight} - \text{initial body weight})/ \text{initial body weight.initial body weight}] \times 100$
2. Specific Growth Rate; $\text{SGR} = 100 \times (\ln \text{final weight} - \ln \text{initial weight})/\text{day}$
3. Feed Conversion Ratio; $\text{FCR} = \text{feed intake}/\text{weight gain}$
4. Coefficient of Variation; $\text{CV} = 100 \times \text{SD}/\text{mean weight of the fish in each tank}$
5. Condition Factor; $\text{K} = 100 \times (\mathit{Weight}/\mathit{Length}^3)$

Data were analyzed by one-way analysis of variance (ANOVA) and differences between means compared by the Tukey's test at a 95 % confidence interval ($P < 0.05$), after confirmation of normality and homogeneity of variance (Zar 1999). The data are presented as mean ± SEM of the replicate groups (3 tanks per treatment; $n = 3$). Statistical analyses were performed with Statistica 7 (Stat Soft. Inc. Tulsa, OK, USA).

Results

Over the course of the feeding trial, the survival rate was 100 % among the groups and final gain was twofold higher than the initial weight. Weight, feed intake and growth of juvenile flounder were significantly affected by feeding regimes. Final mean weight of juveniles fed with F1 was significantly lower than those fed with F2, F3 and F4 ($P < 0.05$, Table 2). The highest SGR was obtained in juveniles fed with F2, F3 and F4. Feeding frequencies did not affect FCR or body size among the groups as $\text{CV}_{\text{weight}}$ ($P > 0.05$) (Table 2). Initial mean K was 1.12, and final means K were 1.19, 1.17, 1.17, and 1.16 for fish in the F1, F2, F3 and F4 groups, respectively.

Different daily food consumption patterns were found among the treatments. Fish fed with F2 ingested significantly more at 0800 h than at 1700 h ($P < 0.05$), whereas fish fed

Table 2 Growth performance and feed utilization of the juvenile flounder fed at different feeding frequencies

Groups	F1	F2	F3	F4	SEM
Final weight (g)	5.06^{b}	5.91^{a,b}	6.24^{a}	6.16^{a}	0.06
Feed intake (g)	109.6^{c}	128.2^{b}	148.8^{a}	146.7^{a}	14.8
WG (%)	109.3^{b}	131.1^{a,b}	146.1^{a}	139.5^{a}	12.6
SGR (%/day)	1.8^{b}	2.0^{a,b}	2.1^{a}	2.1^{a}	0.02
FCR	0.90	0.83	0.87	0.88	0.00
Final CV_{weight}	44.6	48.0	40.0	48.4	5.74

The square root of data is reported as the common SEM and extracted from ANOVA table

Values in the same row with different letters are significantly different ($P < 0.05$)

Weight Gain; $WG = [(\text{final body weight} - \text{initial body weight})/\text{initial body weight}] \times 100$

Specific Growth Rate; $SGR = 100 \times (\ln \text{final weight} - \ln \text{initial weight})/\text{day}$

Feed Conversion Ratio; $FCR = \text{feed intake}/\text{weight gain}$

Coefficient of Variation; $CV = 100 \times SD/\text{mean weight of the fish in each tank}$

with F3 daily consumed significantly more at 0800 h than at 1230 and 1700 ($P < 0.05$). Yet, fish fed with F4 ate similar amounts of feed at each feeding time (Fig. 1).

The results presented in Table 3 show that the moisture and ash contents did not differ significantly among the groups. The feeding frequency had significant effect on lipid and protein content of juvenile flounder. Fish fed four meals a day had the lowest protein content.

Nitrogen and lipid utilizations of the fish fed with different feeding frequencies are presented in Table 4. Nitrogen and lipid intakes were significantly higher in fish fed with F3 and F4 treatment. Similarly, ascending feeding frequency caused significantly declines in the retention of nitrogen ($P < 0.05$). The losses of nitrogen rose with feeding frequency, being significantly different between F1 and F4.

Discussion

In the present study, feeding frequency had significant effects on growth and feed consumption in juvenile flounder. Both parameters seem to increase with the feeding frequency, while further increases in feeding frequency did not result in more growth. Previous studies carried out on other species have also demonstrated that feed consumption and growth increased linearly with an increase in the feeding frequency up to a certain level (Wang et al. 1998; Aydın et al. 2011b). As growth was not significantly enhanced by increasing the number of meals from two to four, feeding three times a day seems to be sufficient for maximum growth in juvenile flounder. The reason for this observation may be attributed to the fact that when the intervals between meals is short, the food passes through the digestive tract more quickly, resulting in less effective digestion (Liu and Liao 1999; Riche et al. 2004; Biswas et al. 2010). Moreover, increasing feeding intervals all day long can increase foraging activity (Johansen and Jobling 1998) and/or oxygen consumption (Guinea and Fernandez 1997), leading to inferior growth rate in fish. However, one should bear in mind that feeding activity of flounder, as other flatfish, is different from pelagic fish. Thus, the current findings for juvenile flounder appear to differ from those

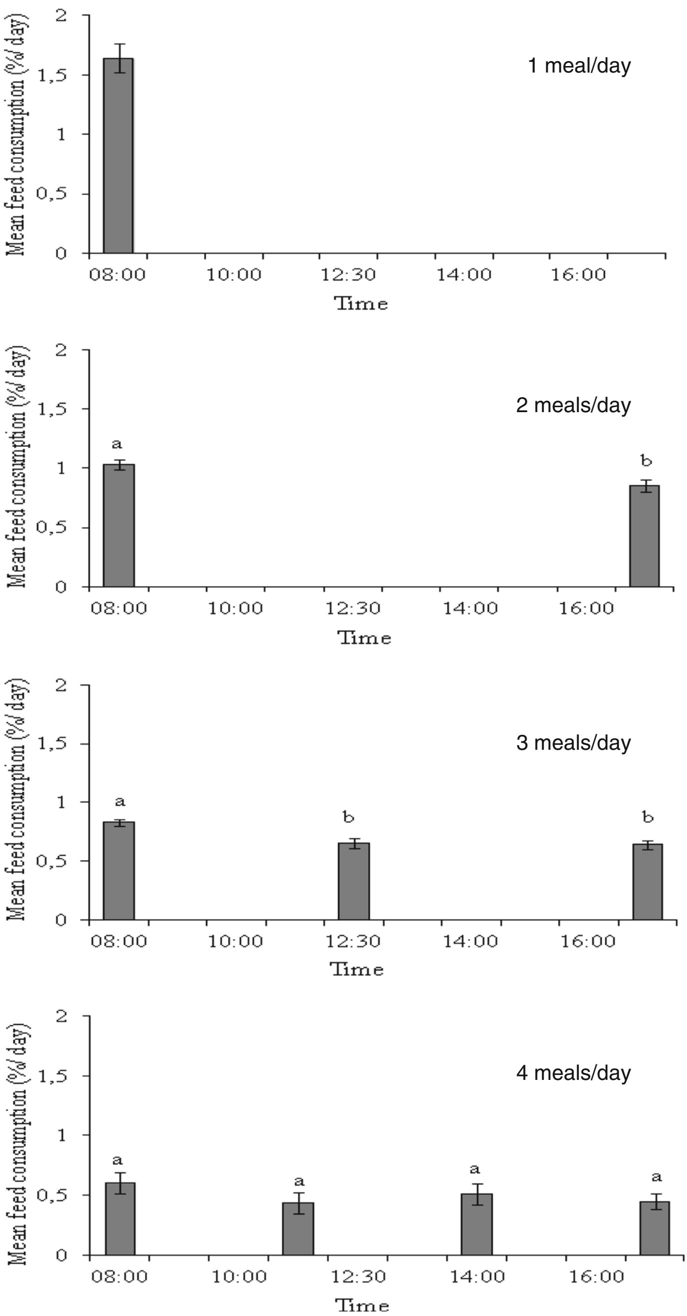

Fig. 1 Mean feed consumption (%/day) per meal at four feeding times of flounder fed at four different feeding frequencies. The data are reported as mean values with their standard error mean ± SEM ($n = 3$). *On top of the bars*, *lower case letters* that are the *same* indicate no significant differences between treatments

Table 3 Proximate composition (%, wet weight basis) of whole body of juvenile flounder fed at four different feeding regimes

	Initial	F1	F2	F3	F4	SEM
Crude protein	15.6 ± 0.29	16.8^a	16.6^a	16.6^a	15.8^b	0.18
Crude lipid	6.4 ± 0.52	5.6^b	6.8^b	8.7^a	8.2^a	0.02
Crude ash	2.1 ± 0.32	3.2	2.6	2.5	2.3	0.31
Moisture	74.8 ± 0.83	74.0	72.6	71.1	72.5	3.97

Initial data are reported as the ± SEM ($n = 3$). The square root of group data is reported as the common SEM and extracted from ANOVA table

Values in the same row with different letters are significantly different ($P < 0.05$)

Table 4 Lipid and nitrogen utilization of juvenile flounder fed with different frequency

	Groups				
	F1	F2	F3	F4	SEM
Lipid					
Intake (g/kg ABW/day)	0.97^c	1.09^b	1.49^a	1.47^a	0.00
Gain (g/kg ABW/day)	0.88^c	1.18^b	1.44^a	1.34^a	0.01
Retention (%)	91.2^b	108.3^a	96.6^{a,b}	91.4^b	2.93
Nitrogen					
Intake (g/kg ABW/day)	0.60^b	0.67^b	0.92^a	0.91^a	0.00
Gain (g/kg ABW/day)	0.47^a	0.46^a	0.39^b	0.35^b	0.00
Retention (%)	78.82^a	68.58^b	42.05^c	38.40^c	7.70
Loss (g/kg/WG)	7.50^b	12.56^b	38.46^a	41.12^a	13.82

The square root of data is reported as the common SEM and extracted from ANOVA table

Values in the same row with different letters are significantly different ($P < 0.05$)

Average Body Weight (ABW) = [final weight (g) + initial weight (g)]/2

Nutrient Gain (g/kg ABW/day) = [(final body nutrient content (g) − initial body nutrient content (g)]/kg ABW)/day

Nutrient Retention (%) = 100 × (Nutrient Gain/Nutrient Intake)

Nutrient Loss (g/kg/WG) = [nutrient fed (g) − nutrient deposited (g)]/WG (kg)

reported for the schooling species, such as Atlantic salmon (*Salmo salar*) and gilthead sea bream (*Sparus aurata*) (Guinea and Fernandez 1997; Johansen and Jobling 1998).

As shown in Table 1, the effects of the feeding frequency on numerous flatfish species depend on several factors including fish size, species, feed composition and type. In general, increasing feeding frequency results in escalations in food consumption (Riche et al. 2004; Türker 2006). This is likely because of the fact that when the interval between meals is short, the food passes through the digestive tract more quickly, resulting in less effective digestion as reported in hybrid striped bass fed at higher frequencies (Liu and Liao 1999). While the assessment of gastric evacuation rate was not the major focus of this study, results of the present study clearly show that juvenile flounder receiving meals at 3-h intervals (F4) exhibited gastric overload, resulting in inefficient digestion and lower utilization efficiency. Our findings on feed intake match with the results obtained from Nile tilapia (*Oreochromis niloticus*) (Riche et al. 2004) and turbot (*P. maxima*) (Türker 2006) but disagree with the report for hybrid sunfish (Wang et al. 1998), in which the authors report an insignificant effect of feeding frequency on feed intake.

In general, feeding frequency influences daily feed intake and feed utilization in fish. However, in line with past studies investigating the feeding frequency and feed efficiency, the results of this study showed that FCR did not differ by feeding frequency in European flounder (Webster et al. 1992; Wang et al. 1998; Türker 2006; Aydın et al. 2011b). Feed efficiency, related to gastric evacuation, is also influenced by physical and chemical properties of the feed (Jobling 1983; Thompson et al. 2000). Lee and Pham (2010) reported that extruded pellets may slow the gastric evacuation rate and in turn delay the return of appetite, thereby increasing feed efficiency in juvenile olive flounder (*P. olivaceus*). However, in our study, no significant FCRs, detected among the feeding groups, suggest that different nutrient utilization may not be affected by feeding frequency in juvenile flounder fed with floating-extruded diet under optimal water temperature. Brown (2010) reported that when access to slow-sinking pellets in shallow water is impeded, aggression and collisions during feeding were more frequently observed in halibut (*H. hippoglossus*). The coefficient of variation of body weight is basically used to distinguish size variations which are induced by competition or hierarchy effects (Jobling 1982; Sunde et al. 1998). In the present study, size variations among the four groups of flounder juveniles fed with floating-extruded diet were identical, indicating that aggressive behavior is low in flounder.

The present study demonstrated that reduced feeding frequency (F1) resulted in low body lipid content of fish, which is in agreement with earlier studies (Andrews and Page 1975; Chua and Teng 1978; Wang et al. 1998; Lee et al. 2000b; Dwyer et al. 2002). In our study, it was observed that whole-body lipid content linearly correlated with feed intake. There is a strong evidence that lipid content of the fish related to dietary energy level, indicating that excess dietary energy is not utilized by fish for growth, it is deposited as lipid (Lee et al. 2000a; Lee and Pham 2010). Lee et al. (2000a) have reported that when feeding frequency is increased up to three times a day, fish better utilize low energy diets (12.7 kJ/g) compared to high energy diets (17.2 kJ/g) in the flounder, *P. olivaceus*. In our study, the energy level of extruded diet was 19.8 kJ/g, even higher than that reported by Lee et al. (2000a).

In line with past studies investigating the effects of feeding frequency, results of the current study clearly showed that nitrogen retention declines with an increase in feeding frequency (Wang et al. 2007). Rates of nitrogen loss (excretion) in our study were markedly elevated with increasing feeding frequency. These findings further support the proposal that flounder vary the balance between ammonia and urea as the major end products of nitrogen excretion. This is supported by Carter and Bransden (2001) in greenback flounder (*Rhombosolea tapirina*) fed on restricted feeding regimes.

In conclusion, the present study provides evidence that, based on nutrient utilization of juvenile flounder (2.5 g), a feeding frequency of two times a day seems to be sufficient for maximal growth and lowest nitrogen losses under our experimental conditions.

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References

Andrews JW, Page JW (1975) The effects of frequency of feeding on culture of catfish. Trans Am Fish Soc 104:317–321

AOAC (1995) Official methods of analysis of the Association of Analytical Chemists. Arlington, VA, USA
Aydın İ (2012) The external abnormalities of hatchery-reared Black Sea flounder (*Platichthys flesus luscus* Pallas, 1814) in Turkey. Turk J Fish Aquat Sci 12:127–133
Aydın İ, Ak O, Küçük E, Polat H, Ceylan B (2011a) Optimum temperature and growth performance of hatchery reared Black Sea flounder (*Platichthys flesus luscus* Pallas, 1814). Turk J Vet Anim Sci 36:101–106
Aydın İ, Küçük E, Şahin T, Kolotoğlu L (2011b) The effect of feeding frequency and feeding rate on growth performance of juvenile Black Sea Turbot (*Psetta maxima* Linneaus, 1758). J Fishscicom 5:35–42
Aydın İ, Şahin T, Polat H, Güneş E (2011c) Reproductive performance of wild and hatchery-reared flounder, *Platichthys flesus luscus*, in the Southern Black Sea Coast. Turk J Zool 35:811–817
Biswas G, Thirunavukkarasu AR, Sundaray JK, Kailasam M (2010) Optimization of feeding frequency of Asian seabass (*Lates calcarifer*) fry reared in net cages under brackishwater environment. Aquaculture 305:26–31
Brown N (2010) Halibut aquaculture in North America. In: Daniels HV, Watanabe WO (eds) Practical flatfish culture and stock enhancement. Wiley-Blackwell, Ames, pp 3–29
Carter CG, Bransden MP (2001) Relationships between protein-nitrogen flux and feeding regime in greenback flounder, *Rhombosolea tapirina* (Günther). Comp Biochem Phys A 130:799–807
Chua TE, Teng SK (1978) Effects of feeding frequency on the growth of young estuary grouper, *Epinephelus tauvina* (Forskål), cultured in floating net-cages. Aquaculture 14:31–47
Dwyer KS, Brown JA, Parrish C, Lall SP (2002) Feeding frequency affects food consumption, feeding pattern and growth of juvenile yellowtail flounder (*Limanda ferruginea*). Aquaculture 213:279–292
Goddard S (1996) Feed management in intensive aquaculture. Chapman and Hall, New York, p 194
Guinea J, Fernandez F (1997) Effect of feeding frequency, feeding level and temperature on energy metabolism in *Sparus aurata*. Aquaculture 148:125–142
Jobling M (1982) Some observations on the effects of feeding frequency on the food intake and growth of plaice, *Pleuronectes platessa* L. J Fish Biol 20:431–444
Jobling M (1983) Effect of feeding frequency on food intake and growth of Arctic charr, *Salvelinus alpinus* L. J Fish Biol 23:177–185
Johansen SJS, Jobling M (1998) The influence of feeding regime on growth and slaughter traits of cage-reared Atlantic salmon. Aquac Int 6:1–17
Kim KD, Kim KM, Kang YJ (2007) Influences of feeding frequency of extruded pellet and moist pellet on growth and body composition of juvenile Japanese flounder *Paralichthys olivaceus* in suboptimal water temperatures. Fish Sci 73:745–749
Kubitza F, Lovshin LL (1999) Formulated diets, feeding strategies, and cannibalism control during intensive culture of juvenile carnivorous fishes. Rev Fish Sci 7:1–22
Lee SM, Pham MA (2010) Effects of feeding frequency and feed type on the growth, feed utilization and body composition of juvenile olive flounder, *Paralichthys olivaceu*. Aquac Res 41:166–171
Lee SM, Cho SH, Kim DJ (2000a) Effects of feeding frequency and dietary energy level on growth and body composition of juvenile flounder, *Paralichthys olivaceus* (Temminck and Schlegel). Aquac Res 31:917–921
Lee SM, Hwang UG, Cho SH (2000b) Effects of feeding frequency and dietary moisture content on growth, body composition and gastric evacuation of juvenile Korean rockfish (*Sebastes schlegeli*). Aquaculture 187:399–409
Liu FG, Liao IC (1999) Effect of feeding regimen on the food consumption, growth, and body composition in hybrid striped bass *Morone saxatilis* × *Morone chrysops*. Fish Sci 65:513–519
Riche M, Oetker M, Haley DI, Smith T, Garling DL (2004) Effect of feeding frequency on consumption, growth, and efficiency in juvenile Nile tilapia (*Oreochromis niloticus*). Isr J Aquacult-Bamid 56(4):247–255
Şahin T (2000) Larval rearing of flounder, *Pleuronectes flesus luscus*, under laboratory conditions. Turk J Mar Sci 6:263–270
Şahin T, Güneş E, Aydın İ, Polat H (2008) Reproductive characteristics and egg development in flounder (*Pleuronectes flesus luscus*) in the southern Black Sea. Isr J Aquacult-Bamid 60:20–26
Schnaittacher G, King WV, Berlins LD (2005) The effects of feeding frequency on growth of juvenile Atlantic halibut, *Hippoglossus hippoglossus* L. Aquac Res 36:370–377
Sunde LM, Imsland AK, Folkvord A, Stefansson SO (1998) Effects of size grading on growth and survival of juvenile turbot at two temperatures. Aquac Int 6:19–32
Thompson KR, Webster CD, Morgan AM, Grisby EJ (2000) Effects of different feeding frequencies on growth, body composition, and fillet composition of juvenile sunshine bass, *Morone chrysops* × *M. saxatilis*. Grown Indoors. J Appl Aquac 10:55–65
Türker A (2006) Effect of feeding frequency on growth, feed consumption, and body composition in juvenile turbot (*Psetta maxima* Linaeus, 1758) at low temperature. Turk J Vet Anim Sci 30:251–256

Wang N, Hayward RS, Noltie DB (1998) Effect of feeding frequency on food consumption, growth, size variation and feeding pattern of age-0 hybrid sunfish. Aquaculture 165:261–265

Wang Y, Kong L, Li K, Bureau DP (2007) Effects of feeding frequency and ration level on growth, feed utilization and nitrogen waste output of cuneate drum (*Nibea miichthioides*) reared in net pens. Aquaculture 271:350–356

Webster CD, Tidwell JH, Yancey DH (1992) Effect of protein level and feeding frequency on growth and body composition of cage-reared channel catfish. Prog Fish Cult 54:92–96

Xie F, Ai Q, Mai K, Xu W, Ma H (2011) The optimal feeding frequency of large yellow croaker (*Pseudosciaena crocea* Richardson) larvae. Aquaculture 311:162–167

Zar JH (1999) Biostatistical analysis, 4th edn. Prentice Hall, New Jersey