Effect of different levels of dietary Betaine on growth performance, food efficiency and survival rate of pike perch 
(Sander lucioperca) fingerlings

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Abstract
A 6-week feeding experiments were carried out to determine the effects of different levels of dietary betaine on growth performance, food efficiency and survival rate of pike perch which has a critical period during transition feeding. Fingerlings of pike perch were fed with live food (treatment A), without betaine added to biomar (treatment B), 1% betaine added to biomar (treatment C) and 2% betaine added to biomar (treatment D) in 1000 l concreted tanks. 200 fingerlings (1.50 g, mean weight) were stocked in each tank and fed up to 7 meals per day. Higher increment in body weight (4.99 ± 0.73 g), specific growth rate (3.90 ± 0.06) and food efficiency (104.42 ± 4.27) were obtained with treatment D (2% betaine added to biomar). This treatment also showed significant (P<0.05) decrease in food conversion ratio (0.93 ± 0.04) in comparison with other treatments. Highest survival rate was observed in treatment B (biomar without betaine) with a 34.5 % rate and greatest cannibalism (1.03 ± 0.01) was found in treatment D, respectively. It was concluded that betaine could increase the palatability and acceptability of food and could be suitable to weaning the pike perch fingerlings to artificial diet.

Keywords: Sander lucioperca, Betaine, Food attractants, Growth, Food efficiency

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Introduction

Pike perch, *S. lucioperca*, is one of the most commercially important species of indigenous ichthyofauna of the Caspian Sea (Sattari et al., 2003). In past decades, the stocks of this fish in Iran have increasingly declined due to overfishing, degradation of spawning ground and etc (Kiabi et al., 1999). Kiabi et al. (1999) considered this fish as a vulnerable species in the southern Caspian Sea basin according to IUCN criteria. Artificial raising and releasing of pike perch fingerlings has done to restocking of this fish in Iran. However, because of high meat quality, fast growth and high acceptability, this species is highly demanded in recent years (Jankowska et al., 2003) and considered as an aquaculture species in European countries such as Hungary, Poland and etc (Steffens et al., 1996). But, there is a critical period in rearing of juvenile pike perch during converting from natural diet to commercial feed which is important to study for introducing it to aquaculture system. According to traditional culturing method, the fingerlings of pike perch rear for 4-6 weeks in nursery pond to reach the length of 3-5 cm, and then keep with carps to produce yearling fish (Antalfi, 1979). Actually, the larvae of pike perch starts feeding on zooplankton such as rotifers and then copepods (Verreth and Kleyn, 1987), switching to higher size foods such as chironomids, crustaceans, mollusks and leeches with growth (Steffens et al., 1996). The transition from planktonivorous to carnivorous habit of pike perch happens when they reach to 4 cm in length (Antalfi, 1979). This period is very critical to fingerlings, because it cause to differences in fish size in the absence of adequate prey fish. This problem leads to cannibalistic habits (Woynarowich, 1960; Steffens et al., 1996). Using dry feed in pike perch rearing is a new idea (Hilge, 1990) which makes possible the culture of this fish in intensive culture system (Zakes, 1997). Due to carnivorous feeding habit and little information of nutritional requirements of this fish, the current recommendations are using the formulated trout and salmon (Brown et al., 1996; Melard et al., 1996) and sparids (Fontaine et al., 1996) feeds. Adding the food attractants to formulated diet has been used to overcoming the problem of transition from natural to formulated diet. Chemo-attractants has been added to formulated diet to overcoming the problems of them rejection by fish (Fast and Lester, 1992), increasing the consumption of poor quality food (Costa-Pierce and Laws, 1985), changing the feeding behavior and making better food acceptance by stimulating the smell and taste systems (Hughes, 1990), minimizing the time that feed remains in water and increasing the possibility of replacement of plants proteins with fish meal and also minimizing the leaching of water soluble nutrients (Polat and Beklevik, 1998). It has been reported that betaine is one of most important food attractants could be used in transition feeding habits of pike perch (Yilmaz and Ablak, 2003). Due to importance of pike perch and little information on effect of food attractant in culture of this fish, this
study aims to investigate the effect of different levels of dietary betaine on growth performance, food efficiency and survival rate of pike perch fingerlings.

**Materials and methods**

This study was conducted in Yosefpour's Fish Breeding and Culture Center, Seyahkal (Guilan, Iran) from May to July 2010. Experiment was performed with four treatments in triplicates, in 12 veniro tanks of 1000 l volume (with dimensions of 0.8 x 0.8 m) with completely randomized design (Baranek et al., 2007). Each veniro contained a volume of 400 l of water with proper aeration. The water inlet with a flow rate of 5-6 l/min, created a slight circulation current in the tank. The initial stocking rate was 2 fish/l, (same size and from same stock) (Bodis, 2007). Light intensity was 40-60 lux for 24 h in rearing saloon (Zakes, 1997). The pond cultured pike perch fingerlings (initial mean weight of 1.49 g) were transported to veniro tanks and acclimated for 2 days. Then fish were feed by four diets as follows: Live food (frozen chironomids) (group 1), starter food of trout (SFT) with pellet size of 1.1 mm for first half of rearing period and 2.5 mm for second half of rearing period (group 2), SFT + 1% betaine (Arya Dalman, Germany) (group 3) and SFT (France) + 2% betaine (group 4) for 6 weeks. The fingerlings were hand-fed from 8:00 to 20:00 with formulated and live food every 2 and 4 h, respectively (Yilmaz and Ablak, 2003). Feeding rate was 8-9 % of actual fish biomass of group 1 and 3-4 % of actual fish biomass for other groups. The amount of trout pellets in the total diet increased from zero through 25, 50, 75 and 100 % in 3-day intervals (Bodis, 2007) Approximate Analytical SFT food is presented in Table 1.

![Table 1: Formulation and proximate composition of experimental diets](image)

<table>
<thead>
<tr>
<th>Composition</th>
<th>0.8-1.1 (mm)</th>
<th>1.5-2.2 (mm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Crude proteine (%)</td>
<td>56</td>
<td>54</td>
</tr>
<tr>
<td>Crude lipid (%)</td>
<td>18</td>
<td>18</td>
</tr>
<tr>
<td>Fiber (%)</td>
<td>0.4</td>
<td>1.5</td>
</tr>
<tr>
<td>Ash (%)</td>
<td>10.5</td>
<td>10.5</td>
</tr>
<tr>
<td>NFE¹</td>
<td>9.5</td>
<td>10.0</td>
</tr>
<tr>
<td>Phosphorus (%)</td>
<td>1.6</td>
<td>1.58</td>
</tr>
<tr>
<td>Vitamin A (IU kg⁻¹)</td>
<td>7500</td>
<td>7500</td>
</tr>
<tr>
<td>Vitamin E (mg kg⁻¹)</td>
<td>400</td>
<td>400</td>
</tr>
<tr>
<td>Vitamin C (mg kg⁻¹)</td>
<td>1000</td>
<td>500</td>
</tr>
<tr>
<td>Gross energy</td>
<td>22.1</td>
<td>22.2</td>
</tr>
</tbody>
</table>

1- Nitrogen-free extracts including crude fiber

Every 7 days 20 fish from each veniro (60 individuals/ treatment) were randomly captured and then length were measured individually, with 0.1 mm accuracy. Fingerlings were weighted collectively from day 0 to day 14, and individually from day 21 to day 42 with 0.01 mg accuracy. The number of sampled
fingerlings was taken into account for survival calculation. Body weight index (BWI), average daily growth (ADG), specific growth rate (SGR), condition factor (CF), food conversion ratio (FCR), food efficiency (FE), survival rate (SR) and cannibalism were assessed as follows (El-Husseiny et al., 2008):

Weight gain (g) = (Wf – Wi)

Average daily growth (ADG, %) = 100 (Wf – Wi) / T

Specific growth rate (SGR, % day\(^{-1}\)) = 100 (LnWf – LnWi) / ∆T

Condition factor (CF) = 100 × Wf × (Lf)\(^{-3}\)

Feed conversion ratio (FCR) = W\(_{TFS}\) / AWG\(^{-1}\)

Feed efficiency (FE) = (FB – IB) × TFC\(^{-1}\)

Counted mortality (%) = 100 (Nd + Nc) / Ni

Survival rate = 100 (Nf – Ni)

Cannibalism (%) = 100 (Nc + Nm) / Ni

Abbreviations in above-mentioned equations are as follow: where \(W_i\) and \(W_f\) are the initial and final body weights (g), \(W_{TFS}\) is the weight of the total feed supplied (g) \(T = \text{duration of experiment (days)}, L = \text{final body length (cm)}, Ni = \text{initial number of fingerlings}, Nf = \text{final number of fingerlings}, Nd = \text{number of dead fish without signs of cannibalism}, Nc = \text{number of dead fish due to cannibalism}, Nm = \text{number of missing fish at the counting (end of experiment)}, C = \text{Cannibalism}, \Delta T = \text{duration of the experiment (days)}, TFC = \text{the total food consumption (g)}, IB = \text{the initial biomass (g)}, FB = \text{the final biomass (g)}.

Statistical analysis:

Results are given as mean and standard deviations. The SPSS (version 13.0) was used for statistical analysis. One-way ANOVA with Duncan's test was used to test the effect of different feed treatments on growth performance, food efficiency and survival rate of pike perch fingerlings. Statistical difference was accepted when \(P < 0.05\).

Results

Weekly monitored water quality parameters are presented in Table 2. Water temperature variations were high and ranged from 23.74 to 29.67 \(^\circ\)C during experiment. Dissolved oxygen concentration was high during experiment and never fell below 6.61 mg O\(_2\)/l.

<table>
<thead>
<tr>
<th>Physico-chemical parameters</th>
<th>Duration of experiment (weeks)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>1</td>
</tr>
<tr>
<td>Water temperature (°C)</td>
<td>23.74 ± 0.18</td>
</tr>
<tr>
<td>DO (mg/l)</td>
<td>6.75 ± 0.39</td>
</tr>
<tr>
<td>pH</td>
<td>8.43 ± 0.21</td>
</tr>
</tbody>
</table>

Data are mean ± S.D.
The results of growth performance of pike perch fingerling were fed with four experimental diets during the first four weeks are shown in Table 3. At the end of week four, the stock of live food finished and from this point to end of experiment, the fingerlings were fed by biomar without betaine (treatment B), 1% betaine added to biomar (treatment C) and 2% betaine added to biomar (treatment D) and comparisons are as presented in Table 4.

**Table 3: Effects of different diets on growth indices of pike perch fingerling until week 4**

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Treatments</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>live food (treatment A)</td>
</tr>
<tr>
<td>Initial weight (g)</td>
<td>1.47 ± 0.18 a</td>
</tr>
<tr>
<td>Final weight (g)</td>
<td>1.75 ± 0.62 b</td>
</tr>
<tr>
<td>Weight gain (g)</td>
<td>0.37 ± 0.54 b</td>
</tr>
<tr>
<td>ADG (%)</td>
<td>1.32 ± 0.63 c</td>
</tr>
<tr>
<td>SGR (% per day)</td>
<td>3.90 ± 0.14 a</td>
</tr>
<tr>
<td>Initial length (mm)</td>
<td>5.68 ± 0.30 a</td>
</tr>
<tr>
<td>Final length (mm)</td>
<td>6.26 ± 0.60 b</td>
</tr>
<tr>
<td>Initial CF</td>
<td>0.76 ± 0.05 a</td>
</tr>
<tr>
<td>Final CF</td>
<td>0.54 ± 0.04 b</td>
</tr>
<tr>
<td>FCR</td>
<td>3.44 ± 0.25 a</td>
</tr>
<tr>
<td>FE</td>
<td>30.25 ± 5.79 c</td>
</tr>
<tr>
<td>Survival rate (%)</td>
<td>8.69 ± 0.08 c</td>
</tr>
<tr>
<td>Cannibalism (%)</td>
<td>0.83 ± 0.02 a</td>
</tr>
</tbody>
</table>

Values (mean ± SD) with different letters in the same line indicate significant differences (P<0.05).

**Table 4: Effects of different diets on growth indices of pike perch fingerlings at week 6**

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Treatments</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>live food (treatment A)</td>
</tr>
<tr>
<td>Initial weight (g)</td>
<td>1.47 ± 0.18 a</td>
</tr>
<tr>
<td>Final weight (g)</td>
<td>0 ± 5.72 ± 0.16 b</td>
</tr>
<tr>
<td>Weight gain (g)</td>
<td>0 ± 4.22 ± 0.18 c</td>
</tr>
<tr>
<td>ADG (%)</td>
<td>0 ± 10.04 ± 1.30 b</td>
</tr>
<tr>
<td>SGR (% per day)</td>
<td>0 ± 3.46 ± 0.03 b</td>
</tr>
<tr>
<td>Initial length (mm)</td>
<td>5.68 ± 0.30 a</td>
</tr>
<tr>
<td>Final length (mm)</td>
<td>0 ± 8.55 ± 0.73 ab</td>
</tr>
<tr>
<td>Initial CF</td>
<td>0 ± 0.76 ± 0.05 a</td>
</tr>
<tr>
<td>Final CF</td>
<td>0 ± 0.87 ± 0.07 a</td>
</tr>
<tr>
<td>FCR</td>
<td>0 ± 1.16 ± 0.03 a</td>
</tr>
<tr>
<td>FE</td>
<td>0 ± 85.92 ± 4.84 b</td>
</tr>
<tr>
<td>Survival rate (%)</td>
<td>0 ± 34.50 ± 0.06 a</td>
</tr>
<tr>
<td>Cannibalism (%)</td>
<td>0.83 ± 0.02 b</td>
</tr>
</tbody>
</table>

Values (mean ± SD) with different letters in the same line indicate significant differences (P<0.05).
Average fish fingerlings weight were 1.75, 3.44, 3.39 and 3.84 g for treatments A, B, D and C, respectively, at the end of week 4 (Table 3). The weight was increased to 5.72, 5.57, 6.52 g for treatments B, D and C, respectively, at the end of feeding trial (Table 4). Daily specific growth rate was significantly ($P<0.05$) higher in treatment D than others at the end of experiment. SGR (% per day was higher in week for than week 6 for all treatments. The final condition factor at week 4, ranged from 0.54 for treatment A (live food) to 0.86 for treatment C (1% betaine + biomar), respectively. There was no significant ($P<0.05$) difference on CF of fingerlings fed different diet at the end of experiment. Food conversion ratio was significantly ($P<0.05$) higher in treatment A (live food). Treatment D showed the lowest FCR during experiment. Highest food efficiency was found in fingerlings fed biomar with 2% betaine (treatment D). Survival rate was significantly ($P<0.05$) increased in fingerlings fed formulated diets. The lowest SR was found in treatment A (live food) with 8.69 %, while highest SR was observed in treatment B with 34.5 %.

**Discussion**

In recent years, interest to culture of pike perch has increased due to high meat quality, fast growing and high acceptability (Jankowska et al., 2003). Recognizing the natural food requirement is necessary for successful culture of fish in intensive culture system (Higuera, 2001). At present, the formulated trout food is used as preferred food for pike perch culture. Chemo-attractants has used for different fish species and showed good results to overcoming some problems such as food rejection by fish, increasing the consumption of poor quality food and etc (Kasumyan and Doving, 2003). Betaine is one of the most important food attractants which could be used in transition feeding habits of pike perch (Yilmaz and Ablak, 2003). Although these compounds did not increase the nutritional value of formulated diet but they are useful to increase the food palatability and acceptance and increase the consumption of poor quality food (Fekrandish et al., 2005).

The present study demonstrated that the addition of betaine in different level to trout starter food (biomar) improve the growth performance and best results found in treatment D (2 % betaine + biomar). Similarly, betaine improved the growth performance (weight gain, specific growth rate and condition factor) in rainbow trout at 3 % level (Sadeghi, 2004), on salmon at 1.5 and 2.0 % level (Polat and Beklevik, 1998) and on yearling Chinook salmon (Clarke et al., 1994). In contrast, several other studies reported that betaine did not improve the growth performance in *Orechromis aureus* (Genc et al., 2006). The effect of betaine-supplemented food on growth improvement in some fish has just observed in food transition period, when increased palatability of food could be an important factor for increasing appetite (Kasumyan and Doving, 2003).

Highest survival rate was found in fingerlings fed biomar without betaine (treatment B) and showed that addition of betaine had not effect on survival rate. The study of changes in survival rate showed that mortality of pike perch fingerlings
was high in first and third weeks. This could be due to changing the feeding behavior at first week and probably to non consumption period and increasing in water temperature, respectively. The pike perch fingerling is very sensitive to higher temperature.

It has been reported that the mortality of fish is increased if there is no enough food in transition period (Hamza et al., 2008). Highest mortality has also reported for pike perch fish during first week of experiment (Kestemont et al., 2007). In current work, the fluctuation of water temperature was high, which affects on metabolism rate, food consumption, growth performance and survival rate. In our experiment, cannibalism behavior was found in second week in treatments A, B and C and in third week in treatment D. The highest cannibalism was found in treatment A at week four. This ratio was increased in treatment D at the end of experiment. It has been demonstrated that cannibalism is influenced by food type, food availability, difference in fish size (Kestemont et al., 2007), difference in food particle size (Szkudlarek and Zakes, 2007) and improper fish fingerling stocking in pond (Adams et al., 2000).

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