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Effects of different photoperiods on the survival and growth of beluga sturgeon (*huso huso*) larvae

Hamid Eshagh Zadeh*[1], Gholamreza Rafiee[1], Soheil Eagderi[1], Rezvanollah Kazemi[2], Hadi Poorbagher[1]

[1]Department of Fisheries, Faculty of Natural Resources, the University of Tehran, P.O. Box: 31585-4314, Karaj, Iran.
[2]International Sturgeon Research Institute, P.O. Box 41635-3464, Rasht, Iran.

Abstract: Effect of different photoperiodic regimes was evaluated on growth performance and survival rate of the Beluga (Huso huso) prelarvae and larvae. Newly hatched prelarvae were stored in 5 round fiberglass 500 L tank with different photoperiod (24L:00D, 18L:06D, 12L:12D, 06L:18 D, 00L:24D) till 50 days post hatch with three replicates. Light intensity was 200 lux during the experiment. Feeding was started from 8 days post hatch using live artemia nauplii. Higher total length, survival rates and lower body area, yolk area for beluga prelarvae obtained in long light photoperiods (24L:00D, 18L:06D). Also, higher growth parameters of the beluga larvae observed in long light photoperiods while different photoperiods had no effect on survival rate. The present study indicated that growth performance and survival rates of larvae are significantly influenced by photoperiod. The photoperiod 18L:06D resulted in the best growth performance and survival rate during early development of the beluga.

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Introduction

Decline of sturgeon stocks in the Caspian Sea has urged the development of artificial propagation programs in hatcheries for both aquacultural and restocking proposes (Bronzi et al., 1999). During artificial propagation of sturgeon fishes, the production of larvae is the most critical and important stage (Yasemi et al., 2011). Among the sturgeon fishes of the Caspian Sea, production of Beluga larvae (*Huso huso*) has drawn a great attention due to its high economic value and suitability for aquacultural propose. Since the main focus of aquaculture is to use the techniques that increase fertilization success, survival rate and growth performance (Verhaegen et al., 2007). The environmental factors such as light intensity and photoperiods, temperature and salinity are considered as important parameters effecting growth and survival of fish larvae (Puvanendran and Brown, 2002; Martínez-Palacios et al., 2004; Van Eenennaam et al., 2005). Light was found an important stimulant during the early development of sturgeon fishes in Chinese sturgeon (*Acipenser sinensis*), Siberian sturgeon (*A. baerii*) and shortnose sturgeon (*A. brevirostrum*) (Zhuang et al., 2002; Richmond and Kynard, 1995). Also, a swim up behavior toward light zone and ontogenetic behavior has been reported in genus *Acipenser*, *Scaphirhynchus* and *Huso* at the time of downward migration indicating important role of light and vision in their ontogenetic behavior (Boglione et al., 1997; Zhuanga et al., 2003).

The role of vision and light preference in prelarvae and larvae of sturgeon fishes have been demonstrated in their behaviors such as orientation, schooling, rheotaxis, phototaxis, avoiding predators and habitat selection. However, there are differences in those behaviors, depending on the ecological

* Corresponding author: Hamid Eshagh Zadeh
E-mail address: hamidshil@yahoo.com

requirements of species, species-specific, their life stage, feeding habitats (OLLA et al., 1996; Kynard and Horgan, 2002; Rodrý´guez and Gisbert, 2002; Arnold, 1974).

During the absorption of yolk-sac, sturgeon larvae have a low density of rod cells in their retina but density of the cells increase later to strengthen the vision under dark conditions (Blaxter and Staines, 1970; Chai et al., 2006). Whereas in most marine fish larvae, light and vision play vital role in early feeding stage because they do not feed at night (Fielder et al., 2002; Villamizar et al., 2009). Hence, this study was conducted to survey the effect of different photoperiodic regimes as an important environmental factor on survival rate and growth performance of farmed-beluga sturgeon from 0 dph (day post hatch) until 50 dph.

Material and Methods

This experiment was accomplished at the Dadman International Sturgeon Research Institute (Guilan, Iran). The specimens were obtained from the artificial propagation of four (three males and one female), nine years old farmed-beluga sturgeon. Newly hatched larvae i.e. eleutheroembryo were immediately introduced into 15 round fiberglass 500-L tanks (105×102×52 cm). The larvae were exposed to the photoperiods D24L:00D, 18L:06D, 12L:12D, 06L:18D and 00L:24D until 50 dph. The experiment was carried out in three replicates with a stocking density of 33 g larvae (with an average weight of 18.2 ± 0.001 mg and total length of 11.20 ± 1.1 mm, [mean ± SD]) per 500 L tank with a water depth of 20 cm till 12 dph and, after that, the water depth increased to 30 cm until the end of experiment. A 60 Watt light fluorescent bulb (Nama noor, Iran) was used as a light source in all treatment. The bulb was suspended over each tank at same height to produce 200 lux light intensity during the experiment. A timer was used to set the dark and light periods. Light intensity was measured using a light meter (TES, model 1336A, Taipei, Taiwan). The water was supplied from ground and river water with a discharge of 400 and 250 mL min^{-1}, respectively. The water temperature, DO and pH of treatments were recorded 15.9, 7.7 and 7.8 during the experiment, respectively. Due to asynchrony in external feeding of prelarvae and to avoid starving and cannibalism, the larvae were fed a diet of *Artemia* nauplii, Daphnia and formulated diets (Biomar 0.5 mm diameter contained 58% protein, 15% fat, 9.8% carbohydrate, 11.9% ash, 1.7% total phosphorus). Feeding was started from 8 dph using live *Artemia franciscana* nauplii (500 nauplii/larvae six times a day). Later, a mixture of Daphnia and Artemia were used from 12 dph to 25 dph. From 20 dph, the percentage of live food decreased gradually and the percentage of formulated diets increased so that from 25 dph to 50 dph, the larvae were fed 100% formulated diets at a rate of 30% total biomass four times a day. Dead larvae were removed and counted daily at each tank to measure the survival rate. Effects of photoperiod on the growth performance and survival rate was investigated in two different stages, i.e. at the end of yolk sac absorption stage (12 dph) and at the end of the experiment (50 dph). At the end of each stage, 30 larvae from each tank were sampled and fixed in 5% buffered formalin. Wet weight of larvae (ww) was measured to the nearest 0.001 g. Left side of specimens were photographed using a stereomicroscope equipped with a 8mp digital camera (Cannon) and their total length (TL), body area (BA) and area of yolk sac (YA), were measured using Image J software. Then, specific growth rate (SGR) and condition factor (CF) were calculated.

Prior to the analysis, normality and homogeneity of variance were checked and percentage data were transformed using arcsine. Data were analysed using one-way ANOVA to examine the effect of photoperiod on growth parameters, survival rate of beluga larvae, followed by Duncan's multiple range test (Zar, 1994). Data are presented as mean ± SD.

Results

At the hatching time (0 dph), YA had a mean of 6.58 mm^2 and decreased to 1.252 mm^2 at 12 dph and there

Table 1. Average (± SD) of growth parameters between photoperiods during yolk sac absorption.

Photoperiod	00L:24D	06L:18D	12L:12D	18L:06D	24L:00D
Final weight (mg)	49.46 ± 2.25[a]	48.80 ± 4.7[a]	49.73 ± 3.2[a]	50.83 ± 4.8[a]	49.56 ± 5.7[a]
Initial weight (mg)	18.2 ± 0.001	18.2± 0. 001	18.2 ± 0.001	18.2 ± 0.001	18.2 ± 0.001
Final total length (mm)	22.83 ± 0.47[a]	23.54 ± 0.88[ab]	23.61 ± 0.62[ab]	23.82 ± 0.47[ab]	24.82 ± 0.56[c]
Initial total length	11.20 ± 1.1	11.20 ± 1.1	11.20 ± 1.1	11.20 ± 1.1	11.20 ± 1.1
Yolk sac area (mm^2)	1.89 ± 0.54[c]	1.68 ± 0.46[bc]	1.68 ± 0.38[bc]	1.43 ± 0.384[a]	1.58 ± 0.37[ab]
Body area (mm^2)	45.36 ± 1.6[b]	42.44 ± 2.4[a]	41.75 ± 1.4[a]	41.43 ± 1.8[a]	41.44 ± 2.5[a]
Specific Growth Rate (%)	8.33 ± 0.97	8.21 ± 1.7	8.37 ± 0.94	8.55 ± 1.2	8.34 ± 1.2
Condition factor (%)	0.416 ± 0.30[c]	0.377 ± 0.55[b]	0.379 ± 0.38[b]	0.374 ± 0.40[b]	0.324 ± 0.36[a]
Survival (%)	60.83 ± 22.27[a]	76.86 ± 10.49[a]	83.71 ± 15.82[ab]	95.86 ± 0.60[b]	91.15 ± 5.23[b]

was no significant difference among treatments ($P < 0.05$). The treatment with longer light period (24L:00D, 18L:06D) had a greater effect on the absorption of the yolk sac than those with shorter light period (12L:12D, 06L:18D, 00L:24D). Maximum and minimum of YA in the prelarvae were recorded in 00L:24D with 1.49 mm^2 and 18L:06D with 1.03 mm^2, respectively.

The mean BA was measured 14 .7160 mm^2 at 0 dph that reached to 42.486 mm^2 in 12 dph and there was a significant difference between photoperiods ($P<0.05$). A higher BA was recorded in 00L:24D, which may be due their larger yolk sacs compared to others. There was a significant difference between photoperiods in TL and CF ($P<0.05$). The maximum and minimum TL were recorded in 24L:00D (24.82 ± 0.56 mm) and 00L:24D (22.83 ± 0.47), respectively. While maximum and minimum CF were recorded in 00L:24D (0.416 ± 0.30) and 24L:00D (0.324 ± 0.36), respectively. There was no significant difference between photoperiods in wet weight of larvae and SGR ($P>0.05$). Mortality of prelarvae started at 0 dph and significant difference was detected between photoperiods in survival rate ($P<0.05$). As shown in Table 1, the long light photoperiods (24L:00D, 18L:06D) increased the survival rate. The maximum and minimum survival rates were found in 18L:006D (95.86%) and 00L:24D (60.83%), respectively.

As shown in Table 2, there were significant differences between photoperiods in growth parameters, whereas no significant difference was detected between photoperiods in survival rate after the absorption of yolk sac. A higher TL and ww was recorded in the photoperiods with longer duration of light (24L:00D, 18L:06D), i.e. in 24L:00D and 18L:06D higher TL (9.82 ± 1.02) and WW (3.47± 1.45) were recorded, respectively. In 12L:12D and 00L:24D lower TL (9.08 ± 0.9) and WW (2.74 ± 1.03) were measured, respectively. 12L:12D and 00L:24D resulted in significantly higher (0.435 ± 0.03) and lower (0.319 ± 0.05) CF ($P < 0.05$). Photoperiod had no significant effect on SGR.

Discussion

The present study indicated that different photoperiods have significant effects on growth parameters and survival rate during and after yolk sac absorption. At the first stage, the earlier yolk sac absorption, lower body surface area, highest total length and survival rates of eleutheroembryo were observed in long light photoperiods, but these photoperiods had no significant effect on the wet weight and SGR. Therefore, according to the results, long light photoperiodic regime result in faster absorption of yolk sacs, i.e. minimum YA and maximum BA in prelarvae. Since the absorption of yolk sac is related to morphogenesis and metabolic processes, long light time photoperiodic regime can increase metabolism and growth performance (Blaxter and Hempel, 1966), which was observed in

Table 2. Average (± SD) of growth parameters between photoperiods after yolk sac absorption.

Photoperiod	00L:24D	06L:18D	12L:12D	18L:06D	24L:00D
Final weight (gr)	2.74 ± 1.03[a]	2.80 ± 0.80[ab]	3.26 ± 1.11[ab]	3.47 ± 1.45[c]	3.37 ± 0.92[bc]
Initial weight (mg)	49.46 ± 2.25[a]	48.80 ± 4.7[a]	49.73 ± 3.2[a]	50.83 ± 4.8[a]	49.56 ± 5.7[a]
Final total length (cm)	9.51 ± 1.20[ab]	9.14 ± 1.24[ab]	9.08 ± 0.9[a]	9.80 ± 1.39[b]	9.82 ± 1.02[b]
Initial total length	22.83 ± 0.47[a]	23.54 ± 0.88[ab]	23.61 ± 0.62[ab]	23.82 ± 0.47[ab]	24.82 ± 0.56[c]
Specific Growth Rate (%)	10.85 ± 1.15	10.94 ± 1.10	11.30 ± 0.90	11.41 ± 1.15	11.40 ± 0.97
Condition factor (%)	0.319± 0.05[a]	0.366± 0.02[ab]	0.435± 0.03[b]	0.369± 0.02[ab]	0.361± 0.01[ab]
Survival (%)	25.46 ± 0.68[a]	25.33 ± 1.45[a]	25.54 ± 1.54[a]	25.62 ± 1.87[a]	26.96 ± 1.04[a]

beluga prelarvae. Our results are consistent with previous studies on the eleutheroembryo of Hadduck (*Melanogrammus aeglefinus*) as those exposed to the photoperiod 24L:00D had smaller BA compared to those exposed to 18L:06D and 12L:12D. In this study, increasing the light intensity decreased YA, but had no effect on BA (Downing and Litvak, 2002). In cod larvae (*Gadus morha*) the different photoperiods had no effect on absorption the yolk sac (Puvanendran and Brown, 2002).

Maximum TL i.e. best growth performance and survival rate were occurred in 24L:00D and 18L:06D during prelarval development, respectively. Whereas the best growth performance observed was not coincided with higher survival rate. This finding is in agree with light preference (positive phototaxis) and selection of white substrates in Siberian sturgeon (*A. baerii)* and Chinese sturgeon (*A. sinensis*) (Chai et al., 2006; Gisbert et al., 1999) Whereas, the low survival of Surubim (*Pseudoplatystoma corruscans*) in the 24L:00D photoperiodic regime has been assigned to increasing swimming activity and energy consumption (Campagnolo and Nuñer, 2008). Also, Puvanendran and Brown (2002) found an increase of survival rate of Cod (*Gadus morha*) larvae in the D24L:00D photoperiods during 0 dph to 28 dph compared with 18L:06D and 12L:12D. However, from 28 dph to 42 dph, there no difference between photoperiods in this species (Puvanendran and Brown, 2002). In addition, our result showed that photoperiod had positive influence on growth and survival rate in prelarval stage, but only on growth performance in larval stage. Similar to previous studies, our study indicated that optimum photoperiods for growth and survival are different (Fielder et al., 2002; El-Sayed and Kawanna, 2004). However in different species continuous light (Campagnolo and Nuñer, 2008), dark (Piaia et al., 1999) and intermediate (Solbakken and Pittman, 2004) photoperiod may act better in comparison to other photoperiods. Other studies on 6-month and one-year old Beluga showed that 12L:12D had the best influence on growth performance and survival rate (Bani et al., 2009; Ghomi et al., 2010). Finally, it is need to be mentioned that dark-light cycles in fish farming changes depend on species based on its habitat and life history. Marine fish larvae dependent on light and vision (Fielder et al., 2002) but sturgeon feeding is not dependent on vision. Nevertheless, presence of light is essential in addition to other factors such as chemical and electrical for optimum growth and feeding.

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