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Optimal stocking density for beluga, *Huso huso*, and ship sturgeon, *Acipenser nudiventris* during the grow-out phase

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1. INTRODUCTION

Sturgeon aquaculture has largely increased during the last decades due to the high market values of caviar and meat (Bronzi & Rosenthal, 2014). As sturgeon farming is a must for the sustainability of natural stocks of these group of endangered fish species (Rosten, 2017), it is critical to optimize rearing and husbandry conditions. In particular, stocking density (SD) is an important factor in the intensive culture of farmed fish, since it does not only affect the profitability of the production system (Ellis et al., 2002), but also affect water quality and fish welfare (Rafatnezhad et al., 2008; Szczepkowski et al., 2011; Ni et al., 2014). Therefore, the aim of this study was to evaluate the effects of different SD during three grow-out phases on growth performance and feed utilization in beluga (*Huso huso*) and ship sturgeon (*Acipenser nudiventris*).

2. MATERIALS AND METHODS

This study was conducted in a commercial sturgeon farm (Dezful, Khuzestan, Iran) in an open flow-through system. Three different production stages (S1, S2 and S3) differing in the initial body weight (BW) were considered for beluga ($140 < BW_{S1} < 500$ g; $500 < BW_{S2} < 1800$ g; and $1500 < BW_{S3} < 3500$ g) and ship sturgeon ($90 < BW_{S1} < 200$ g; $270 < BW_{S2} < 620$ g; $625 < BW_{S3} < 1250$ g). For each production stage, three different SD were tested in concrete square tanks (2.0×1.0×1.2 m^3) with 3 replicates per SD. Tested SD for beluga were as follows: S1: 1.5, 3 and 6 kg m^{-2}; S2: 3, 6 and 9 kg m^{-2}; and S3: 6, 9 and 12 kg m^{-2}. In the case of ship sturgeon, SD for S1 and S2 were similar to beluga, but SD for S3 were 6, 8 and 10 kg m^{-2}. Both species were fed a compound diet (Effico Sigma 840, Biomar, Nersac, France; proximate composition:

43% crude protein, 18% crude fat, 8.1% ash, 3.7% fiber; 17.1 MJ/Kg digestible energy; pellet size: 6.5 and 9 mm for S1 and S2-3, respectively) four times per day (08:00, 12:00, 17:00 and 22:00) during grow-out phases. The S1 lasted for 60 days but S2 and S3 each lasted for 12 weeks. Feed was supplied at a feeding ration of 1–1.5% BW. The first phase Uneaten pellets were drained off and counted every day to calculate the total feed intake per tank using a mean dry pellet weight. Flow rate in experimental tanks was 0.6 L sec^{-1}. Water temperature, pH and dissolved oxygen levels were measured daily in the outlet of each experimental tank by means of a pH meter (HI 81143, Hanna, Cluj-Napoca, Romania) and oxygen meter (HI 9142, Hanna), respectively. Water CO_2, NH_3, NO_2, and NO_3 levels were quantified weekly by spectrophotometric methods (DR/2000 Spectrophotometer, Hach, Loveland, CO, USA) and according to the manufacturer's instructions. Biological oxygen demand (BOD) was measured according to the Winkler titration method (Carpenter, 1965). Photoperiod was natural (32˚22′N, 48˚24′E).

At the end of each rearing stage, all fish were fasted for 24 h and their BW and total length (TL) were measured individually to the nearest 0.1g and 1 mm, respectively. The following standard formulae were used to assess growth performance and feed utilization parameters: weight gain (WG, %) = $[(BW_f – BW_i) / BW_i] \times 100$; specific growth rate (SGR, % day^{-1}) = $[(\ln BW_f – \ln BW_i) / t] \times 100$, where BW_i and BW_f are initial and final body weight (g) of fish and t is experimental period in each S (day); Fulton's condition factor (K, %) = $[BW_f$ (g) / (body length (cm))$^3] \times 100$; Feed intake (FI) = (total ingested feed at the end of each phase (g) / number of fish); Feed conversion ratio (FCR) = FI (g) / WG (g). Data were analyzed using SPSS version 16.0 (Chicago, IL, USA). All the data are presented as mean ± SEM calculated from three biological replicates. Arcsine transformations were conducted on all data expressed as percentages. One-way analysis of variance was performed following the confirmation of normality (Shapiro–Wilk's test) and homogeneity of variance (Levene's test). Tukey's test was used for *post-hoc* multiple comparisons. Significance was determined at $\alpha = 0.05$.

3. RESULTS

As fish were kept in an open-flow water system, water quality parameters were not affected by different SD in different grow-out stages (Table 1). Survival rate was 100% in all experimental groups at all tested production stages. In both species, growth performance was significantly reduced with increasing SD at the end of S1 (Tables 2 and 3; $P < 0.05$). During S2, growth performance decreased in beluga stocked at 9 kg m^{-2} (BW_f, $P = 0.022$), whereas FI was reduced in beluga kept at 6 and 9 kg m^{-2}, when compared to those at 3 kg m^{-2} ($P = 0.001$). In contrast, neither growth nor FI were affected by increasing SD in ship sturgeon during S2 and S3. Interestingly, growth performance and FI did not change with increasing SD during S3 in beluga, which indicated the adaptability of this species to high SD at higher body sizes ($1500 < BW < 3500$). Values of K and FCR were not affected by different SD in either sturgeon species, regardless of the production stage considered (Tables 2 and 3; $P > 0.05$).

4. DISCUSSION

Several studies have reported a reduction in somatic growth with increasing SDs in several sturgeon species due to a deterioration in water quality (Yang et al., 2011; Ni et al., 2016), chronic stress (Ni et al., 2016) and enhancement of social interactions (Rafatnezhad et al., 2008).

Under present experimental conditions, such changes in growth and FI may not be attributed to a reduction in water quality parameters, as no changes were found regardless on the SD considered; thus, the above-mentioned results found during S1 might be attributed to the hierarchical behavior among specimens and competition for feed that resulted in lower FI at higher SD. Results from the S2 revealed that there existed species-specific differences regarding the effect of SD on growth performance and FI between beluga and ship sturgeon. Reduction in growth performance may be attributed to changes in expression of genes related to the growth hormone/insulin-like growth factor axis, as well as changes in lipid and protein metabolism, as described in Amur sturgeon (*A. schrenckii*) (Ren et al., 2018). In this sense, beluga juveniles were more sensitive to high SD than ship sturgeon. These results showed that ship sturgeon (270 < BW < 620 g) is more tolerant to stressing conditions than beluga (500 < BW < 1800 g), although the reasons for such differences between both species need to be further explored.

Finally, it should be highlighted that no changes were found in the FCR among the three tested SD regardless of the sturgeon species and production stage considered. These results may indicate that feed digestion and absorption were not affected under crowding conditions (high SD), regardless of changes in FI in some cases (S1-2 for beluga, and S1 for ship sturgeon). These results are in contrast with those reported in other sturgeon species like Amur sturgeon, beluga and Atlantic sturgeon where adverse effects of increasing SD with a reduction in the feed efficiency associated with growth reduction have been described (Rafatnezhad et al., 2008; Yang et al., 2011; Szczepkowski et al., 2011). These differences among different sturgeon species indicated that there exist species-specific variations regarding crowding tolerance among this group of primitive fishes.

5. CONCLUSION

Considering results from growth performance and FI, the appropriate SD for beluga with body sizes ranging from 140 < BW < 500 g and 500 < BW < 1800 g would be 1.5 and 6 kg m^{-2}, respectively, whereas in larger fish (1500 < BW < 3500 g), beluga specimens were able to withstand up to 12 kg m^{-2} with no negative impacts on somatic growth and feed efficiency parameters. Regarding ship sturgeon, medium and high BW sizes (S2 and S3) adapted well to high SD (9 and 16 kg m^{-2} for fish weighting 270 < BW < 620 g and 625 < BW < 1250 g, respectively), whereas in smaller specimens (90 < BW < 200 g) higher SD than 1.5 kg m^{-2} negatively influenced growth performance and FI. Species-specific differences between both tested sturgeon species with regard to crowding tolerance under different production stages needs to be further investigated in order to provide insight into the physiological mechanisms underlying this differential response between beluga and ship sturgeon.

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