Notas Científicas

Growth and stress of dourado cultivated in cages at different stocking densities

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Abstract – The objective of this work was to evaluate the growth and the stress levels of juvenile dourado (*Salminus brasiliensis*) cultivated in cages. Fish stocked at densities of 15 (D15) and 30 (D30) fish per square meter were evaluated in a completely randomized design with three replicates. Fish were fed twice a day with extruded ration (42% crude protein). Density influenced only biomass and daily food intake, and glucose and lactate concentrations increased over time. D15 and D30 did not influence the growth of dourado. However, the increase of glucose and lactate levels over time indicates that cultivation in cages is a stressful condition for this species.

Index terms: *Salminus brasiliensis*, glucose, hematocrit, hemoglobin, lactate.

Crescimento e estresse de dourados criados em tanques-rede em diferentes densidades de estocagem

Resumo – O objetivo deste trabalho foi avaliar o crescimento e os níveis de estresse de juvenis de dourado (*Salminus brasiliensis*) cultivados em tanques-rede. Peixes estocados nas densidades de 15 (D15) e 30 (D30) peixes por metro quadrado foram avaliados em delineamento inteiramente casualizado, com três repetições. Os peixes foram alimentados duas vezes ao dia com ração extrusada (42% de proteína bruta). A densidade influenciou apenas a biomassa e a ingestão diária, e as concentrações de glicose e lactato aumentaram ao longo do tempo. D15 e D30 não influenciaram o crescimento do dourado. No entanto, o aumento dos níveis de glicose e de lactato ao longo do tempo indica que o cultivo em tanques-rede é uma condição estressante para esta espécie.

Termos para indexação: *Salminus brasiliensis*, glicose, hematócrito, hemoglobina, lactato.

Aquaculture in cages is a well-recognized strategy to meet the growing demand for food worldwide. In this intensive system of cultivation, high densities are essential to increase productivity (Barcellos et al., 2004; Demétrio et al., 2012). Studies indicate that there is an inverse relationship between density and growth of fish when stocked at a very high density, and that maximum growth is generally obtained at an intermediate stocking density that also provides a reasonable survival percentage (Chakraborty et al., 2010). The ideal density is one that does not cause substantial reduction in growth rates or in environmental quality (Van de Nieuwegiessen et al., 2008).

Density can also affect growth rates and may cause changes in the metabolism of fish due to the increase of stress levels (Braun et al., 2010). Stress can become chronic, leading to a decrease in productivity, and can cause fatal outbreaks (Montero et al., 1999). Information concerning the physiological responses associated with stressors may indicate the health status of the fish and can be useful in identifying less stressful conditions, allowing quantitative and qualitative increase in fish production (Barcellos et al., 2004).

Therefore, stocking density is an important stress factor in fish culture. Despite this, stress responses of South American native species (Barcellos et al., 2004;

DOI: 10.1590/S0100-204X2013000800050

Braun et al., 2010) growing in captivity are still not well known. A study related to the density of dourado [*Salminus brasiliensis* (Cuvier, 1816)] in cages showed that survival and growth were similar at densities of 10, 20, and 30 fish per square meter in squared or circular cages (Beux et al., 2008); however, the stress response was not measured.

The objective of this work was to evaluate the growth and the stress levels of juvenile dourado cultivated in cages.

The experiment was conducted at the reservoir of the Itá hydropower plant, located in the upper Uruguay River, state of Santa Catarina, Brazil, for 40 days, from November 17 to December 27, 2007. Juvenile dourado were obtained from induced spawning of a couple of breeders. Fish were distributed in 1 m^3 cages at stocking densities of 15 and 30 fish per square meter (0.81 and 1.63 kg m^{-3}), in a completely randomized design with three replicates. Initial mean weight, standard length, and biomass are presented in Table 1. Fish were fed with extruded ration, containing 42% crude protein, offered twice daily to apparent satiation. At the densities of 15 and 30, fish consumed 1,708.0±43.8 g and 2,316.7±54.2 g, respectively.

The following parameters were measured to analyze growth: weight (g) = (final mean total weight - initial mean total weight); length (cm) = (final mean standard length - initial mean standard length); final biomass (kg) = (final mean weight of fish per treatment x final number of fish per treatment); apparent feed conversion rate = [total weight of feed supplied / (final biomass - initial biomass)]; and apparent daily food intake = (total weight of feed provided in cages / number of fish per cage / number of days per experiment).

Table 1. Weight, standard length, biomass, apparent feed conversion, and apparent daily food intake (mean ± standard deviation) of juvenile dourado (*Salminus brasiliensis*) cultivated in cages in the reservoir of the Itá hydropower plant, state of Santa Catarina, Brazil, at the densities of 15 and 30 fish per square meter, for 40 days[(1)].

Variable	Densities	
	15 fish per m^3	30 fish per m^3
	0 day	
Weight (g)	49.7±2.0a	48.4±2.1a
Standard length (cm)	16.0±0.5a	15.5±0.4b
Biomass (kg m^{-3})	0.75±0.01a	1.45±0.02b
	40 days	
Weight (g)	61.8±7.7a	62.2±8.9a
Standard length (cm)	16.9±0.7a	16.5±12.3a
Biomass (kg m^{-3})	0.93±0.05a	1.87±0.13b
Apparent feed conversion	9.38±4.2a	5.58±1.48a
Apparent daily food intake	5.7±0.1a	3.9±0.1b

[(1)]Means followed by equal letters do not differ by the t test, at 5% probability.

Dourado stress was assessed at 20 and 40 days by analyzing hematocrit, hemoglobin, glucose, and lactate concentrations at the Laboratório de Biologia e Cultivo de Peixes de Água Doce, Universidade Federal de Santa Catarina, SC, Brazil, sampled from three fish per cage. These samples were collected 2 hours before feeding in fish anaesthetized with eugenol (Biodinâmica, Ibiporã, PR, Brazil) at a concentration of 50 mg L^{-1} (Vidal et al., 2006). That study was registered at the Animal Ethics Committee under Protocol PP00128.

Blood was obtained from the tail vein using heparin (Cristália, SP, Brazil) and kept on ice until use. A blood aliquot was used in the hematocrit and hemoglobin concentration analyses, while another was centrifuged (3,000 *g* for 5 min) to obtain plasma, which was stored at 4°C for glucose and lactate analyses. The blood was also centrifuged for 5 min in capillary tubes in a microcentrifuge (Evlab, Londrina, PR, Brazil) to obtain the hematocrit. Hemoglobin concentration was determined by the method of cyanmethemoglobin (Van Kampen & Zijlsta, 1965). Glucose and lactate concentrations were determined with kits from Biotécnica (Varginha, MG, Brazil) and Kovalente (São Gonçalo, RJ, Brazil), respectively.

The fish sampled in the stress analyses were not used in the growth analysis since they were replaced by others with same mean weight and standard length (for density of 15 fish per square meter: 51.0±5.2 g; 15.6±0.4 cm; for density of 30 fish per square meter: 55.1±7.9 g; 15.7±0.5 cm).

Dissolved oxygen concentration, temperature, pH, and electrical conductivity were monitored daily with a Hach multi-parameter (Hach, Loveland, CO, USA), and water transparency was assessed with a Secchi disk. Throughout the study, those variables presented the following means±standard deviation: 7.95±2.43 mg L^{-1}, 25.5±2.5ºC, 7.89±0.47, 43.70±3.13 µS cm^{-1}, and 1.18±0.21 m, respectively.

Growth and physiological parameters were analyzed with the t test (Zar, 2009), at 5% probability, for comparison between the two stocking densities.

Stocking density did not influence weight, standard length, and feed conversion rate of dourado (Table 1). However, the increase in density influenced biomass and the apparent daily food intake. This increase in biomass was similar to that recorded for other species, such as Chitralada strains of *Oreochromis niloticus*, 150, 200, and 250 fish per square meter (Sampaio & Braga, 2005).

Juvenile dourado showed lower performance in cages than fish cultivated in controlled conditions. The feed conversion rates in fish reared in laboratory ranged from 1.5 to 2.0 (Braun et al., 2010), and in cages from 2.5 to 5.0 (Beux et al., 2008), a result that was dependent on the handling intervals associated with the stocking densities. The feed conversion rate obtained in the density of 30 fish per square meter was similar to that recorded by Beux et al. (2008) when cultivating dourado in the same density, with handling intervals of 60 days.

The increase in glucose and lactate concentrations in the densities of 15 and 30 fish per square meter was registered (Figures 1 A and 1 B). These results

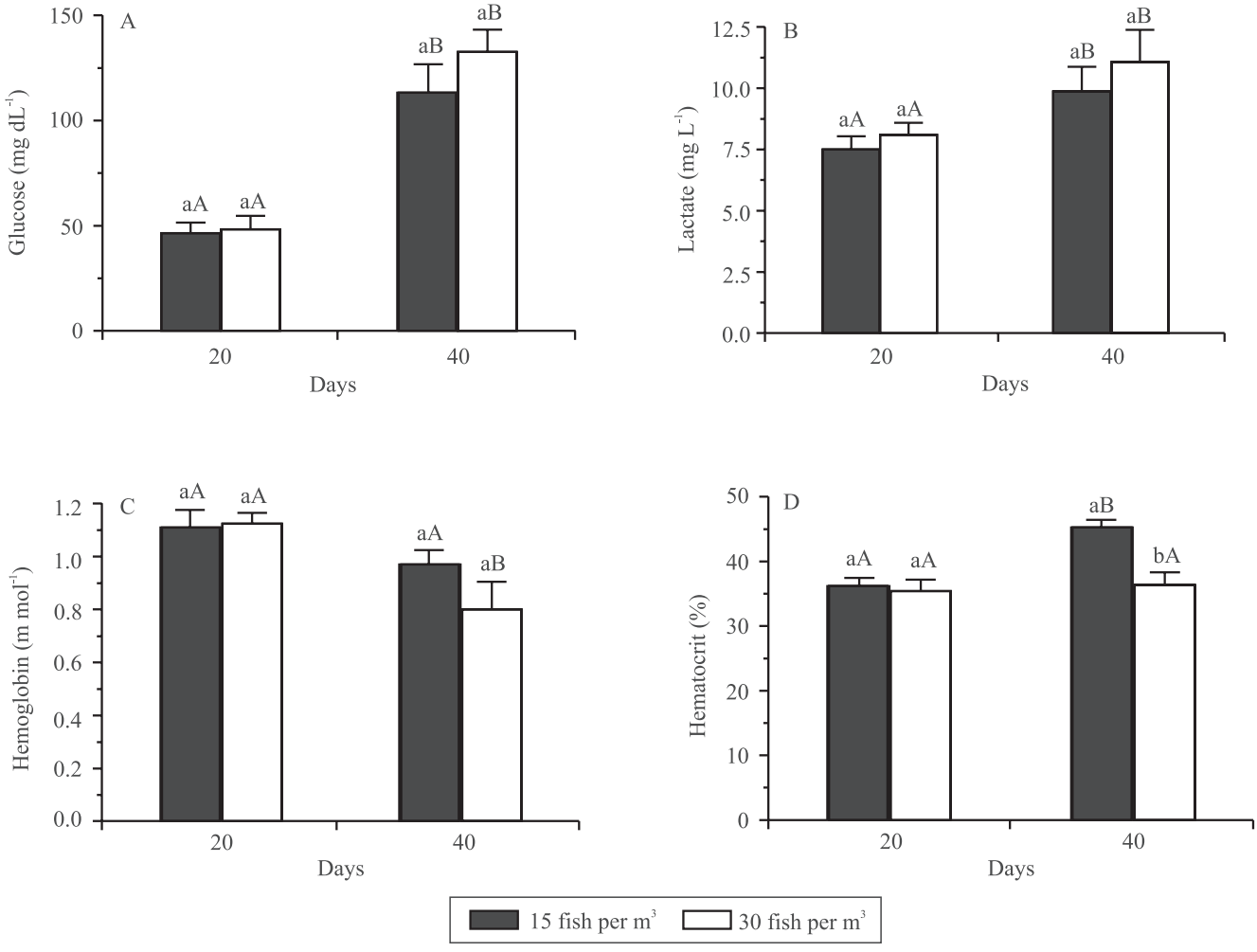

Figure 1. Concentrations (mean±standard deviation) of plasma glucose (A), lactate (B), hemoglobin (C), and hematocrit (D) of juvenile dourado (*Salminus brasiliensis*) cultivated in cages in the reservoir of the Itá hydropower plant, state of Santa Catarina, Brazil, at the densities of 15 and 30 fish per square meter, for 40 days. Density means followed by equal lowercase letters do not differ by the t test, at 5% probability, on day 20 or 40. Means of days 20 and 40 followed by equal uppercase letters do not differ by the t test, at 5% probability, at the densities of 15 or 30 fish per square meter.

DOI: 10.1590/S0100-204X2013000800050

indicate that the experimental condition stressed fish since hyperglycemia contributes to supply the energy demand (Braun et al., 2010), in this case, through the activation of glycogenolysis/glycogenesis, inhibition of glycolysis, and the increase of hepatic glycogenesis, processes that result in glucose production (Montero et al., 1999). The temporal increase of plasma lactate confirms the presence of stress (Figure 1 B). That increase may be attributed to the presence of hypoxia in tissues, which activates the ATP production through anaerobic glycolysis, using glycogen stored in liver and muscles, forming lactate as a final product (Lehninger et al., 2008). This response may be related to constant exposure of fish to stressors caused by the location of the cages in artificial reservoirs (Barcellos et al., 2004; Beux et al., 2008).

Hemoglobin concentration reduced with time in the density of 30 fish per square meter (Figure 1 C), a condition that can be associated with chronic stress (Barcellos et al., 2004). At lower density, however, the increase in the volume of circulating red blood cells did not affect hemoglobin content. Hemoglobin and hematocrit are linked and associated with stress response, and the hematocrit variation is related to the change in the erythrocyte volume. The adrenaline produced during stress causes in vivo swelling in erythrocytes (Nikinmaa & Huestis, 1984), apparently due to the retention of Na^+ and Cl^- in the intracellular environment. Because of the increased intracellular Na^+ and Cl^- concentration, water enters into the erythrocyte and, consequently, increases its volume (Railo et al., 1985). On day 20, when the hematocrit was lower, the hemoglobin concentrations were higher than on day 40, when the hematocrit concentration increased (Figures 1 C and 1 D).

Under the tested conditions, the stocking densities of 15 and 30 fish per square meter do not influence the growth of dourado in cages, but the increase of glucose and lactate levels over time, regardless of the stocking density, indicates that the cultivation in cages is a stressful condition for this species.

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Received on August 30, 2011 and accepted on May 24, 2013