

Influence of stocking density on water quality and growth performance in production of juvenile pirarucu, *Arapaima gigas*, in irrigation canals**Influência da densidade de estocagem na qualidade da água e na performance de crescimento na produção de juvenis de pirarucu, *Arapaima gigas*, em canais de irrigação**

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ABSTRACT

The pirarucu is reared in cages under intensive systems or ponds under semi-intensive systems. This research evaluates the effect of stocking density on water quality, survival and growth of juvenile pirarucu reared in net pens located in irrigation canals. Pirarucu (193.3 ± 37.3 g; 31.5 ± 2.1 cm) were stocked at densities of 3.6, 6 or 10 fish m^{-3} in 4.2 m^3 net pens installed at the Irrigation District of the Coastal Tablelands (DITALPI), Parnaíba (Piauí, Brazil), with three replicate for each density. Fish were fed a formulated diet containing 40% crude protein and 14.2 MJ kg^{-1} of feed and cultured for 180 days. The physicochemical parameters of the water were at satisfactory levels for fish culture. Survival was 100.0% in all density tested. The final weight, weight gain and absolute growth rate (AGR) were higher for the medium stocking density (6 fish m^{-3}) than for fish at low stocking density (3.6 fish m^{-3}) and high stocking density (10 fish m^{-3}). Final weight gain, survival, and productivity have demonstrated that the production of pirarucu in net pens using water from irrigation canals can become a viable strategy and a cheaper alternative to increase fish production. In addition, it improves the efficient use of water by integrating two productive activities, agriculture and aquaculture.

Keywords: Pirarucu, net pens, stocking density, irrigation canals.

RESUMO

O pirarucu é cultivado em sistemas intensivos, em tanques-rede, ou semi-intensivos, em viveiros. Essa pesquisa avaliou o efeito da densidade de estocagem na qualidade da água, sobrevivência e crescimento de juvenis de pirarucu cultivados em hapas instaladas em canais de irrigação. Pirarucu ($193,3 \pm 37,3$ g; $31,5 \pm 2,1$ cm) foram estocados em densidades de 3,6, 6 ou 10 peixes m^{-3} em hapas de 4,2 m^3 instaladas no Distrito de Irrigação Tabuleiros Litorâneos (DITALPI), Parnaíba (Piauí, Brasil), com três repetições para cada densidade. Os peixes foram alimentados com uma dieta formulada com 40% de proteína bruta e 14,2 MJ kg^{-1} de ração e cultivados por 180 dias. Os parâmetros físico-químicos da água apresentaram níveis satisfatórios para o cultivo de peixes. A sobrevivência foi 100,0% para todas as densidades testadas. O peso final, o ganho de peso e a taxa de crescimento absoluto (TCA) foram maiores para a densidade de estocagem média (6 peixes m^{-3}) que para os peixes em baixa densidade de estocagem (3,6 peixes m^{-3}) e alta densidade de estocagem (10 peixes m^{-3}). O ganho de peso final, a sobrevivência e a produtividade têm demonstrado que a produção de pirarucu em hapas usando a água dos canais de irrigação pode ser uma estratégia viável e uma alternativa viável para incrementar a produção de peixes. Em adição, o uso eficiente da água é incrementado pela interação das duas atividades produtivas, a agricultura e a aquicultura.

Palavras-chave: Pirarucu, hapas, densidade de estocagem, canais de irrigação.

1 INTRODUCTION

In Brazil, there are more than 3,000 fish species and a considerable part of these can be used for aquaculture, such as the pacu, *Piaractus mesopotamicus* (ABIMORAD *et al.*, 2009), pintado, *Pseudoplatystoma corruscans* (SANTOS and LUZ, 2009), dourado, *Salminus brasiliensis* (BRAUN *et al.*, 2010), pirarucu, *Arapaima gigas* (Oliveira *et al.*, 2012), jundiá, *Rhamdia quelen* (Silva *et al.*, 2012), and tambaqui, *Colossoma macropomum* (FIÚZA *et al.*, 2015). Despite this diversity, native species accounted for less than 20% of national aquaculture production in 2012 (BRAZIL, 2014).

Accordingly, the potential of aquaculture for native fish species, such as the pirarucu has been studied (GANDRA *et al.*, 2007; NÚÑEZ *et al.*, 2011; MATTOS *et al.*, 2016; MALHEIROS *et al.*, 2016; ANDRADE-PORTO *et al.*, 2017). This species is an example of a native species with high potential for intensive production (CAVERO *et al.*, 2003a; PEREIRA-FILHO *et al.*, 2003; OLIVEIRA *et al.*, 2012; OLIVEIRA *et al.*, 2013; RODRIGUES *et al.*, 2019). Pirarucu, a member of the Arapaimidae family (Osteoglossiformes), is endemic from the Amazonian basin and one of the largest fresh water fishes in South America (ROUBACH *et al.*, 2003). This carnivorous fish grows very fast in various aquaculture systems, reaching up to 27-41 g day⁻¹, and 10-15 kg year⁻¹ (PEREIRA-FILHO *et al.*, 2003; NÚÑEZ, 2009).

In Brazil, the pirarucu is raised in cages under intensive systems or in ponds under semi-intensive systems (REBAZA *et al.*, 2010). However, little is known about the rearing of pirarucu in other aquaculture environments, such as integrated agriculture, aquaculture systems, aquaponics and recirculating aquaculture systems (RAS).

The increasing pressure on world water resources requires imperative actions towards integration of water uses. This is especially true in the case of agriculture and aquaculture, which are in many ways, complementary (PHILLIPS *et al.*, 1991). The integration of aquaculture in irrigation systems or integrated irrigated aquaculture (IIA) has been defined as an activity capable of producing fish in the water on its way to or from agriculture (FAO, 2006). The inclusion of aquaculture in irrigation systems is a key strategy for a most efficient use of water. Aquaculture can be established in floating cages in the reservoirs, in irrigation canals or in ponds, using agricultural land. The use of net pens may be an alternative to the cage culture, where the canal is blocked off at intervals by barriers to form a series of pens (REDDING and MIDLEN, 1990). The culture of fish in irrigation canals requires cooperation between the water authorities that manage the systems, and fish farmers (EDWARDS, 2015). Moreover, control of water level and flow, use of pesticides and other

agricultural chemicals can represent a serious problem for the production of fish (REDDING and MIDLEN, 1990).

The Northeast of Brazil has 52 irrigated perimeters, with approximately 190,000 hectares of irrigable areas and a network of open canals ranging over 2,900 km (OLIVEIRA and SANTOS, 2015). Furthermore, the rational use of irrigation canals can be a viable alternative to increase aquaculture production in arid areas such as the Northeast of Brazil. Therefore, the aim of this study was to determine the effect of stocking density on water quality, survival and growth of juvenile pirarucu, *A. gigas*, and contribute to aquaculture production in irrigation canals.

2 MATERIALS AND METHODS

Pirarucu juveniles from a single spawn were obtained from Aquaculture Research Center of National Department of Works Against Droughts (DNOCS), Pentecoste, Ceará, Brazil, and transferred to Irrigation District of the Coastal Tablelands (DITALPI), Parnaíba, Piauí State, Brazil. DITALPI is an irrigated perimeter with an irrigable area of 8,000 ha and a network of open canals ranging 14,800 meters. This irrigated perimeter consists of the main canals and main cross structure devices, secondary canals and secondary cross structure devices, and of a tertiary system and corresponding tertiary canals. All canals have a trapezoidal shape.

This study was conducted in a secondary canal. For each treatment, part of the secondary canal was divided into compartments to form a series of three net pens. The cross-section area of the secondary canal was 2.1 m² (width of water surface of the secondary canal = 4.2 m, bottom width of the secondary canal = 0.5 m and average depth of water flow in secondary canal = 0.9 m), with a length of 2 meters, horizontal sectional surface of 8.4 m² and a volume of 4.2 m³ (Figure 1). The distance between each series of three net pens was 10 m. The flow rate in the secondary canal was 0.6 m³ s⁻¹ during the operation of the irrigation electropumps (21:00 h – 09:00 h) and in the 0.2 m³ s⁻¹ complementary period. The net was brushed every two weeks to facilitate water exchange between the compartments. The net pens were enclosed with 20-mm galvanized wire mesh coated with UV resistant PVC and fixed in an inverted concrete arch. The net pens were covered with a polyethylene mesh to prevent the escape of the fish and the entry of predators.

Prior to stocking, 280 fish were transferred to a 2000 L indoor tank for 60 days and were fed until satiety with a commercial extruded diet. Subsequently, total body weight and length were measured and a total of 246 fish (193.3±37.3 g; 31.5±2.1 cm; mean±SD) were randomly selected, counted and assigned to 9 net pens, at three densities of 3.6 (low stocking density, LSD, 0.8 kg m⁻³), 6 (medium stocking density, MSD, 1.2 kg m⁻³) or 10 (high stocking density, HSD, 1.9 kg m⁻³) fish

m⁻³ with three replicate net pens for each density. The net pens were disposed in the same direction of the water flow from the highest density (HSD) to the lowest (LSD).

Pirarucu juveniles were hand-fed four times a day a commercial pelleted feed with 40% crude protein and 14.2 MJ kg⁻¹ of feed (TC 40; Purina®, São Paulo, Brazil) over a period of 180 days. Fish were fed at rates of 2.5% body weight/day for the first 90 days and 2.0% body weight/day from the 91th day until termination. Fish were sampled every 30 days to evaluate development in weight and length. For this, all fish in each net pen were captured, anesthetized with benzocaine (100 mg L⁻¹), weighed and measured. After each sampling period the amount of feed given was adjusted to the mean weight and biomass in each net pen.

At the end of the trial, fish were harvested and went under calculating of final weight and length, condition factor ($K = \text{body weight/body length}^3 \times 100, \%$), specific growth rate ($\text{SGR} = [(\ln \text{ final weight} - \ln \text{ initial weight})/\text{days}] \times 100, \% \text{ day}^{-1}$), absolute growth rate (AGR, g fish⁻¹ day⁻¹), fish survival (%), yield (kg m⁻³) and feed conversion ratio (FCR, weight of feed/gain in wet weight of fish) for each net pen and treatment.

Temperature, dissolved oxygen (DO) and pH were monitored twice daily at 08:00 h and 17:00 h with an oximeter (YSI model 55) and digital pH meter, respectively. Water samples were collected fortnightly at 08:00 h and 17:00 h from inside of all the net pens. Additionally, water samples were also obtained before the first series of net pens (HSD), between the first (HSD) and second series (MSD), between the second series (MSD) and the third series (LSD), and after the third series of net pens (LSD). Water quality parameters were analyzed in the water samples using the methodologies described by APHA (1995). The parameters included: dissolved oxygen (DO), pH, ammonia, nitrate, nitrite, orthophosphate, total alkalinity and turbidity.

All data were expressed as mean±SD. One-way analysis of variance (ANOVA) was used to determine the effects of stocking density on the water quality and growth performance and Tukey test was used for post hoc comparisons.

3 RESULTS

At the end of the 180 day period, the LSD, MSD, and HSD groups reached a total length and body weight ranging from 83.4±2.5 to 86.6±2.8 cm and 4939.5±40.7 to 5421.4±101.4 g, respectively (Figure 2). Growth performance values are shown in Table 1. Survival rate was 100.0% and not significant differences between the treatments ($P>0.05$). In contrast, significant differences ($P<0.05$) were detected in the parameters with the increasing of stocking density. Thus, the final weight and weight gain of pirarucu at MSD were higher than that in the fish at LSD and HSD. In the same way, stocking density significantly affected the productivity and feed conversion ratio ($P<0.05$). There

were no significant differences in condition factor between treatments during the study ($P>0.05$). The condition factors for the LSD, MSD and HSD treatments to begin the study were 0.6 ± 0.1 , 0.6 ± 0.1 and 0.8 ± 0.1 , respectively. The condition factors at the end of the study were 0.8 ± 0.1 for the LSD and MSD treatments and 0.9 ± 0.1 for the HSD treatment (Figure 2).

The average values of water quality parameters in pirarucu rearing under varying density levels are presented in Table 2. The evolution over time for dissolved oxygen, ammonia, nitrite and nitrate are shown in Figures 3, 4, 5 and 6. Not significant differences between the collection points ($P<0.05$).

4 DISCUSSION

In this century, a critical challenge is the resolution of the water crisis, with less water available for agriculture, including aquaculture (MOLDEN, 2007). More efficient use of water could be attained by integration of aquaculture with natural and constructed water bodies (EDWARDS, 2015). This study represents the first attempt to evaluate growth performance of pirarucu using net pens in irrigation canals.

Stocking density is one of the main factors affecting growth and water quality parameters in aquaculture. Survival was similar to that observed for pirarucu reared in ponds (PEREIRA-FILHO *et al.*, 2003), cages (CAVERO *et al.*, 2003b; OLIVEIRA *et al.*, 2012; OLIVEIRA *et al.*, 2013) and RAS (PEDROSA *et al.*, 2019).

High stocking densities can act as stressors, resulting in reduced growth and increased feed conversion rate (RAHMAN *et al.*, 2005; SUZIKI *et al.*, 2001). Therefore, it is very important to adjust the stocking density with the carrying capacity of the aquaculture system (MAGOUZ *et al.*, 2019). In the present study, the stocking density did affect growth parameters. Final weight, weight gain and absolute growth rate (AGR) at 6.0 fish m^{-3} (MSD) were higher than at both 3.6 (LSD) and 10.0 fish m^{-3} (HSD). Those results agree with Oliveira *et al.* (2012), who observed significant effect of the stocking density on final weight, weight gain and absolute growth rate (AGR) for pirarucu juveniles cultured for 140 days in cages, although these authors did not observe effects on production and feed conversion rate (FCR). Cavero *et al.* (2003b) observed that these parameters were not affected during a culture of pirarucu in cages at densities of 15, 20 and 25 fish m^{-3} . On the other hand, results of the present study showed that the increasing stocking density led to a decreased feed conversion rate (FCR). Additionally, increased stocking density significantly affected the production (kg m^{-3}). The highest stocking density in treatment HSD (10 fish m^{-3}) gave the better production compared with treatments LSD (3.6 fish m^{-3}) and MSD (6.0 fish m^{-3}).

The culture of fish in cages in irrigation canals requires coordination between the water authorities that manage the system and fish farmers. There was a recent mass mortality of caged fish due to a water shortage in an irrigation canal in Thailand in 2012 (EDWARDS, 2015). Cage aquaculture in irrigation canals is prohibited in some countries because of concerns that it may reduce water flow and increase sedimentation in the canal (DUGAN *et al.*, 2007). In this work, the parameters of water quality remained within ideal range to an adequate water quality (BEVERIDGE, 1996; POPMA and LOVSHIN, 1996), with the exception of dissolved oxygen levels during the 2th month of cultivation, which did not interfere with the growth of pirarucu. It is possible that early morning dissolved oxygen levels in this experiment were within the range that can delay the growth in teleosts (BOYD and TUCKER, 1998). However, an advantage of pirarucu in relation to other species of teleosts is represented by its adaptation to oxygen poor environments, breathing atmospheric air, a habit that they demonstrate from the early stages of the life cycle (BALDISSEROTTO and GOMES, 2013). The 100% survival rate for all treatments confirms the fact that the pirarucu is very resistant to variations in water quality.

An important parameter in the quality of pirarucu juvenile water is the level of nitrogen compounds. Even though, according to Cavero *et al.* (2004), the pirarucu is a species that tolerates high levels of ammonia and nitrite concentration in water, due to its ability for aerial respiration. This does not reduce the importance of maintaining an adequate quality of water during rearing in intensive systems, mainly because in environments with high concentrations of nitrogen compounds, the physiology of fish is compromised, and consequently its growth. Throughout the experiment period, the average ammonia, nitrite and nitrate levels were kept within ideal levels, with the exception of some morning readings to ammonia levels in the final months of cultivation. However, it did not affect the growth of the fish, as the final average weight gain of pirarucu was around 5 kg. This was similar to that observed in the literature, where the pirarucu has an annual weight gain of 10 kg (IMBIRIBA, 2001). On the other hand, the fact of fish tolerate high concentrations of ammonia does not mean that it is favorable to its development. Apparently, this fact was described by Pereira-Filho and collaborators (2003), which observed the increase of ammonia and nitrite levels proportional to the increase of biomass of pirarucu juveniles in ponds, but the final average weight of the fish was lower than that obtained by other authors for the same culture period.

5 CONCLUSIONS

The present results that the stocking density did not affect the parameters of water quality analyzed, as well as survival. However, growth and all other parameters related to the zootechnical performance of pirarucu juvenile culture in net pens installed in irrigation canals were affected by the

variation in stocking density, making this an important factor to be accurately determined, in order to better assist the growing potential producers of this species of fish.

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Figure 1 Net pens (4.2 m³) in irrigation District of the Coastal Tablelands (DITALPI) (Parnaíba, Piauí State, Brazil), for cultivation of pirarucu for 180 days at 3.6, 6 and 10 fish/m³ (LDS, MSD and HSD) in triplicate.

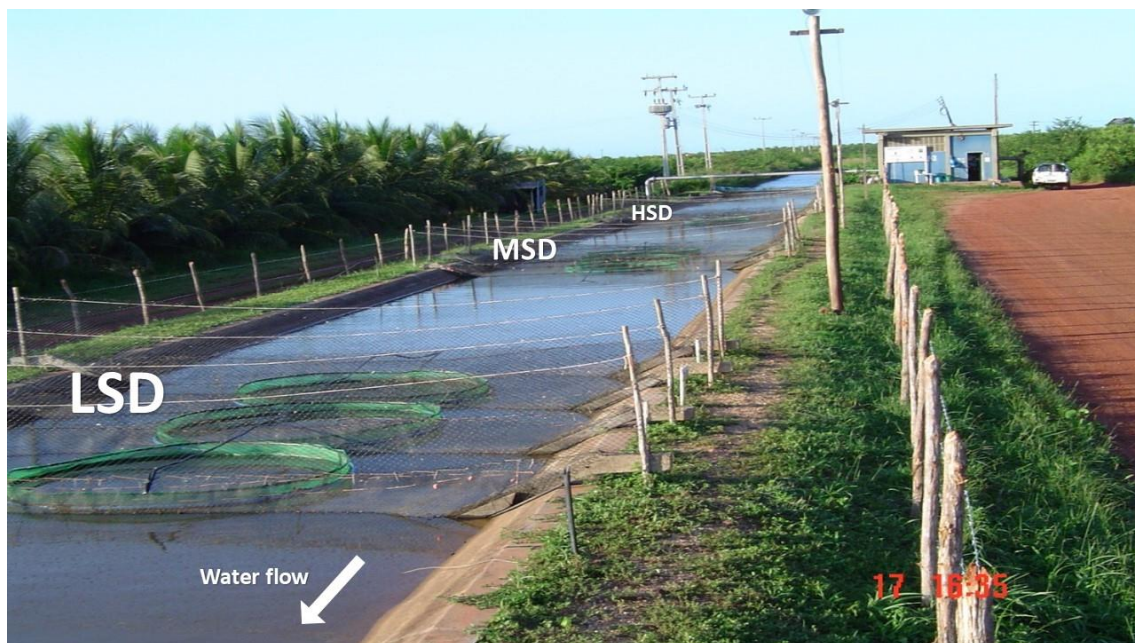


Figure 2 Evolution of total length, body weight, and condition factor of pirarucu juveniles cultured for 180 days at three stocking densities in net pens. HSD = high density stocking (10.0 fish m⁻³), MSD = medium density stocking (6.0 fish m⁻³), LSD = low density stocking (3.6 fish m⁻³).

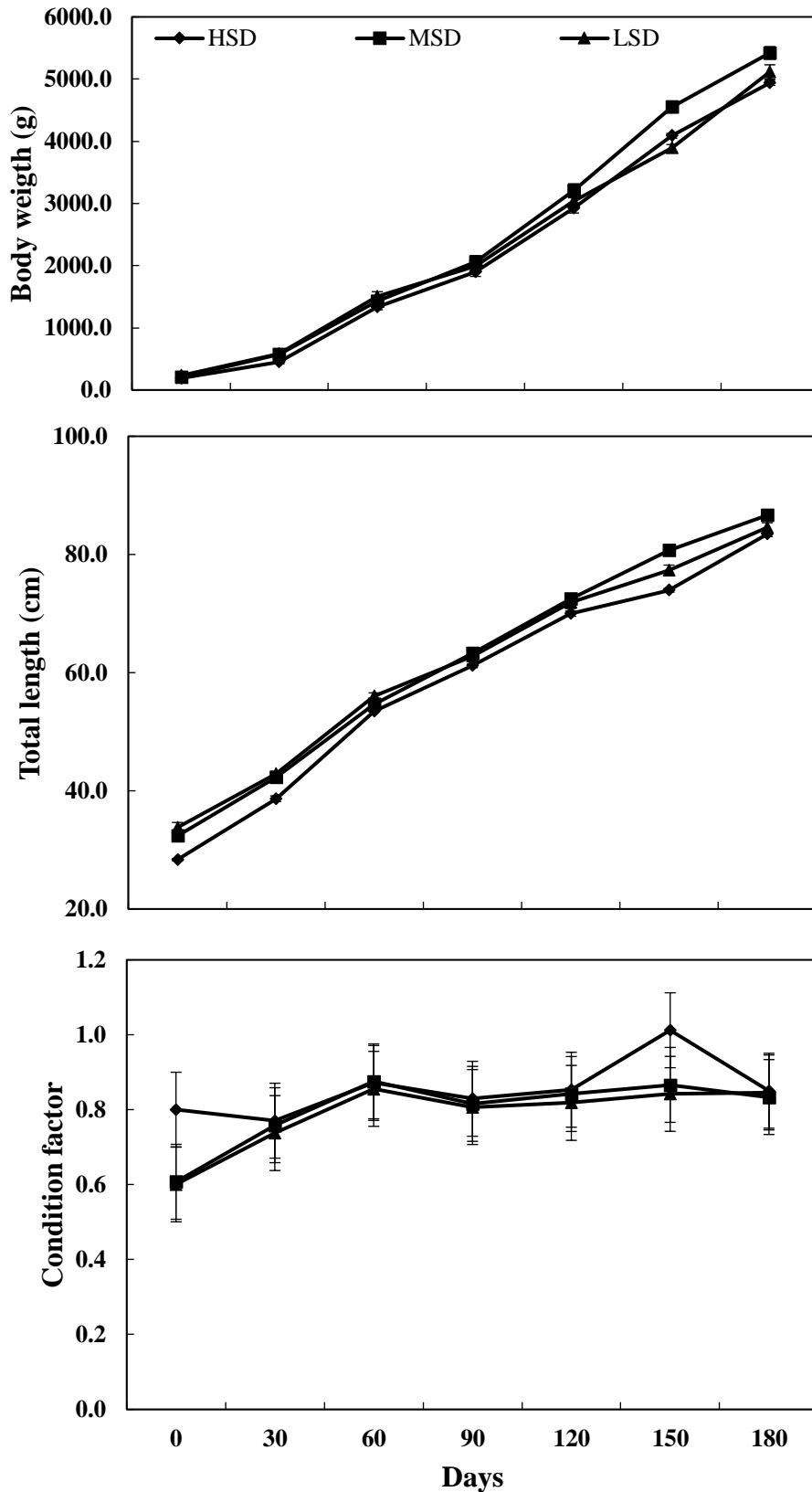


Table 1 Survival, initial and final weights, weight gain, specific growth rate (SGR), absolute growth rate (AGR), yield and feed conversion ratio (FCR) of pirarucu juveniles (initial mean weights, 189.9–231.9 g) cultured for 180 days at three stocking densities in net pens.

Parameter	Stocking densities (fish m ⁻³)		
	3.6	6.0	10.0
Survival (%)	100.0	100.0	100.0
Initial weight (g)	231.9±11.0	205.9±18.0	189.9±24.1
Final weight (g)	5116.8±112.6 ^a	5421.4±101.4 ^b	4939.5±40.7 ^a
Weight gain (g)	4884.9±102.8 ^a	5215.5±114.1 ^b	4749.6±42.9 ^a
SGR (% day ⁻¹)	1.72±0.05 ^a	1.82±0.05 ^b	1.81±0.01 ^b
AGR (g day ⁻¹)	27.1±0.6 ^a	29.0±0.7 ^b	26.4±0.6 ^a
Yield (kg m ⁻³)	17.6±1.7 ^a	31.3±1.3 ^b	47.5±0.4 ^c
FCR	2.1±0.1 ^a	1.8±0.1 ^b	1.4±0.1 ^b

Data are means ± S.D. of three replicate cages. For each row, means with different letters as superscripts are significantly different ($P < 0.05$).

Table 2 Physicochemical parameters of net pens in irrigation canals during cultivation of pirarucu juveniles. Average values \pm S.D. collected in the morning (8 h) and afternoon (17 h) every 30 days up to 180 of total cultivation, before the first series of net pens (HSD), between the first (HSD) and second series (MSD), between the second series (MSD) and the third series (LSD), and after the third series of hapas (LSD).

Average values (08:00 h)							
Parameters	Before	Between	After	LSD	MSD	HSD	Ideal range ¹
Temperature (°C)	29.1 \pm 0.4	29.3 \pm 0.4	29.4 \pm 0.9	29.3 \pm 0.4	29.0 \pm 0.5	29.0 \pm 1.0	25 - 32° C
Oxygen (mg L ⁻¹)	5.4 \pm 1.2	5.3 \pm 1.1	5.0 \pm 1.0	5.4 \pm 1.0	5.3 \pm 0.9	5.2 \pm 1.1	> 5.0 mg L ⁻¹
pH	7.5 \pm 0.3	7.5 \pm 0.2	7.3 \pm 0.4	7.5 \pm 0.2	7.4 \pm 0.2	7.3 \pm 0.4	6.0 - 9.0
Turbidity (cm)	29.5 \pm 13.6	29.4 \pm 12.6	28.3 \pm 11.8	30.0 \pm 13.4	28.7 \pm 11.9	28.0 \pm 9.9	20 - 60 cm
Ammonia (mg L ⁻¹)	0.040 \pm 0.036	0.092 \pm 0.083	0.164 \pm 0.131	0.088 \pm 0.065	0.117 \pm 0.112	0.142 \pm 0.131	< 0.1 mg L ⁻¹
Nitrite (mg L ⁻¹)	0.013 \pm 0.005	0.014 \pm 0.005	0.014 \pm 0.004	0.013 \pm 0.005	0.014 \pm 0.004	0.015 \pm 0.004	< 0.3 mg L ⁻¹
Nitrate (mg L ⁻¹)	0.073 \pm 0.045	0.072 \pm 0.051	0.092 \pm 0.047	0.072 \pm 0.036	0.082 \pm 0.043	0.083 \pm 0.036	0.2 - 10 mg L ⁻¹
Orthophosphate (mg L ⁻¹)	0.032 \pm 0.019	0.037 \pm 0.019	0.038 \pm 0.013	0.034 \pm 0.018	0.040 \pm 0.016	0.042 \pm 0.018	0.005 - 2.0 mg L ⁻¹
Alcalinity (mg L ⁻¹)	20.5 \pm 5.9	19.6 \pm 3.9	19.9 \pm 5.1	19.7 \pm 4.7	19.6 \pm 4.7	20.5 \pm 5.8	50 - 300 mg L ⁻¹
Average values (17:00 h)							
Parameters	Before	Between	After	LSD	MSD	HSD	Ideal range ¹
Temperature (°C)	30.4 \pm 0.5	30.4 \pm 0.4	30.6 \pm 0.5	30.4 \pm 0.4	30.4 \pm 0.4	30.4 \pm 0.4	25 - 32° C
Oxygen (mg L ⁻¹)	6.9 \pm 0.9	6.8 \pm 0.7	6.9 \pm 0.7	6.8 \pm 0.7	6.8 \pm 0.7	7.0 \pm 0.6	> 5.0 mg L ⁻¹
pH	7.7 \pm 0.3	7.7 \pm 0.3	7.4 \pm 0.5	7.7 \pm 0.3	7.7 \pm 0.3	7.7 \pm 0.4	6.0 - 9.0
Turbidity (cm)	33.8 \pm 13.9	35.9 \pm 13.8	34.8 \pm 13.5	34.9 \pm 14.2	35.0 \pm 14.4	35.5 \pm 13.0	20 - 60 cm
Ammonia (mg L ⁻¹)	0.015 \pm 0.007	0.019 \pm 0.010	0.029 \pm 0.013	0.014 \pm 0.006	0.021 \pm 0.010	0.021 \pm 0.009	< 0.1 mg L ⁻¹
Nitrite (mg L ⁻¹)	0.011 \pm 0.007	0.012 \pm 0.007	0.013 \pm 0.008	0.012 \pm 0.007	0.012 \pm 0.007	0.013 \pm 0.008	< 0.3 mg L ⁻¹
Nitrate (mg L ⁻¹)	0.072 \pm 0.034	0.070 \pm 0.044	0.071 \pm 0.047	0.071 \pm 0.039	0.067 \pm 0.033	0.069 \pm 0.037	0.2 - 10 mg L ⁻¹
Orthophosphate (mg L ⁻¹)	0.033 \pm 0.015	0.032 \pm 0.018	0.034 \pm 0.023	0.029 \pm 0.018	0.031 \pm 0.020	0.034 \pm 0.022	0.005 - 2.0 mg L ⁻¹
Alcalinity (mg L ⁻¹)	21.6 \pm 5.2	21.7 \pm 4.0	20.2 \pm 3.4	20.4 \pm 3.5	20.2 \pm 3.1	20.3 \pm 3.4	50 - 300 mg L ⁻¹

¹Boyd and Tucker (1998); Popma and Lovshin (1996).

Figure 3 Average values month of the oxygen dissolved in the rearing net pens. Collected in the morning (8 h) and afternoon (17 h) every days up to 180 of total cultivation, “before” = before the first series of net pens (HSD), “between” = in the first (HSD) and second series (MSD) + between the second series (MSD) and the third series (LSD), and “after” the third series of hapas (LSD).

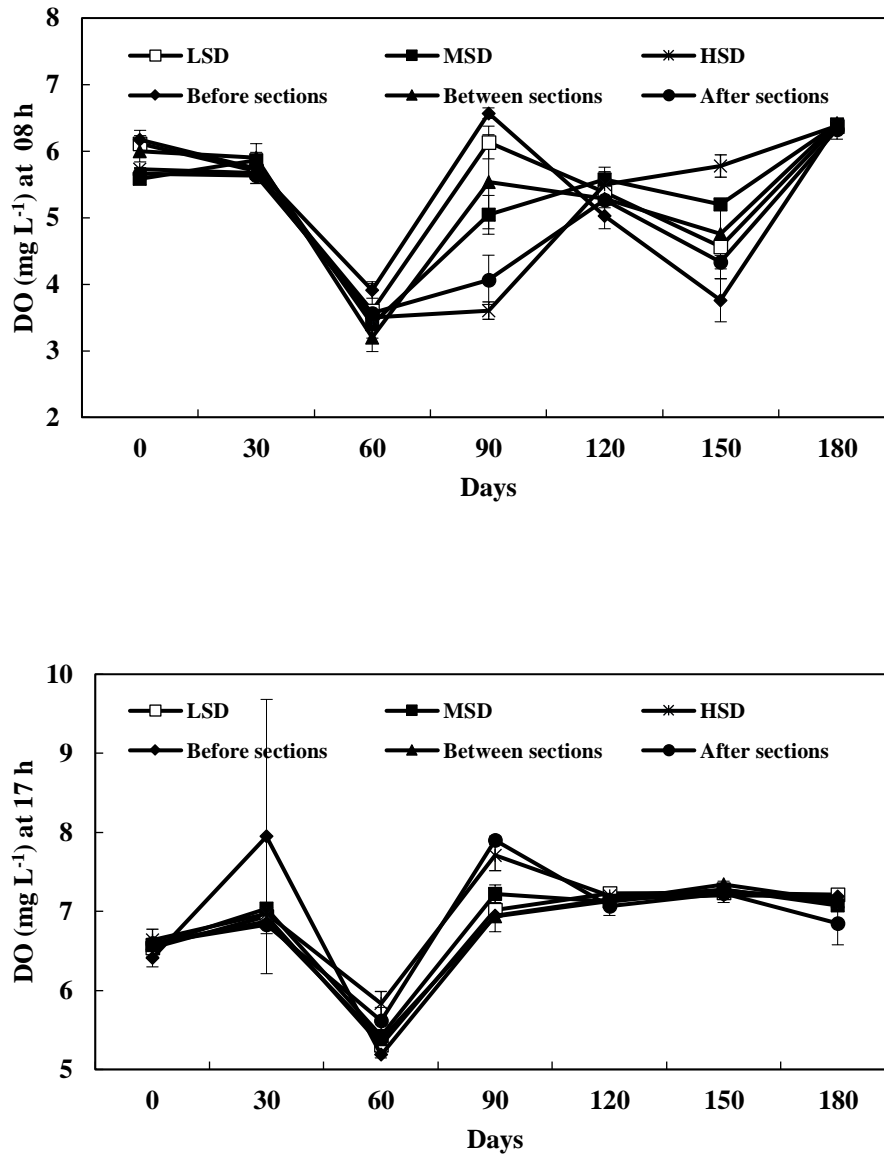


Figure 4 Average values month of the ammonia in the rearing net pens. Collected in the morning (8 h) and afternoon (17 h) every days up to 180 of total cultivation, “before” = before the first series of net pens (HSD), “between” = in the first (HSD) and second series (MSD) + between the second series (MSD) and the third series (LSD), and “after” the third series of hapas (LSD).

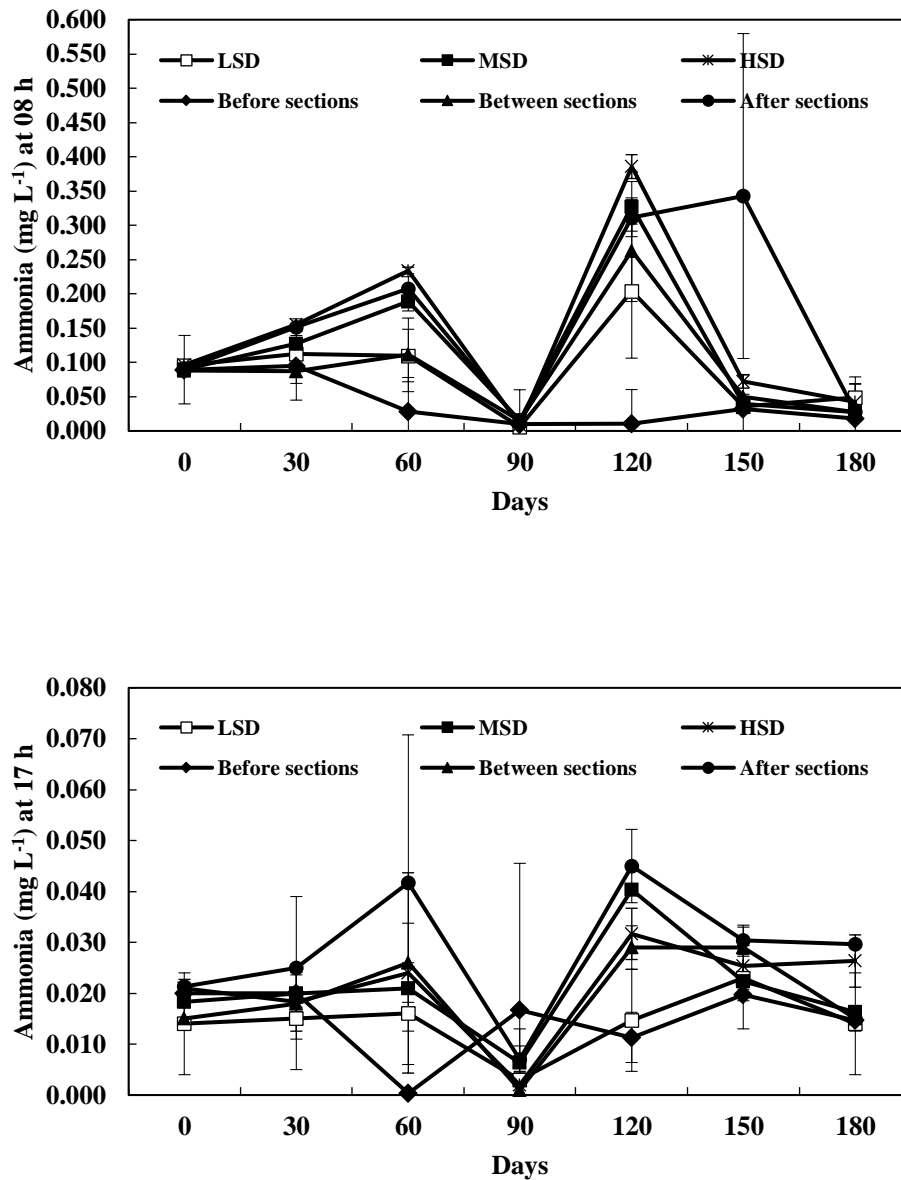


Figure 5 Average values month of the nitrite in the rearing net pens. Collected in the morning (8 h) and afternoon (17 h) every days up to 180 of total cultivation, “before” = before the first series of net pens (HSD), “between” = in the first (HSD) and second series (MSD) + between the second series (MSD) and the third series (LSD), and “after” the third series of hapas (LSD).

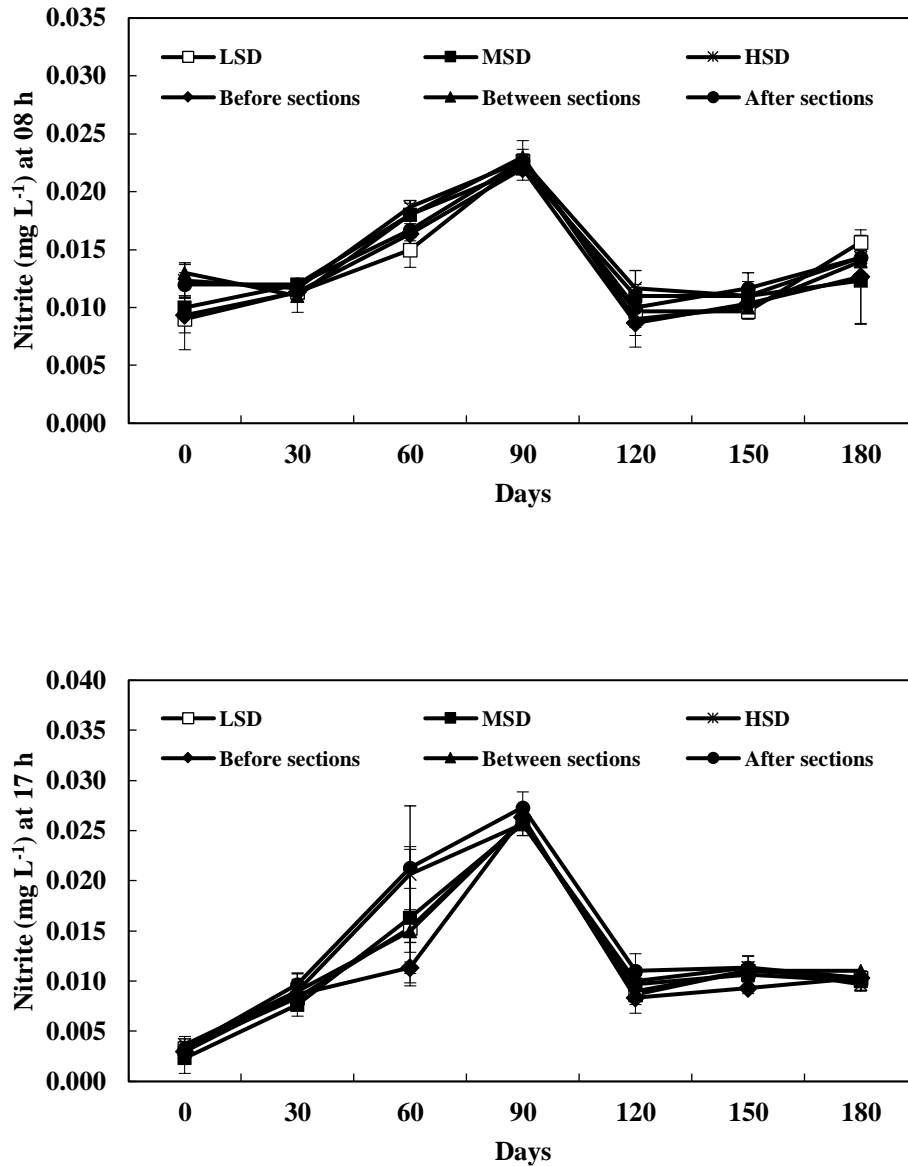
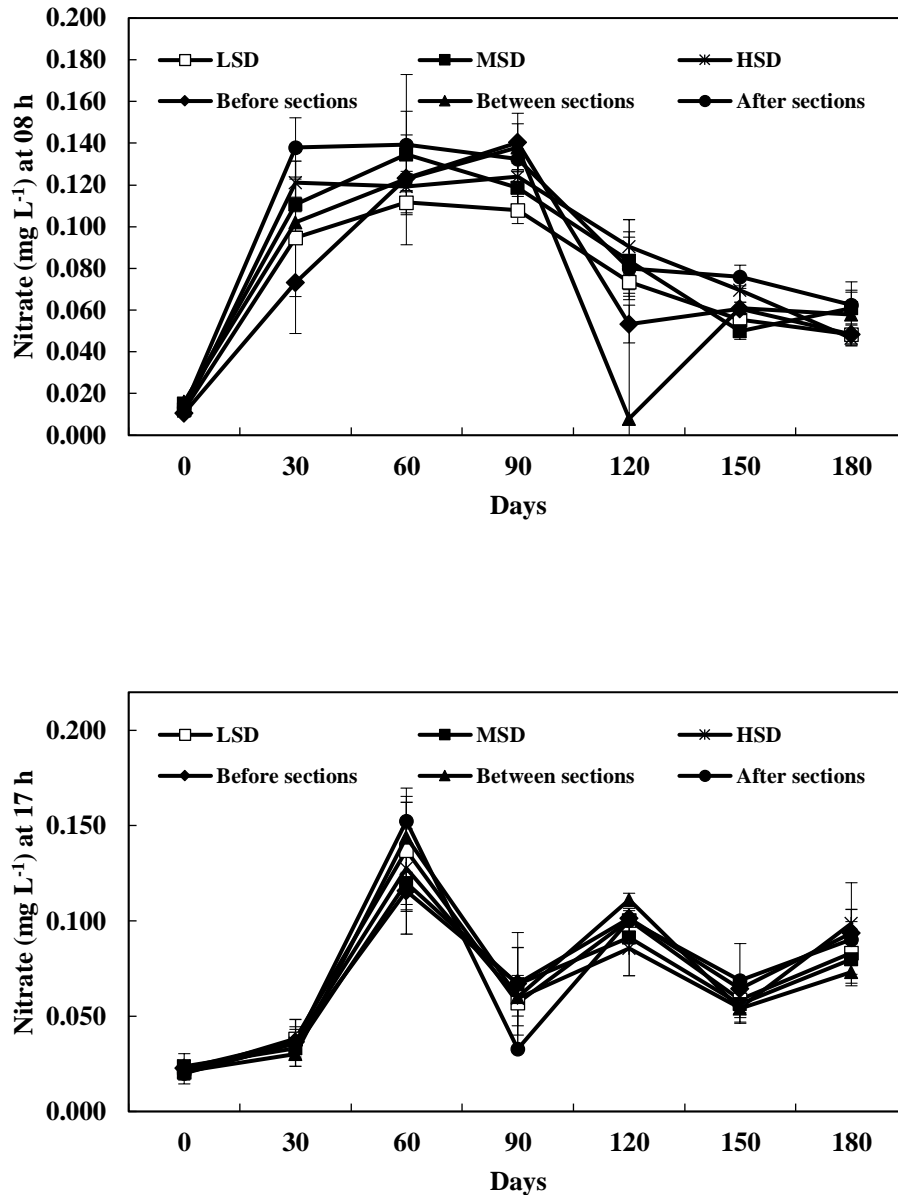


Figure 6 Average values month of the nitrate in the rearing net pens. Collected in the morning (8 h) and afternoon (17 h) every days up to 180 of total cultivation, “before” = before the first series of net pens (HSD), “between” = in the first (HSD) and second series (MSD) + between the second series (MSD) and the third series (LSD), and “after” the third series of hapas (LSD).



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