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Original Article

The growth performance and parasite load of angelfish juveniles *Pterophyllum scalare* kept at different stocking densities in two rearing systems

Desempenho de crescimento e carga parasitária de juvenis acará bandeira *Pterophyllum scalare* mantidos em diferentes densidades de estocagem em dois sistemas de criação

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Abstract

This study evaluated the growth performance and parasite load of angelfish juveniles *Pterophyllum scalare* kept at different stocking densities using two rearing systems. The experiment was conducted in a factorial design (4x2) with four stocking densities (0.1, 0.4, 0.7, and 1.0 g/L), two type of aquarium tanks (glass and ceramic aquariums), and four replicates. The experiment lasted 60 days using 148 juvenile fish (3.05 ± 0.09 g) randomly placed in 32 aquariums (50 L) equipped with filters and aeration. All fish received two meals a day *ad libitum* (8:00 and 16:00). Water quality parameters such as temperature, dissolved oxygen, pH, and total ammonia were measured. At the end of the experiment, all fish were measured and weighed to determine growth performance and then subjected to parasitological analysis. The data were analyzed with a two-way ANOVA with post-hoc Tukey test (p<0.05). No effects on growth performance at different stocking densities were observed. However, there was an increase in *Capillaria pterophylli* infestation in the high stocking density within ceramic aquariums. Thus, this study recommends the use of 1.0 g/L for the intensive aquaculture system of freshwater angelfish, and applying cleaning management to avoid parasite infestation, particularly in ceramic aquariums.

Keywords: Amazonian fish, *Capillaria pterophylli*, freshwater angelfish, ornamental aquaculture.

Resumo

Este estudo avaliou o desempenho de crescimento e a carga parasitária de juvenis de peixe anjo *Pterophyllum scalare* mantidos em diferentes densidades de estocagem usando dois sistemas de criação. O experimento foi conduzido em esquema fatorial (4 x 2) com quatro densidades de estocagem (0,1, 0,4, 0,7 e 1,0 g/L), dois aquários (aquários de vidro e cerâmica) e quatro repetições. O experimento durou 60 dias utilizando 148 peixes juvenis (3,05 ± 0,09 g) distribuídos aleatoriamente em 32 aquários (50 L) equipados com filtros e aeração. Todos os peixes receberam duas refeições diárias ad libitum (8h e 16h). Parâmetros de qualidade da água como temperatura, oxigênio dissolvido, pH e amônia total foram medidos. Ao final do experimento, todos os peixes foram medidos e pesados para determinação do desempenho de crescimento e posteriormente submetidos à análise parasitológica. Os dados foram analisados por meio de ANOVA a dois fatores com teste post-hoc de Tukey (p<0,05). Não foram observados efeitos no desempenho do crescimento em diferentes densidades de estocagem. Entretanto, houve um aumento na infestação por *Capillaria pterophylli* na alta densidade de estocagem dentro dos aquários cerâmicos. Assim, este estudo recomenda o uso de 1,0 g/L para o sistema intensivo de aquicultura de acarás de água doce, e aplicação de manejo de limpeza para evitar a infestação parasitária, principalmente em aquários cerâmicos.

Palavras-chave: Peixe amazônico, *Capillaria pterophylli*. Acará bandeira, aquicultura ornamental.

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1. Introduction

The global ornamental fish trade has become a billion-dollar industry growing 14% annually since the 1970s (Jones et al., 2022). Brazil occupied the ninth position in the world ranking of ornamental fish exportation, generating approximately US$18 million. Among the various native fish species, the angelfish *Pterophyllum scalare* belongs to a unique group with the 30 most commercialized ornamental fishes worldwide (Eiras et al., 2022; Perera et al., 2023).

The popularity of *P. scalare* among fish farmers and hobby fishers relates to captivity adaptation, ease of reproduction, and acceptability as industrial food, making it economically viable (Gallani et al., 2016). For these reasons, the captive rearing of *P. scalare* in intensive production systems has been growing annually (Perera et al., 2023). However, some handling procedures from these systems, such as inadequate stocking densities, can reduce fish health and impair profits for fish farmers (Pinheiro-Junior et al., 2023).

High stocking densities cause stress (Eiras et al., 2022), which promotes immunosuppression by the fish (Gallani et al., 2016), consequently increasing diseases (Khanjani et al., 2023). The transmission of pathogens is another important consequence of inadequate management (Cardoso et al., 2024; Hoseinifar et al., 2023). An inadequate stock density creates a perfect field for transmission and parasite infestation (Dominguez et al., 2023; Eiras et al., 2022; Çağiltay et al., 2017; Abe et al., 2016).

Monogenea *Sciadicleithrum iphithimum* and the nematode *Capillaria pterophylli* have been reported in angelfish *Pterophyllum scalare*, causing a reduction in growth and increase in mortality (Fujimoto et al., 2006). However, detailed scientific reports on the relationship between high stocking densities and parasite load are lacking. Thus, this study evaluated the growth performance and parasite load of angelfish juveniles *Pterophyllum scalare* kept at different stocking densities in two rearing systems.

For the angelfish production system to become efficient, it is necessary to optimize the space by increasing the stocking rate without promoting fish imbalance, which promotes less resistance to farming diseases.

2. Materials and Method

The experiment was carried out at the Aquaculture Production Laboratory of the School of Agricultural Sciences of Federal University of Grande Dourados (UFGD), Brazil. The Brazilian guidelines on the care and use of animals for scientific and teaching purposes (DBCA) and the National Council for Animal Experimentation Control (CONCEA) were followed. The assay has been approved by the Ethics Committee for Animals Use of Universidade Federal da Grande Dourados (Protocolo 03/2019).

This study used a factorial design (4x2) with four stocking densities (0.1, 0.4, 0.7, and 1.0 g/L), two rearing systems (glass and ceramic aquariums), and four replicates. The experiment used 148 juvenile fish (3.05 ± 0.09 g) randomly placed in 32 aquariums (50L) equipped with filters and forced aeration. The fish were purchased from a commercial fish farm and the sanitary quality of the batch was certified.

The system was water recirculation with weekly replacement with water within potability parameters. All fish received two meals a day *ad libitum* avoiding food losses (8:00 and 16:00) of extruded ration (Poytara® crude protein 35%, crude fiber 4%, mineral material 9%, moisture 10%, calcium 2%, and phosphorus 1%). Each day, we measured the water temperature (YSI 550A), dissolved oxygen (YSI 550A), and pH (YSI 60). Total ammonia (NH_4) was also measured weekly using Hanna HI93715 and converted to non-ionized ammonia (NH_3).

At the end of the experiment (60 days), we measured and weighed all fish to determine growth parameters such as weight gain (WG: final weight – initial weight), biomass gain (BG: final biomass – initial biomass), feeding conversion rate (FCR), specific growth rate (SGR: ln(final weight) – ln(initial weight)/days × 100), final density (FD: final biomass/L), and survival (S: final number of fish/initial number of fish ×100).We also performed the parasitological analysis ofthe gills and midgut (proximal, middle, and distal intestine) from six fish per treatment to determine parasite prevalence, mean intensity, and abundance (Bush et al., 1997). All parasites were fixed and identified following the methods of Amato et al. (1991) and Eiras et al. (2006).

The data were subjected to the Shapiro Wilk normality test and Bartlett homoscedasticity tests, followed by a two-way analysis of variance (Two-Way Anova) with post hoc Tukey test ($p<0.05$).

3. Results

The use of a glass aquarium and a floor aquarium does not present a significant difference in water quality parameters and angel fish development. Most of the water quality parameters did not differ significantly among the treatments (Table 1). In addition, no interaction was observed between different stocking densities and rearing systems. Only toxic ammonia showed a difference with the increase in stocking density (Table 1).

The weight gain, specific growth rate, and feeding conversion rate did not differ among the stocking densities. Further, no mortalities occurred in the higher stocking densities (Table 2).

The parasite infected gill was *Sciadicleithrum iphithimum* (Dactylogyridae) and the gastrointestinal tract was *Capillaria pterophylli* (Capillariidae). The prevalence and mean intensity of monogenea were not influenced by the stocking densities tested, however an increase in the nematoda parasitism was observed along the increases of stocking density. The densities of 0.7 and 1.0 g/L presented the highest prevalence and mean intensities of *C. pterophylli* (Table 3). The rearing systems (glass or ceramic aquarium) did not have any effect on the prevalence ofeither parasite.

However, the mean intensity showed a statistical interaction between stocking density and rearing systems (Table 4).

Different uppercase letters in the row and lowercase in the column indicate statistical differences by Tukey test

Table 1. The water parameters and growth performance for angelfish *Pterophyllum scalare* reared at rearing systems.

Parameters	Rearing systems	
	Glass Aquarium	Cramic Aquarium
Waterparameters		
TemperatureºC	28.28 ± 0.11	28.10 ± 0.11
Dissolvedoxygen (mg/L)	5.66 ± 1.10	5.77 ± 0.52
pH	4.68 ± 0.82	5.14 ± 1.02
Toxic ammonia (µg/L)	5.45 ± 1.27	4.94 ± 0.53
Growth performance		
Biomassgain	36.23 ± 2.90	34.53 ± 2.90
Weightgain (g·fish-1)	1.78 ± 0.49	1.59 ± 0.61
Specific growth rate (g·day-1)	0.62 ± 0.13	0.55 ± 0.31
Feedingconversion rate	2.38 ± 1.34	2.91 ± 1.81
Final density	0.74 ± 0.06	0.76 ± 0.05
Survival (%)	97.50 ± 10.32	91.90 ± 17.67

Mean ± standard deviation. Different letters in the row indicate statistical differences for Tukey test (p≤0.05).

Table 2. The water parameters and growth performance for angelfish *Pterophyllum scalare* reared at different stocking densities.

Parameters	Different stocking densities			
	0.1 g/L	0.4 g/L	0.7 g/L	1.0 g/L
Waterparameters				
TemperatureºC	28.11 ± 0.50	28.37 ± 0.13	28.12 ± 0.16	28.16 ± 0.11
Dissolvedoxygen (mg/L)	5.99 ± 0.20	5.77 ± 0.41	5.64 ± 0.40	5.45 ± 0.25
pH	5.02 ± 0.72	5.06 ± 0.85	5.25 ± 0.93	4.30 ± 0.47
Toxicammonia (µg/L)	4.08 ± 0.24b	5.22 ± 1.04ab	5.73 ± 0.17ab	5.76 ± 0.61a
Growth performance				
Biomassgain	8.87 ± 1.70d	26.17 ± 1.80c	41.97 ± 3.70b	64.52 ± 4.30a
Weightgain (g·fish-1)	2.23 ± 1.41	1.40 ± 0.17	1.43 ± 0.19	1.67 ± 0.44
Specific growth rate (g·day-1)	0.74 ± 0.50	0.50 ± 0.15	0.51 ± 0.15	0.58 ± 0.12
Feedingconversion rate	3.44 ± 1.90	2.73 ± 0.20	2.36 ± 0.90	2.05 ± 1.49
Final density	0.19 ± 0.03c	0.58 ± 0.03b	0.92 ± 0.08ab	1.31 ± 0.09a
Survival (%)	87.25 ± 25.0	100.0 ± 0.0	95.00 ± 10.0	96.4 ± 7.50

Mean ± standard deviation. Different letters in the row indicate statistical differences for Tukey test (p≤0.05).

Table 3. Mean ± standard deviation of the parasite prevalence values in angelfish *Pterophyllum scalare* reared at different stocking densities and two types of aquariums.

	Differentstockingdensities				Typesofaquarium	
	0.1 g/L	0.4 g/L	0.7 g/L	1.0 g/L	Glass	Ceramic
*Gill	25.0 ± 50.0a	16.6 ± 33.0a	0.0 ± 0.0b	0.0 ± 0.0b	20.8 ± 39.0a	0.00 ± 0.00a
DI	0.0 ± 0.0b	12.5 ± 25.0b	100.0 ± 0.0a	100.0 ± 0.0a	56.2 ± 49.0a	50.0 ± 53.0a
MI	12.5 ± 2.5b	15.0 ± 5,7b	29.2 ± 24.0b	33.3 ± 27.0b	43.7 ± 42.0a	18.7 ± 27.0a
PI	0.0 ± 0.0a	37.5 ± 1.7a	12.5 ± 1.0b	16.6 ± 1.3b	22.9 ± 1.4a	10.4 ± 1.1a

Different lowercase letters in the row indicate statistical difference by Tukey test (p≤0.05). DI = intestine distal section; MI = intestine middle section; PI = intestine proximal section; *infestation of *S. iphithimum* in gills and *Capillaria pterophylli* infestation in midgut.

Table 4. Mean ± standard deviation of the mean intensity for angelfish *Pterophyllum scalare* reared at different stocking densities and two rearing systems (glass and ceramic aquariums).

Infestedorgan	Rearing system	0.1 g/L	0.4 g/L	0.7 g/L	1.0 g/L
*Gill	Ceramic	7.50 ± 1.12	60.00 ± 7.20	0.00 ± 0.00	0.00 ± 0.00
	Glass	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00
DI	Ceramic	0.00 ± 0.00Aa	2.50 ± 1.25Aa	7.00 ± 0.01Aa	2.95 ± 0.84Aa
	Glass	0.00 ± 0.00Ba	0.00 ± 0.00Ba	3.25 ± 3.12Aa	1.65 ± 0.84ABb
MI	Ceramic	0.50 ± 0.50	3.80 ± 0.08	5.75 ± 2.80	1.50 ± 4.50
	Glass	0.00 ± 0.00	0.00 ± 0.00	2.00 ± 8.00	3.00 ± 8.00
PI	Ceramic	0.00 ± 0.00	2.30 ± 3.30	0.00 ± 0.00	1.50 ± 4.50
	Glass	0.00 ± 0.00	0.00 ± 0.00	1.00 ± 2.00	1.50 ± 4.50

DI = intestine distal section; MI = intestine middle section; PI = intestine proximal section; *infestation of S. iphithimum in gills and Capillaria pterophylli infestation in the midgut.

($p \leq 0.05$), DI- intestine distal section, MI- intestine middle section; PI- intestine proximal section. *infestation of *S. iphithimum* in gills and *Capillaria pterophylli* infestation in the midgut.

4. Discussion

The freshwater angelfish is one of the most traded species of ornamental fish, however, there is a lack of scientific descriptions of the diseases that affect these animals (Gallani et al., 2016). Previous studies describe parasitic infestations caused by parasite (Florindo et al., 2017) and environmental factors are generally the main causes of stress in fish (Perera et al., 2023) decreasing their resistance to adverse factors (Ribeiro et al., 2023).

Most water physicochemical attributes were adequate for freshwater angelfish rearing, and the variables did not interfere with the growth performance of the fish (Abe et al., 2016; Çağiltay et al., 2017; Pinheiro-Junior et al., 2023). However, non-ionized ammonia increased the values at higher stocking densities. The fish growth can be affected by elevated levels of non-ionized ammonia (Çağiltay et al., 2017). In intensive aquaculture systems with high stocking densities, ammonia is a major problem in fish production (Barros et al., 2014). The lethal form (NH_3) provokes behavioral and physiological disorders such as oxidative stress or immunosuppression (Schroeder et al., 2011). Even at sub-lethal doses, growth reduction occurs over long exposure (Gonçalves et al., 2017). However, non-ionized ammonia values in this study remained stable and adequate for the angelfish *P. scalare,* with harmless effects on growth performance (Serezli et al., 2016). Elevated ammonia levels have also been linked to increased vulnerability to different parasitic, viral, bacterial diseases in fishes (Serezli et al., 2016; Neissi et al., 2020; Khanjani et al., 2023; Motta et al., 2021).

Currently, the greatest difference between food fish aquaculture and ornamental fish aquaculture is the commercialization method, where the ornamental fish are sold per unit and not by weight (Igarashi et al., 2004; Faria et al., 2019). Therefore, increasing stocking density makes the production system economically viable (Novák et al., 2020; Perera et al., 2023). The high stocking densities in the present study did not affect the growth of fish, making it more profitable for ornamental fish farmers (Çağiltay et al., 2017; Abe et al., 2016). Some reports such as in *Guppy Poecilia reticulata* and angelfish *Pterophyllum scalare* (Adel et al., 2013; Gonçalves Júnior et al., 2013) indicate 0.6g/L as an adequate stocking density to maintain performance and health, which is lower when compared to the present study of 1.0 g/L.

In addition, the lowest parasitological indexes for monogenean in this study could be explained by lower values of ammonia, even at high stocking densities. According to Florindo et al., (2017), monogenea prevalence increases in ornamental fishes subjected to greater levels of ammonia. The water quality is more factor that production. Moreover the level ammonia is influence that in all phase production (Barros et al., 2024; Cardoso et al., 2024), discarding the relationship between high stocking densities and monogenea infestation for angelfish.

In angelfish juvenile observed the gregarious conduct (Eiras et al., 2022) that facility the accidental ingestion of feces (Gallani et al., 2016) could be affecting nematode infestation in the intestine. Nematodes of *Capillaria* sp. (Capillariidae) present a direct cycle and the fish are contaminated by the ingestion of embryonated eggs that were expelled in the faeces from parasitized fish (Adel et al., 2013). Thus, the proximity between the animals, as well as the group's increase in seeking food may have increased parasitic infestation at higher densities. The reduction of the nematode mean intensity observed at the higher density can be explained by a factor known as the dilution effect, where the increase in advance density and a reduction of the parasite number due to a higher parasite distribution among the hosts (Samsing et al., 2014; Zhang et al., 2023; van Walraven et al., 2021).

In glass aquariums, due to the flat and smooth surface, cleaning is easier and more effective (Ullah et al., 2023), removing the eggs to avoid consumption. In aquariums with ceramic floors, due to porosity (Souza et al., 2008) there is greater adhesion of organic matter. The infestation increases in ceramic aquariums, probably because the eggs adhere to the bottom of the aquarium, making them easier to discover and consume. A ceramic aquarium is similar to a glass aquarium glass in terms of size and usefulness, but at a lower cost of production, which increases profitability. However, if a ceramic aquarium is chosen for the *P. scalare*rearing, cleaning should be intensified to remove the adhered eggs.

5. Conclusion

This study recommends that intensive rearing of angelfish can be carried out in glass or ceramic aquariums without harming development. That is essential the importance of the best of management practices in intensive systems in aquaculture and the relationship with the technical viability. The proper utilization on stocking density the 0.7g/L until 1.0 g/L

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