

Effects of feeding rate and feeding frequency on water quality and growth performance of juvenile tambaqui in a biofloc system

Efeitos da taxa e da frequência de arraçoamento sobre a qualidade da água e o desempenho de juvenis de tambaqui em sistema de bioflocos

Efectos de la tasa y la frecuencia de alimentación sobre la calidad del agua y el rendimiento de juveniles de tambaqui en un sistema de biofloc

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Abstract

Biofloc technology (BFT) stands out as a promising strategy for intensive aquaculture, where feeding rate and frequency are key management factors. This study aimed to evaluate the short-term effects of different feeding rates and feeding frequencies on water quality and growth performance of juvenile tambaqui (*Colossoma macropomum*) during the initial rearing phase in a biofloc system over 14 days. One hundred and twenty fish (2.42 ± 0.20 g) were used, distributed in a completely randomized design, in a 2×4 factorial scheme, with three replicates. The physicochemical parameters of the water remained generally stable; however, the feeding rate influenced dissolved oxygen and nitrite, with less favorable conditions at a rate of 10%. Feeding frequency affected total ammonia nitrogen, indicating interference in nitrogen dynamics. No significant differences were observed in growth parameters between treatments. However, the feeding rate impacted feed intake and feed conversion, with the 5% rate being the most efficient. Feeding frequency did not influence growth performance, indicating physiological and feeding plasticity of the species. A feeding rate of 5% is more suitable, as it reduces environmental impacts without compromising growth.

Keywords: Aquaculture; Intensive production; Feed management; Feed efficiency.

Resumo

A tecnologia de bioflocos (BFT) destaca-se como uma estratégia promissora para a aquicultura intensiva, na qual a taxa e a frequência de arraçoamento são fatores determinantes no manejo. Este estudo teve como objetivo avaliar os efeitos a curto prazo de diferentes taxas e frequências de alimentação na qualidade da água e no desempenho de

crescimento de juvenis de tambaqui (*Colossoma macropomum*) durante a fase inicial de cultivo em um sistema de bioflocos ao longo de 14 dias. Foram utilizados 120 peixes ($2,42 \pm 0,20$ g), distribuídos em delineamento inteiramente casualizado, em esquema fatorial 2×4 , com três repetições. Os parâmetros físico-químicos da água mantiveram-se dentro de faixas adequadas; no entanto, a taxa de arraçoamento influenciou o oxigênio dissolvido e o nitrito, com condições menos favoráveis na taxa de 10%. A frequência alimentar afetou o nitrogênio amoniacal total, indicando interferência na dinâmica do nitrogênio no sistema. Não foram observadas diferenças significativas nos parâmetros de crescimento entre os tratamentos. Entretanto, a taxa de arraçoamento impactou o consumo de ração e a conversão alimentar, sendo a de 5% mais eficiente. A frequência alimentar não influenciou os parâmetros zootécnicos, evidenciando flexibilidade fisiológica e alimentar da espécie. Conclui-se que a taxa de 5% é mais adequada, por reduzir impactos ambientais sem comprometer o crescimento.

Palavras-chave: Piscicultura; Produção intensiva; Manejo alimentar; Eficiência alimentar.

Resumen

La tecnología de biofloc (BFT) se destaca como una estrategia prometedora para la acuicultura intensiva, donde la tasa y frecuencia de alimentación son factores clave de manejo. Este estudio tuvo como objetivo evaluar los efectos a corto plazo de diferentes tasas y frecuencias de alimentación sobre la calidad del agua y el rendimiento de crecimiento de juveniles de tambaqui (*Colossoma macropomum*) durante la fase inicial de cría en un sistema de biofloc durante 14 días. Se utilizaron ciento veinte peces ($2,42 \pm 0,20$ g), distribuidos en un diseño completamente aleatorio, en un esquema factorial 2×4 , con tres réplicas. Los parámetros físicoquímicos del agua se mantuvieron generalmente estables; sin embargo, la tasa de alimentación influyó en el oxígeno disuelto y el nitrito, con condiciones menos favorables a una tasa del 10%. La frecuencia de alimentación afectó el nitrógeno amoniacal total, lo que indica interferencia en la dinámica del nitrógeno. No se observaron diferencias significativas en los parámetros de crecimiento entre tratamientos. Sin embargo, la tasa de alimentación impactó la ingesta de alimento y la conversión alimenticia, siendo la tasa del 5% la más eficiente. La frecuencia de alimentación no influyó en los parámetros zootécnicos, lo que demuestra la flexibilidad de la especie. Se concluye que una tasa de alimentación del 5% es la más adecuada, ya que reduce el impacto ambiental sin comprometer el crecimiento.

Palabras clave: Acuicultura; Producción intensiva; Manejo alimentario; Eficiencia alimenticia.

1. Introduction

Tambaqui (*Colossoma macropomum*) stands out as one of the most important species in Brazilian aquaculture, being one of the main native species cultivated in the country (FAO, 2022). Its economic, social, and productive relevance is significant, especially in the North and Northeast regions, where a large portion of production is concentrated, due to characteristics such as hardiness, rapid growth, and broad market acceptance (Rodrigues et al., 2023). According to IBGE (2023), tambaqui production reached approximately 113.6 thousand tons in 2022, corresponding to about 17% of national aquaculture production and generating a value exceeding R\$ 1.2 billion, consolidating its strategic role in the development of Brazilian aquaculture.

Simultaneously, national and global aquaculture have shown continuous expansion, driving the intensification of production systems (FAO, 2024). Although this process is necessary to meet the growing demand for fish, it is associated with relevant environmental challenges, such as increased nutrient loading in systems, effluent generation, and deterioration of water quality (Ahmad et al., 2017). In this context, the development of technologies that reconcile productivity and sustainability becomes essential.

Biofloc technology (BFT) emerges as one of the most promising strategies for modern aquaculture, as it integrates microbiological, physicochemical, and nutritional processes in systems with low or no water exchange (Avnimelech, 2015; Nakayama et al., 2022; Serafini et al., 2025; Santos et al., 2023; Yu et al., 2023b). In this sense, manipulation of the carbon:nitrogen (C:N) ratio stimulates the growth of heterotrophic bacteria capable of assimilating toxic nitrogenous compounds, such as ammonia, converting them into reusable microbial biomass (Crab et al., 2007; Dauda et al., 2018). In addition, nitrification occurs through autotrophic bacteria, converting total ammonia nitrogen (TAN) into nitrite and subsequently into nitrate (Luo et al., 2020). Thus, the balance between nutrient input and microbial assimilation capacity is determinant for system stability.

In this context, feeding management plays a central role, since feed constitutes the main source of nutrient input into the system (Tarigan et al., 2025). The amount and method of feed supply directly influence the formation, composition, and microbial activity of flocs, affecting nitrogen dynamics in the system (Adineh et al., 2025; Liu et al., 2025). Therefore, inadequate feeding strategies may compromise both water quality and fish productive performance.

Feeding rate expresses the percentage of biomass supplied daily as feed and is one of the main parameters of feeding management (Huang et al., 2025). High rates may result in the accumulation of organic matter, increased concentration of nitrogenous compounds, and reduction of dissolved oxygen, due to increased microbial activity and biochemical oxygen demand (Blatt et al., 2025). On the other hand, reduced rates may limit growth, although, in biofloc systems, part of this limitation may be mitigated by the ingestion of natural food available in the system (Aliabad et al., 2022). Recent studies indicate that reducing feeding rate may maintain or even improve feed efficiency and physiological parameters in biofloc systems, highlighting the importance of adjusting this factor (Adineh et al., 2025; Villes et al., 2024; Yadav et al., 2025). However, there is still a lack of studies focused on the initial rearing phase, especially for native species.

Feeding frequency, in turn, is related to the number of daily meals and the interval between them, influencing voluntary feed intake, digestibility, and fish metabolism (Baloi et al., 2016). Fractionating feeding throughout the day may favor greater nutrient utilization, reducing digestive overload and optimizing feed efficiency (Xu et al., 2020). In addition, feeding frequency may influence metabolic parameters, such as glucose levels, especially in juveniles, which have a high metabolic rate (Villes et al., 2024). However, most studies on feeding frequency have been conducted in conventional or recirculating systems, and information for biofloc systems remains limited, where nutritional dynamics are complex (Assis et al., 2025; Marinho-Pereira et al., 2025).

The interaction between feeding rate and feeding frequency is particularly relevant, as these factors act in an integrated manner, simultaneously influencing intake, metabolism, and nutrient utilization efficiency (Huang et al., 2025). While feeding rate defines the total amount of nutrients introduced into the system, feeding frequency regulates how these nutrients are distributed over time. Studies indicate that this interaction may influence both zootechnical performance and water quality, being especially important in intensive systems (Eiras et al., 2025; Rodrigues et al., 2024). However, investigations addressing this approach in biofloc systems, especially for tambaqui juveniles, are still scarce.

Given this context, this study aimed to evaluate the short-term effects of different feeding rates and feeding frequencies on water quality and zootechnical performance of tambaqui juveniles during the initial rearing phase in a biofloc system.

2. Methodology

An experimental laboratory study with a quantitative approach was conducted, employing calculations (Risemberg et al., 2026; Pereira et al., 2018), as well as simple descriptive statistics, including data classes, mean values, and absolute frequencies (Shitsuka et al., 2014), in addition to statistical analysis (Costa Neta & Bekman, 2009).

The experiment was carried out at the Aquaculture Laboratory (LAQUI) of the Federal University of Western Pará (Ufopa), Monte Alegre Campus, over a period of 14 days. The study was conducted in accordance with the guidelines of the National Council for the Control of Animal Experimentation and approved by the Ethics Committee on the Use of Animals of Ufopa. A total of 120 tambaqui juveniles (*Colossoma macropomum*), obtained from a commercial fish farm, were used. After transport to the laboratory, the animals were subjected to a seven-day acclimation period in a circular polyethylene tank with a useful volume of 500 L, containing water and continuous aeration.

Rectangular polyethylene tanks with a useful volume of 65 L were used as experimental units. Each unit was stocked

with five tambaqui juveniles, with an initial mean weight of 2.42 ± 0.20 g, corresponding to a stocking density of 77 fish m^{-3} , resulting in a biomass of 186 g m^{-3} . The experimental design was completely randomized in a 2×4 factorial arrangement, consisting of two feeding rates (5% and 10% of biomass per day) and four feeding frequencies (1, 2, 3, and 4 times per day), totaling eight treatments with three replicates, resulting in 24 experimental units. A commercial feed containing 42% crude protein, 8% ether extract, 3% fiber, 14% mineral matter, and 12% moisture was used. The feed was previously weighed using a digital scale and provided daily at 08:00, 11:00, 13:30, and 16:30 h, according to the evaluated treatment, with the amount per feeding adjusted based on the established feeding rate and frequency for each treatment.

The animals were reared in a biofloc system (BFT). Each experimental unit received 20 L of water from a previously matured biofloc fish culture, with total ammonia nitrogen (TAN) and nitrite concentrations below 0.5 mg L^{-1} (Zhang et al., 2016), used as the initial inoculum. The remaining volume (45 L) was completed with dechlorinated tap water. The units were maintained under continuous aeration, ensuring adequate water oxygenation, medium homogenization, and suspension of microbial flocs in the water column. The photoperiod was set at 12 h light:12 h dark.

Water quality monitoring included daily measurements of dissolved oxygen, temperature, and pH. The first two parameters were measured using a portable meter (BLE 91-00), and pH was measured using a portable multiparameter device (EZ-9909). TAN and nitrite concentrations were determined twice a week using a colorimetric method with commercial kits (Labcon Test). Weekly, alkalinity, salinity, and settleable solids volume were evaluated. Alkalinity was determined by a titrimetric method using a commercial kit, salinity with a portable multiparameter device (EZ-9909), and settleable solids using Imhoff cones, according to the methodology described by Avnimelech (2007). When TAN levels reached values equal to or higher than 1 mg L^{-1} , corrections were performed by adding a carbon source (sugar) at a ratio of 6 g of carbon per 1 g of TAN, according to Ebeling et al. (2006). To maintain alkalinity, sodium bicarbonate was applied at a dose of 30 g m^{-3} of water. During the experimental period, no water exchange or total suspended solids adjustments were performed; only dechlorinated water was added to compensate for evaporation and sampling losses.

Growth performance was evaluated through periodic biometric measurements, including an initial measurement, an intermediate measurement at 7 days, and a final measurement at the end of the experiment (14 days), using a precision digital scale, with the aim of monitoring animal growth and adjusting feed supply. At the end of the experimental period, the data were organized and used to calculate the zootechnical indices, as described below:

$$\text{Initial mean weight (g)} = \text{Initial biomass (g)} / \text{Initial number of fish}$$

$$\text{Final mean weight (g)} = \text{Final biomass (g)} / \text{Final number of fish}$$

$$\text{Specific growth rate (\% day}^{-1}\text{)} = [\ln(\text{final mean weight}) - \ln(\text{initial mean weight})] / \text{Time (days)} \times 100$$

$$\text{Survival (\%)} = (\text{Final number of fish} / \text{Initial number of fish}) \times 100$$

$$\text{Feed intake (g tank}^{-1}\text{)} = \sum \text{Daily feed provided (g)}$$

$$\text{Biomass gain (g)} = \text{Final biomass (g)} - \text{Initial biomass (g)}$$

$$\text{Feed conversion ratio} = \text{Feed provided (g)} / \text{Biomass gain (g)}$$

$$\text{Weight gain (g)} = \text{Final mean weight (g)} - \text{Initial mean weight (g)}$$

$$\text{Daily weight gain (g)} = \text{Weight gain (g)} / \text{Time (days)}$$

The obtained data were initially subjected to tests of residual normality (Shapiro–Wilk) and homogeneity of variances (Levene). For variables that met the assumptions, a two-way analysis of variance (Two-Way ANOVA) was performed. When effects were observed ($p < 0.05$), means were compared using Tukey's test and, when appropriate, interaction effects between

factors were further analyzed. Variables that did not meet normality and homogeneity assumptions were analyzed using the non-parametric Kruskal–Wallis test. When differences among treatments were detected ($p < 0.05$), medians were compared using Dunn’s test. All statistical analyses were performed using R software, version 4.4.2 (R Core Team, 2025).

3. Results

During the rearing of tambaqui juveniles in a biofloc system over 14 days, overall stability of water quality parameters was observed. However, specific effects of feeding rate and feeding frequency were detected on some variables. Regarding zootechnical performance, the observed differences were mainly associated with feed intake and feed conversion ratio.

The physicochemical parameters temperature, pH, alkalinity, settleable solids, and salinity were not influenced ($p > 0.05$) by feeding rates, feeding frequencies, or their interaction. However, total ammonia nitrogen (TAN) was significantly affected by feeding frequency. In contrast, feeding rate significantly influenced ($p < 0.05$) dissolved oxygen and nitrite levels. The interaction between factors showed a significant effect only for nitrite (Table 1).

Table 1. Physicochemical parameters of water of tambaqui juveniles (*Colossoma macropomum*) reared in a biofloc system for 14 days under different feeding rates and feeding frequencies.

Factor	Temperature (°C)	Dissolved oxygen* (mg L ⁻¹)	pH	TAN** (mg L ⁻¹)	Nitrite** (mg L ⁻¹)	Alkalinity (mg L ⁻¹)	SS (mL L ⁻¹)	Salinity (ppm)
Feeding rate								
5%	27.77	5.81a	7.74	0.13	0.03a	41.39	10.42	274.31
10%	27.70	5.65b	7.67	0.18	0.13b	45.69	11.36	278.32
p-value	0.415	0.018	0.151	0.283	<0.001	0.135	0.352	0.251
Feeding frequency								
1	27.75	5.78	7.72	0.15ab	0.07	44.44	10.47	278.28
2	27.72	5.74	7.72	0.23a	0.05	42.78	11.00	276.33
3	27.71	5.71	7.69	0.15ab	0.09	40.00	10.92	271.67
4	27.75	5.69	7.71	0.10b	0.10	46.94	11.17	278.97
p-value	0.973	0.775	0.979	0.011	0.835	0.338	0.964	0.437
Feeding rate × feeding frequency								
T5 F1	27.84	5.89	7.77	0.15	0.03	45.56	10.17	281.22
T5 F2	27.76	5.82	7.74	0.15	0.03	40.00	10.61	272.44
T5 F3	27.72	5.79	7.73	0.15	0.02	37.78	10.72	273.11
T5 F4	27.74	5.74	7.73	0.07	0.03	42.22	10.17	270.44
T10 F1	27.66	5.66	7.67	0.15	0.10	43.33	10.78	275.33
T10 F2	27.68	5.66	7.69	0.30	0.07	45.56	11.39	280.22
T10 F3	27.69	5.63	7.66	0.16	0.17	42.22	11.11	270.22
T10 F4	27.76	5.63	7.68	0.13	0.17	51.67	12.17	287.50
p-value	0.849	0.355	0.970	0.060	0.034	0.456	0.938	0.103
CV	0.74	2.66	1.42	52.05	96.55	18.53	22.14	2.98

* Means followed by different letters in the same column differ from each other by Tukey’s test ($p < 0.05$), after analysis of variance (ANOVA).

** Means followed by different letters in the same column differ from each other by Dunn’s test ($p < 0.05$), after application of the Kruskal–Wallis test.

TAN: total ammonia nitrogen; SS: settleable solids; T5 and T10: feeding rates of 5% and 10% of biomass per day; F1, F2, F3, and F4: feeding frequencies of 1, 2, 3, and 4 times per day; CV: coefficient of variation.

Source: Authors.

Dissolved oxygen was higher in experimental units subjected to a feeding rate of 5% of biomass per day, showing a difference of 2.8% compared to the 10% rate. In turn, nitrite concentration was approximately three times higher in units subjected to the 10% rate compared to those under the 5% rate. Regarding feeding frequency, a significant effect ($p < 0.05$) was observed only for TAN, with a concentration 130.0% higher at the frequency of two feedings per day compared to the lowest concentration recorded at four feedings per day.

For the interaction between feeding rate and feeding frequency, a significant effect ($p < 0.05$) was observed only for nitrite, with values ranging from 0.03 to 0.17 mg L⁻¹, corresponding to a variation of 466.7%. Higher concentrations were observed in treatments subjected to the 10% feeding rate, with a global effect identified by the Kruskal–Wallis test ($p < 0.05$). However, no pairwise differences were detected using Dunn’s test.

The results for the zootechnical performance of tambaqui juveniles reared in biofloc systems under different feeding rates and feeding frequencies are presented in Table 2.

Table 2. Growth performance of tambaqui juveniles (*Colossoma macropomum*) reared in a biofloc system for 14 days under different feeding rates and feeding frequencies.

Factor	IW (g)	FW (g)	SGR (% day ⁻¹)	Survival (%)	FI* (g tank ⁻¹)	BG (g tank ⁻¹)	FCR**	DWG (g day ⁻¹)
Feeding rate								
5%	2.43	3.81	3.19	100.00	0.12b	6.88	1.20b	0.10
10%	2.40	3.93	3.49	98.33	0.24a	7.62	2.42a	0.11
p-value	0.777	0.508	0.388	0.317	<0.001	0.330	<0.001	0.330
Feeding frequency								
1	2.44	3.82	3.20	100.00	0.19	6.88	1.94	0.10
2	2.28	3.66	3.41	96.67	0.16	7.03	1.60	0.10
3	2.50	3.95	3.26	100.00	0.19	7.23	1.87	0.10
4	2.45	4.00	3.49	100.00	0.18	7.75	1.68	0.11
p-value	0.306	0.609	0.518	0.391	0.221	0.848	0.858	0.848
Feeding rate × feeding frequency								
T5 F1	2.38	3.70	3.15	100.00	0.12	6.58	1.24	0.09
T5 F2	2.26	3.43	3.00	100.00	0.11	5.87	1.26	0.08
T5 F3	2.56	3.98	3.16	100.00	0.12	7.12	1.22	0.10
T5 F4	2.52	4.11	3.47	100.00	0.12	7.95	1.05	0.11
T10 F1	2.50	3.93	3.24	100.00	0.26	7.18	2.64	0.10
T10 F2	2.30	4.00	4.03	93.33	0.23	8.77	2.10	0.13
T10 F3	2.45	3.92	3.36	100.00	0.26	7.34	2.53	0.10
T10 F4	2.38	3.89	3.50	100.00	0.25	7.56	2.31	0.11
p-value	0.685	0.484	0.796	0.428	0.857	0.479	0.014	0.479
CV	8.68	11.27	19.18	4.11	9.19	24.09	26.00	24.09

* Means followed by different letters in the same column differ from each other by Tukey’s test ($p < 0.05$), after analysis of variance (ANOVA).

** Means followed by different letters in the same column differ from each other by Dunn’s test ($p < 0.05$), after application of the Kruskal–Wallis test.

IW: initial weight; FW: final weight; SGR: specific growth rate; FI: feed intake; BG: biomass gain; FCR: feed conversion ratio; DWG: daily weight gain; T5 and T10: feeding rates of 5% and 10% of biomass per day; F1, F2, F3, and F4: feeding frequencies of 1, 2, 3, and 4 times per day; CV: coefficient of variation.

Source: Authors.

Feeding rate significantly influenced ($p < 0.05$) feed intake and feed conversion ratio. Fish subjected to the 10% feeding rate showed approximately twice the feed intake compared to those fed at 5%, corresponding to an increase of 100.0%. Similarly, the 5% feeding rate provided higher feed efficiency, as indicated by better feed conversion ratio values, whereas the 10% rate resulted in lower efficiency, with feed conversion ratio 101.7% higher compared to the 5% rate.

Regarding feeding frequency, no significant differences ($p > 0.05$) were observed for any of the evaluated performance variables, indicating no effect of this factor on fish growth during the evaluated period. As for the interaction between feeding rate and feeding frequency, a significant effect ($p < 0.05$) was observed only for feed conversion ratio, with values ranging from 1.05 to 2.64, corresponding to a variation of 151.4% among treatments. A trend of poorer feed conversion ratios was observed in treatments subjected to the 10% feeding rate.

Overall, the results indicate that although the growth of tambaqui juveniles did not differ among the evaluated feeding strategies, the effects of feeding management were more evident on feed efficiency and water quality.

4. Discussion

Feeding rate and feeding frequency are key factors in the feeding management of tambaqui juveniles during the initial rearing phase. In the present study, feeding rate exerted a greater influence on feed efficiency and system limnological dynamics, mainly reflected in the concentrations of nitrogenous compounds and dissolved oxygen levels. In contrast, feeding frequency showed a more limited effect, restricted to total ammonia nitrogen (TAN). These results indicate that increasing the feeding rate promoted greater organic matter input into the system, possibly altering the balance between nutrient loading and microbial assimilation capacity (Chen et al., 2025), with direct implications for processes such as nitrification and microbial respiration within the system (Wu et al., 2024).

Despite the observed changes in water quality parameters and feed efficiency, juvenile growth did not differ among treatments over the 14-day period. This result suggests that, in the short term, tambaqui juveniles exhibit tolerance to moderate variations in environmental conditions, possibly associated with the species' high plasticity and the nutritional contribution of bioflocs as a complementary food source (Santos et al., 2023). Additionally, it indicates that the effects of feeding management on zootechnical performance may become more evident over longer experimental periods.

The overall stability of water quality observed throughout the experiment highlights the proper functioning of the biofloc system. Physicochemical parameters remained within the recommended ranges for the species, such as temperature between 27 and 32 °C, pH between 6.5 and 8.5, and dissolved oxygen above 4 mg L⁻¹ (Amanajás et al., 2018; Wood et al., 2018; Oliveira et al., 2017), conditions considered suitable for tambaqui culture in intensive systems. The absence of effects on variables such as pH, temperature, and alkalinity reinforces the system's capacity to maintain environmental balance even under different feeding strategies. This behavior is associated with the characteristic microbial activity of biofloc systems, in which heterotrophic bacteria assimilate nitrogenous compounds, reducing their toxicity and promoting greater environmental stability (Khanjani et al., 2024; Jan et al., 2026). Similar results were reported by Nakayama et al. (2022), who observed stability of nitrogen parameters during tambaqui juvenile culture in biofloc systems under different feeding regimes.

The increase in feeding rate from 5% to 10% of biomass per day resulted in reduced dissolved oxygen levels and increased nitrite concentrations, highlighting the impact of organic loading on system dynamics. This effect may be attributed to the greater input of organic matter derived from excess feed and excreta, which intensifies microbial respiration and increases biochemical oxygen demand (Li et al., 2023). Simultaneously, intensification of the nitrogen cycle occurs, with increased production of intermediate compounds such as nitrite. This pattern has been reported in intensive systems under conditions of overfeeding or high feeding rates (Yadav et al., 2025; Jan et al., 2026). In biofloc systems, although there is a

capacity for nitrogen assimilation, this capacity is limited and may be temporarily exceeded under conditions of high organic loading (Emerenciano et al., 2017; Li et al., 2023; Khanjani et al., 2025).

Regarding feeding frequency, the observed effect on TAN suggests that the distribution of feed throughout the day influences nitrogen excretion dynamics in the system. Lower feeding frequencies tend to generate higher TAN peaks due to concentrated feed intake, whereas higher frequencies promote a more gradual release of nitrogenous compounds (Hargreaves, 2013). This behavior is directly related to protein metabolism in fish, especially in juveniles, which exhibit high metabolic rates and nitrogen excretion proportional to feed intake (Mes et al., 2023; Zimmer et al., 2024). Thus, feed fractionation may contribute to greater stability of ammonia concentrations in biofloc systems (Luo et al., 2020; Chen et al., 2023).

The interaction between feeding rate and feeding frequency demonstrates that the effects of feeding frequency depend on the total amount of feed supplied. At lower feeding rates, the system exhibited greater stability, indicating that microbial assimilation capacity was not exceeded. Conversely, at higher feeding rates, feeding frequency was insufficient to compensate for increased organic loading, indicating a limit in the system's processing capacity (Villes et al., 2024; Adineh et al., 2025). This result reinforces that, in biofloc systems, environmental balance is more directly related to the total amount of nutrients added than to how these nutrients are distributed over time (Hisano et al., 2021; Mabroke et al., 2021; Marinho-Pereira et al., 2025).

The absence of effects on fish growth may be attributed to the short duration of the experiment (14 days). In short-term trials, fish tend to exhibit limited growth responses, even when subjected to different feeding management strategies (Assis et al., 2025). In addition, the presence of bioflocs as a supplemental food source may have contributed to reducing differences among treatments by providing nutrients continuously (Kuhn et al., 2009). Similar results were reported by Souza et al. (2014), who also observed no differences in the growth of tambaqui juveniles reared over a 30-day period.

The increased feed intake observed at the 10% feeding rate, without a corresponding increase in growth, resulted in poorer feed conversion, indicating lower efficiency in nutrient utilization. This pattern suggests that part of the supplied feed was not converted into biomass, being diverted to the culture environment and likely assimilated by the microbial community. Excess feed may reduce digestive efficiency and increase nutrient excretion, diverting energy that could otherwise be allocated to growth (Dalbem Barbosa et al., 2024). Studies with tambaqui and other tropical species have shown that feeding rates above optimal levels tend to reduce feed efficiency (Silva et al., 2007; Eiras et al., 2025).

Feeding frequency, when analyzed independently, did not influence productive performance, indicating that within the evaluated range, tambaqui exhibit flexibility in feed utilization. However, this effect may become more evident in longer-term experiments, in which differences in feed distribution over time may impact growth and feed efficiency. Rodrigues et al. (2024), evaluating tambaqui with an average weight of 95 g in earthen ponds, also observed no differences in growth between feeding frequencies of two and three meals per day, suggesting the ability of fish to adjust feed intake. Similarly, Leal et al. (2024) demonstrated that tambaqui juveniles reared in recirculating aquaculture systems (RAS) exhibited similar performance under different feeding regimes, reinforcing the species' feeding plasticity.

The interaction between feeding rate and feeding frequency for feed conversion ratio highlights that production efficiency depends on the appropriate combination of these factors. The best feed conversion ratios were observed in treatments with lower feeding rates, indicating that feeding frequency does not compensate for excess feed supply. This result emphasizes the importance of adjusting feeding rate as one of the main management strategies in biofloc systems.

From a practical perspective, the results indicate that using the lower feeding rate (5% of biomass per day) may represent greater potential feed savings and reduced environmental impacts without compromising growth in the short term. This strategy is particularly relevant during the initial rearing phase in biofloc systems, in which water quality stability is

critical for production success (Khanjani et al., 2024). Changes in limnological parameters may negatively affect zootechnical performance and survival of cultured juveniles (Putra et al., 2020; Yu et al., 2023a).

As limitations of the study, the short experimental duration (14 days) should be highlighted, as it may not reflect responses observed in longer production cycles, in which cumulative nutritional effects tend to become evident. Additionally, the stocking density used (77 fish m⁻³) is relatively low for biofloc systems, which may have reduced the intensification of competition for feed. Furthermore, no physiological analyses were performed, such as hematological or histomorphometric evaluations, which could contribute to elucidating underlying responses to treatments, as observed by Leal et al. (2024), who reported intestinal morphology changes associated with feeding management.

Overall, the results demonstrate that feeding management in biofloc systems should consider not only zootechnical performance but also its effects on water quality. In the short term, feeding rate proved to be more determinant than feeding frequency, directly influencing production efficiency and system dynamics.

5. Conclusion

A feeding rate of 5% of biomass per day provided higher feed efficiency and lower impact on water quality compared to the 10% rate, without compromising the growth of tambaqui juveniles (*Colossoma macropomum*) reared in a biofloc system over 14 days. Feeding frequency (1 to 4 times per day) did not influence zootechnical performance, indicating species flexibility under these conditions. Thus, in the short term, lower feeding rates are more efficient from both productive and environmental perspectives, while feeding frequency can be adjusted according to operational aspects without negatively affecting the animals.

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