



## Article

# Comparative Analysis of the Culture of Pink Shrimp *Farfantepenaeus brasiliensis* and Pacific White Shrimp *Litopenaeus vannamei* in Biofloc System

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**Abstract:** Shrimp farming in the Biofloc Technology System (BFT) is already considered an alternative to the traditional culture. The bioflocs maintain the water quality and can be used as a food supplement for shrimp. The Pacific white shrimp *Litopenaeus vannamei* forms the basis for most of the production in BFT. However, its culture is limited by the low temperatures. Thus, the BFT culture potential of native species, such as the pink shrimp *Farfantepenaeus brasiliensis*, should be considered. The present study aimed to compare the cultures of *F. brasiliensis* and *L. vannamei* in the grow-out phase in the BFT system. The experiment comprised two treatments: (FB), grown out of *F. brasiliensis*, and (LV), grown out of *L. vannamei*. The study lasted 70 days and was conducted at the Marine Station of Aquaculture at the Federal University of Rio Grande, Rio Grande do Sul State, Brazil. The stocking density was 100 shrimp/m<sup>2</sup> for both species. The shrimp were fed twice a day with commercial food. The physicochemical parameters of the water were monitored throughout the experimental period. The results showed that all physicochemical parameters of the water remained within the tolerated limits for both species. However, during the growth phase in the BFT, it was observed that the *L. vannamei* shrimp showed a better zootechnical performance than *F. brasiliensis*. The results indicate that *L. vannamei* has a higher capacity to catch bioflocs as supplementary food, demonstrating a better response of that species to the BFT system in the grow-out phase compared to *F. brasiliensis*.

**Keywords:** shrimp production; biofloc; pink shrimp; Pacific white shrimp



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## 1. Introduction

The crustaceans have stood out among the large cultured groups and presented essential growth in the last few years, contributing to 11 million tons of aquaculture production [1]. Most of this contribution comes from *Litopenaeus vannamei* shrimp farming, representing about 52% of the world's shrimp farming [1]. *L. vannamei* is a native species from the Eastern Pacific distributed from Mexico to the Northern coast of Peru. This species presents excellent robustness and easy adaptation to different culture systems, besides having one of the best zootechnical indices and great market acceptance [1–3]. Besides achieving high growth and survival rates, it has lower nutritional requirements in terms of protein when compared to other penaeid species such as *Farfantepenaeus paulensis* and *Farfantepenaeus brasiliensis* (Brazilian native species) [4–6].

Despite their higher nutritional requirements, native species can be interesting in aquaculture. *F. brasiliensis*, for example, has the potential to grow in confined environments,

as has already been shown in some studies [7,8]. The species ranges from North Carolina (USA) to the coast of Rio Grande do Sul (Brazil) [9,10] and constitutes one of the primary fishing resources of the southeastern Brazilian coast [10]. However, one of the main limitations to its production is the lack of a specific commercial feed that meets its nutritional requirements [5], as it is the lack of a complete technological package that would guarantee the economic viability of production [11].

Regardless of its expansion and economic growth, global aquaculture has still been questioned regarding its sustainability due to its impacts [12]. These nitrogen- and phosphate-rich nutrients can accelerate the eutrophication of water bodies or be a source of pathogens [13]. Due to this concern, some technologies have already been studied and used to reduce the impact generated by this sector; among them is the Biofloc Technology System (BFT), which aims at minimum or zero water exchange [14]. In this system, the carbon:nitrogen ratio of the water is manipulated to stimulate the emergence of microorganisms capable of assimilating and converting the nitrogen excreted by the cultured animals and from the leftover feed offered [15]. Several researchers have already reported that this type of system guarantees success in production [16–18]; besides decreasing the discharge of effluents from the environment, it can be classified as a more sustainable method of culturing. In addition to the reduction in effluent generation, the microorganisms present in bioflocs can serve as food [19–21], making it possible to reduce the protein levels in commercial feeds or even to act as a protein supplement for species that require higher values. The ability of penaeid shrimp to consume microorganisms can cause this type of food and culture system to have a high success rate. The authors of [22] reported the importance of bacteria, ciliates, and flagellates in the feeding and survival of *F. paulensis* larvae, and in [23], using protozoa, rotifers, and nematodes as live food for *L. vannamei* led to favorable results in terms of the zootechnical parameters. However, the aggregate forms of microorganisms can provide better consumption efficiency by the cultured shrimp [24].

*L. vannamei* has been one of the main species used for studies in BFT systems, with an already-consolidated technological package. The authors of [25,26] reported that bioflocs are important food sources for this species for reproduction and the post-larvae stage. It was stated in [21] that the contribution of this food source can vary from 63 to 86% in the growth phase. Few studies have evaluated the zootechnical performance of native species in a BFT system. Some studies have shown the potential of *F. brasiliensis* in this system [27–29], but they focused on the nursery phase, lacking further research to assess the potential and economic viability of *F. brasiliensis* and other native shrimps in biofloc system, especially in the grow-out phase. In this sense, the objective of this study was to evaluate the potential of rearing pink shrimp *F. brasiliensis* in a BFT system in the grow-out phase.

## 2. Material and Methods

### 2.1. Study Area

The study was conducted at the Marine Station of Aquaculture Professor Marcos Alberto Marchiori (EMA-FURG), Institute of Oceanography, Federal University of Rio Grande (FURG), located in the city of Rio Grande, Rio Grande do Sul State, Brazil (32° S, 52° W).

### 2.2. Origin of Shrimps

The research utilized two species of marine shrimp. The species *Litopenaeus vannamei* was purchased as nauplii from Aquatec® (Cangarueta, RN, Brazil). Upon arrival at the EMA-FURG, they were kept in the hatchery and nursery sectors until juveniles reached the weight of about 0.7 g for the experiment. The second species used in the study was the native *F. brasiliensis*. Shrimps were captured on the northern coast of Santa Catarina State and transported to the maturation sector of EMA-FURG. After seven days of acclimation, the shrimp were induced to undergo gonadal maturation through unilateral eye stalk ablation to obtain fertilized eggs. After 24 h of incubation, nauplii were transferred to the hatchery and nursery sectors until they weighed about 0.7 g, which was necessary to start the experiment.

### 2.3. Experimental Design

The study was conducted in a 582 m<sup>2</sup> greenhouse from March to June (summer/autumn). The experiment had two treatments that compared the cultures of the two species in a biofloc system. The treatment labeled LV = *Litopenaeus vannamei* and FB = *Farfantepenaeus brasiliensis*. Juveniles were stocked in twelve tanks (six tanks for each species) of 35 m<sup>3</sup> lined with HDPE (1.5 mm). Each tank was equipped with a diffused air aeration system, with the air injected by a blower and distributed throughout the tank through micro-perforated hoses (Aerotube<sup>®</sup>, Sandy Springs, GA, USA). Each tank was equipped with a feeding tray to check the daily feed consumption. The stocking density was 100 shrimp/m<sup>2</sup>. The animals were stocked with initial weights of  $0.72 \pm 0.37$  (*F. brasiliensis*) and  $0.78 \pm 0.29$  (*L. vannamei*). The shrimp were fed twice a day using a commercial diet (Potimar GUABI<sup>™</sup>) with 38% crude protein (feeding rate followed the methodology described in [30]).

### 2.4. Biofloc Formation

Before stocking the animals, each experimental unit was inoculated with about 10% of its volume with an inoculum of biofloc water from a previous culture that had already gone through the nitrification process and had a microbial community formed to avoid the high levels of nitrogen [31]. To maintain the C:N ratio of the water, organic fertilizations were carried out with sugarcane molasses, as proposed by [14,32].

### 2.5. Water Quality Management

Throughout the experiment, the temperature and dissolved oxygen parameters were monitored daily using an oximeter (YSI<sup>®</sup> 55, Yellow Springs, OH, USA), and pH using a digital pH meter (YSI<sup>®</sup> 60). When the pH was below the levels indicated for the species [33], hydrated lime [Ca(OH)<sub>2</sub>] was added to the water according to the methodology proposed in [34]. Total ammonia nitrogen (TAN) and nitrite (N-NO<sub>2</sub><sup>-</sup>) levels were monitored daily, according to [35]. Nitrate (N-NO<sub>3</sub><sup>-</sup>) and phosphate (P-PO<sub>4</sub><sup>-3</sup>) levels were determined weekly, following the methodology in [36]. The concentration of total suspended solids (TSS) and the alkalinity were determined three times a week, according to [35,37], respectively.

The levels of total suspended solids (TSS) were monitored and maintained (500 mg L<sup>-1</sup>) according to [38] through the clarification process of the culture water, according to the methodology of [39], which was adapted in [40]. The water turbidity was measured once a week using a turbidimeter (Hach<sup>®</sup> model 2100P, Loveland, CO, USA). The salinity and transparency of the water were measured twice a week, the first using an optical refractometer (Atago) and the second with a Secchi disk. The volume of settleable bioflocs was quantified three times a week using the Imhoff cone, as described by [41]. To help maintain the quality of the culture water, applications of a commercial probiotic (INVE<sup>®</sup>, Belgium) were made through two application routes: one applied directly to the water (0.5 ppm/week) and the other mixed into the feed (3.0 g kg<sup>-1</sup> feed).

### 2.6. Zootechnical Performance

Weekly shrimp weights were taken from 100 individuals per experimental unit using a 0.01 g precision scale (MARTE As 1000 C). After biometrics, the feed adjustments were calculated according to the methodology described in [30]. The zootechnical parameters evaluated were:

Final weight (g);

Weekly weight gain (g) (WWG) = (final weight – initial weight)/weeks of culture;

Productivity (kg/m<sup>2</sup>) = (final biomass – initial biomass)/volume of tank or culture area;

Survival (%) = (final biomass/average individual final weight)/number of individuals stocked) × 100;

Feed conversion rate (FCR) = Amount of feed supplied during the entire culture/(final biomass-initial biomass).

### 2.7. Structure Analysis of the Maxillipeds

The third maxilliped of each species was prepared for scanning electron microscopy (SEM). The maxilliped samples were washed thrice with 0.2 M sucrose and 0.1 M sodium cacodylate buffer solution for 15 min. Soon afterward, they were fixed in a 1.0% osmium tetroxide ( $\text{OsO}_4$ ) solution (1:1 2.0% osmium tetroxide solution and 0.4 M sodium cacodylate) for 3 h at 4 °C and then washed in water bidistilled for 15 min twice. After the fixation process, the samples were dehydrated with increasing ethanol concentrations. Then, for SEM, the maxilliped samples were dried in the critical point apparatus (Emitech K850, Montigny-le-Bretonneux, France) mounted on copper supports (stubs) and plated in liquid silver to allow for the conduction of electrons. The samples were dried at 26 °C and then taken to a metallizer (Emitech K 550) to be covered with a gold surface layer. The sample was prepared using the EMBRAPA Scanning Electron Microscopy Sample Processing Manual [42]. At the end of the process, the samples were observed using a Zeiss DSM 940 microscope (Baden-Württemberg, Germany).

### 2.8. Statistical Analysis

The data obtained were submitted to normality (Shapiro–Wilk) assumptions and homoscedasticity (Levene) tests to verify the data distribution. If assumptions were met, the data were submitted to one-way ANOVA ( $\alpha = 0.05$ ), and when significant differences were detected, Tukey's test was applied with a 95% reliability level [43].

## 3. Results

### 3.1. Water Quality

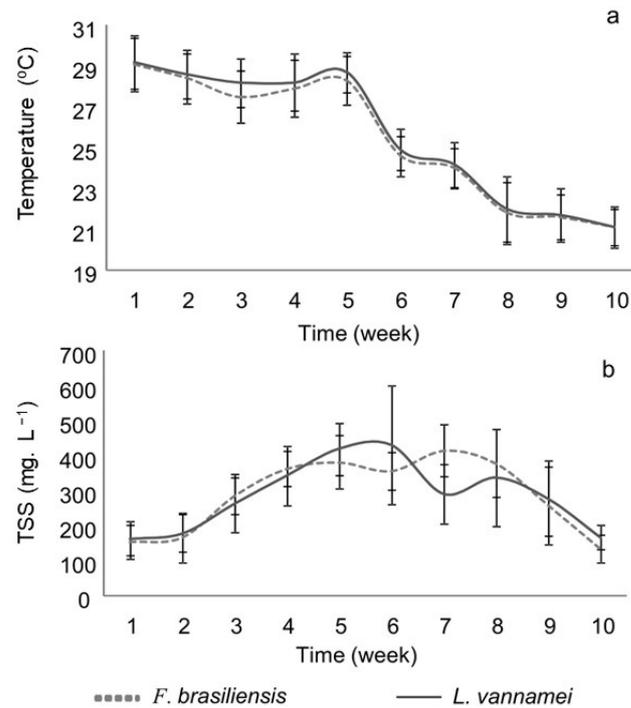
The mean values ( $\pm$ SD) of the water quality parameters are described in Table 1.

**Table 1.** Means ( $\pm$ standard deviation) of the physical and chemical parameters of the water in the culture of *Litopenaeus vannamei* and *Farfantepenaeus brasiliensis* in a biofloc system.

Parameters	Treatments	
	<i>F. brasiliensis</i>	<i>L. vannamei</i>
Temperature (°C)	25.4 $\pm$ 3.1	25.7 $\pm$ 3.2
Dissolved oxygen (mg L <sup>-1</sup> )	6.6 $\pm$ 0.5	6.5 $\pm$ 0.4
pH	8.1 $\pm$ 0.2	8.0 $\pm$ 0.2
Salinity	32.8 $\pm$ 0.7	33.12 $\pm$ 0.8
Alkalinity (mg of CaCO <sub>3</sub> L <sup>-1</sup> )	157.7 $\pm$ 23.3	143.56 $\pm$ 22.1
Total suspended solids (mg L <sup>-1</sup> )	298.5 $\pm$ 119.7	299.7 $\pm$ 128.4
Turbidity (NTU)	105.0 $\pm$ 83.9	110.8 $\pm$ 90.2
TA-N (mg L <sup>-1</sup> )	0.15 $\pm$ 0.11	0.20 $\pm$ 0.18
NO <sub>2</sub> <sup>-</sup> N (mg L <sup>-1</sup> )	0.3 $\pm$ 0.2	1.1 $\pm$ 1.30
NO <sub>3</sub> <sup>-</sup> N (mg L <sup>-1</sup> )	17.9 $\pm$ 8.8	19.1 $\pm$ 9.6
PO <sub>4</sub> <sup>3-</sup> P (mg L <sup>-1</sup> )	1.43 $\pm$ 0.93	1.30 $\pm$ 1.0

The temperature showed no significant differences between the treatments ( $p > 0.05$ ). It remained stable and above 28 °C until the fifth week of the experiment, and a decrease in temperature was observed until the end of the culture, obtaining average values below 22 °C (Figure 1a). The dissolved oxygen concentrations in both treatments remained above 6.0 mg L<sup>-1</sup>. Despite the lack of significant differences ( $p > 0.05$ ) between treatments during the entire culture, the FB treatment showed average oxygen levels above the LV treatment after the fifth week. The pH did not differ significantly ( $p > 0.05$ ) between treatments and maintained an average value of 8.05  $\pm$  0.2. Differences in mean salinity values between the FB (32.8  $\pm$  0.7) and LV (33.12  $\pm$  0.82) treatments were non-significant ( $p > 0.05$ ). The concentrations of total suspended solids showed a gradual increase from the beginning of the experiment (Figure 1b), with a subsequent decrease in these levels from the sixth week for LV treatment and from the seventh week for FB treatment due to the clarification process performed in the experimental units. TSS levels did not show statistical differences

between treatments ( $p > 0.05$ ). The turbidity values did not differ statistically ( $p > 0.05$ ) and showed the same variation pattern as the TSS. The treatments did not present significant differences regarding alkalinity ( $p > 0.05$ ). The mean concentrations were  $157.70 \pm 23.31$  and  $143.56 \pm 22.15 \text{ mg} \cdot \text{L}^{-1} \text{ CaCO}_3$  in the FB and LV treatments.

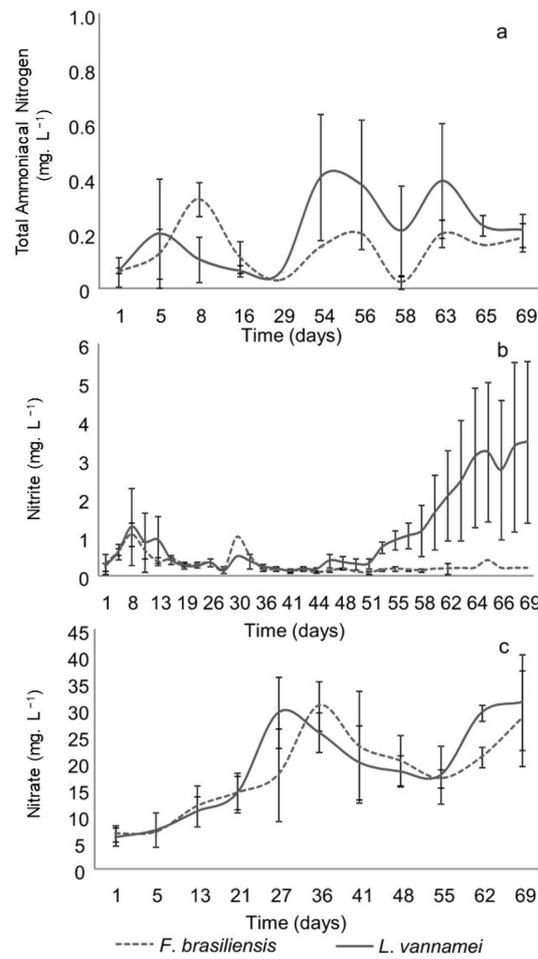


**Figure 1.** Variations (means  $\pm$  standard deviation) in (a) temperature and (b) total suspended solids concentration during the culture of *Litopenaeus vannamei* and *Farfantepenaeus brasiliensis* in a biofloc system.

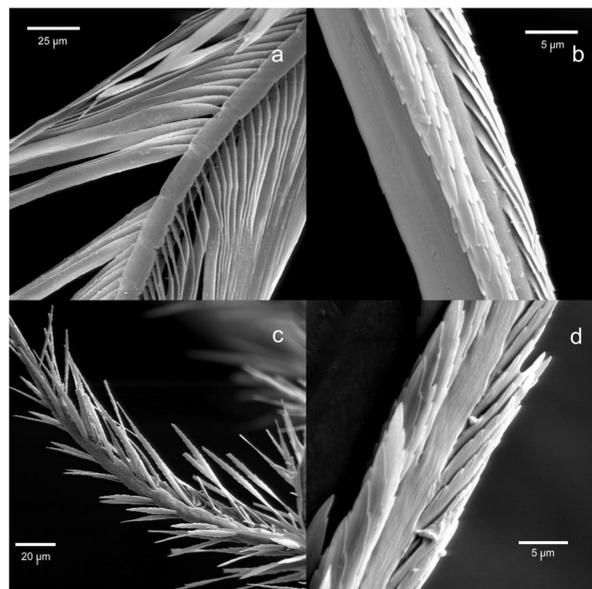
The concentration of total ammonia nitrogen (TA-N) showed oscillations in both treatments during cultivation, with higher levels for treatment LV (Figure 2a), though the differences were non-significant ( $p > 0.05$ ). Nitrite levels showed no significant differences between treatments ( $p > 0.05$ ). However, it was observed that, after the 50th day of the experiment, the concentration of this nutrient increased until the end of the experiment in the LV treatment only (Figure 2b). Nitrate and phosphate concentrations showed no significant differences between treatments ( $p > 0.05$ ). Nitrate gradually increased throughout the cultivation, with a maximum value of  $30 \text{ mg L}^{-1}$  at the end of the culture (Figure 2c). Low phosphate levels were found throughout the experiment.

### 3.2. Structure Analysis of the Maxillipeds

Thirty scanning electron microscopy photos of the third maxilliped of the *Litopenaeus vannamei* (15 images) and *Farfantepenaeus brasiliensis* (15 images) shrimp were analyzed. The photos had magnification levels from  $32\times$  to  $5000\times$ . Endopodites presented longer and feathery bristles in *L. vannamei* and shorter and tighter bristles in *F. brasiliensis* (Figure 3).



**Figure 2.** Variations (means  $\pm$  standard deviation) in concentrations of (a) total ammonia nitrogen, (b) nitrite, and (c) nitrate during the culture of *Litopenaeus vannamei* and *Farfantepenaeus brasiliensis* in a biofloc system.



**Figure 3.** Scanning electron microscopy photographs of the third maxilliped. (a) *Litopenaeus vannamei* 575 $\times$  magnification; (b) *Litopenaeus vannamei* 3000 $\times$  magnification; (c) *Farfantepenaeus brasiliensis* 600 $\times$  magnification; (d) *Farfantepenaeus brasiliensis* 3000 $\times$  magnification. Samples were cultured in biofloc system.

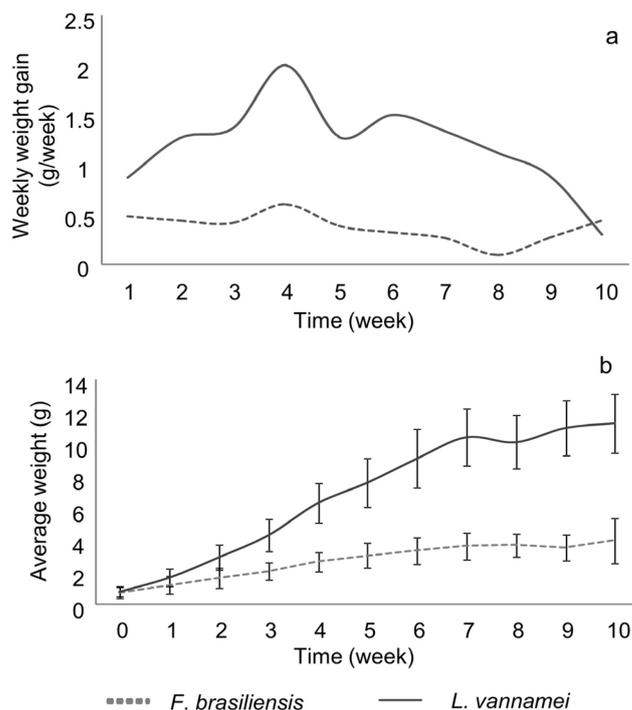
### 3.3. Zootechnical Performance

The indices of zootechnical performance for both treatments are presented in Table 2. The initial weights (g) of *F. brasiliensis* ( $0.72 \pm 0.37$ ) and *L. vannamei* ( $0.78 \pm 0.29$ ) were similar ( $p > 0.05$ ). However, the final weights of *L. vannamei* ( $11.28 \pm 1.89$ —LV treatment) were significantly higher ( $p < 0.05$ ) compared to the *F. brasiliensis* ( $3.96 \pm 1.40$ —FB treatment) (Figure 4b). Similarly, the weekly growth rate (Figure 4a) was also higher ( $p < 0.05$ ) for *L. vannamei*.

**Table 2.** Mean values ( $\pm$ standard deviation) of the zootechnical performance of *Litopenaeus vannamei* and *Farfantepenaeus brasiliensis* shrimp cultured in a biofloc system. Different superscript letters on the same lines indicate significant differences ( $p < 0.05$ ) between treatments.

Parameters	Treatments	
	<i>F. brasiliensis</i>	<i>L. vannamei</i>
Initial weight (g)	$0.72 \pm 0.37^a$	$0.78 \pm 0.29^a$
Final weight (g)	$3.96 \pm 1.40^a$	$11.28 \pm 1.89^b$
Weekly growth ( $\text{g week}^{-1}$ )	$0.32 \pm 0.20^a$	$1.05 \pm 0.62^b$
Final biomass (kg)	$9.34 \pm 1.68^a$	$40.49 \pm 2.55^b$
Survival (%)	$64.50 \pm 9.68^a$	$98.12 \pm 6.12^b$
Productivity ( $\text{kg m}^{-2}$ )	$0.26 \pm 0.05^a$	$1.12 \pm 0.07^b$
Feed conversion ratio	$5.45 \pm 1.34^a$	$1.43 \pm 0.06^b$

Significant differences ( $p < 0.05$ ) were found between the two species in terms of biomass, survival rate, and productivity, with the means of these parameters being  $9.34 \pm 1.68$  kg,  $64.50 \pm 9.68\%$ , and  $0.26 \pm 0.05$   $\text{kg/m}^2$  for the FB treatment and  $40.49 \pm 2.55$  kg,  $98.12 \pm 6.12\%$ , and  $1.12 \pm 0.07$   $\text{kg m}^{-2}$  for the LV treatment, respectively. The FCR was significantly ( $p < 0.05$ ) higher for *F. brasiliensis*.



**Figure 4.** Mean values (means  $\pm$  standard deviation) of zootechnical performance: (a) weekly weight gain and (b) mean weight during the culture of *Litopenaeus vannamei* and *Farfantepenaeus brasiliensis* in a biofloc system.

### 4. Discussion

The water quality parameters remained within the range suitable for the growth of both species [33]. Temperature is one of the most critical parameters, as it can affect the food consumption, growth, and survival of the species in culture environments [28,44].

According to [33], the Pacific white shrimp *L. vannamei* can survive in a wide temperature range (15–35 °C); however, for the best zootechnical performance, it should be between 28–32 °C [45]. In [46], it was found that the growth of *L. vannamei* was negatively affected at temperatures below 23 °C, and in [47], it was observed that temperatures of 20 °C negatively affected the species' food consumption and growth.

More information is needed regarding the ideal temperature range for the growth and survival of *F. brasiliensis*, especially in the grow-out phase. However, it is known that native species, both *F. brasiliensis* and *F. paulensis*, show greater tolerance to low temperatures when compared to *L. vannamei* [8,28,48]. Evaluating the effect of temperature in the nursery stage revealed that the best temperature for their growth is 27 °C. The average temperature of the present study was 25.5 °C, within the acceptable range for the growth of both cultivated species. During the first weeks of the culture, the average temperature in both treatments was near 29 °C, in the optimal range for growth. However, the temperature gradually decreased starting in the fifth week, reaching 21 °C at the end of the experiment. The progressive decline in temperature throughout the second half of the experiment period was one of the main factors responsible for the decrease in weekly weight gain and growth of both species, especially *L. vannamei*. The reported temperature drop was caused by successive cold fronts (coming from the south) passing through the region where the experiment was conducted.

Dissolved oxygen (DO) is a crucial parameter for thriving shrimp culture [49], and DO concentrations above 5.0 mg L<sup>-1</sup> are recommended [33,50]. In BFT systems, the DO requirement is higher, as besides the consumption by the shrimp, there is additional consumption by the microorganisms present in the bioflocs [51–53]. The present study's average DO concentration was 6.5 mg L<sup>-1</sup>, above the recommended level for shrimp cultures, throughout the experiment. The pH, alkalinity, and salinity were within the values recommended in [33].

The continuous monitoring of the TSS concentration in BFT systems is essential for the maintenance of water quality as well as for the zootechnical performance of the shrimps [15,40,54]. In this study, there were no significant TSS differences ( $p > 0.05$ ) between treatments, and they were within the recommended range for penaeid species [38,55]. Although there is not a recommended value for *F. brasiliensis*, the TSS values were similar to those found by [27,28,56] for a culture in biofloc system. From the sixth and seventh weeks of the LV and the FB treatments, the clarification process was started, aiming to decrease the TSS concentration, as suggested by [40,57]. According to [58], turbidity is directly related to TSS concentration, as an escalation in suspended particles increases turbidity in the culture water, decreasing light penetration. In the present study, turbidity did not lead to significant differences between treatments; it followed the variation of TSS throughout the experiment.

Ammonia is the main nitrogenous form of crustacean excreta [59], and, at high concentrations, can be toxic and harmful to shrimp, as can its metabolites (nitrite and nitrate) [60–62]. The values of these nitrogenous compounds found in this study remained within tolerable limits for both species [33,63], causing no adverse effects on the zootechnical performance of the farmed shrimp. Although, from the 50th experimental day, the nitrite concentration in the LV treatment gradually increased, remaining higher compared to the FB treatment until the end of the experiment (Figure 2b), these levels did not exceed the recommended limit for *L. vannamei* [62]. The continuous clarification process may have caused this increase in nitrite concentration in the LV treatment. From the 6th week of the culture, the nitrite concentration also increased when the solids removal process started in this treatment. The intense clarification before removing TSS may have also been responsible for eliminating heterotrophic and nitrifying bacteria in the biofloc, leading to an increase in nitrite in the system. It was observed in [64] that removing suspended solids decreases the bacterial abundance from the microbial community in the BFT system. Therefore, the intense clarification process from the 6th week onwards likely caused a

reduction in bacteria and a more significant increase in the total ammonia nitrogen in the water, as the bacteria are responsible for its removal [32].

The concentration and behavior of orthophosphates in culture are related to the constant nutrient input, with the decomposition of uneaten feed and the excretion of cultured organisms being the primary source of phosphorus in the system [65,66]. The authors of [66] reported that they found orthophosphate concentrations of 32 mg L<sup>-1</sup> in culture farms without harming the shrimp's performance. In this study, the average orthophosphate concentration in both treatments was much lower than [66] reported.

The results regarding zootechnical performance showed significant differences among treatments, with higher values of final weight, weekly weight gain, biomass, survival, and productivity for *L. vannamei* when compared to *F. brasiliensis* treatment. Some studies have shown good zootechnical performance results for *F. brasiliensis* cultured in a biofloc system. However, these encouraging results have been reported only in the nursery phase [27,28,56]. Information on growth in the BFT system is lacking for both *F. brasiliensis* and *L. vannamei*. Several studies corroborate the results regarding the adaptability of later species in this culture system [17,31,67].

Stocking density is a factor that plays a vital role in survival rate and other zootechnical parameters. This study found significant differences in survival rates between the LV and FB treatments. In other comparative studies, survival rates were not statistically different, e.g., [68,69], though stocking densities were lower compared to the present study. In [70,71], higher stocking densities than those used in this study (for *L. vannamei*) were tested. They found no significant differences in survival rates between treatments. The authors of [29] observed that *F. brasiliensis* could be reared at up to 600 m<sup>-2</sup> densities in a BFT system during the nursery phase without affecting survival. Although no studies have demonstrated the effects of different stocking densities for this species in the grow-out phase, in [72], different stocking densities were analyzed for the native pink shrimp, *F. paulensis*, a species of the same family. The authors recommended rearing them at stocking densities ranging from 40 to 120 shrimp m<sup>-2</sup> in the grow-out phase to obtain satisfactory zootechnical indices. However, ref [72] used a different system to the one presented in this study, using rearing cages in a natural environment. The stocking density used in this study was 100 shrimp per m<sup>-2</sup>, a low density when compared to other research on the BFT system [73–77]. The BFT system aims for high productivity in small areas, which requires high operating costs [78]; production at very low stocking densities could make it economically unviable. The density did not affect the growth of the *L. vannamei* shrimp. However, as there have been no previous studies on the growth of *F. brasiliensis*, more studies are needed in order to indicate the effects of stocking density on this species so that the economic and zootechnical indices in the BFT system are suitable.

Determining the protein digestibility of feedstuffs that comprise shrimp feed is essential for developing balanced, high-quality diets [79–81]. In addition, effective diets based on the responses of different shrimp species can offer many advantages, such as a better feed conversion rate and faster growth [79,82]. In this study, both treatments were fed with commercial feed specifically developed for *L. vannamei* based on the nutritional requirements of this species. This may have favored lower AFCR rates for *L. vannamei* compared to *F. brasiliensis*.

Regarding utilizing biofloc as a food supplement for these species, the contribution was more significant for *L. vannamei*. In [19], a substantial contribution of biofloc was also shown for the nutrition of this species. The structure of its third maxilliped contributed to the better nutrition and overall zootechnical performance of *L. vannamei*. This appendix is the largest and outermost buccal apparatus in decapod crustaceans [83,84], and it performs several functions, including the manipulation of food particles [85,86]. In *L. vannamei*, the endopods of the third maxilliped are covered by more extended, more abundant, and feathered bristles (Figure 3a,b) that facilitate the capture of biofloc particles, whereas *F. brasiliensis* (Figure 3c,d) has more straightforward and shorter bristles, making capturing particles more difficult. The endopods of *F. brasiliensis* may also be adapted to its feeding

habits. The authors of [87] analyzed the stomach contents of this species. They obtained results showing that the majority of its diet is made up of insects and mollusks, and that only 0.6% of its stomach contents were made up of debris. It was also observed in [88] that crustaceans and mollusks represented the majority of prey consumed and that microorganisms had only minor contributions to the diet, which leads us to think that this species may be less adapted to consuming bioflocs, especially during the grow-out phase. The authors of [89] observed the exact difference between the third maxilliped of *L. vannamei* and other penaeid species. They showed that the former has a higher efficiency in terms of capturing biofloc. Using the stable isotope analyses, ref. [21] showed that bioflocs can contribute to up to 86% of the feed of *L. vannamei* in the grow-out phase. These results corroborate those found in this study and show that *L. vannamei* can perform better in this culture system.

## 5. Conclusions

Based on the results obtained in the present study, *F. brasiliensis* shrimp present lower zootechnical indices compared to *L. vannamei*, showing that this species still needs to be adapted to this system during the grow-out phase. However, significant advances are still required in order to make the cultivation of native species viable, such as new studies on their nutritional needs, protein digestibility, stocking densities, genetic improvement and consumption, and use of bioflocs. Furthermore, this study corroborates research on the adaptability and production success of *L. vannamei* when reared in a biofloc culture system.

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