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EFFECT OF ALKALINITY ON FOOD CONSUMPTION OF JUVENILE PACIFIC WHITE SHRIMP REARED IN CLEAR WATER AND BIOFLOC SYSTEM*

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ABSTRACT

This study aimed to evaluate the effect of alkalinity on food consumption and other performance parameters of juvenile *Litopenaeus vannamei* reared in water containing biofloc and clear water. For this purpose, shrimp of 4.06 ± 0.34 g were kept in containers of 3 L of biofloc and clear water during 3 days, with the concentrations of 50, 100 and 200 mg L^{-1} of alkalinity, plus a Control group, with 5 replicates each group. Food consumption was verified once a day and other performance parameters were evaluated at the end of the experiment. The food consumption and survival rates of the shrimp was not affected by the different levels of alkalinity and clear water and biofloc systems. The best results of weight gain and specific growth rate were observed in the highest concentrations of alkalinity in the biofloc system. Survival, as well as food consumption, was not affected between levels of alkalinity and in clear water and biofloc. The exposure to inappropriate alkalinity concentrations over long periods of time can adversely affect the animals, thus emphasizing the importance of maintaining adequate levels of alkalinity to the cultivated species.

Key words: calcium carbonate; feeding; *L. vannamei*; microbial floc.

EFEITO DA ALCALINIDADE NO CONSUMO ALIMENTAR DE JUVENIS DE CAMARÃO BRANCO DO PACÍFICO CULTIVADOS EM SISTEMA DE ÁGUA CLARA E BIOFLOCOS

RESUMO

Esse estudo objetivou avaliar o efeito da alcalinidade no consumo alimentar e demais parâmetros de desempenho de juvenis de *Litopenaeus vannamei* cultivados em água contendo bioflocos e água clara. Para tanto, durante 3 dias, camarões de 4,06 ± 0,34 g foram mantidos em recipientes de 3 L, sob as concentrações Controle, 50, 100 e 200 mg L^{-1} de alcalinidade, com 5 repetições cada, em bioflocos e água clara. O consumo alimentar foi verificado uma vez ao dia e os demais parâmetros de desempenho foram avaliados ao final do experimento. Nesse estudo, verifica-se que o consumo alimentar dos camarões não é afetado entre os níveis de alcalinidade e nos sistemas de água clara e bioflocos. Já o ganho em peso e a taxa de crescimento específico são afetados positivamente nas maiores concentrações de alcalinidade, no sistema de bioflocos, onde demonstram os melhores resultados. E a sobrevivência, assim como o consumo alimentar, não é afetada entre os níveis de alcalinidade e nos sistemas de água clara e bioflocos. Contudo, a possibilidade de exposição à concentrações de alcalinidade inapropriadas, durante longos períodos de tempo, pode afetar negativamente os animais, assim, ressaltando a importância da manutenção da alcalinidade em níveis adequados à espécie cultivada.

Palavras-chave: carbonato de cálcio; alimentação; *L. vannamei*; flocos microbianos.

INTRODUCTION

In a shrimp farm, the food management is an important factor to be considered, since its management can affect, for example, growth, feed conversion rate and survival of animals (NUNES and PARSONS, 1998; CUZON *et al*., 2004; BARBIERI *et al*., 2016). Appropriate feeding practices include observing the weight and survival (*e.g.* biomass) of the shrimp (PONTES *et al*., 2008; BARBIERI *et al*., 2015). Both biotic and abiotic parameters affect food consumption of the Pacific white shrimp *Litopenaeus vannamei*

(JORY *et al*., 2001). The abiotic parameter of alkalinity represents the concentration of bases able to neutralize the acids present in water. The main bases responsible for alkalinity are the carbonates (CO_3^{-2}) and bicarbonates (HCO_3^-), expressed in calcium carbonate ($CaCO_3$) equivalent (FURTADO *et al*., 2011). According to VAN WYK and SCARPA (1999), alkalinity is an important factor in a cultivation system, as its buffering capacity (*e.g.* to maintain the balance of acid ↔ base) decreases pH variations throughout the day. In superintensive cultivation systems without water renewal, the concentrations of alkalinity and pH may decrease due to consumption of alkalinity by heterotrophic and nitrifying bacteria (EBELING *et al*., 2006). According to CHEN *et al*. (2006), for each gram of ammonia ($N\text{-}NH_3$ + $N\text{-}NH_4^+$) oxidized to nitrate ($N\text{-}NO_3^-$), 7.07 g of alkalinity and 4.18 g of dissolved oxygen are consumed, 0.17 g of which are from bacterial biomass. However, EBELING *et al*. (2006) affirmed that for each gram of ammonia assimilated to the bacterial protein, 3.57 g of alkalinity, 4.71 g of dissolved oxygen and 15.17 g of carbohydrates are consumed, whereas 8.07 g of bacterial biomass are formed and 9.65 g of carbon dioxide are produced. When comparing the performance of heterotrophic and nitrifying bacteria, the heterotrophic consume around half the amount of alkalinity and constitute around eight times more bacterial biomass with almost the same consumption of dissolved oxygen in relation to the nitrifying bacteria (EBELING *et al*., 2006). FURTADO *et al*. (2011) recommend to maintain the alkalinity levels above 100 mg L^{-1} of $CaCO_3$, in cultivation systems without water renewal, for the best performance of nitrifying bacteria. Along with phytoplankton, zooplankton and detritus, this bacterial biomass constitutes the microbial floc or biofloc (BURFORD *et al*., 2003; AVNIMELECH, 2007; SCHRYVER *et al*., 2008; RAY *et al*., 2010b). The biofloc represent an additional food source to commercial feed (HARGREAVES, 2013), offering better performance results of penaeid shrimps (MOSS and PRUDER, 1995; EPP *et al*., 2002; ARNOLD *et al*., 2009; MEGAHED, 2010; KRUMMENAUER *et al*., 2014). The *L. vannamei* is the most widely cultivated species throughout the world, especially in the Western Hemisphere (SAOUD *et al*., 2003), and it shows a great performance in superintensive cultivation systems without water renewal (CUZON *et al*., 2004; WASIELESKY JUNIOR *et al*., 2006a). Thus, this study evaluates the effect of alkalinity on the food consumption and other performance parameters such as weight gain, specific growth rate, feed conversion rate and survival of *L. vannamei* cultured in water containing biofloc and in clear water.

METHODS

Location

The experiment was conducted in the Laboratory of Shrimp Farming at the Marine Aquaculture Station "Prof. Marcos Alberto Marchiori" of the Federal University of Rio Grande, located at the Cassino Beach (Rio Grande/RS/Brazil - 32°11'S, 52°10'W).

Biologic material

The postlarvae (PL) of the shrimp *Litopenaeus vannamei* were acquired in the PL10 stage from the commercial laboratory Aquatec (Canguaretama/RN/Brazil) and kept in the hatchery, in fiber tanks with sea water renewed every 2 days (28°C and 30‰), with commercial feed (40% crude protein-Guabi) and nauplii of *Artemia* sp. for 40 days. The animals were then transferred to wooden tanks (35 m^2), covered with high-density polyethylene geomembrane in an agricultural greenhouse, with water coming from a superintensive cultivation without water renewal (29°C and 30‰) and commercial feed (PotiMar/38% crude protein-Active-Guabi) for 27 days. After this period, the juvenile shrimp were reallocated for the acclimatization to the experimental conditions.

Acclimatization

Prior to the beginning of the experiment, the animals were acclimated to the experimental conditions for 4 days. In a room with 12-h photoperiod, one of two polyethylene tanks (163 L of usable volume) was filled with sea water filtered in sand filter and cuno (5 µm), chlorinated water (15 mL sodium hypochlorite/1000 L water) and dechlorinated (1 mg ascorbic acid/1000 L water) (29°C and 33‰) (Clear Water, called CW system). The other tank was filled with water coming from a superintensive cultivation without renewal of water containing suspended solids (28°C and 35‰, 0.18 mg L^{-1} ammonia, 0.54 mg L^{-1} nitrite, 461 mg L^{-1} total suspended solids and 120 mg L^{-1} alkalinity) (Biofloc, named BF system). Both tanks had 130 shrimp each. The water from the tanks was constantly aerated with air diffusers attached to silicone hoses connected to a radial air compressor with 4 HP. The water from CW system was daily renewed in 50%. The animals were fed a commercial feed (PotiMar/38% crude protein - Active - Guabi), through the trays, twice a day (9:00 and 15:00 h), according to JORY *et al*. (2001) (initial rate of 5% of the biomass). An excess of over 50% of feed was offered, as the authors recommend this to ensure full satiety of the shrimp. However, the unconsumed feed was removed from the trays the day after, in order to maintain a good water quality. The animals were kept in the circumstances described during 3 days. After the acclimatization, the shrimp were acclimated to the experimental conditions for over a day. For this stage of acclimatization, 40 polyethylene containers (3 L of usable volume) were filled, 20 with Clear Water - CW system (27°C and 34‰) and other 20 with Biofloc - BF system (27°C and 37‰, 0.18 mg L^{-1} ammonia and nitrite 0.08 mg L^{-1}, 511 mg L^{-1} total suspended solids, 115 mg L^{-1} alkalinity), and one animal was randomly transferred (4.06 ± 0.34 g) each. The aeration of the containers and the shrimp feeding were similar to the previously exposed at the initial stage of acclimatization.

Experimental protocol

The animals were maintained in the same containers and reared for an experimental period of 3 days. Aliquots of hydrochloric acid and calcium hydroxide solutions were added to each group of containers (20 in CW system and 20 in BF system) in order to obtain different predetermined concentrations of alkalinity. So, based on the level of 100 mg L^{-1} indicated by VAN WYK and SCARPA (1999), the concentrations of 50, 100 and 200 mg L^{-1} were used (in the form of calcium carbonate, $CaCO_3$), plus a Control group (without hydrochloric acid and calcium hydroxide), with 5 repetitions each, totaling 40 experimental units. The water in the container

was aerated and alkalinity concentrations were adjusted on a daily basis with aliquot parts of the solutions. The shrimp were fed as cited previously for the acclimatization, twice per day (13:00 and 15:00 h), according to JORY *et al*. (2001) (initial rate of 5% biomass). An excessive amount of feed of 50% from the recommended by the researchers was offered, in order to provide enough food for the complete satiety of the animals.

Water physical and chemical parameters

Temperature and dissolved oxygen in experimental media were verified with an oximeter Oxi 315i-WTW/USA, twice a day (9:00 and 15:00 h). Salinity and pH were daily measured (9:00 h) with the refractometer Salt Refractometer w/ATC-Sper Scientific/USA and the pHmeter S20 SevenEasy™ - Mettler Toledo/USA, respectively. The total suspended solids were daily evaluated with the Gravimetric method of Volatilization, according to STRICKLAND and PARSONS (1972). Alkalinity levels, ammonia and nitrite were measured daily by Titrimetry, Indophenol Blue and Griess Reaction, described by APHA (1998), UNESCO (1983) and AMINOT and CHAUSSEPIED (1983), respectively.

Food consumption of the shrimp

Food consumption of the shrimp was verified with the method of Suppression and Provision of Food, adapted from SOARES *et al*. (2005). To this end, the ration was: (1st) suppressed, from 17:00 h until 13:00 h of the following day, totaling 20 hours of suppression to allow the evacuation of the stomach and presumably stimulate the appetite of the animals; (2nd) provided, at 13:00 h; (3rd) removed at 14:00 h, totaling 1 hour of provision, in order to measure, from the food unconsumed, the amount of feed effectively consumed by the shrimp; and (4th) provided, from 15:00 h to 17:00 h, totaling 2 hours of provision, to ensure full satiety, prior to the next period of suppression. The unconsumed food was placed in aluminum crucibles and dried until constant weight in a chamber (Odontobrás/Brazil) at 60°C, and weighed once a day with an analytical scale (ED-Sartorius/USA). The feed of the respective container was not considered for the assessment of food consumption if exuviae was observed.

The food consumption was assessed with the following equation:

$$FC = \left\{\left[FP_{NH}^{*} - \left(FRC_{NH}^{*} - C_{NH}^{*}\right)\right] - L\right\} - SS \quad (1)$$

Where, FC = food consumption (g), FP_{NH} = feed provided (g), FRC_{NH} = feed removed and crucible (g), C_{NH} = crucible (g), L = leaching (1.5%), SS = suspended solids possibly present in the unconsumed food in BF system containers (5%) and $_{NH}$* = no humidity (8%).

Performance parameters of the shrimp

The weight gain, specific growth rate, feed conversion rate and survival were verified by the following formulae at the end of the experiment:

$$WG = W_F - W_I \quad (2)$$

Where, WG = weight gain (g), W_F = final weight of the shrimp at the end of the experiment (g) and W_I = initial weight of the shrimp at the beginning of the experiment (g).

$$SGR = \left[\left(\ln W_F - \ln W_I\right) \times 100\right] / ED \quad (3)$$

Where SGR = specific growth rate (% day^{-1}), $lnWF$ = neperian logarithm of the final weight of the shrimp (g), $lnWI$ = neperian logarithm of the initial weight of the shrimp (g), DE = number of experimental days.

$$FCR = FC / WG \quad (4)$$

Where FCR = feed conversion rate, FC = food consumption (g) and WG = weight gain (g).

$$S = \left(L_F / L_I\right) \times 100 \quad (5)$$

Where, S = survival (%), L_F = number of live shrimp at the end of the experiment and L_I = number of live shrimp at the beginning of the experiment.

Statistical analyses

Prior to performing the statistical analysis, the performance parameters of the shrimp, such as specific growth rate and survival, expressed in percentage, were arcsine transformed. All values of water physical and chemical parameters and performance parameters of the shrimp were verified for the assumptions of normality (Kolmogorov-Smirnov) and homoscedasticity (Levene), and transformed in case the assumptions were not met. Finally, the values were submitted to an analysis of variance, *ANOVA*, Factorial, and means were compared with Tukey's *HSD* test when significant differences were detected (ZAR, 1996). The statistical analyses were performed with the software STATISTICA 7.0.

RESULTS

Water physical and chemical parameters

Some physical and chemical parameters showed significant differences between treatments ($p<0.05$) (Tables 1a and 1b).

Shrimp food consumption

Some of the performance parameters of the shrimp showed significant differences between treatments ($p<0.05$) (Tables 2a and 2b, Figure 1). The food consumption did not differ significantly between the levels of alkalinity and the systems CW and BF (0.06, 0.05, 0.06 and 0.07 g feed shrimp^{-1} hour^{-1} and 0.08, 0.07, 0.07 and 0.09 g feed shrimp^{-1} hour^{-1}, respectively) (Figure 1).

Table 1a. Water physical and chemical parameters of juvenile *Litopenaeus vannamei* reared under different concentrations of alkalinity (mg L^{-1} of $CaCO_3$), in clear water system[1,2].

	*Control***	*50*	*100*	*200*
T* morning (°C)	27.11 ± 0.37[a]	26.94 ± 0.28[a]	27.03 ± 0.30[a]	27.00 ± 0.34[a]
T afternoon (°C)	27.49 ± 0.18[a]	27.17 ± 0.12[a]	27.57 ± 0.23[a]	27.56 ± 0.24[a]
DO* morning (mg L^{-1})	6.23 ± 0.13	6.30 ± 0.11	6.36 ± 0.20	6.30 ± 0.13
DO afternoon (mg L^{-1})	6.19 ± 0.09	6.44 ± 0.09	6.29 ± 0.15	6.35 ± 0.21
Salinity (‰)	34.40 ± 0.51[b]	34.33 ± 0.49[b]	34.80 ± 0.41[b]	34.53 ± 0.52[b]
pH	8.30 ± 0.04[a]	7.78 ± 0.14[a]	8.18 ± 0.06[a]	8.37 ± 0.05[a]
Ammonia (mg L^{-1})	1.98 ± 0.79[a]	2.33 ± 1.03[a]	2.23 ± 1.02[a]	2.18 ± 1.03[a]
Nitrite (mg L^{-1})	0.04 ± 0.03[b]	0.02 ± 0.02[b]	0.02 ± 0.02[b]	0.05 ± 0.04[b]
TSS* (mg L^{-1})	-	-	-	-

*T = temperature, DO = dissolved oxygen, and TSS = total suspended solids. [1]Means ± standard deviations of five repetitions. [2]Different superscript letters in a row indicate significant differences ($p<0.05$). **According to the treatments Control, 50, 100 and 200, the verified concentrations are, respectively, 147.33 ± 3.72, 51.33 ± 2.29, 107.00 ± 7.02 and 188.67 ± 3.52 mg L^{-1}.

Table 1b. Water physical and chemical parameters of juvenile *Litopenaeus vannamei* reared under different levels of alkalinity (mg L^{-1} of $CaCO_3$), in biofloc system[1,2].

	*Control***	*50*	*100*	*200*
T* morning (°C)	25.41 ± 0.32[b]	25.49 ± 0.29[b]	26.21 ± 0.68[ab]	25.78 ± 0.38[ab]
T afternoon (°C)	26.33 ± 0.21[b]	26.25 ± 0.26[b]	26.61 ± 0.10[b]	26.59 ± 0.18[b]
DO* morning (mg L^{-1})	6.52 ± 0.05	6.52 ± 0.06	6.40 ± 0.11	6.47 ± 0.06
DO afternoon (mg L^{-1})	6.63 ± 0.05	6.68 ± 0.06	6.58 ± 0.08	6.60 ± 0.06
Salinity (‰)	38.13 ± 0.92[a]	38.36 ± 1.45[a]	37.80 ± 1.08[a]	37.87 ± 0.83[a]
pH	8.12 ± 0.26[a]	7.00 ± 0.40[b]	7.88 ± 0.12[a]	8.39 ± 0.08[a]
Ammonia (mg L^{-1})	0.08 ± 0.04[b]	0.67 ± 0.30[ab]	0.25 ± 0.18[b]	0.14 ± 0.09[b]
Nitrite (mg L^{-1})	0.22 ± 0.16[a]	0.06 ± 0.03[b]	0.14 ± 0.06[a]	0.13 ± 0.03[a]
TSS* (mg L^{-1})	518.17 ± 140.45[a]	391.69 ± 86.26[b]	478.29 ± 92.41[ab]	506.67 ± 98.28[a]

*T = temperature, DO = dissolved oxygen, and TSS = total suspended solids. [1]Means ± standard deviations of five repetitions. [2]Different superscript letters in a row indicate significant differences ($p<0.05$). **According to the treatments Control, 50, 100 and 200, the verified levels are, respectively, 135.33 ± 8.76, 43.93 ± 4.46, 89.00 ± 3.38 and 185.67 ± 4.17 mg L^{-1}.

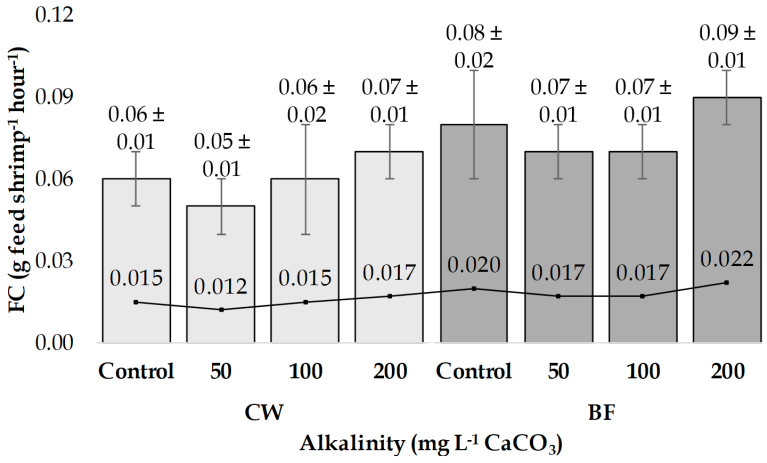

Figure 1. Food consumption (FC) of juvenile *Litopenaeus vannamei* reared under different concentrations of alkalinity, in clear water (CW) and biofloc (BF) systems. Means ± standard errors of five repetitions. Values within the bars (-▪-) correspond to the average food consumption expressed in g feed g shrimp^{-1} hour^{-1}.

Table 2a. Performance parameters of juvenile *Litopenaeus vannamei* reared under different concentrations of alkalinity (mg L^{-1} of $CaCO_3$), in clear water system[1,2].

	*Control***	*50*	*100*	*200*
WG* (g)	0.41 ± 0.13[ab]	0.13 ± 0.08[c]	0.30 ± 0.09[b]	0.45 ± 0.06[ab]
SGR* (% day^{-1})	2.23 ± 0.72[ab]	0.81 ± 0.55[b]	1.79 ± 0.42[ab]	2.57 ± 0.38[ab]
FCR*	0.46 ± 0.09[b]	1.39 ± 0.28[a]	0.64 ± 0.13[a]	0.61 ± 0.12[a]
Survival (%)	100.00	100.00	100.00	100.00

*WG = weight gain; SGR = specific growth rate, and FCR = feed conversion rate. [1]Means ± standard deviations of five repetitions. [2]Different superscript letters in a row indicate significant differences ($p<0.05$). **According to the treatments Control, 50, 100 and 200, the verified concentrations are, respectively, 147.33 ± 3.72, 51.33 ± 2.29, 107.00 ± 7.02 and 188.67 ± 3.52 mg L^{-1}.

Table 2b. Performance parameters of juvenile *Litopenaeus vannamei* reared under different levels of alkalinity (mg L^{-1} of $CaCO_3$), in biofloc system[1,2].

	*Control***	*50*	*100*	*200*
WG* (g)	0.50 ± 0.10[a]	0.35 ± 0.10[b]	0.39 ± 0.08[ab]	0.49 ± 0.07[a]
SGR* (% day^{-1})	3.18 ± 0.70[a]	1.80 ± 0.53[ab]	2.31 ± 0.59[ab]	3.13 ± 0.81[a]
FCR*	0.32 ± 0.07[b]	0.65 ± 0.13[a]	0.45 ± 0.09[b]	0.44 ± 0.09[b]
Survival (%)	100.00	100.00	100.00	100.00

*WG = weight gain; SGR = specific growth rate, and FCR = feed conversion rate. [1]Means ± standard deviations of five repetitions. [2]Different superscript letters in a row indicate significant differences ($p<0.05$). **According to the treatments Control, 50, 100 and 200, the verified levels are, respectively, 135.33 ± 8.76, 43.93 ± 4.46, 89.00 ± 3.38 and 185.67 ± 4.17 mg L^{-1}.

Performance parameters of the shrimp

In both systems, the weight gain differed significantly between the concentrations of alkalinity. This parameter was higher in the treatments Control and 200 mg L^{-1} (0.50 and 0.49 g, respectively), in the BF system ($p<0.05$). In relation to the specific growth rate, this differed significantly between the levels of alkalinity and the systems CW and BF, being greater in Control and 200 mg L^{-1} (3.18 and 3.13% day^{-1}, respectively), in the BF system ($p<0.05$). In both systems, the feed conversion rate differed significantly between the concentrations of alkalinity. The parameter was lower in the Control group (0.46), in the CW system, and in the concentrations Control, 100 and 200 mg L^{-1} (0.32, 0.45 and 0.44, respectively) in the BF system ($p<0.05$). Survival was not significantly different, being equal (100.00%) between the different concentrations of alkalinity and the systems CW and BF.

DISCUSSION

Water physical and chemical parameters

In the system of clear water and biofloc, in all concentrations of alkalinity, the temperature in the morning and afternoon were slightly out of the 28 to 32°C range indicated by WALKER *et al*. (2011) for the highest growth and survival of juvenile *L. vannamei*. The dissolved oxygen in the morning and afternoon, at all levels of alkalinity in both systems, were around 5 mg L^{-1}, which was suggested by CARBAJAL-HERNÁNDEZ *et al*. (2013) for the cultivation of marine shrimps. In clear water and biofloc systems, at all levels of alkalinity, salinity was within the range of 33 to 40‰, advocated by PONCE-PALAFOX *et al*. (1997) for greater growth and survival of juvenile *L. vannamei*. The pH, at all concentrations of alkalinity in both systems, remained within the range of 7 to 9, suggested by CHEN *et al*. (2006) for the best development of heterotrophic and nitrifying bacteria. In the clear water and biofloc systems, at all levels of alkalinity, ammonia and nitrite concentrations were below the levels of safety of 3.95 and 25.7 mg L^{-1}, in the salinity of 35‰, recommended by LIN and CHEN (2001, 2003), respectively, for the cultivation of *L. vannamei* juveniles. In all alkalinity levels, total suspended solids were slightly out of the range of 453 to 465 mg L^{-1} indicated by RAY *et al*. (2010a) for the best growth of juvenile *L. vannamei* reared in a superintensive system with minimum water renewal.

Concentration of alkalinity

Food consumption of the shrimp

Food consumption presented similar values between levels of alkalinity and in clear water and biofloc systems.

Performance parameters of the shrimp

However, the weight gain showed the lowest value in the concentration 50 mg L^{-1} in the clear water system, and the highest levels in Control and 200 mg L^{-1} groups in the biofloc system. Similarly, FURTADO *et al*. (2011) cultivated juvenile *L. vannamei* with pH and alkalinity corrected with additions of Na_2CO_3, $Ca(OH)_2$ and $NaHCO_3$. The shrimp were exposed to 78, 100, 144.5 and 162.2 mg L^{-1} of $CaCO_3$ for 60 days, and the lowest weight gain was observed in the lowest alkalinity level (6.2 g). In the same way, FURTADO *et al*. (2015) reared juveniles

of the same species with 70.26, 145.50, 224.33 and 299.13 mg L^{-1} of $CaCO_3$ for 49 days, also observing a decreased weight gain in the lowest alkalinity level (4.57 g). The specific growth rate exhibited the lowest value in the level of 50 mg L^{-1} in the clear water system, and the highest value in the Control and 200 mg L^{-1} of the biofloc system. Similar to this study, FURTADO *et al*. (2015) registered the lowest specific growth rate in the lowest concentration of alkalinity (6.41% day^{-1}). However, unlike our study, FURTADO *et al*. (2014) cultivated juveniles of *L. vannamei* under different doses of $Ca(OH)_2$, exposed to levels of 82.05, 90.65, 132.17 and 159.27 mg L^{-1} of $CaCO_3$ for 56 days, and verified the lowest specific growth rate in the highest concentration of alkalinity (5.07% day^{-1}). The feed conversion rate showed the highest values at 50, 100 and 200 mg L^{-1} in the clear water, and at 50 mg L^{-1} in the biofloc system, and the lowest levels in the Control of the clear water system and at the Control, 100 and 200 mg L^{-1} of the biofloc system. FURTADO *et al*. (2011) and (2015) observed a greater feed conversion rate in the lowest concentration of alkalinity (3.0 and 1.15, respectively). However, FURTADO *et al*. (2014) recorded a higher feed conversion rate in the highest level of alkalinity (1.79). Survival showed equal values between the alkalinity concentrations and the clear water and biofloc systems. These values (100.00%) were higher than those recorded by FURTADO *et al*. (2011, 2014, 2015) cultivating *L. vannamei* (from 80.0 to 85.0%, 85.03 to 93.72% and 88.00 to 92.12%, respectively) under different levels of alkalinity. However, it is noteworthy that the present study had much smaller duration than the studies cited. Along with alkalinity, the pH of the water is able to affect the growth of animals, which could be signaled in the current study. PAN *et al*. (2007) cultivated *L. vannamei* postlarvae, exposing them to pH 7.1, 7.6, 8.1, 8.6 and 9.1 for 96 hours, and observed the lowest weight gain in the lowest pH, but similar survival among all treatments. WANG *et al*. (2012) clarify that pH can affect growth, metabolism, homeostasis, osmoregulation and immunity of penaeids, such as the *L. vannamei*. ALLAN and MAGUIRE (1992) affirmed that low pH levels decrease the growth of *Penaeus monodon*. WASIELESKY JUNIOR *et al*. (2006b) observed that the pH reduction to values below 7 in cultivation systems without water renewal also difficult the growth of *L. vannamei*. In addition, WICKINS (1976) explains that low values of pH reduce the frequency of ecdysis of *P. monodon*. ABBINK *et al*. (2011) cultivated juvenile fish of the species *Seriola lalandi*, exposed to pH 6.58, 7.16 and 7.85 for 27 days and observed a decreased final weight and specific growth rate in the lowest pH. The authors explain that the acidification of the water of cultivation may cause acidosis in the blood of the animals, leading to the disruption of their homeostasis to a higher demand/expenditure of energy.

Cultivation system

Food consumption of the shrimp

The food consumption had equal values between levels of alkalinity and the CW and BF systems. WASIELESKY JUNIOR *et al*. (2006a), in a cultivation of juvenile *L. vannamei* in a superintensive system without water renewal, with commercial feed containing 35% of crude protein, observed the lowest food consumption in the control of the clear water system (145.89 g $replica^{-1}$) in comparison to the biofloc system (158.81 g $replica^{-1}$). In the study cited, although food consumption was assessed with a different feeding schedule from our study, it serves as a comparison of the feeding behavior of *L. vannamei* reared in a system without water renewal, containing biofloc, and in clear water.

Performance parameters of the shrimp

The weight gain showed the smallest value in the concentration 50 mg L^{-1} of the clear water system, and the highest level at the Control and 200 mg L^{-1} groups of the biofloc system. Similar to our study, XU and PAN (2012), rearing juveniles of *L. vannamei* in a system without water renewal, under C:N relations of 15:1 and 20:1, observed minor weight gain in the clear water system (control) (41.7%), which was different from the biofloc system (56.5%). The specific growth rate exhibited the lowest value in the level of 50 mg L^{-1} in the clear water system, and the highest value in the Control and 200 mg L^{-1} of the biofloc system. Similarly to the current study, XU *et al*. (2012), reared juveniles of *L. vannamei* in a system without water renewal, with a ration containing 35% crude protein, and verified the lowest specific growth rate in the control of clear water system (1.38% day^{-1}), when compared to the biofloc system (1.52% day^{-1}). KRUMMENAUER *et al*. (2011, 2014) cultivated juveniles of *L. vannamei* in a biofloc system for 120 and 30 days and observed growth of up to 0.92 and 1.14 g $week^{-1}$ or 2.39 and 2.93% day^{-1}, respectively. HARGREAVES (2013) explains that the biofloc present in farming systems without water renewal represent an additional food source (besides the commercial ration) for the animals. Several studies have evaluated the centesimal composition of biofloc, registering levels from 14.50 to 52.05% protein (essential and non-essential amino acids), 0.10 to 9.90% of lipids (saturated, monounsaturated and polyunsaturated fatty acids) and 28.25 up 36.40% carbohydrates, as well as pigments (carotenoids and chlorophylls) and minerals (WASIELESKY JUNIOR *et al*., 2006a; AZIM and LITTLE, 2008; AZIM *et al*., 2008; JU *et al*., 2008; KUHN *et al*., 2009, 2010; BECERRA-DÓRAME *et al*., 2012; MAICÁ *et al*., 2012; XU and PAN, 2012; XU *et al*., 2012). Such composition offers excellent nutritional quality, improving the zootechnic indices of the shrimp, as it might be registered to weight gain and specific growth rate in this study. However, CRAB *et al*. (2010) underline that the nutritional properties of biofloc are influenced by factors like the carbon source added to the cultivation to improve the natural productivity. The researchers explain that each carbon source stimulates the development of certain species of bacteria, protozoa and microalgae, which constitute, as such, specific microbial communities and ultimately confer to a proximal composition to the biofloc with attractiveness, palatability and digestibility. The feed conversion rate showed the highest values at 50, 100 and 200 mg L^{-1} in the clear water, and at 50 mg L^{-1} in the biofloc system, and the lowest levels in the Control of the clear water system and at the Control, 100 and 200 mg L^{-1} of the biofloc system. VINATEA *et al*. (2010) and SCOPEL *et al*. (2011) cultivated juveniles of *L. vannamei* in systems without water renewal for 21 weeks and 76 days,

respectively, and observed mean feed conversion rates of 1.36 and 1.57, higher than the overall mean among treatments in biofloc system in the present study (0.47). Survival showed equal values between alkalinity and concentrations in clear water and biofloc systems. These results (100.00%) were higher than those observed by AUDELO-NARANJO *et al*. (2012) and FRÓES *et al*. (2013) which cultivated juvenile *L. vannamei* in systems without water renewal for 40 days and 3 months (values from 97 to 95%), respectively.

CONCLUSIONS

Food consumption of juvenile *L. vannamei* is not affected between levels of alkalinity (Control, 50, 100 and 200 mg L^{-1}) and in clear water and biofloc. The weight gain and specific growth rate are affected positively by the highest concentrations of alkalinity, Control and 200 mg L^{-1}, biofloc system, with the best results. Survival and food consumption are not affected between levels of alkalinity and in clear water and biofloc. However, the possibility of exposure to inappropriate alkalinity concentrations over long periods of time can adversely affect the animals, thus emphasizing the importance of maintaining appropriate levels of alkalinity to the cultivated species.

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