INFLUENCE OF STOCKING DENSITY ON THE ZOOTECHNICAL PERFORMANCE OF *Litopenaeus vannamei* DURING THE NURSERY PHASE IN A BIOFLOC SYSTEM

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ABSTRACT

This study evaluated the zootechnical performance of the marine shrimp *Litopenaeus vannamei* reared at different stocking densities during the nursery phase in a biofloc culture system. The experiment consisted of four treatments with three replicates, corresponding to the densities of 1,500, 3,000, 4,500 and 6,000 shrimp m^{-2}. Twelve 300 L circular plastic tanks (microcosms) with a bottom area of 0.5 m^2 were randomly stocked with shrimp (0.004 g) according to the treatments and maintained for 35 days. These tanks were supplied by biofloc water from a 200 m^2 matrix tank (macrocosm), where *L. vannamei* was reared with the stocking density of 300 shrimp m^{-2}. The water quality parameters remained within acceptable limits throughout the experiment. The zootechnical performance results showed significant differences (P<0.05) among treatments. The values of final weight were higher in shrimp reared at the densities of 1,500, 3,000 and 4,500 m^{-2}, whereas survival was significantly reduced at 4,500 and 6,000 shrimp m^{-2}. The density of 3,000 shrimp m^{-2} was considered the most appropriate considering the zootechnical performance for the nursery phase of *L. vannamei* in a biofloc technology system in northeastern Brazil.

Keywords: shrimp; bft; growth; survival

INFLUÊNCIA DA DENSIDADE DE ESTOCAGEM NO DESEMPENHO ZOOTÉCNICO DE *Litopenaeus vannamei* DURANTE A FASE DE BERÇÁRIO EM SISTEMA DE BIOFLOCOS

RESUMO

O presente estudo avaliou o desempenho zootécnico do camarão marinho *Litopenaeus vannamei* quando cultivado em diferentes densidades de estocagem na fase de berçário em sistema de bioflocos. O experimento foi composto de quatro tratamentos com três repetições, correspondendo às densidades de 1.500, 3.000, 4.500 e 6.000 camarões m^{-2}. Camarões com peso médio inicial de 0,004 g foram estocados aleatoriamente, conforme os tratamentos, em 12 tanques plásticos circulares, com área de fundo de 0,5 m^2 e volume útil de 300 L (microcosmos) durante um período de 35 dias. Estes tanques foram conectados a um sistema de recirculação abastecido pela água de um tanque retangular de 200 m^2 (macrocosmo), utilizado para o cultivo de *L. vannamei* na densidade de 300 camarões m^{-2} em sistema de bioflocos. Os parâmetros de qualidade de água se mantiveram dentro dos limites aceitáveis durante o experimento. Os resultados referentes ao desempenho zootécnico apresentaram diferenças significativas (P<0,05) entre os tratamentos. Os valores de peso final foram superiores nos camarões cultivados nas densidades de 1.500, 3.000 e 4.500 m^{-2}, enquanto que a sobrevivência foi significativamente reduzida nas densidades de 4.500 e 6.000 camarões m^{-2}. A densidade de 3.000 camarões m^{-2} foi considerada a mais adequada, considerando o desempenho zootécnico, para o cultivo de *L. vannamei* na fase de berçário em sistema de bioflocos na região nordeste do Brasil.

Palavras chave: camarão; bft; crescimento; sobrevivência

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INTRODUCTION

The traditional shrimp farming has, among other characteristics, high rates of water renewal to maintain water quality. However, it also releases effluents with various organic and inorganic compounds into the environment. These effluents are composed mainly by ammonia, phosphorus and dissolved carbon, which are water pollutants (MCINTOSH *et al.*, 2000; JACKSON *et al.*, 2003; COHEN *et al.*, 2005). Moreover, the drained water may introduce and increase the occurrence of pathogenic microorganisms in the environment (PIEDRAHITA, 2003). The current development model of shrimp cultivation should follow the premises of sustainable development.

The shrimp culture in biofloc technology systems (BFT) is based on the formation of microbial aggregates, high productivity through the use of high stocking densities, and little or no water renewal, which reduces the emission of effluents to the environment and increases the biosafety levels in the culture (MCINTOSH *et al.*, 2000). The bioflocs are formed by adding a carbon source in the water, and represent an alternative source of energy and high protein food for the shrimp (CRAB *et al.*, 2007; OTOSHI *et al.*, 2011; XU *et al.*, 2012). Furthermore, the bacterial community forming bioflocs is able to recycle the organic matter accumulated during cultivation, as well as convert nitrogen compounds into microbial biomass (SAMOCHA *et al.*, 2007; DE SCHRYVER *et al.*, 2008).

Among several variables related to the management of biofloc systems, the choice of the most appropriate shrimp stocking density for the different culture phases is critical because it exerts great influence on the productivity (JACKSON and WANG, 1998; MOSS and MOSS, 2004). The nursery phase corresponds to the intermediate stage between the larval rearing and shrimp growout, generally realized in tanks with higher stocking densities than those used in the growout phase. This procedure enables a better management control and biosecurity in the early stages of cultivation, which has a positive reflection on the zootechnical performance of the animals during the growout phase, as well as benefits related to area and yield optimization in the culture (COELHO *et al.*, 2007; FOES *et al.*, 2011; WASIELESKY *et al.*, 2013). However, the excessive increase of stocking density during this phase may reduce the growth and survival of shrimp (MOSS and MOSS, 2004; KRUMMENAUER *et al.*, 2011). This fact is related to a combination of factors such as reduction in the available space and natural food, increased cannibalism, degradation of water quality and accumulation of organic matter in the bottom of the tank (MAGUIRE and LEEDOW, 1983; PETERSON and GRIFFITH, 1999; ARNOLD *et al.*, 2006).

This study aimed to evaluate the influence of stocking densities on growth and survival of the shrimp *Litopenaeus vannamei* during the nursery phase in a biofloc system in northeastern Brazil.

MATERIAL AND METHODS

The experiment was performed in the marine shrimp farm Aquacultura Campo Novo, located in Rio Formoso, southern coast of Pernambuco, Brazil (08°39’01.01"S and 35°06' 52.31"W).

The 35 day experiment consisted of four treatments with three replicates, corresponding to the densities of 1,500, 3,000, 4,500 and 6,000 shrimp m^{-2}.

The shrimps (0.004 ± 0.01 g) were obtained as post-larvae (12 days after metamorphosis) from a commercial laboratory and randomly distributed in the experimental units. The system included twelve 300 L circular plastic tanks with a bottom area of 0.5 m^2, provided with constant aeration through the use of a radial blower (0.5 HP) and porous stones. The animals were fed twice a day (8:00 h and 15:00 h) with commercial feed (42% of crude protein), adjusted according to the feeding table proposed by JORY *et al.* (2001).

The daily rate of water recirculation in the experimental tanks was approximately 20 times their volumes. The water supply was pumped from an adjacent matrix (macrocosm) tank (200 m^2) where *L. vannamei* different batch as those used in the experiment were cultured (300 shrimp m^{-2}) in a biofloc system. This tank was partially covered to reduce light intensity and benefit the heterotrophic community. For the formation of bioflocs, the water was fertilized by the addition of wheat bran, sugar cane molasses and the feed supplied to the animals, according to the methodology described by EBELING *et al.* (2006) and AVNIMELECH (2009).

The parameters of water quality were recorded directly in the matrix tank. Water temperature, pH, salinity and dissolved oxygen were measured daily using a multiparameter analyzer (model 556 MPS, YSI Inc, USA). Whereas, ammonia (NH_3-N), nitrite (NO_2-N), nitrate (NO_3-N), alkalinity and volume of bioflocs were measured every three days. Nitrogen compounds and alkalinity were analyzed with a photocolorimeter. Imhoff cones were used to obtain the volume of bioflocs (mL L^{-1}) in the water (AVNIMELECH, 2007).

Shrimp growth was measured weekly, when 25 shrimp per treatment were sampled and individually weighed in an analytical digital balance (Marte®; precision of 0.1 mg), and then returned to their original tanks. The survival rate (S) was calculated using the formula: S% = (final number of shrimp / initial number of shrimp) x 100. The feed conversion rate (FCR) was calculated from the following formula: FCR = food offered / biomass increment. After the homogeneity of variance and normality were verified, the data were submitted to an analysis of variance (ANOVA). If significant differences were detected among the treatments, the Tukey's test was applied for mean separation. The differences were considered significant at 95%. The software Statistica 7.0 was used for all the statistical analyses.

RESULTS

The values of water quality parameters recorded during the experiment are presented in the Table 1.

Table 1. Mean (± SD), minimum and maximum values of water quality parameters recorded during the nursery phase of *Litopenaeus vannamei* in a biofloc system.

Variable	Means ± (SD)	Minimum	Maximum
Temperature (°C)	28.34 ± 0.60	26.88	29.69
pH	8.17 ± 0.33	7.68	8.69
Dissolved Oxygen (mg L^{-1})	4.38 ± 2.6	1.89	9.9
Salinity	11.36 ± 0.25	10.84	11.77
Volume of floc (ml L^{-1})	7.00 ± 0.5	6.00	8.00
Ammonia (TAN, mg L^{-1})	0.17 ± 0.08	0.08	0.24
Nitrite (NO_2-N, mg L^{-1})	0.65 ± 0.18	0.44	0.8
Nitrate (NO_3-N, mg L^{-1})	1.55 ± 0.50	1.17	2.28
Alkalinity (mg L^{-1})	148.50 ± 4.43	142.01	152

There were no significant differences in shrimp growth among stocking density treatments until the 4th week of culture (Figure 1). However, the mean weight of shrimp at a density of 6,000 m^{-2} was significantly lower than all other treatments in the 5th week (Figure 1).

At the end of the experiment, survival of shrimp was significantly higher in the treatments with 1,500 m^{-2} and 3,000 m^{-2}. Feed conversion rate values were significantly higher in the treatment 6,000 m^{-2} when compared to the other treatments (Table 2).

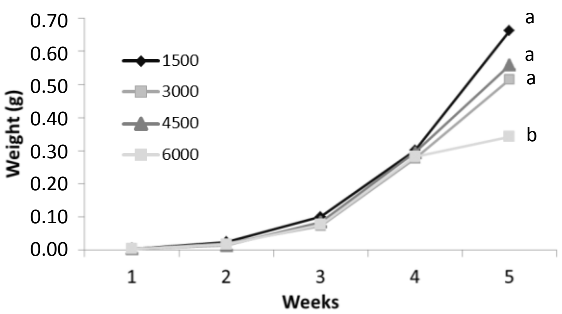

Figure 1. Growth in mean weight (g) of *Litopenaeus vannamei* reared for 5 weeks at different stocking densities (1,500, 3,000, 4,500 and 6,000 m^{-2}) in the nursery phase in a biofloc system. Different letters represent significant differences (P<0.05).

Table 2. Zootechnical performance (mean ± SD) of *Litopenaeus vannamei* cultured in different stocking densities at the nursery phase for five weeks in a biofloc system.

Density (shrimp m^{-2})	Final weight (g)	Survival (%)	FCR
1,500 m^{-2}	0.66 ± 0.07^a	95.70 ± 2.79^a	1.18 ± 0.09^a
3,000 m^{-2}	0.52 ± 0.10^a	94.90 ± 2.02^a	1.34 ± 0.29^a
4,500 m^{-2}	0.56 ± 0.12^a	54.26 ± 3.44^b	2.19 ± 0.38^a
6,000 m^{-2}	0.34 ± 0.04^b	46.26 ± 3.75^b	3.99 ± 0.61^b

Different superscript letters in the same column represent significant differences (P<0.05); FCR - Feed conversion rate.

DISCUSSION

The physico-chemical parameters of water quality showed no significant differences between treatments during the experiment. The macrocosm-microcosm recirculation system maintained the water quality parameters similar and stable for all treatments due to the high water renewal rates. This experimental design, originally proposed by MOSS and MOSS (2004), proved to be efficient in separating the effects of population stress and degradation of water quality on the performance of shrimp.

Temperature is one of the most influential factors on shrimp growth performance. According to PONCE-PALAFOX *et al.* (1997), the shrimps are relatively inactive at temperatures below 25 °C, because they reduce feed intake and consequently their growth. Therefore, the minimum values observed during the experiment stayed within acceptable levels.

MUGNIER and SOYEZ (2005) verified that dissolved oxygen concentrations below 2.8 mg L^{-1} cause hypoxia, which may adversely affect growth and survival of *L. vannamei*. In the experiment, dissolved oxygen values remained above this value. Similarly, pH value was within acceptable levels, as according to WASIELESKY *et al.* (2006), pH values below 7 decrease the growth and feed conversion rates of *L. vannamei*.

MAICÁ *et al.* (2011) recommend salinity values above 25 for *L. vannamei* juveniles reared in a zero-water-exchange super-intensive system. However the species under study is typically euryhaline and has the ability to tolerate salinity concentrations ranging from 0.5 to 40 (MCGRAW *et al.*, 2002).

The concentrations of total ammonia, nitrite, nitrate, alkalinity and volume of bioflocs in the matrix tank remained below the levels considered detrimental to the growth and survival of the species in this stage of life (VAN WYK and SCARPA, 1999; LIN and CHEN, 2001; 2003; FURTADO *et al.*, 2011).

Several studies report an inverse relationship between the growth performance of cultured penaeid shrimp and stocking densities (WILLIAMS *et al.,* 1996; WASIELESKY *et al.*, 2001; MOSS and MOSS, 2004; PEIXOTO *et al.*, 2013; WASIELESKY *et al.*, 2013). In the present study, final mean weight values were higher in the densities 1,500, 3,000 and 4,500 m^{-2}. Similar results were found by WASIELESKY *et al.* (2013) using the same stocking densities in the nursery phase of *L. vannamei* in biofloc systems in southern Brazil

The survival of shrimp was affected in the densities of 4,500 and 6,000 m^{-2} from the 4th week of cultivation, probably reflects the fall in the space viability. These results may compromise the viability of shrimp farming at these stocking densities during the nursery phase in biofloc systems, taking into consideration the high costs of postlarvae and feed during the cultivation (WASIELESKY *et al.*, 2013). However, overall shrimp survival in lower densities (1,500 and 3,000 m^{-2}) was similar to those reported (70-99%) in studies that evaluated the nursery phase of *L. vannamei* in a biofloc system with limited water exchange (SAMOCHA *et al.*, 2007; MISHRA *et al.*, 2008; WASIELESKY *et al.*, 2013). FOES *et al.* (2011) observed a decrease in survival of *Farfantepenaeus paulensis* postlarvae when reared in biofloc system at the highest density treatment (2.000m^{-2}). Furthermore, ARNOLD *et al.* (2006) argued that survival is the most important parameter to be considered in intensive production of *Penaeus monodon* during the nursery.

The low feed conversion rate in the treatment with 6,000 shrimp m^{-2} was probably a reflection of low survival rates. MISHRA *et al*. (2008) reported feed conversion rates of 1.5 for *L. vannamei* in the nursery phase in a system with water exchange. According to WASIELESKY *et al.* (2006), FCR values ranging from 1.29 to 2.49 can result in significant financial costs, considering that feed is the major cost in the production of *L. vannamei* in many countries. However, in the present study only shrimp reared in lower densities (1,500 and 3,000 m^{-2}) showed FCS values close to the lower limit proposed by these authors. The reduced growth and survival of cultured penaeid shrimp at high densities is related to a combination of factors such as reduced availability of space and natural productivity, degradation of water quality and sediment accumulation (MAGUIRE and LEEDOW, 1983; PETERSON and GRIFFITH, 1999). In this study, water quality and food availability were equally maintained in all treatments. Therefore, zootechnical performance indexes were probably a consequence of the limitated space caused by the high densities used.

CONCLUSIONS

The results of the zootechnical performance of *L. vannamei* suggest that the density of 3,000 shrimp m^{-2} is more suitable for the nursery phase in a biofloc technology system in northeastern Brazil.

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