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Performance of *Litopenaeus vannamei* postlarvae reared in indoor nursery tanks at high stocking density in clear-water versus biofloc system

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ABSTRACT

The aim of this study was to compare two recirculating culture systems, clear-water (CW) vs. biofloc (BFT) system, on the performance of *Litopenaeus vannamei* postlarvae reared in indoor nursery tanks at four stocking densities (1500, 3000, 6000, and 9000 orgs/m^3) during 42 days. In the study, the mean final growth weight fluctuated between 0.34 and 1.26 g, with a survival range of 85.0–98.4% at the four stocking densities. In both systems, survival was not affected by stocking density. The mean final weight was higher in the culture with CW (≈0.64, 0.41, 0.31, and 0.17 g/org at the stocking densities of 1500, 3000, 6000, and 9000 orgs/m^3, respectively) than in the BFT system. Specific growth rate (SGR) values (CW = 9.7–11.8 and BFT system = 8.6–10.1% weight increase/day) were higher at all treatments in the CW system. Our results indicate that in CW-recirculation system it is possible to obtain good production results in shrimp postlarvae nurseries under intensive farming condition.

1. Introduction

The nursery phase for shrimp production was first introduced in 1974 (Parker et al., 1974) and evaluation of small ponds and raceways as a nursery was conducted in the 1980s (Sturmer and Lawrence, 1987; Lawrence and Huner, 1987; Seidman and Issar, 1988; Samocha and Lawrence, 1992). Although, in the 1990s most farmers had reverted to direct stocking, at present the use of nurseries is on the rise, because a nursery phase contributes to the rapid growth of the cultured organisms (Emerenciano et al., 2012). In the nursery phase, it is possible to manage higher stocking densities and cultivate in the cooler months to reduce production costs and increase the number of crops per year in shrimp farms (Samocha et al., 2000; Yta et al., 2004; Fóes et al., 2011; Wasielesky et al., 2013).

Super-intensive production of shrimp is gaining increased attention worldwide as a potential means to improve aquaculture production via application as a transitional nursery system between the hatchery and grow-out ponds (Arnold et al., 2006b; Wasielesky et al., 2013; Correia et al., 2014). However, an increase in stocking density in the nursery phase may affect the growth and survival of shrimp due to an increase in cannibalism and degradation of water quality (Abdussamad and Thampy, 1994; Peterson and Griffith, 1999; Nga et al., 2005; Arnold et al., 2006a; Wasielesky et al., 2013).

Different studies with some penaeid species confirm that the use of nurseries in the biofloc system (BFT) contributes to the rapid growth of the cultured organisms (Fóes et al., 2011; Emerenciano et al., 2012; Wasielesky et al., 2013). The BFT is an environmental and sustainable technology used in aquaculture to maintain water quality through converting nitrogenous waste into bacterial proteinaceous biomass, after the addition of carbohydrate sources (Crab et al., 2012; Xu et al., 2013), and which is subsequently consumed by the cultivated aquatic organisms (Avnimelech, 2005).

The BFT system is formed predominantly by aerobic and heterotrophic bacteria, protozoa, metazoan, microalgae, exoskeletons, feces, and remains of dead organisms (Schryver et al., 2008). Thus, the ability of shrimp to obtain additional nutrients from biofloc has been suggested as one of the causes for the better growth of *Litopenaeus vannamei* shrimp reared in the BFT system. In contrast, in the clear-water (CW) system the development of microorganisms

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able to reintegrate to the system the generated nitrogenous waste and contribute with nutrients for the cultured organisms is limited. Although, it has been reported that, when used in intensive systems, water-recirculation may increase its viability by reducing the large release of nutrients associated with intensive cultures (Reid and Arnold, 1992; Mishra et al., 2008). These factors are essential to avoid fast water quality degradation in a CW system, because in the intensive culture without recirculation or water exchange, the organisms' feces, food waste, dead microorganisms, toxic metabolites, such as ammonia and nitrite, can accelerate water quality degradation in a short time, up to the limit of killing the reared organisms, even more so when stocking densities are high.

Hence, it is important to know whether a high stocking density in the culture with CW-recirculation produces similar results to those reported with the BFT system, because a BFT system involves a high microorganism's concentration and a poor management strategy (Ray et al., 2011); besides, failure in water oxygenation (Vinatea et al., 2009) could impact the shrimp postlarvae performance in the nursery. Tacon et al. (2002), Izquierdo et al. (2006), and Moss et al. (2006) reported that shrimp reared with CW have a lower growth rate than those reared in water with high total suspended solids (TSS) concentration, but Emerenciano et al. (2007, 2013) observed the contrary. Therefore, the present work was aimed at investigating the performance of *L. vannamei* postlarvae reared in indoor nursery tanks at four stocking densities in two recirculating culture systems, CW vs. BFT. The study was conducted simultaneously in both systems.

2. Material and methods

2.1. Experimental design

The experiments were performed in the indoor tank facilities of the Marine Aquaculture Station (EMA), of the Federal University of Rio Grande, Southern Brazil (32°12′S, 52°10′W). The biological material used in this study was acquired from the Aquatec Ltd. laboratory (Canguaretama, Rio Grande do Norte, Brazil). After the *L. vannamei* nauplii arrived at the EMA facilities, they were kept in the shrimp hatchery until reaching the post-larval stage (PL′25). The shrimp postlarvae (mean initial weight = 0.009 ± 0.002 g) were transferred to the experimental tanks, where the experimental design was randomized with four treatments (four stocking densities) per experimental system: culture with CW-recirculation: 1500 (T_{CW1500}), 3000 (T_{CW3000}), 6000 (T_{CW6000}), and 9000 (T_{CW9000}) orgs/m^3; and culture with BFT: 1500 ($T_{BFT1500}$), 3000 ($T_{BFT3000}$), 6000 ($T_{BFT6000}$), and 9000 ($T_{BFT9000}$) orgs/m^3. Three replicates were randomly assigned to each stocking density. Shrimp were fed twice a day (≈0800 and 1600 h) with commercial feed (40% protein). Initially, the feeding rate was established according to Jory et al. (2001), and posteriorly the feed was adjusted daily according to their consumption. The study lasted 42 days.

2.2. Shrimp culture systems

To evaluate the effects on *L. vannamei* postlarvae of stress caused by population density in the cultures with CW-recirculation or BFT system, two similar systems of indoor nursery tanks were designed (Fig. 1). These recirculating systems were used to maintain the same water quality in all experimental units, to observe only the effects of stocking density in the nursery phase. Each system included twelve 0.15-m^3 circular tanks (microcosms) with a bottom area of 0.5-m^2 diameter, supplied by intense individual aeration with air stones. Each experimental tank was associated with water input pumped from a matrix tank (4 m^3 each), referred to as macrocosm. In each system, a submersible pump distributed water to the tanks, and this water was returned via gravity to a drain directed to the macrocosm tanks. In both systems, the water was completely recirculated from the macrocosm to the microcosm ≈20 times each day (flow rate ≈ 2.1 L/min/tank).

The systems were filled with seawater filtered through a sand filter, and posteriorly through a 5-μm-pore cartridge. Before starting the study, the matrix tank of CW-recirculation was supplied with a biological filter located inside the tank (Fig. 1), consisting of two 100-L tanks filled to 40% with Bio-Balls each (Coralife®, WI, USA) and intense aeration, where the water was recirculated by pumping during the whole experiment. The matrix tank of the BFT system received an inoculum of 50% of its total volume, being this water from a grow-out tank that showed a mature biofloc. The matrix tank of BFT was stocked with 34 shrimp *L. vannamei*/m^3 (mean body weight = 14.5 ± 0.41 g) to strengthen the maintenance and formation of bioflocs. In both cases, there was no water renewal during the study, only replacement of what was lost due to evaporation by adding dechlorinated freshwater. During experimentation with the BFT system, the use of sugar cane molasses as a source of organic carbon was not necessary because the levels of total ammonia did not reach 1 mg/L (Avnimelech, 1999).

2.3. Water quality parameters analyses

The photoperiod for the experimental room was 12/12 h light/dark cycle, with a 200 lx intensity at the surface of the water provided by artificial lighting. The water temperature was maintained with two heaters immersed in each of the matrix tanks. During the study, the physicochemical parameters were monitored in both matrix and experimental tanks. The dissolved oxygen (DO; mg/L), temperature (°C), pH, and salinity (g/L) were monitored twice a day (≈0800 and 1600 h) utilizing a multiparameter analyzer (model 556 MPS, YSI Inc., Yellow Springs, OH, USA). The concentrations of total ammonia-N (mg/L), nitrite-N (mg/L), nitrate-N (mg/L), and orthophosphate-P (mg/L) were measured weekly, according to the methods recommended by UNESCO (1983). Total suspended solids (TSS, mg/L) and alkalinity (mg $CaCO_3$/L) were determined once per week through gravimetry by filtering aliquots of 20 mL of water through GF 50-A glass fiber filters (Strickland and Parsons, 1972), and following the methodology proposed by APHA (American Public Health Association, 1998), respectively.

2.4. Productive shrimp performance

During the study, biometrics was performed weekly, weighing 50 shrimp from each experimental tank individually using a digital balance (precision 0.001 g, Sartorius®). The shrimp were returned to their original tanks after weighing.

At the study end, all the shrimp that survived in each experimental tank were weighed and counted to evaluate their growth [final weight, specific growth rate (SGR)], survival, feed conversion ratio (FCR), and final density per treatment. The SGR (% weight increase/day) was calculated using the formula reported by Ricker (1979), and the FCR with the formula reported by Hari et al. (2004). Survival (%) data were transformed (arcsine of the square root) before their analysis (Zar, 1996).

2.5. Statistical analyses

Once the homoscedasticity and normality of the data were verified, the shrimp biological performance data at the different stocking densities, within each system (CW-recirculation or BFT system), were analyzed with one-way analysis of variance