Aquacultural Engineering 45 (2011) 127–136

Contents lists available at SciVerse ScienceDirect

Aquacultural Engineering

journal homepage: www.elsevier.com/locate/aqua-online

Water quality dynamics and shrimp (*Litopenaeus vannamei*) production in intensive, mesohaline culture systems with two levels of biofloc management

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ARTICLE INFO

Article history:
Received 13 June 2011
Accepted 3 September 2011

Keywords:
Biofloc
Sustainable aquaculture
Marine shrimp
Minimal exchange
Solids management

ABSTRACT

A dense microbial community develops in the water column of intensive, minimal-exchange production systems and is responsible for nutrient cycling. A portion of the microbial community is associated with biofloc particles, and some control over the concentration of these particles has been shown to provide production benefits. To help refine the required degree of control, this study evaluated the effects of two levels of biofloc management on water quality and shrimp (*Litopenaeus vannamei*) production in commercial-scale culture systems. Eight, 50 m^3 raceways were randomly assigned to one of two treatments: T-LS (treatment-low solids) and T-HS (treatment-high solids), each with four replicate raceways. Settling chambers adjacent to the T-LS raceways had a volume of 1700 L with a flow rate of 20 L min^{-1}. The T-HS raceways had 760 L settling chambers with a flow rate of 10 L min^{-1}. Raceways were stocked with 250 shrimp m^{-3}, with a mean individual weight of 0.72 g, and shrimp were grown for thirteen weeks. Raceways in the T-LS treatment had significantly reduced total suspended solids, volatile suspended solids, and turbidity compared to the T-HS treatment ($P \leq 0.003$). The T-LS raceways also had significantly lower nitrite and nitrate concentrations, and the T-HS raceways had significantly lower ammonia and phosphate concentrations ($P \leq 0.021$). With the exception of nitrate, there were no significant differences between the change in concentration of water quality parameters entering and exiting the settling chambers in the T-LS versus the T-HS treatment. Nitrate never accumulated appreciably in the T-LS raceways, possibly due to denitrification in the settling chambers, bacterial substrate limitations in the raceways, or algal nitrate assimilation. However, in the T-HS raceways nitrate did accumulate. The T-HS settling chambers returned a significantly lower nitrate concentration and significantly greater alkalinity concentration than what entered them ($P \leq 0.005$), indicating that denitrification may have occurred in those chambers. There were no significant differences in shrimp survival, feed conversion ratio, or final biomass between the two treatments. However, shrimp in the T-LS treatment grew at a significantly greater rate (1.7 g wk^{-1} vs. 1.3 g wk^{-1}) and reached a significantly greater final weight (22.1 g vs. 17.8 g) than shrimp in the T-HS treatment ($P \leq 0.020$). The results of this study demonstrate engineering and management decisions that can have important implications for both water quality and shrimp production in intensive, minimal-exchange culture systems.

1. Introduction

Intensive, minimal-exchange shrimp culture systems have little, if any, water exchange and high animal stocking densities (Ray et al., 2009). Decreased water exchange reduces pollutant discharge, disease exchange between wild and captive stocks, and introductions of exotic species to the wild. With little water exchange and the tolerance of *Litopenaeus vannamei* to low and moderate salinities, these systems can be sited at inland locations, preserving coastal ecosystems and offering fresh marine shrimp to areas that otherwise could not access such a commodity (Browdy and Moss, 2005).

High animal stocking densities reduce the footprint of culture systems, but also necessitate large nutrient inputs. These nutrients lead to eutrophication within the systems and, in response, a dense microbial community develops in the water column, much of which is contained on and within biofloc particles (Avnimelech, 2009; Ray et al., 2010b). The microbial community in intensive, minimal-exchange culture systems is responsible for cycling nutrients, most importantly nitrogen compounds. Feed decomposition and animal excretions contribute to ammonia, which is toxic to shrimp. Algae and heterotrophic bacteria can directly assimilate ammonia to build cellular proteins, and nitrifying bacteria can oxidize ammonia to form nitrite and nitrate (Ebeling et al., 2006). Each of these three groups contribute to detoxifying nitrogenous waste, but each has drawbacks: algae are limited in the amount of nitrogen they can remediate (Brune et al., 2003), heterotrophic

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doi:10.1016/j.aquaeng.2011.09.001

bacteria require substantial amounts of oxygen to assimilate ammonia (Browdy et al., in press), and nitrifying bacteria can be slow to establish, resulting in spikes of toxic ammonia and nitrite (Ray et al., 2009).

To stimulate the rapid uptake of ammonia by heterotrophic bacteria, labile organic carbon sources such as sucrose can be added to the culture water (Avnimelech, 2009; Crab et al., 2007; De Schryver et al., 2008). A carbon:nitrogen ratio (C:N) of system inputs (feed and carbohydrates) above approximately 10 should result in efficient ammonia assimilation (Avnimelech, 1999; Ebeling et al., 2006). To effectively assimilate ammonia, these bacteria must expand in biomass; however, the nitrogen they assimilate is not taken out of the system unless the bacteria are removed.

The microbial community not only detoxifies nutrients, but can recycle those nutrients and provide benefits for animal growth and feed conversion ratios (FCR) (Ju et al., 2009; Moss, 1995; Wasielesky et al., 2006). Although there are clear benefits to having an in situ microbial community, some control over these organisms and the biofloc particles they are associated with may be necessary. Using 6200-L outdoor tanks, half with simple settling chambers and half without, Ray et al. (2010a) demonstrated that managing biofloc concentration could significantly improve shrimp growth rate, FCR, and biomass production. Also, the authors showed that settling chambers contributed to significantly decreased nitrate and phosphate concentrations and significantly increased alkalinity concentration in the shrimp culture systems.

The purpose of the current project was to help refine optimal biofloc concentration and evaluate simple management and engineering considerations for regulating that concentration to achieve advantageous water quality dynamics and shrimp production in commercial-scale systems. A detailed analysis of the effects that settling chambers can have on important water quality parameters is provided. Mesohaline conditions were used to facilitate the sustainability and potential inland development of intensive minimal-exchange systems.

2. Materials and Methods

2.1. Experimental setting

This project was conducted at the University of Southern Mississippi's Thad Cochran Marine Aquaculture Center (CMAC), a part of the Gulf Coast Research Laboratory, located in Ocean Springs, Mississippi, USA. At the CMAC is a commercially scaled minimal-exchange, intensive shrimp culture facility which was described by Ogle et al. (2006). Briefly, it consists of twelve, 3.2 m × 30.1 m, rectangular, cement block, high density polyethylene (HDPE)-lined raceways, eleven of which were used for this project, including those used during the nursery phase. The raceways are covered by six dome-shaped greenhouse structures covered in clear plastic sheeting (two raceways per greenhouse structure), each connected to a central, wood-frame structure that houses a harvest basin. Each raceway has a dirt floor beneath the liner, which is gently sloped toward the harvest basin.

2.2. Shrimp source, nursery, and feeds

Litopenaeus vannamei postlarvae (PL 12) were obtained from Shrimp Improvement Systems, LLC (Islamorada, Florida, USA). These shrimp were stocked into three of the above mentioned raceways at a density of 2986 shrimp m^{-3} to begin a nursery phase. The nursery raceways were maintained at a volume of 60 m^3 and a salinity of between 19 and 24 g L^{-1} with no water exchange. The water used for the nursery phase had been used the previous year for culturing shrimp, but solids were settled from the water and it was passed continuously through a foam fractionator at a flow rate of approximately 150 L min^{-1} for one month prior to use.

Each nursery received blown air from a 746 W regenerative blower (Sweetwater®, Aquatic Ecosystems Inc., Apopka, Florida, USA) delivered through thirty-six, 15.2 cm long ceramic air diffusers. Shrimp were fed PL Raceway Plus #1 between stages PL 12 and PL 18, and PL Raceway Plus #2 between stages PL 19 and PL 30 (Zeigler™ Brothers Inc., Gardners, Pennsylvania, USA). Both of these feeds were guaranteed by the manufacturer to provide a minimum of 50% protein and 15% fat, and a maximum of 1% fiber, 12% moisture, and 7.5% ash. Shrimp were then fed Zeigler™ Hyperintensive-35 for the remainder of the nursery phase and throughout the duration of this project. The Hyperintensive feed was analyzed by the Clemson University's Agricultural Services Laboratory (Clemson, South Carolina, USA) and found to contain 33.4% crude protein, 10.4% fat, 8.6% moisture, and 6.6% ash.

During the nursery phase, dissolved total ammonia nitrogen (TAN) and nitrite–nitrogen (NO_2–N) concentrations were monitored. TAN was assessed using Hach method 8155 (Hach Company, 2003) and NO_2–N was measured using the spectrophotometric procedure outlined by Strickland and Parsons (1972). Absorbance was measured at 655 nm for TAN and 543 nm for NO_2–N using a Hach DR 3800 spectrophotometer (Hach Company, Loveland, Colorado, USA). Shrimp were cultured in the nursery raceways for 39 days and then stocked into the experimental raceways. In response to NO_2–N concentrations above 2 mg L^{-1} during the nursery, sucrose was added to stimulate nitrogen assimilation by heterotrophic bacteria.

2.3. Experimental systems

Eight of the raceways described in Section 2.1 were used for this experiment with the following modifications. Each experimental system had a central wall of plastic sheeting suspended between two pieces of PVC pipe. The top pipe was suspended with ropes from the greenhouse structure and the bottom pipe was weighted internally with rubber-coated iron bars and water. Water was propelled around the central wall of each raceway using a combination of four airlift mechanisms and a 560 W water pump delivering water to eighteen, 1.3 cm diameter Venturi nozzles (Turbo-Venturi®, Kent Marine, Franklin, Wisconsin, USA) throughout the raceway (Fig. 1) and to the settling chambers. The airlift mechanisms consisted of three, 15.2 cm long ceramic air diffusers oriented parallel to the water flow and receiving air from a blower described in Section 2.2. The airlifts were constructed of a 2.5 cm diameter PVC frame which held the diffusers approximately 6 cm above the raceway floor. Above the diffusers was a sheet of EPDM rubber held by the PVC frame and oriented at an approximately 35° angle relative to the water movement. Air from the diffusers traveled vertically and contacted the EPDM, which served as a deflector to project the air, and the water traveling with it, horizontally forward.

The Venturi nozzles were located near the bottom of each raceway, each connected to a 1.3 cm diameter vertical pipe that was connected to a 5 cm diameter pipe that circumvented the raceway. Each venturi had tubing attached to the gas injection point which then attached to another pipe, 2.5 cm in diameter that circumvented the raceway. The pipe had two valves to allow ambient air to be drawn in and a point where pure oxygen gas could be injected; this allowed air, pure oxygen, or a combination of the two to be injected into the raceway water through the Venturi nozzles.

2.4. Experimental design

The eight raceways used for this experiment were each randomly assigned to one of two treatments, each treatment containing four replicate raceways. The low solids treatment (T-LS)