"Farming Marine Shrimp in Freshwater Systems: An Economic Development Strategy for Florida: Final Report"

FDACS Contract #4520

Principle: Investigator: Peter M. Van Wyk Harbor Branch Oceanographic Institution 5600 Highway U.S. 1 North Ft. Pierce, Florida 34946

Introduction:

In recent years there has been renewed interest in shrimp culture here in Florida due to technological developments that now make it possible to culture *Litopenaeus vannamei* indoors in near-freshwater recirculating aquaculture systems. Harbor Branch and others have demonstrated in recent years that *L. vannamei* can be successfully produced in water with chloride concentrations as low as 300 ppm.

Water with chloride levels this low is generally classified as freshwater and can be used to irrigate most crops. The significance of this is that shrimp production can now be practiced on cheaper, non-coastal agricultural land. New advances in the technology for producing L. vannamei indoors in high-density recirculating aquaculture systems now allows for yearround production of this species even in temperate climates with relatively cold winters. Year-round production improves the economic potential of an enterprise in several ways. The annual revenues of the operation are increased because year-round production increases annual productivity. Continuous harvesting facilitates direct marketing to retail markets, which may allow for a higher price to be received for the product. Producing shrimp indoors in recirculating systems benefits the producer by significantly reducing the risk of exposing the shrimp to the viral diseases that have wreaked havoc in open coastal ponds throughout the world. In the wake of devastating epidemics of Taura Syndrome Virus (TSV) and White Spot Syndrome Virus (WSSV), some shrimp farm managers in Latin America are considering switching to intenstive tank-based production systems because of the additional biosecurity these systems can provide. Indoor production systems provide the additional benefit of reducing crop loss due to predation. In addition, these systems significantly reduce the risk of accidental release of non-native shrimp into Florida's coastal waters.

The objective of the current study was to demonstrate the production technology required to successfully cultivate the marine shrimp, *Litopenaeus vannamei*, in freshwater recirculating aquaculture systems, and to evaluate the economic potential of this approach to shrimp culture.

Production Systems:

Two different production systems were evaluated in this study: 1) a single-phase (direct stock) production system, and 2) a three-phase, partitioned production system. In a singlephase production system shrimp postlarvae are stocked into a culture tank and remain in that same tank until final harvest. The stocking density in the culture tank is based on the desired final harvest density plus overstock to compensate for expected mortalities. Initially, the system biomass is extremely low relative to the carrying capacity of the system, which is In a three-phase production system, the only reached at the end of the production cycle. production process is divided into three distinct phases, each carried out in a different culture tank, or in different sections of a partitioned culture tank. The shrimp typically spend onethird of the total culture period in each of the three sections of the tank. Postlarval shrimp are initially stocked into a small nursery tank, representing 10-13% of the total culture area of the complete three-phase system. At the end of the nursery period (after 50-60 days) the juvenile shrimp are transferred to the second section of the tank, called the intermediate growout section. This section is larger than the nursery section, representing about 27-30% of the total culture area. The shrimp remain in the intermediate growout section for another 50-60 days before being transferred to the final growout section, which occupies 60% of the total culture area. After another 50-60 day period the shrimp are harvested for market.

The objective of a three-phase production system is to utilize the available production area more efficiently by operating closer to the carrying capacity of the system for a greater percentage of the culture period. In a single-phase system the biomass is very low relative to the carrying capacity of the tank for the first two-thirds of the culture cycle. The number of postlarvae stocked into the nursery section is determined by the projected harvest density of the final growout section, with overstocking to account for expected mortalities. The amount of area devoted to each phase is calculated to allow the shrimp to continue to grow until they reach the end of that phase. When the shrimp are ready to be transferred to the next section they should be approaching the carrying capacity for the section they are in.

The three-phase system permits higher production levels than can be achieved in a singlephase system. Each section of a three-phase system is stocked at a density which will grow to the carrying capacity for the alloted area in one-third the amount of time it takes the shrimp in a single-phase system to reach the carrying capacity. Tank space is used more efficiently than in a single-phase system in which culture tanks are maintained at low densities throughout the early part of the growout cycle. The production of a three-phase system should, theoretically, be 1.8 times greater than the production in a single-phase assuming survival and growth rates are equivalent between the two systems. system. Although the area harvested for each crop is only 60% of the area harvested in a single-phase system, the final growout section of the 3-phase system is harvested three times for every harvest of the single-phase system. Increasing the harvest frequency has obvious advantages from a marketing standpoint, and may also smooth out the cash flow for the business. The potential disadvantages of a three-phase production system are the increased risk of mortality during the transfer process and potential density-dependent reduction in shrimp growth rates and survivals.

Greenhouse Recirculating Aquaculture Systems:

The objective of the Harbor Branch Oceanographic Institution (HBOI) shrimp culture program has been to develop a cost-effective indoor, freshwater production system based on a recirculating water treatment system. The principle that guided HBOI in the development of new system designs is: "Keep It Simple". The ideal system should be simple to build using inexpensive, readily available materials, and should be operable by individuals with limited training specific to systems operation. With this in mind, HBOI focused its efforts on designing an inexpensive system capable of growing shrimp at moderately high densities ($>600 \text{ shrimp/m}^2$) that have been reported for *L. vannamei* in more sophisticated recirculating systems (Davis and Arnold, 1998).

The shrimp production systems utilized in this project represent two generations of system design. First generation systems at HBOI (System A) feature above-ground raceways and sand filters. Given their simplicity, these systems perform surprisingly well, supporting loading rates of up to 2.25 kg shrimp/m³. However, sand filters are expensive to operate because they require inefficient, high-head pumps to push the water through the compacted sand filter media and because sand filter maintenance is very labor intensive. The cost of operating the pumps on these systems can be quite high because they operate on a continuous basis. The second generation systems at HBOI (System B) feature in-ground raceways and low-head water treatment systems. The in-ground raceway should be less prone to heat-loss, reducing heating costs in the winter. Pumping costs in System B are lower because the low-head system design cuts the horsepower requirements by more than half.

A second key objective of this study was to compare the productivity and economics of these two types of recirculation systems.

System Descriptions:

System A

System A is housed in a 30' x 152' Quonset-style greenhouse. The greenhouse consists of a series of arches or bows made of 2" diameter galvanized steel pipe. The bows are anchored in concrete at their bases. The arches are supported by purlins running the length of the greenhouse connected by clamps to each rib. Cross-struts span every second arch providing additional support. A double layer of 6-mil clear UV-resistant polyethylene plastic material covers the greenhouse. The space between the two layers of plastic is inflated by means of a small blower. The two layers of plastic are highly efficient at collecting and retaining solar heat. The dead air space between the layers of plastic functions as an insulating layer. During the night, greenhouses with a single layer covering lose much of the heat collected during the daytime. Nighttime heat loss is greatly reduced when a double layer of plastic is used to cover a greenhouse. The improved heat retention justifies the added expense of the second layer of plastic and the inflation system. During the summer months an 80% shade cloth covers the outside of the greenhouse. The shade cloth minimizes algal growth within the raceways. The greenhouse is ventilated by two 1.5 hp extractor fans and by one 0.5-hp

extractor fan mounted on one end of the greenhouse. The ventilating air enters at the greenhouse at the opposite end through two mechanical louver windows. The fans and the louvers are thermostatically activated, providing a measure of automated temperature control to the greenhouse.

The System A greenhouse contains four culture tanks, each operating on separate filter systems (Figure 1). Two of the culture tanks (H5-NE and H5-SE) are set up as single-phase culture systems and two tanks (H5-NW and H5-SW) are set up as three phase systems . H5-NE (single-phase), and H5-NW (three-phase) both measure 13.5' x 56'., while (H5-SE and H5-SE (single-phase) and H5-NW (three-phase) both measure 13.5' x 64'. The three-phase systems are subdivided into three sections. The nursery section of H5-NW (H5-NW1) measures 13.5' x 6.5', with the long axis of the raceway perpendicular to the long axis of the overall growout area. The intermediate growout section (H5-NW2) measures 13.5' x 13.5'. The final growout section (H5-NW3) measures 13.5' x 36'. The nursery section of H5-SW (H5-SW1) measures 13.5' x 13.5' x 13.5'. The final growout section (H5-NW3) measures 13.5' x 38.5'. Four-inch diameter bulkhead fittings positioned at the bottom of the walls dividing the three sections all shrimp to be transferred from one section to the next without being handled.

The culture tanks consist of a wooden frame supporting a black 30-mil high-density polyethylene liner. The wooden frame is two board widths high and is built using 2"x12" boards of pressure-treated lumber supported by galvanized pipe set vertically in a concrete anchor. The vertical pipe supports are set on 4-ft centers. The arches forming the frame of the greenhouse support the outside walls of the culture tanks.

The culture tanks are rectangular in shape and have been set up with a "racetrack" configuration. The racetrack configuration is essentially a hybrid between a circular tank and rectangular tank. Each culture tank is set up with two drain outlets at either end of the tank, centered between the end wall and the sides of the tank. A center divider baffle has been positioned between the two drain outlets, and functions to separate water flowing down one side of the "racetrack" from the water flowing down the opposite side. The water in the tank flows in an elongated oval pattern, travelling down one side of the tank, circling around the drain outlet at one end, then travelling up the other side of the raceway and circling around the opposite drain outlet. Baffles have been placed in the corners of the tank to prevent



Figure 1: Culture tank layout in System A. Upper two culture tanks are single phase systems. Lower tanks are three-phase systems.

eddies from developing in the corners. The baffles help create a semi-circular flow pattern at each end of the tank so that the water pivots about the drain outlets. This flow pattern generates centrifugal forces as the water circles the drain, concentrating the suspended solid wastes in the area around the drains. Water is introduced into the tank at the head of the straight runs. The incoming water mixes with the water circling the racetrack, creating relatively uniform water quality throughout the tank. The water enters the tank through spray bars spanning the width of the straight run of the tank.

All drain outlets are 4 inches in diameter, with the exception of those in the nursery tank, which are 2 inches in diameter. All drain outlets consist of bulkhead fittings which pass through the liner and feed into a common 4-inch central drainage pipe. A PVC standpipe is set in each drain outlet. The height of the standpipe sets the minimum water level in the tank. The top of the standpipe is fitted with a cylindrical screen extending to 6 inches above the maximum water level in the tank. The purpose of this screen is to exclude shrimp from the drain outlets. An outer sleeve is placed over the standpipe to allow the water flowing out of the drain outlet to be drawn from the bottom of the tank. The outer sleeve consists of a PVC pipe with a slightly larger diameter than that used for the standpipe. The pipe is scalloped or screened at the bottom to allow bottom water to pass through it. Water passing through the drain outlets empties into a 4-inch diameter central drainage pipe. The central drainage pipe discharges into a 4' x 3' x 4' polyethylene sump located on the outside of the tank at one end of the raceway. The sump serves as a settling basin and pump well.

A 2-hp centrifugal pool pump circulates water through the system. The intake for the pump is located near the bottom of the sump and is fitted with a check valve to prevent the pump from losing its prime when it is turned off. A 36-inch diameter high-rate downflow sand filter serves as both the solids filter and the biofilter for the system. The sand filters are loaded with 500 lbs of Number 20 silica sand.

A 2.5-hp regenerative blower supplies air to the system. Each culture tank is provided with forty 1" x 3" medium pore diffusers. Each diffuser supplies approximately 0.3 standard cubic feet per minute (scfm) of air to the raceway, providing each culture tank with a total of 12.0 scfm of air. The airstones are distributed at 3-foot intervals along the sidewalls of the culture tanks. A beltdrive blower powered by a 9-hp diesel motor serves as an emergency backup. The backup blower has a pressure-actuated switch that starts the blower motor whenever the pressure in the air system drops to zero.

System B:

System B represents a second generation in HBOI shrimp production system design. The design objectives for this system were to:

- 1) reduce the cost of the greenhouse structure
- 2) reduce the construction costs for the culture tanks
- 3) make the systems more energy efficient
- 4) reduce the labor required to maintain the systems
- 5) increase the carrying capacity of the systems, while keeping system costs down
- 6) provide for consistent circulation of water throughout the system.

The System B culture tanks are housed in two 30' x 96' Quonset-style greenhouses . These greenhouses are similar to the System A greenhouses described above, but are less expensive. The System A greenhouses are rated to be able to withstand winds of up to 120 mph, while the System B greenhouses are only rated for winds of up to 80 mph. During the summer months a 95% shade cloth was placed on the outside of the greenhouse. This provided significantly more shade than the System A shade cloth, which provided only 80% shading. The greenhouse ventilation system consists of two 42-inch x 3/4-hp exhaust fans and two 51-inch shuttered windows. A single thermostat controls both the windows and the exhaust fans. An 8' x 8' sliding door is located at one end of the greenhouse. This door allows large pieces of equipment or harvest boxes to be easily moved into the greenhouse.

The culture tanks in System B are similar to those in System A, except that they are partially excavated below ground level. Instead of having the floor of the culture at ground level and the tank depth determined by the height of the wooden frame, the floor of the System B culture tanks is excavated to a depth of 18-inches below grade. A wooden frame surrounds the perimeter of the excavated area, adding an additional 12-inches to the depth of the raceway. The wooden frame is similar to the frame used to create the System A culture tanks. A berm with a 1:1 slope extends from the bottom of the wooden frame down to the floor of the tank. The overall tank depth, when filled with water is 24-inches, or 6-inches deeper than the System A tanks. The culture tank is lined with the same 30-mil high density polyethylene liner material as is used in System A. There are several advantages to this approach to raceway construction. Because much of the volume of the raceway is below ground level there should be less heat loss from the raceways during the winter. The raceways can be made slightly deeper without appreciably increasing the cost of construction. A deeper tank will sustain a higher biomass, and will also have more stable temperature and water quality characteristics.

Each of the System B greenhouses is occupied by two culture tanks, each with its own water treatment system. The culture tanks lie side by side in the greenhouse, sharing a common central wall. The 3-foot wide walkway between the tanks has been replaced with a 1-foot wide catwalk above the tanks. In this configuration approximately 90% of the available area in the greenhouse is under cultivation, compared to about 80% in System A. Reducing the number of systems per greenhouse from four to two reduces the labor requirement by half,



Figure 2: System B Single-Phase Raceways with Axial Flow Pump

without sacrificing any production.

One greenhouse in System B (Figure 2) contains two single-phase shrimp production systems (J1 and J2). Each single-phase culture tank measures 14.5' x 88'. Except for the fact that they are in-ground culture tanks, most of the details of their construction are essentially the same as in System A. The tracks are configured in "racetrack" configuration with a central baffle and corner baffles.

The second greenhouse in System B (Figure 3) contains two three-phase shrimp production systems (J3 and J4). The culture tanks in the three-phase system are layed out like the single-phase culture tanks, except that they are separated into three discrete sections by divider walls. The nursery sections measures 10' x 14.5' (11% of the culture area). The intermediate growout section measures 14.5' x 27' (31% of the culture area) and the final growout section measures 14.5' x 51' (58% of the culture area). Four-inch diameter bulkhead fittings positioned at the bottom of the walls dividing the three sections all shrimp to be transferred from one section to the next without being handled.

One of the design objectives was to build a system that was more energy efficient than our sand filter-based systems. Towards this end it was decided to incorporate into the design a low-head filtration system that flowed by gravity through the solids filter and biofilter.

An upflow bead filter is used in System B to filter out solid wastes as well as for biofiltration. The upflow bead filter consists of cylindro-conical sump (4' diameter x 4' deep, 1,200-liter capacity), filled with 16 ft^3 of biofilter beads. The beads are polyethylene cylinders 7 mm long by 10 mm in diameter with radiating fins that provide additional surface area. These beads are positively buoyant. The tank is plumbed so that the raw water from the culture tank enters the filter tank through a 4-inch bulkhead fitting cut into the conical portion of the tank. A second bulkhead fitting is cut into the sidewall about 12-inches below the culture tank water level and connects the solids filter to the biofilter tank. A 4-inch pipe with 5-mm slots cut into its upper surface is inserted into upper bulkhead fitting on the inside of the tank. This pipe collects filtered water from near the surface of the water and allows it to pass into



Figure 3: System B 3-Phase Production Systems, powered by a 3/4 hp centrifugal pump.

the biofilter tank. The water must take a tortuous pass through the filter bed before it reaches the collecting pipe at the top of the water column. In the process settleable solids and larger suspended solids are trapped on the sticky surfaces of the beads. The solids are flushed from the system once a day by plugging the raw water inlet and the filtered water and removing a 2-inch standpipe from the drain outlet in the bottom of the cone. A 6-inch diameter outer standpipe keeps the beads from beads from draining out of the system. Windows covered by a 1/4" mesh screen are located near the bottom of the outer standpipe and allow water and solid wastes to pass through the outer standpipe and out of the tank when the central standpipe is removed.

An aerated, submerged biofilter receives water after it flows out of the low-head bead filter. The biofilter uses the same beads as are used in the solids filter, but in this application the beads are tumbled by air bubbles introduced into the bottom of the filter bed through a grid of 10 medium pore airstones. The beads are contained within a $3.5' \times 5' \times 4'$ cage made of 1" Schedule 40 PVC pipe and 1/4" square-mesh polyethylene mesh screen. The cage serves to contain the beads so that they do not get sucked into the pump or go out down the drain. The cage sits in a rectangular, polyethylene sump (4' x 6' x 4'), two inches above the bottom of the sump.

The biofilter sump doubles as a pump reservoir for the main system pump. The amount of head required to return the water to the culture tanks is minimal since there are no filter components between the pump and the culture tanks, and the elevation head that must be overcome is less than 12-inches. Two different types of low-head pumps are being used in System B. A 1/4-hp axial flow pump (designed and built by Harbor Branch personnel) performs the pumping duties in the single-phase culture systems. This pump utilizes a plastic propeller as an impeller. The pump column is made of 4-inch PVC pipe. A tee halfway up the column directs the flow out of the pump and into the culture tank. The pump inserts into a bulkhead fitting that passes through the wall of the sump and the culture tank just below the tank water surface. There is essentially zero head pressure. These axial flow pumps are capable of moving large volumes of water with very little energy expenditure. The pump discharges 160 gpm of water in this application.

Despite the high discharge volume of these axial flow pumps, the water is discharged with very little pressure or velocity. As a result, the return flow does not generate a great deal of circulation within the culture tank. Nor does the return flow provide any additional aeration or degassing.

The axial flow pump could not be used in the three-phase systems because the return flow had to be piped to the opposite end of the greenhouse to the nursery and intermediate growout sections. The discharge out of these axial flow pumps drops off rapidly as head pressure increases. At only 18 inches of head the pump discharge is less than one-quarter of the discharge at zero feet of head. For this reason a centrifugal pump was used with the three-phase production systems. The pump selected was a low-head, high-efficiency 3/4-hp centrifugal pump. This pump will push 150 gpm of water against 10 feet of head and is much more efficient than the high-head 2-hp pool pump used in System A. The System A pump will only pump 100 gpm at this head and requires more than twice the horsepower.

Using a centrifugal pump in the three-phase systems permits the return flow to be introduced through spray bars, which span the raceways. Each spray bar consists of a 2" diameter PVC pipe drilled with 1/4" diameter orifices. The momentum of the water passing at high velocity out of these orifices is transferred to the mass of water circulating in the tank. We observed that, although the discharge of the centrifugal pumps is slightly less than that of the axial flow pumps, the velocity of water flow rate in the three-phase culture tanks is much higher. This is because the water enters the culture tank at a high velocity and its momentum sets the entire mass of water in the tank moving. This is very important because it prevents the solid wastes from settling out and accumulating on the floor of the culture tank. Another important benefit derived from spray bars is that the water is aerated as it enters the tank and excess carbon dioxide is de-gassed as the water passes out of the spray bar. Whichever pumping system is used, configuring the system so that there are no filter components between the pump and the culture tank guarantees that the flow rates through the system are constant over time. This is in sharp contrast to System A, where flow rates declined by 50-75% between sand filter backwashes.

A single 2.5-h.p regenerative blower supplies the air supply for the four culture tanks in System B. This blower supplies approximately 100 scfm of air against a head pressure of 50-inches of water. Each system is supplied with 25 scfm of air, which is delivered through submerged 3"x1" medium pore diffusers. A total of 44 diffusers are positioned in the culture tanks at 4-ft intervals on either side of the central baffle. Ten additional airstones are set into an air manifold at the bottom of the biofilter cage and serve to aerate and tumble the biofilter media.

A belt-drive blower powered by a 9-hp diesel engine provides emergency backup aeration. This blower is twice as large as it needs to be, delivering 200 scfm of air at 50-inches of water, but was sized to accommodate future expansions. A pressure switch turns the diesel engine on whenever it detects a loss of air pressure in the air system.

Freshwater and seawater are both supplied by wells. Wellwater is a desirable water source because it virtually free from bacterial, viral, or parasitic pathogens. The wellwater does, however, have some undesirable chemical characteristics. Like a lot of wellwater, HBOI's wellwater is high in hydrogen sulfide, carbon dioxide, and ammonia, and is low in oxygen. Before it can be used the water must pass through a series of pretreatments.

The first step in the pretreatment process is to remove the supersaturated gases such as hydrogen sulfide and carbon dioxide by passing the water through a degassing tower. The degassing tower consists of an eight-foot tall polyethylene tank with a six-foot diameter. Inside the tank is a screened plate spanning the entire cross-sectional area of the tank. This plate supports plastic coiled packing media, which fills the volume of the tank above the plate. The water distributed over the packing media at the top of the tank trickles down through the media in thin sheets and small droplets. A 1/4-hp blower pumps air into the bottom of the column. The column is open at the top, allowing the air to escape. By increasing the area of the air-water interface, gas-exchange between the air and the water occurs at an accelerated rate. Supersaturated gases in the well water are transferred from the

water to the air, and gases that are under-saturated in the water, such as oxygen, are transferred from the air to the water. The water passing out of the degassing tower should be close to the saturation level for all of these gases.

The next step in the treatment process is to remove the majority of the ammonia that is in the water. This is accomplished by passing the water through a 12,000 liter biofilter tank. The biofilter tank contains numberous barrels of oyster shell. Each of the barrels is provided with an airlift, which functions to circulate water through the oyster shell biofilter media. Oyster shell is lifted from the bottom of the bed by the airlift and deposited again at the top of bed. This circulation of the oyster bed through the airlift serves to slough off biofloc from the surface of the oyster shell. The flow rate through the biofilter tank is approximately 200 liters per hour. The total residence time in the biofilter is approximately one hour. During this time the ammonia is reduced from nearly 1 ppm to about 0.05 ppm. The nitrite concentration of the water leaving the biofilter is typically less than 0.01 ppm.

The treated water flows by gravity into one of two 20,000-liter water storage reservoirs. The water storage reservoirs are enclosed polyethylene chemical storage tanks that have been given a double coat of paint to keep them dark inside to prevent algal growth. A 2-hp centrifugal pool pump draws water from the reservoirs and pumps it through a sand filter and out to the culture systems. The water delivery pump operates continuously so that water is available on a demand basis. A return to the reservoir tank is provided to protect the pump when there is little or no demand.

The effluent from the shrimp production tanks discharges into a sump containing chlorine tablets to kill any escaping shrimp. A submersible trash pump with a mercury float switch pumps water from the chlorination sump to a series of retention ponds. The retention ponds for the Harbor Branch aquaculture park consist of three one-quarter acre ponds connected in series. All effluent from the facility discharge into one corner of the first pond in the series. Overflow pipes pass through the levees separating each of the three ponds. The first retention pond is the primary solids settling pond, and typically has the densest growths of algae and aquatic plants. The aquatic plants absorb nitrogenous wastes from the water. Evaporation and seepage account for virtually all of the losses of water from the retention ponds. The second and third ponds in the series provide extended residence time for the water to guarantee that the water has enough time to evaporate, or seep out of the ponds. Every few years it will be necessary to pump out the sludge that collects in the bottom of the ponds. One of the advantages associated with producing shrimp in near-freshwater systems is that this sludge can be used as fertilizer for certain vegetable or row crops. Retention ponds similar to the ones used by Harbor Branch are likely to be required of all feed-based aquaculture operations in the state of Florida.

Production Trials

Methods

Three sets of paired production trials were conducted during the project. In each of the paired trials a single-phase and a three-phase culture system were stocked at the same time with postlarvae from the same cohort to permit comparisons to be made. One of the three-

phase culture tanks, H5-NW, was stocked on 4/12/99 in a non-paired trial. No single-phase culture tanks were available for stocking at that time. System H5-NE (single-phase) and system H5-NW (three-phase) were paired in the first trial (System A Winter Trial). These tanks were stocked November 27, 1998 and harvested on May 25, 1999, after 180 days. Systems H5-SE (single-phase) and H5-SW (three-phase) were paired in the second trial (System A Spring Trial). These tanks were stocked on February 23, 1999 and harvested on August 21, 1999, after 179 days. A third production trial paired single-phase and three-phase culture tanks in System B (System B Spring Trial). This study was to have been initiated at the same time as the System A Spring Trial, but was delayed by two months by unforeseen problems in obtaining building permits to construct the System B greenhouses. Post-larvae were stocked into System B during the last week of April. The System B trials were terminated on September 14, after only 135-140 days, in anticipation of possible landfall of Hurricane Floyd. These tanks were harvested early to prevent accidental escape of the shrimp in the event of flooding.

In each of the trials stocking densities were calculated to achieve a harvest density of 150 shrimp/m², with overstock to compensate for the expected 35% mortality from PL to harvest. The target stocking density in the single-phase systems was 230 shrimp/m². The target stocking density in the nursery section of the three-phase systems was 1,250 shrimp/m². Table 1 summarizes both stocking and harvest data for each of the three trials. The System B culture systems were stocked at densities that were approximately 30% less than the targeted initial densities. This was because of heavy pre-stocking mortality among postlarval shrimp held in the hatchery from the originally scheduled stocking date in late February until the actual stocking date in April. We wanted to give the postlarvae a head start in the hatchery to make up for the delayed startup date. Cannibalism in the hatchery tanks resulted in heavy losses, so not enough of the large postlarvae were available to permit stocking at the targeted densities.

All systems were stocked with Specific Pathogen Free (SPF) postlarvae produced for the study at Harbor Branch. SPF postlarvae are guaranteed to be free from the known viral diseases, including Baculovirus, Infectious Hypodermal Hematopoetic Necrosis Virus (IHHN), Taura Syndrome Virus (TSV), and WSSV. The postlarvae were acclimated to near-freshwater conditions in the hatchery prior to stocking. Acclimation to near-freshwater conditions is possible only after the shrimp have sufficient gill development to permit osmoregulation, usually after they reach PL12.

All crops were reared in near-freshwater conditions. Salinities in the growout tanks averaged 0.7 pppt, chloride concentrations averaged 400 mg Cl^2/L , total hardness averaged 400 mg/L as CaCO₃, and alkalinity averaged 150 mg/L as CaCO₃.

The feeds used in this study were specially formulated with elevated levels of calcium, phosphorus, potassium, Vitamin C, and other vitamins and minerals. The elevated levels of these ingredients are necessary for normal growth and development of shrimp in high density freshwater recirculating systems. A variety of feeds are required to raise the shrimp from postlarvae to harvest size. During the nursery phase we fed postlarval and juvenile diets manufactured at HBOI by Rolland Laramore. For the first five days the shrimp were fed

J400, a 50% protein 400 μ m postlarval diet. The shrimp were fed J1000, a 50% protein juvenile diet (particle size 850-1200 μ m) until they reached a size of 0.2 g/shrimp. When they reached that size they were weaned onto a 1.6 mm pellet (J1600). J1600 has a protein content of 45%. The shrimp remained on J1600 until they reached a size of about 0.8 grams/shrimp, when they were switched to a 3/32" 45% protein juvenile pellet. The shrimp remained on this diet until the end of the nursery phase. In the intermediate and final phases of the growout the shrimp were fed a 3/32" grower pellet. Until April we were feeding a diet manufactured by Burris Mill and Feed that contained 38% protein . This diet was not particularly attractive to the shrimp, and we were dissatisfied with the growth of the shrimp on this diet. In April we switched to a 35% protein Rangen diet formulated for intensive culture. This diet was more palatable to the shrimp and produced better growth rates.

The shrimp were fed four times per day by hand at 8:00 A.M., 11:00 A.M., 2:00 P.M., and 5:00 P.M. The shrimp were fed according to their appetite. Feeders were instructed to monitor feed consumption and adjust feeding rates upward by 10% if all of the feed was consumed in a 3-hour period, and downwards by 10% if significant quantities of feed remained from the previous feeding.

Throughout the project records were kept on feed consumption, temperature, salinity, dissolved oxygen, total ammonia (TAN), unionized ammonia, nitrites, alkalinity, and hardness. Shrimp from each culture system were weighed on a biweekly basis to monitor growth.

The daily maintenance routine included twice daily jetting and backwashing of sand filters and upflow bead filters. Dead shrimp were removed and counted whenever they were observed.

Results

The production results from all trials are summarized in Table 1.

System A Winter Trial

Survival in the single-phase system (H5-NE) stocked on 11/27/98 was a surprising 88%, with nearly 13,800 shrimp surviving out of 15,750 shrimp stocked. The harvest density was 201 shrimp/m². A total of 142 kg of shrimp were harvested. The biomass loading in the system was 2.25 kg/m², which is very close to what we had hoped to produce from the system. However, the average size of the shrimp at harvest was only 10.3 grams after 180 days (Figure 4). With such a high survival, the carrying capacity for the system was reached before the shrimp reached an acceptable harvest size. The average growth rate for this crop was a very disappointing 0.4 grams/week.

System breakdowns may also have contributed to the small size of the shrimp in the singlephase tank (H5-NE). Beginning in March we encountered problems with the H5-NE sand filter. One of the laterals in the bottom of the sand filter broke. The broken lateral caused all of the sand to be lost from the sand filter and a complete loss of biofiltration. Feed rates were sharply reduced until new sand became biologically active. The following month two valves on the sand filter broke off on different occasions. The resulting downtime forced

additional reductions in feed rates. Throughout April and May ammonia levels were often high (Figures 6) and oxygen levels (Figure 7) were routinely less than 5.0 mg/l.

The 47% survival rate of shrimp in the three-phase system, H5-NW, was almost half that of the single-phase system. The relatively low survival resulted from high rates of cannibalism during the nursery and intermediate phases, and from *Vibrio* infection. Cold weather in December and February was followed by relatively warm period (Figure 5). These temperature fluctuations may have stressed the shrimp, precipitating the outbreak of *Vibrio*. With less crowding and generally good water quality conditions, the shrimp in H5-NW grew much better than those in H5-NE, reaching the size of 15.1 grams in 180 days. This corresponds to a growth rate of 0.58 grams/week. System H5-NW did not experience the same water quality problems late in the study that were experienced in H5-NE. Ammonia (Figure 6) levels remained low throughout the trial, and dissolved oxygen levels (Figure 7) remained high. Nevertheless, the growth rates were slower than expected. Previous experience led us to expect the shrimp would reach a harvest size of 18 grams in 180 days.

A major factor contributing to the slower than expected growth rates in both systems was the cold weather in December and February. Neither the tanks nor the greenhouses were heated, so passive solar heating of the greenhouses was the only means of maintaining temperatures. In the first month of the trial a shade cloth covered the outside of the greenhouse, cutting down on the warming effect of solar radiation during the day, but doing little to prevent heat loss at night. Cold weather in December and in February (Figure 5) resulted in water temperatures of 22°C or less for extended periods of time. Temperatures rarely rose above 26°C for the first 60 days of the culture period. When the water temperature is less than 22° C, the shrimp do not grow. Growth rates are significantly reduced when temperatures drop below 26°C.

System A Spring Trial:

As was the case in the Fall production trials, the survival in the single-phase production system, H5-SE (76%) was significantly higher than the survivals achieved in either of the two three-phase systems stocked at the same time (Table 1). The survival in the two three-phase studies stocked in February were 61% (H5-SW) and 40% (H5-NW). The survival in H5-NW was estimated at 65% until July 7, when the air supply to the raceway was interrupted due the failure of a pipe connection. Over two thousand shrimp (20% of the population) died as a result of the low dissolved oxygen condition. Cannibalism was frequently observed in the nursery and intermediate sections of the three-phase culture tanks.

After 179 days the shrimp in the single-phase culture tank, H5-SE averaged 14.6 g and a total of 194.2 kg of shrimp were harvested. Growth rates averaged 0.57 g/shrimp/week. Shrimp harvested from the three-phase culture tanks averaged 15.3 g/shrimp (H5-SW) and 13.6 g/shrimp (H5-NW) after 180 days. The growth rates in H5-SW averaged 0.6 g/shrimp/week, while the shrimp in H5-NW grew at an average rate of 0.53 g/shrimp/week.

Culture System	Date Stocked	Number of Shrimp Stocked	Initial Average Wt. (g)	Culture Period (Days)	Number of Shrimp Surviving	Percent Survival	Final Average Wt. (g)	Final Biomass (kg)	Final Density (Shrimp/m ²)	Feed Conversion Ratio	
Single-Phase											
H5-NE	11/27/98	15,750	.005	180	13,798	88%	10.3	142	201	1.76	
H5-SE	2/23/99	17,400	.01	179	13,300	76 %	14.6	194.2	169	1.83	
J1	4/28/99	19,000	.003	135	12,807	67%	9.0	115.3	109	1.36	
J2	4/26/99	20,000	.03	137 ¹	18,074	90%	9.5	171.7	153	1.41	
Three Ph	nase	•								•	
H5-NW	11/27/98	10,000	.005	180	4,680	47%	15.1	70.7	108	1.91	
H5-SW	2/23/99	11,362	.005	179	6,938	61%	15.3	106.2	142	1.61	
H5-NW	2/5/99	10,450	.008	180	4,153	40%	13.7	56.9	95	2.05	
H5-NW	4/12/99	9,900	.02	154	6,224	63%	14.6	90.9	144	1.67	
J3	4/25/99	10,000	.02	138	8,184	82%	15.0	122.8	121	1.70	

Table 1: Summary of production results

1) Nearly all of the shrimp initially stocked on 4/26/99 died on 6/1/99 due to high nitrite levels resulting from stocking the PLs before the biofilter was conditioned. The tank was restocked on 6/14/99 with 20,000 shrimp of the same age as the survivors from the initial stocking. The re-stocked shrimp were approximately the same size (0.90 g vs. 0.99 g) as the surviving shrimp from the initial stocking. Because the restocked shrimp were the same age and size as the surviving shrimp from the initial stocking, the culture period for J2 given in the table is counted from the initial stocking date (4/26/99).

Growth rates in all three of the tanks stocked in February were extremely slow during the first ninety days after stocking (Figure 8), averaging less than 0.3 g/shrimp/week. The shrimp in all three tanks measured 4 grams or less after 90 day. For the second 90 days of the culture period, growth rates averaged between 0.80 and 0.85 g/shrimp/week in each of the three tanks.

Low temperatures may be partly responsible for the slow growth observed in the spring production trials. Weekly temperature averages were less than 28°C for more than half of the culture period (Figure 9). Growth rates begin to be affected when temperatures drop below 28°C.

No major ammonia or nitrite problems were observed in any of the three culture tanks. Total ammonia nitrogen levels were maintained, for the most part, at a concentration less than or equal to 0.4 mg TAN/L (Figure 10). TAN concentrations were elevated in H5-NW for a period of three weeks in June, ranging between 0.8 and 1.2 mg TAN/L. Concentrations of toxic unionized ammonia ranged between 0.05 - 0.08 mg NH₃-N/L during this period. These concentrations of unionized ammonia are well below the lethal limit for juvenile shrimp, but could have had an impact on the growth rates of the shrimp. Nitrite levels were generally less than 0.4 mg NO₂-N/L in all three culture tanks. Nitrite levels were slightly elevated (0.8 – 1.2 mg NO₂-N/L) in H5-SE during the last 2 weeks before the shrimp were harvested. However, these levels were more than an order of magnitude less than the lethal limit for adolescent shrimp.

Dissolved oxygen levels dropped to about 1.5 ppm in H5-NW on July 7 when the air supply to the tank was interrupted for an undetermined period of time because of the failure of a pipe connection. This incident resulted in the loss of 2,000 shrimp. Aside from this incident, dissolved oxygen levels were maintained above 5 mg/L throughout the culture period in both of the three-phase tanks (Figure 11). The dissolved oxygen concentrations in the single-phase tank, H5-SE, were maintained above 5 mg/L except for the final 7 weeks, when dissolved oxygen concentrations averaged between 4 and 5 mg/L (Figure 11). This may explain why the growth rate of the shrimp in H5-SE slowed during the final three weeks of the culture period (Figure 8).

System B Summer Trials

The System B trials were terminated 6 weeks early on September 13 and 14 because of the threat posed by Hurricane Floyd. As a result, the survival and final harvest size data presented for tanks J1, J2, and J3 in Table 1 are not directly comparable to the System A data because the culture period was much shorter. At the time these tanks were harvested, however, trends were already emerging.

Survival in J1 after 135 days was 67%, slightly lower than had been observed in other single-phase culture systems. The large majority of the mortality was observed during the fourth and fifth week of the study, when nitrite levels peaked at 10 mg NO₂-N/L. This was due to the fact that, because of the construction delays, we stocked the system without preconditioning the biofilters. Nitrite levels were even higher J2, which was stocked with

larger postlarvae and was receiving more feed. J2 experienced massive mortality four weeks after it was stocked. We restocked J2 on June 17 with juvenile shrimp of the same age as the shrimp that had been initially stocked. Survival of these animals was 90% to the end of the study.

One of the three-phase systems, J4, also experienced nitrite levels above 20 mg NO_2 -N/L during the fourth week of the study and suffered nearly 100% mortality. There were no animals available to restock this tank, so we continued the study with only one three-phase tank in System B.

The survival in the remaining three-phase tank, J3, was 82% after 138 days (Table 1). This survival was achieved despite the fact that these shrimp experienced nitrite levels as high as 6.0 mg NO₂-N/L during the biofilter conditioning process. In contrast to what was observed in the three-phase tanks in System A, very little cannibalism was observed in J3. The reduction in cannibalism may be related to two system modifications that reduced the encounter frequency between the shrimp. The tank depth in the nursery section of J3 is nearly twice as deep as in the System A nursery sections. In addition, current velocities were higher in the nursery and intermediate sections of J3. Higher current velocities cause the shrimp to move off the bottom and swim in the water column. This, combined with the increased tank depth, reduced the encounter frequency between the shrimp.

The shrimp in the single-phase tanks, J1 and J2, averaged 9.0 and 9.5 g/shrimp, respectively, after 136 days (Table 1, Figure 12). These average weights are comparable to the weights observed for shrimp in the System A studies after the same time period (Figures 4 and 8). Growth rates were particularily slow during the first 90 days of the culture period. This was most likely due to the fact that throughout the second month the shrimp were on a restricted diet because of the high nitrite concentrations in the tanks. In addition, these greenhouses were covered with a 95% shadecloth, which virtually eliminated algal growth. Without adequate feed input and without significant natural productivity in the tanks, the shrimp were not adequately nourished during the first half of the two and half months of the study. In contrast, the shrimp in the three-phase tank, J3, grew very rapidly and reached an average weight of 15.0 g/shrimp after only 138 days. The average growth rate of these shrimp over the final 72 days of the growout period was 1.0 g/shrimp/week. Projecting this growth rate out over the next 42 days (to the expected harvest at 180 days), the predicted harvest weight of these shrimp would be 21 g/shrimp.

Without carefully controlled experiments, it is difficult to say with certainty why the shrimp in J3 grew at a much faster growth rate than did all of the other tanks in this study. It is possible that the lower stocking density (about 30% less than the stocking density of the System A three-phase tanks) allowed for faster growth rates. However, with the higher survival rate, the final harvest density (122 shrimp/m²) was intermediate in the range of final harvest densities (Table 1) for the three-phase tanks in System A (95-144 shrimp/m²). Yet the growth rates in J3 far outpaced the growth rates in any of the System A tanks. System differences such as tank depth and filtration systems might be partially responsible for some of the observed differences in growth rates. While the differences in the tank depth and water filtration systems could explain differences between the growth rates between J3 and the System A tanks, they do not explain the pronounced difference in growth rates between J3 and the single-phase System B tanks (Figure 12). One important difference existed between the greenhouse enclosing J3 and the greenhouse enclosing J1 and J2. The shadecloth covering the J3 greenhouse was removed in the middle of June. After the loss of the shrimp in J4, system J4 was put into algae production for the clam hatchery. This required removal of the shadecloth. Following removal of the shadecloth, a dense algal bloom developed in J3, which was maintained until the end of the study. It is possible that the phytoplankton provided a supplemental food supply for the shrimp.

Temperatures in the System B greenhouses (Figure 13) were much warmer than the temperatures that were maintained in the System B winter and spring trials (Figures 5 and 9). Temperatures were maintained above 30°C in J3 from July until the end of the study. The temperatures in J1 and J2 were slightly warmer than in J3 through the first 60 days. During this period average weekly temperatures in J3 were less than 28°C. Average weekly temperatures in J1 and J2 remained above 28°C from the end of the first month to the end of the study (Figure 13).

Ammonia was not a problem during the System B trials, despite the fact that the systems were started up without preconditioning of the biofilters. Ammonia concentrations spiked, as expected, about three weeks after the systems were started up (Figure 14). The maxiumum total ammonia nitrogen (TAN) concentrations observed in any of the tanks was 1.2 mg TAN/L. TAN concentrations declined to low levels following the initial peak, and remained low throughout the rest of the studies (Figure 14). TAN levels in J1 never peaked at all.

High nitrite concentrations presented the major water quality problem in the System B trials, and resulted in the loss of two crops of shrimp. Establishment of the *Nitrobacter* population on the biofilter media lagged far behind the establishment of the *Nitrosomonas* population. For much of the first 60 days, the majority of the ammonia that was converted into nitrite by the *Nitrosomonas* bacteria, remained in the system. During this period nitrite levels were controlled primarily by water exchange. Initially we were rinsing and tumbling the beads in the solids filter twice a day to remove the solid wastes. In addition, the beads in the biofilter tank were tumbled continuously by aeration. It appears that the vigorous tumbling the beads were subjected to during this procedure interfered with establishment of the *Nitrobacter* population. Near the end of June we reduced rinses of the solids filter to once every third day. This procedural change was quickly followed by a reduction in the nitrite levels to less than 0.5 mg NO₂-N/L in all three tanks. The solids filter thereafter functioned as a biological solids filter, and was the principal site for nitrification of nitrite to nitrate by *Nitrobacter*.

Dissolved oxygen concentrations were maintained above 6.0 mg/L for the first 90 days in all of the System B tanks (Figure 15). Dissolved oxygen concentrations were maintained above 5.0 mg/L throughout the study in J1. Dissolved oxygen (DO) concentrations dropped below 5.0 mg/L in both J2 and J3 during the final month of the study. Morning DO levels occasionally dropped to as low as 3.5 mg/L in system J3, but typically would rise to about 7.0 mg/L in the afternoon. This diurnal swing in DO levels was related to the presence of an

algal bloom in this system. Dissolved oxygen concentrations never dropped to dangerous levels in any of the System B tanks.

System A Summer Trial

The three-phase culture system H5-NW was stocked for the third time during the year on April 12. This crop was not paired with a single-phase System A crop because no single-phase culture systems were available for stocking at that time. To a certain extent, the crop serves as a System A counterpart or control to the summer production trials in System B. This study was also terminated early on September 14 due to the threat of landfall by hurricane Floyd.

Production data for this crop are summarized in Table 1. Survival was 63%, with a harvest density of 144 shrimp/m². Cannibalism during the nursery phase and shrimp jumping out of the tank account for most of the observed mortality. Based on observations of mortality patterns in other crops, very little additional mortality would be expected if the crop had been carried on for three more weeks. Based on the high survival observed during the last three weeks in other tanks, it is likely the survival for a 180-day growout would have been above 60%.

The average weight after 159 days was 15.1 grams. While growth rates averaged only 0.67 grams per week for the entire culture period, over the last 72 days of the culture period growth rates averaged 1.0 gram per week (Figure 16). If growth rates had continued at this pace for another 3 weeks, the shrimp would have averaged approximately 18 grams after 180 days. While this is slightly smaller than the projected harvest size of the System B three-phase crop in J3, it is much better than the growth rates that were observed in any of the winter or spring trials in System A.

During the first 80 days, weekly average temperatures (Figure 17) were maintained between 26°C and 28°C. Weekly average temperatures were maintained between 28°C and 29°C during the last 80 days of the study. Warm temperatures during the latter half of the growout period coincide with the 1.0 gram/week growth rates.

For the majority of the culture period, TAN concentrations were maintained below 0.4 mg TAN/L (Figure 18). For a two-week period during the second month of the culture period TAN concentrations averaged 0.8 mg TAN/L. During this time frame the pH averaged between 8.0 and 8.3, and unionized ammonia levels rose as high as 0.08 mg NH₃-N/L. While these concentrations are well below the lethal level, they are more than double the desired upper limit of 0.03 mg NH₃-N/L. Had the unionized ammonia levels remained at this level for long, growth rates would likely have been affected. Nitrite levels were generally less than 0.6 mg/L throughout the study. For a short period in early June, nitrite levels rose to 1.4 mg NO₂-N/L. This level is slightly above the desireable upper limit for nitrite, but well below the lower lethal limit for juvenile shrimp.

Dissolved oxygen concentrations were maintained above 5.0 mg/L throughout the study (Figure 19). During the first half of the study dissolved oxygen concentrations were generally maintained above 6.0 mg/L.

Discussion

One of the objectives for this study was to compare the productivity of a single-phase production system with that in a three-phase production system. As part of the analysis, a comparison was made of the potential annual production of shrimp in a single-phase and a three-phase production system (Table 2). This comparison was based on average stocking densities, survival rates, and harvest weights achieved during the course of this project. This analysis indicated that, despite lower average survival rates, the total annual production of a three-phase production system should be 60% higher per unit of production area than the annual production of a single-phase system. The average weight of shrimp harvested from three-phase systems during this study was actually larger than for single-phase systems. Even if the harvest weights of shrimp from three-phase and single-phase systems were equal, the annual productivity of the three-phase system would be 40% higher than that of a single-phase system. If survival and final harvest weight of the two systems were equal, the three-phase system would out-produce a single-phase system by 80%. These results strongly favor the use of three-phase production systems over the traditional single-phase approach.

The primary objective for this project was to quantify the key production parameters and associated costs for growing shrimp in greenhouse-enclosed freshwater recirculating systems. An economic analysis based on the data that were collected during the course of this project was performed for a hypothetical 12-greenhouse enterprise (see Appendix A). Because the production potential for the three-phase system was so much greater than that of a single-phase system, only the three-phase system was modeled.

Darameter	Culture System			
i alametei	Single-Phase	Three-Phase		
Number of shrimp stocked/m ² of culture area/crop	200	120		
Average survival rate to 180 days ¹	77%	60%		
Average number of shrimp harvested/m ² of culture area/crop	154	72		
Average weight of individual shrimp harvested (g) ²	14.1	15.9		
Average total weight harvested/m2 of culture area/crop (kg)	2.17	1.14		
Potential crops/year	2	6		
Potential total harvest weight/m ² of culture area/ year (kg)	4.34	6.87		

Table 2: Comparison of the annual production potential a single-phase and three-phase system.

- 1 Predicted 180-day survival rates were used rather actual survival rates for the tanks that were harvested early due to hurricane Floyd. The 180-day survival rate was predicted by projecting a straight-line survival curve from the initial stocking date to the actual harvest date out to the date when the culture period would have reached 180 days. This probably underestimates 180-day survival rates because most of the mortality occurs during the first 90 days after stocking. The survival rate of the spring trial of H5-NW, which lost 2,000 shrimp due to a disconnected airline, was adjusted upward by assuming that 80% of the shrimp lost in that event would have otherwise survived.
- 2 Predicted 180-day harvest weights were used rather than actual harvest weights for the tanks that were harvested early due to hurricane Floyd. The 180-day harvest weights were predicted by assuming that during the time period between the actual harvest date and 180 days the shrimp would continue to grow at the same rate as was observed for the 10-week period prior to the actual harvest.

Based on the costs and production parameters estimated in this study, the economic model shows that culturing shrimp in systems like those demonstrated in this study would not be profitable. However, the sensitivity analysis shows that if the survival can be improved to 70%, and the growth rate improved so that 18 gram shrimp can be grown in 150 days, a 12-greenhouse enterprise could generate an internal rate of return of nearly 50% (assuming the shrimp can be sold as a fresh, heads-on product for \$5.24/lb). How likely is this scenario?

There is good reason to believe that growth rates can be improved. As was discussed earlier, the slow growth rates that were observed in many of our production trials were, at least partly related to cool temperatures in the culture tanks during significant portions of the culture period. However, the production tanks were not heated and temperatures were not optimal for growth throughout much of the winter and spring trial culture periods. With optimal culture temperatures there is little doubt the shrimp could be grown to a minimum harvest size of 18 g/shrimp in 180 days. This was demonstrated in the summer trials in tanks J3 and H5-NW (Table 1). We did not, however, demonstrate that shrimp can be grown in tank culture systems to 18 grams in 150 days.

It is well known that in ponds, *L. vannamei* grows best in ponds with high levels of natural productivity (Scura, 1995). Phytoplankton and organic detritus are both important components of the shrimp's diet (Moss, 1992). *L. vannamei* has a very inefficient digestive system consisting of short, straight gut. Evidence is accumulating that *L.vannamei* does not utilize prepared diets efficiently, especially if their feces are rapidly filtered from the system. However, if their feces are allowed to remain in the system, heterotrophic bacteria will colonize the fecal material and convert feed protein into bacterial protein. Shrimp consume the decaying fecal material and the associated bacteria. The shrimp derive significant nutritional benefits from the bacterial proteins and partially digested feed proteins during this second pass.

The importance of the detrital food chain to shrimp growth was not fully appreciated until this study was nearly over. The culture tanks were shaded to control algae growth, and solid wastes were quickly removed from the system in the interest of maintaining optimal conditions for biofiltration. As a result, the shrimp were almost completely dependent upon the nutrition they could absorb from the prepared feeds in a single pass through the gut. Recent unpublished work at Harbor Branch has demonstrated that the growth rates of shrimp grown in tanks managed for optimization of the detrital food chain have been up to 50% faster than the growth rates observed in the systems modeled in this report. Similarly, Moss (1999) reported growth rates of *L.vannamei* cultured in a high density tank-based culture system at the Oceanic Institute (OI) in Hawaii that were double the growth rates observed in the HBOI system. The primary difference between the OI system was a "clearwater" system. The presence of algae and organic detritus in the tanks was credited by Moss for the rapid growth rates that were observed in the OI system.

These results suggest that the mediocre growth rates observed in this study were not strictly a function of the tank environment, or the high densities that were used. Rather, the slow

growth may be related to the scarcity of detritus in the system. Better growth rates might be realized with alternative management strategies. Sand filters are probably too efficient at removing algae and fecal wastes. The filtration system used in System B allows some of the finer particulates remain in the system. These are broken down by bacteria and are potentially reprocessed by the shrimp. Removing the shade cloth and allowing phytoplankton blooms to develop appears to allow for much faster growth during the first 90 days of the culture period. Work is needed to learn how to manage systems with dense phytoplankton blooms so that they are stable. Nevertheless, it is clear that it is possible to grow the shrimp to a size of 18 grams in 150 days.

The possibility exists that growth rates are reduced in a freshwater culture environment. Based on our results in this study, we have no basis for evaluating the possibility that the stress of the freshwater culture environment somehow inhibits shrimp growth, because we do not have saltwater controls to compare our results to. Reports from pond culture systems shows that excellent growth is possible at salinities down to 2 ppt. At 0.5 ppt, however, the shrimp are closer to their physiological limits. It is certainly plausible that the increased energy expended on osmoregulation comes at the expense of growth. This is an important question for future investigation.

There is good reason to believe that survival rates can be improved in three-phase systems. A major problem encountered in our three-phase culture tanks was the high rate of cannibalism in the nursery and intermediate sections of the three-phase raceways. This is, at least in part, a density-related phenomenon. Post-larvae are very cannibalistic in high density environments, especially when underfed. The high density increases the encounter rate between individuals, increasing the opportunities for aggression to occur. Design modifications such as deeper tanks and greater water movement should help reduce cannibalism by reducing the encounter rate between postlarvae. Increasing the frequency of feedings is another management strategy that may help reduce cannibalism. Shrimp are more likely to cannibalize their peers when they are hungry. This problem can be overcome by feeding aggressively and more frequently to make sure the shrimp are never hungry.

There is growing evidence that artificial substrates can help improve both survival and growth rates of shrimp in both ponds and raceways. Artificial substrates provide additional surface area, which lets the shrimp spread out more. In addition, periphyton growing on artificial substrates provides a nutritious supplement to the artificial feeds.

The average survival rates achieved in our single-phase culture tanks was 77%. Survival of shrimp in our unshaded System B three-phase system was 82%. There is good reason to believe that improved system design, use of artificial substrates, promotion of algal and detrital food chains and increased feeding frequencies can improve survival rates to 70%. If both growth and survival can be improved, and shrimp can be sold for at least \$5.00 per pound, greenhouse culture of shrimp in freshwater recirculating systems could be a profitable business.

Recommendations

While it is clear improvements are needed, we still are confident this is a technology for the future. The following are some recommendations for improving the technology, based on the experiences of this year:

- 1. Heating systems are required to obtain consistent growth year-round. Studies are necessary to determine the most economical system for heating a raceway.
- 2. Deeper raceways with higher velocity should help improve survival, and obtain more uniform growth rates.
- 3. Artificial substrates should be investigated to determine if they help reduce cannibalism and if epiphytic growth can provide an additional source of natural foods.
- 4. Research should be conducted to determine how to create stable production systems with rich algal and detritus-based food chains.
- 5. More research is needed to develop nutritionally complete diets, especially for young juveniles.
- 6. Work on improving low-head biofiltration and solids removal systems.
- 7. More research is needed to determine whether or not near-freshwater systems inhibit growth due to chronic osmotic stress. Additional research is need to determine if mineral supplements to the water are needed or beneficial.
- 8. Market research is needed to determine the nature of direct markets for freshwater shrimp.
- 9. Work is needed to determine if other types of biofiltration systems are better suited to shrimp culture (for example, systems utilizing heterotrophic bacteria, in addition to autotrophic bacteria, to control ammonia and nitrite concentrations.).

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Figures



Figure 4: System A Winter Production Trial Growth Curve.



Figure 5: System A Winter Production Trial Temperature Data



Figure 6: System A Winter Production Trial Total Ammonia Nitrogen Concentrations



Figure 7: System A Winter Production Trial Dissolved Oxygen Concentrations



Figure 8: System A Spring Production Trial Growth Curves.



Figure 9: System A Spring Production Trial Temperature Data.



Figure 10: System A Spring Production Trial Total Ammonia Nitrogen Concentrations.



Figure 11: System A Spring Production Trial Dissolved Oxygen Concentrations.



Figure 12: System B Summer Production Trial Growth Curves.



Figure 13: System B Summer Production Trial Temperature Data.



Figure 14: System B Summer Production Trial Ammonia Concentrations.



Figure 15: System B Summer Production Trial Dissolved Oxygen Concentrations.



Figure 16: System A Summer Production Trial Growth Curve



Figure 17: System A Summer Production Trial Temperature Curve.



Figure 18: System A Summer Production Trial Total Ammonia Nitrogen Concentrations.



Figure 19: System A Summer Production Trial Dissolved Oxygen Concentrations.