Improving Greenhouse Cucumber Production in De-coupled Aquaponic Systems

by

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Abstract

Cucumbers are a major greenhouse crop, usually grown in hydroponic systems, but can be adapted to aquaponics— the integration of fish and plant production. Aquaponic production has shifted from coupled, recirculating systems to de-coupled, non-recirculating systems due to the ability to adjust water quality parameters, namely pH, in each de-coupled unit to optimize production. In order to effectively adapt cucumber production to aquaponics, determining production practices that minimize production costs and increase yield is necessary. Cucumber production requires energy and labor inputs for the duration of the production cycle, which means costs are likely to increase as production continues even with continued revenue from longer production lengths. To determine a production-cycle length that would maximize profit, four production cycle-lengths were evaluated for cucumbers in a de-coupled aquaponic system. Revenue from cucumber production increased linearly as production cycle length increased, providing the highest revenue at a 95-day cycle length at 17.80 USD per m^2. Estimated costs increased as cycle length increased, but the highest profits were obtained at shorter cycle lengths. In aquaponic systems, shortening production cycle length during the traditional off-season for cucumber production would maximize profits.

To determine the effect of pH in a de-coupled aquaponic system, a study was conducted using aquaculture effluent from tilapia culture tanks at 4 pH treatments: 5.0, 5.8, 6.5, and 7.0 to irrigate a cucumber crop. Growth and yield parameters, nutrient content of the irrigation water, and nutrients incorporated into the plant tissue were collected over two growing seasons. pH did not have a practical effect on growth rate, internode length or yield over the two growing seasons.

Availability and uptake of several nutrients were affected by pH, but there was no over-arching effect that would necessitate its use in commercial systems. Nutrient concentrations in the aquaculture effluent would be considered low compared to hydroponic solutions; however elemental analysis of leaf tissue was within the recommended ranges. Research into other nutrient sources provided by the system (i.e. solid particles carried with the irrigation water) would provide further information into the nutrient dynamics of this system.

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Chapter I

Literature Review

Feeding a Global Population

Agriculture is an integral part of society, reaching into nearly every aspect of daily life from textiles to medicine to biofuels, and perhaps most importantly, diet. By the year 2050, the United Nations estimates that the global population will reach 9 billion people and 11.2 billion by 2100 (U.N., 2017). An increasing world population calls for an increase in agricultural food production to satisfy the demands of a growing population. Estimates of the increase in food demand range from a need for a 45-71% increase from the years 2010-2050 (Keating et al., 2014). Innovative solutions are required to meet the needs of a society and to obtain food security as populations increase.

Aquaculture

Aquaculture is the cultivation of aquatic species for human use. Many consider it to become a major factor in supplying global food production in the coming years. A large portion of the worldwide seafood supply is obtained from capture fisheries, however as new technologies for capturing wild fish emerge and the demand for fresh seafood increases, overharvesting and stock depletion are becoming problems for production.

According to the Food and Agriculture Organization (FAO), aquaculture's contribution to the world fish supply has been increasing over the years with 44.1% of the total worldwide fish production coming from aquaculture in 2014. The aquaculture industry over-took production from capture-fisheries for the first time, supplying 50% of the global fish supply (FAO, 2016), and is still considered to be a growing sector in the agriculture industry. In 2010 the FAO reported that the aquaculture industry was growing at a faster rate than capture fisheries and terrestrial meat production—6.6%, 1.2%, and 2.8% respectively (FAO, 2010). The United States ranked 16th in global aquaculture production in 2016 (NMFS, 2018), with Alabama as the 4th largest state for aquaculture production in the U.S. (USDA-NASS, 2017). Taking advantage of the growing aquaculture industry could make the United States competitive with the leading aquaculture producing countries (i.e. China, India, Indonesia, Vietnam, and Bangladesh) (NMFS, 2018).

Fish and other aquaculture products are gaining attention as an alternative protein source to terrestrial meat. Crude protein accounts for 18.5% of fish fresh weight, and fish provide all amino acids essential for the human diet as well as vitamins A, B, C, D, E, and K (Haard, 1995). As a growing industry supplying nutrient-rich protein, optimizing aquaculture production in the United States could be one way to establish food security for the U.S. and the world in the coming years.

Terrestrial Aquaculture Production

Aquaculture production systems range in their system designs form low-intensity systems (10 kg biomass•ha^{-1})), which require little-to-no human input to high intensity systems where a high stocking density (up to 100,000 kg biomass•ha^{-1}) is contained in one area with a large degree of human intervention used to run the system (Tidwell, 2012). Aquaculture systems can be open, semi-closed, or closed depending on the ways oxygen supply, waste removal, and temperature control are handled in the system. Low intensity, open systems rely on natural

processes to provide these functions, whereas in high-intensity, closed systems, humans monitor and control the oxygen supply, waste removal, and temperature balance in the system.
Outdoor pond systems have been used to cultivate fish for the aquaculture industry, however recirculating aquaculture systems (RAS) provide the potential for producers to control all environmental aspects of production, especially if the system is located indoors.

If left unattended, water quality in aquaculture production systems decline over time due to the build-up of ichthyotoxic substances in the water. These substances include carbon dioxide, and nitrites, but the main concern for aquaculture producers is the build-up of ammonia that is released from fish gills and through fish excrement. In RAS production, biological filtration is used to filter the water of wastes and harmful compounds that build up in the system as a result of production. Biological filtration is employed through the use of biofilters where bacteria convert the harmful compounds into tolerable forms for production. The water is then disinfected before being re-circulated through the system. Although these systems are termed as "re-circulating" water does need to be exchanged from the system, but at a lower rate than the water that is exchanged in other systems. Not only do these systems allow for a greater degree of environmental control, but their capability to support higher production densities make them an attractive option for increasing aquaculture production. In non-recirculating systems, water quality is maintained by discharging aquaculture effluent into the environment and adding fresh water back into the system.

Impacts of Increasing Aquaculture Production

Feed conversion ratio (FCR) is used as a measure of production efficiency and is defined as the mass of dry feed required to produce one unit of biomass (Fry et al., 2018). A low FCR is more desirable as it indicates a higher efficiency in feed utilization. Fish, in general, have a lower FCR compared to other protein sources. While FCR varies with species, tilapia commonly have an FCR of 1-2.5 depending on the growth stage of the fish and the type of feed given (El-Sayed, 2013). FCR for poultry, beef -cattle, and swine are 2.19, 14.97, and 2.94, respectively (Peters et al., 2014). While fish have a lower FCR, production cost of commercial aquaculture feed has been increasing due to increases in the cost of aquaculture feed ingredients. The rising cost of fish feed, which accounts for upwards of 50% of production costs in aquaculture enterprises, limits the potential for fish to become a main contender with terrestrial species for human consumption (Rana, 2009).

Another impediment to increasing aquaculture production is the environmental impact associated with the discharge of aquaculture effluent into the environment, primarily with concern to water quality and, in particular, nitrogen levels. Water quality parameters such as pH, alkalinity, dissolved oxygen, and temperature are often monitored daily and can be easily adjusted to obtain the correct levels for production; however, nitrogen levels are managed by exchanging nitrogen-rich water with fresh water. When fish feed enters the aquaculture system, it is digested by the fish, incorporated into their tissues, and enters the tank solution as either ammonia excreted directly from fish gills or nutrients made available by microorganisms living in the system (Rakocy et al., 2016). Neto and Ostrenksky (2015) estimated nitrogen flow in Nile tilapia as 35% of feed-nitrogen incorporated into tissues, 33% passing through gills as unionized ammonia, 18% as uneaten feed, and 13% contained in the solid waste. Ammonia is present in two forms in solution: unionized ammonia (NH_3) and ionized ammonia (NH_4^+), which combined, make up total ammonia nitrogen (TAN). Water quality parameters such as pH, temperature, and salinity dictate the ratio of NH_3:NH_4^+ (Timmons et al., 201; Lekang, 2013) in the system. NH_3 is toxic to fish, and levels as low as 0.6 ppm can lead to fish death (Durborow, 1997). It is recommended to keep NH_3 levels below 0.05 ppm (Timmons et al., 2001) to avoid toxicity. The greater the concentration of TAN the greater the potential for NH_3 toxicity as a sudden pH swing could rapidly convert NH_4^+ to its unionized and more toxic form.

To decrease NH_3 levels, NH_3 is converted into the less toxic form, nitrate (NO_3) through the process of nitrification. In RAS systems, nitrification is promoted through the use of either biofilters or bioflocs to convert ammoniacal waste into the safer nitrate form through oxidation in a two-step process by nitrifying bacteria. NH_3 is first converted into an intermediate, nitrite (NO_2^-), by *Nitrosomonas spp.*, and ultimately into nitrate (NO_3) by *Nitrobacter spp*. Nitrate, while less toxic to fish, must still be removed from the system to avoid reaching harmful levels. Recommended nitrate levels are between 0-400 $mg \bullet L^{-1}$ (Timmons et al., 2012). Growth of the nitrifying bacteria in the system is promoted by providing a media with relatively high surface area or by suspending solids into the water column through agitation (Lekang, 2013). The success of an established biofilter depends on water quality parameters discussed above. Temperature, pH, dissolved oxygen, and organic matter all play into the ability of the bacteria population in the biofilter to grow and filter the water. Bioflocs are another type of biological filters where microorganisms and organic matter are aggregated and suspended in the water column (Hargreaves, 2013). A biofloc system allows for minimal water exchange, thus decreasing the spread of disease. It has also been shown that bioflocs increase the immune response of tilapia (Menaga et al. 2019) and increase fish biomass (Liu, 2019) compared to conventional methods.

Along with harmful compounds, solids accumulate in aquaculture systems due to uneaten feed, feces, and micro-organisms from the biofilter or biofloc (Timmons et al., 2012). If the amount of solids in the water exceeds 25 mg•L^{-1}, it can harm not only the fish but also the bacteria in the biofilter (Timmons et al., 2012). Solids can also become places for pathogens to accumulate and spread disease throughout the system. In most systems the removal of solids, by either settling or filtration, is necessary to ensure water quality and maintain system performance.

Once free of harmful compounds, the water, now rich in nitrates and other nutrients, is discharged from the system into the environment and the discharged water is replaced. The discharge of this nutrient-rich water into the environment has the potential to cause eutrophication of surface waters. Aquaculture operations have been shown to contribute high loads of N and P to surface waters creating areas of eutrophication (Axler et al., 1996; Zhang et al., 2006).

Increasing aquaculture production alone would exacerbate the economic and environmental impacts associated with it. Several methods of extracting pollutants from wastewater exist, however, integrating aquaculture with plant production through aquaponics is the only method that produces a salable product. Aquaponics combines the environmental stewardship of wastewater treatment with a way of providing food security and generating a secondary income from aquaculture inputs.

Hydroponics

Hydroponics is the technique of growing crops using water as the carrier for all nutritional elements required for plant growth (Resh, 2008). Plants require 16 elements in sufficient quantities to maintain normal metabolic functions and complete the reproductive cycle. These “essential” nutrients can be divided into two groups, macro-nutrients and micro-nutrients, depending on the relative amounts required for plant growth. The macro-nutrients, nitrogen (N), phosphorus (P), potassium (K), calcium (Ca), magnesium (Mg) and sulfur (S), are required in the largest amounts with N, P, and K being of particular importance. The micro-nutrients, boron (B), chlorine (Cl), copper (Cu), iron (Fe), manganese (Mn), molybdenum (Mo) and zinc (Zn) are required in much smaller amounts (Foth and Ellis, 1997). Insufficient or excessive quantities of any of these elements will result in deficiency or toxicity symptoms, resulting in poor plant growth, damage, mortality or the inability of the plant to complete its reproductive cycle. Hydroponic nutrient solutions supply all essential plant elements required for growth in measured amounts. The solution can be recirculated through the system to increase water and nutrient use efficiency.

A major factor that governs the availability of essential nutrients in of hydroponic systems, as well as conventionally produced plants, is solution pH. Low pH can damage root cell walls, which can lead to electrolyte leakage and cell death. At higher pH, some plant nutrients have the potential to precipitate out of solution in the form of insoluble salts. This is mainly a concern with phosphorus, which forms an insoluble salt with free calcium in solution as calcium phosphate at slightly alkaline pH and readily at pH above 8.0. Many of the micro-nutrients also become unavailable at high pH including B, Cl, Cu, Fe, Mn, and Zn (Foth and Ellis, 1997). Therefore, most commercial hydroponic systems are maintained at a pH of 5.8 (Bugbee, 2003), within the 5.5-6.5 pH range for conventional crop production in soil bound systems.

Hydroponic systems are typically housed within greenhouses where environmental conditions such as temperature and other factors such as pH can be monitored and controlled.

Cucumber (*Cucumis sativus* L.*)* is a warm season annual vegetable that thrives at temperatures between 26.7 and 29.4°C (80-85°F) and is a major greenhouse vegetable crop

(Hochmuth, 2015; Wittwer and Honma, 1979). Cucumbers were ranked second among the highest-valued greenhouse-grown crops in the United States behind tomatoes (USDA-NASS, 2014). Nearly 1.02 million m^2 of cucumbers were grown under protected-culture structures such as greenhouses and high tunnels in 2014 amounting to $78M in production value. Of all protected-culture cucumbers grown, 91% were grown using hydroponic systems. Because of the similarities between hydroponics and aquaponics, cucumber production can be easily adapted to aquaponic production.

Cultivars for greenhouse production are primarily parthenocaropic and gynoecious, producing seedless fruit on pistillate flowers. Miniature cucumbers range in size between 12 to 20 cm at market-size (WISS, 2016) and are a popular greenhouse type (Hochmuth, 2015). Plants are typically spaced 30.5-45.7 cm apart in rows and grown vertically on a trellis system in the greenhouse. Several pruning and training systems have been developed to maximize production and efficiency in the greenhouse. Two popular methods are the umbrella method where plants are pruned so that one main stem grows up the trellis twine up to the main trellis cable after which the meristem is removed and the upper two lateral stems trained to grow back down to the ground. The growing points of the remaining lateral stems are removed once they reach the ground. It is recommended that the hydroponic solution pH for greenhouse cucumber cultivation by maintained at 5.5-6.0 for optimum production (Hochmuth, 2015; Resh, 2008; Papadopoulos, 1994). Harvesting takes place over a 10- to 12-week period when the cucumbers have reached a marketable size (Johnson and Hickman, n.d.).

Aquaponics

Aquaponics is the integration of aquaculture fish production with hydroponic plant production (Rakocy et al., 2016). Anderson, et al. (1989) stated that hydroponic systems afford greater crop yields using less space, water and labor, as well as reducing pollution to the environment compared to in-ground soil production. Combining hydroponic systems with aquaculture production not only yields the benefits of hydroponic production, but further lends itself to sustainability by harnessing the byproducts of one industry for use in another.

Aquaponic systems consist of three biological components: fish, plants, and microbes- primarily nitrifying bacteria. The bacteria bridge the gap between the aquaculture unit and the horticulture unit by converting ammonia, which is toxic to fish, into the plant-available nitrogen source—nitrate (Rakocy et al., 2016). Nitrate is a form of nitrogen that is less toxic to fish and is also one of the primary forms of nitrogen taken up by the plant as an essential element.

Combining aquaculture and horticulture production into an integrated aquaponic system has both environmental and economic benefits (Rakocy et al., 2016). To maximize the value obtained from aquaculture inputs, using aquaculture effluent as fertilizer for horticultural crops has the potential to offset the cost of fish feed by generating secondary income through the sale of horticultural crops (Rakocy et al., 2004). In addition to the economic benefits associated with aquaponics, the aquaculture effluent that would otherwise be discharged into the environment would be re-used as a fertilizer in the horticulture component, thus reducing the potential for environmental damage due to eutrophication from the discharge of the nutrient-rich effluent into the environment.

Various aquaponic systems were developed that make use of the nutrient-rich aquaculture effluent as fertigation for crop species (Diver, 2006). Most notably, Dr. J.E. Rakocy at the University of the Virgin Islands is known for his contributions to aquaponic research. He has developed a re-circulating aquaponic system that produces a variety of horticultural crops including lettuce and basil using floating rafts and staggered production as well as producing Nile Tilapia in the aquaculture component (Diver, 2006; Rakocy et al., 2004). Other systems include the North Carolina State University system developed by Dr. Mark McMurtry (Diver, 2006). The NCSU system is comprised of underground fish tanks that water above-ground vegetable beds. Gravity returns the filtered water back to the aquaculture component.

Early aquaponic system design focused on recirculating systems where the horticulture component served as a biofilter to extract ichthyotoxic compounds from the water and incorporate them into plant tissue. The filtered water is then returned to the aquaculture unit. Recirculating systems not only re-cycle water, but also increase nutrient use efficiency and reduce the discharge of waste into the environment (Rakocy et al., 2004). Fish and plant culture are linked together in these systems known as “coupled systems”. Drawbacks to these systems are that water quality parameters need to accommodate all species within the system.

A newer approach to aquaponic production systems unlinks the fish culture unit from the plant culture unit. These “de-coupled” systems were first developed as non-recirculating systems, which do not return the water back to the aquaculture unit once it has passed through the horticulture unit (Monsees et al., 2017). By adding more horticulture units to the system, the aquaculture effluent can flow until it is used completely by the plants, eliminating effluent runoff. Because these systems are non-recirculating, water quality for each unit can be adjusted separately and optimized for the specific species in each component. Re-circulation of water in each unit separately has been incorporated into de-coupled systems (Kloas, 2015). Not only does de-coupling allow for water quality optimization, it also allows growers to use pesticides that contain ichthyotoxic compounds in the plant unit without affecting the fish in the aquaculture unit. Additionally, there is more flexibility with de-coupled systems in the type of horticulture units that can be used to grow plants. Coupled systems commonly employ floating raft or deep-water culture hydroponic techniques since large amounts of water need to be moved at one time. Drip irrigation and NFT systems can be used in the newer, de-coupled systems, which allows for more variety in the types of crops that can be grown. Where floating rafts and deep-water culture are more suited for production of leafy greens, drip irrigation allows larger, vining crops to be grown with the nutrient-rich water.

pH in Aquaponics

While providing benefits to both the aquaculture and horticulture industries, there are some challenges associated with aquaponic production. As in conventional aquaculture systems water quality is an important factor in aquaponic production as it plays a large role in the ability to produce large fish, fast. As mentioned previously, parameters interrelate to make up the water quality of the aquaculture component of the aquaponic system. These parameters include pH, alkalinity, dissolved oxygen, temperature, and concentrations of various forms of nitrogen and organic matter in the water (Timmons et al., 2001).

A major challenge in managing an aquaponic system is balancing pH between the three biological components since each component operates at specific pH optima. Nitrifying bacteria are most active in wastewater treatment at neutral to slightly basic pH ranges: 7.0-8.5 (Kim et al., 2007). Depending on the species, fish can tolerate a wider range of pH. Tilapia are especially tolerant of lower pH as compared to other fish species (Lim and Webster, 2006), which is one reason why they are a popular choice for aquaponic production. A slightly basic pH is more favorable for fish production (Zou et al., 2016). Plants, on the other hand, depend on pH for nutrient availability and thrive in conditions that are slightly acidic: 5.5-6.5 (Resh, 2008). Several studies have established a reconciling pH for recirculating aquaponic systems between 7.0- 8.0 to favor nitrification at the expense of an optimal plant pH range (Tyson, 2007; Tyson et al., 2008a).

De-coupled aquaponic systems are in a unique position to allow for the pH to be adjusted to suit each unit separately, optimizing production for both units. Monsees (2017) conducted a study that looked at yield differences between coupled and de-coupled systems. The authors observed a 36% increase in tomato production in de-coupled systems (pH 6.4) compared to coupled systems (pH ~7.3). Kloas et al. (2015) also found increased yields in decoupled systems when compared to coupled systems and noted that the de-coupled system had the potential for even higher yields than what was observed over the course of the initial study.

Because pH is one of the main differences between aquaponics and hydroponics, researchers have tried to elucidate its importance in plant production by comparing the two production systems. Wortman (2015) conducted a study comparing the yields of crops grown under conventional hydroponic conditions and simulated-aquaponic conditions (low EC and high pH ~7.5). The study found that the simulated-aquaponic treatment produced lower yields than conventional hydroponics. The author attributed this decrease in yields to the neutral to slightly alkaline pH commonly used in aquaponic operations to accommodate both fish and plant species. Although the study addressed the yield differences in crops at different pH conditions for conventional hydroponics compared to aquaponics, it did not consider nutrient solution compositional differences between the systems. Hydroponic growth solutions contain a complete nutrient solution; however, aquaponic solutions have been observed to contain lower levels of certain nutrients (Rakocy et al., 2004). Common deficiency problems in aquaponic systems are potassium, calcium, magnesium, and iron. Because of the inherent differences in solution composition for aquaponics compared to hydroponics, the difference in growth may not only be a factor of solution pH. It may even be possible that pH influences which nutrients are more available or less available in aquaponic solution.

RAS-type aquaculture systems remove solid wastes before the water moves to the plants, leaving the soluble nutrients in the irrigation water for plant uptake. Liquid aquaculture effluent does not necessarily contain all 16 essential plant elements in sufficient concentrations to support optimum plant growth. Additionally, the pH of aquaculture effluent is commonly held at levels higher than those recommended for plant growth. Several studies address the differences in pH between hydroponic and aquaponic production by using hydroponic solution at various pH levels to simulate the effects of nitrification in aquaponic solution. (Tyson, 2008b; Wortman et al., 2015). Tyson (2008b) concluded that a pH of 7.0 would be a suitable compromising pH for aquaponic cucumber production even though early marketable fruit yields were limited. His results may be best applied to clearwater aquaponic systems in which aquaculture effluent is filtered and solid particles removed. Nutrient availability in solution has the potential to increase or decrease according to solution pH. Phosphorus is often a limiting nutrient in aquaponic production. Cerozi and Fitzsimmons (2016) found decreased phosphorus availability at increasing aquaponic solution pH due to the formation of insoluble calcium phosphate at higher pH. Adjusting solution pH of AE has the potential to make more nutrients, especially those typically lacking in aquaponic production, available for plant uptake.

Conclusion

Aquaponic production has the potential to provide a source of both protein and vegetables in one, integrated system. This not only increases food security, but also decreases the discharge of aquaculture waste into the environment, recycling the nutrients that are already there and would otherwise be released into the environment. In order for aquaponic cucumber production to become a viable option for aquaponic producers, the lack of information regarding its economic feasibility and optimum operating conditions needs to be addressed, especially when it comes to newer system designs. A comprehensive study has yet to be undertaken to address aquaponic cucumber production in a de-coupled, biofloc-type aquaponic system. Based on the need for a better understanding of the best production practices for greenhouse cucumbers in these systems, the effects of production cycle length and pH in aquaponic cucumber production employing these types of systems in east Alabama will be the focus of this research.

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Chapter II

Effect of production cycle length on profitability of cool season cucumber production in a de-coupled aquaponic system

Abstract

Aquaponic systems provide a way for the aquaculture industry to mitigate environmental and economic impacts associated with production, mainly high production costs and effluent runoff. Cucumbers are a major greenhouse crop primarily produced in hydroponic systems. Adapting cucumbers to aquaponic production requires determining a production cycle length that minimizes production costs. Sixteen plots of cucumbers were grown in a de-coupled aquaponic system at four cycle lengths. Yield and input costs were determined for each cycle length: 65, 75, 85, and 95 days after transplant. Revenue from cucumber production increased linearly as production cycle length increased, providing the highest revenue at a 95-day cycle length at 17.80 USD per m^2. The number of cucumbers harvested per plot per day increased by one cucumber every 12.5 days after harvesting began. Estimated costs increased as cycle length increased, but the highest profits were obtained at shorter cycle lengths. In aquaponic systems, shortening production cycle length during the traditional off-season for cucumber production would maximize profits.

Introduction

According to USDA-NASS (2014), cucumbers are the second highest-valued greenhouse-grown crop in the United States. Over 250 acres of greenhouse production in the

United States produced 36.3 thousand tons of cucumbers amounting to $77 million USD of total sales in 2014. Hydroponic cucumbers accounted for 91% of total cucumber production under protected culture. Due to similarities in nutrient-delivery between hydroponic and aquaponic systems, strategies can be adapted to transition hydroponic production to aquaponic production.

Cucumbers are a warm season annual, thriving at temperatures between 26.7 and 29.4°C (80-85°F) with an optimum growing temperature of 27.7°C (82°F) (Hochmuth, 2015; Swaider and Ware, 2002). During the colder months in eastern Alabama, heaters are used to keep greenhouse temperatures high enough for optimum cucumber production in the off-season, which increases production costs. Because cucumbers have a vining growth habit, they will continue to produce fruit on the main and lateral stems until the meristems are removed. The combination of protected culture and the vining growth habit of cucumbers allows the potential for extended growing seasons for cucumber production beyond the traditional 10-12 week harvesting period as well as production in the off-season of traditional field production (Cantliffe et al., 2008). Thus, cucumber production in the off-season has the potential to increase profits from cucumber production due to higher retail costs in the winter months.

Although greenhouse cucumbers are a very popular hydroponic crop, few studies have focused on aquaponic cucumber production. To date, most research on aquaponics that addresses cucumber production has focused on economics of aquaponic systems in general. Several studies have investigated the profitability of aquaponics for both small- and commercial-scale operations. Location and species choice, for both fish and plants, impact the profitability of aquaponics operations as a whole (Asciuto et al, 2019; Bosma et al., 2017). Small scale systems have been shown to be profitable, with labor costs accounting for the main variable cost (Asciuto et al, 2019). On a commercial basis, profitability of aquaponics is more in line with small-scale farming operations (Love et al., 2015). Using aquaculture effluent provides a free source of nutrients to the plants, lowering production costs and potentially increasing profits. Extending the growing season with longer production cycle lengths provides even more potential for increase profits. The objective of this study is to determine if production-cycle lengths alone are able to offset production costs, energy costs in particular, by the additional revenue generation in the traditional off-season for aquaponic cucumber production.

Materials and Methods

Cucumber (*Cucumis sativus* L. 'Delta Star') seeds were sown in 72-count round (58 mL) cells seeding trays (Hydrofarm, Pentaluna, CA) filled with a professional formula germination mix (Sungro, Agawarm, MA). Trays were placed on a heating mat (Phytotronics Inc., Earth City, Missouri) set to 82°F, covered with a humidity dome. and watered by hand as needed. One week after seeding, after the appearance of the first true leaves, seedlings were fertilized once per day until transplant with a nutrient solution containing 150, 80, 200, 150, and 35 mg• L^{-1} N, P, K, Ca, and Mg, respectively from water-soluble 8N-6.5P-30K (Gramp's Original Hydroponic Lettuce Fertilizer, Ballinger, TX), calcium nitrate (15.5N-0P-0K), and magnesium sulfate (10% Mg). On 10 September 2018, seedlings were transplanted into 11-L Dutch buckets (CropKing, Lodi, Ohio) filled with horticultural-grade perlite (Sungro, Agawarm, MA) in a 9.1m x 29.3m double-polyethylene-covered greenhouse with a N to S orientation located at E.W. Shell Fisheries Center, Auburn, Alabama, USA. For the first 31 days of the experiment, the greenhouse was covered with 55% shade cloth.

The experiment was a one-way treatment design composed of four pre-set termination times of 65, 75, 85, and 95 days after transplant (DAT). Plants were arranged in a completely randomized block design to account for a temperature gradient in the greenhouse. Four blocks were arranged in the greenhouse with each block containing one experimental unit per treatment. Each experimental unit consisted of four Dutch buckets containing one cucumber plant each. Two additional Dutch buckets each containing one cucumber plant were placed on each end of the experimental unit to account for shading effects on the edge plants of the plot for a total of 96 cucumber plants.

Cucumber plants were trained and pruned according to the umbrella method (Hochmuth, 2015). The main stem was trained vertically 2.1 m up to a trellis cable. The main meristem was removed when two nodes had grown above the trellis cable. All lateral stems were pruned from the main stem as they appeared except for the two lateral stems directly below where the meristem was removed. Those two lateral stems were allowed to grow back to the ground.

Plants were irrigated with aquaculture effluent (AE) from an aquaculture unit located in an adjacent greenhouse. Irrigation frequency and duration were controlled using an irrigation controller (Sterling 30, Superior Controls, Torrance, CA) and was adjusted throughout the experiment based on plant demand. Water requirements averaged 6 and 8 L water plant^{-1} d^{-1}. Water quality was monitored each morning for pH, electrical conductivity (EC), and NO_3-N using handheld meters (LAQUA twin, Horiba, Kyoto, Japan). An integrated pest management (IPM) program was established prior to the start of the experiment primarily for whitefly control. Mycotrol™ was applied on 2 DAT and again on 56 DAT to control whiteflies.

Fish production was conducted in a 9.1m x 29.3 m greenhouse, covered with a double layer of 6-mL polyethylene glazing, which contained two 102,000-L rectangular tanks each holding an average of 6,000 tilapia (*Oreochromis niloticus)*. The aquaculture unit utilized a modified biofloc-type biofiltration system in which approximately 5% of the tank volume (5,100 L) was exchanged and replaced daily with fresh water which was sourced from a reservoir and fed via gravity to the aquaculture system. Tilapia were fed twice daily until satiation with a commercial aquaculture feed containing 36% crude protein (Cargill, Franklinton, LA). Water quality in the tank was monitored daily for pH, ammonia, and dissolved oxygen levels. Water pH in the aquaculture unit was maintained at approximately 6.5 by adding a hydrated lime slurry several times per week, as needed. Ammonia and dissolved oxygen levels remained within acceptable levels for tilapia production for the duration of the experiment. Suspended solids, including uneaten feed, feces, and biofloc were settled and removed from aquaculture effluent (AE) using two passive clarifiers connected in a series. The clarifiers were 1,500-L cone-bottom tanks located adjacent to the aquaculture unit outside the greenhouse. Aquaculture effluent was continuously pumped into the first clarifier from the aquaculture unit using an air lift and was forced to pass under a solid baffle, which separated the tank into two halves, before being moved to the second clarifier which was used as the irrigation reservoir for the plant greenhouse. The clarifiers removed an average of 50% of suspended solids from AE before it was used to irrigate plants.

Total variable costs of production were estimated for each treatment based on data collected throughout the experiment (Table 2.1). Costs were allocated to each input collected based on the total potential growing area at a planting density of 1.32 plants • m^{-2}. Each day, electricity and propane usage were recorded for the plant production greenhouse. Estimated labor hours were calculated based on average labor hours for greenhouse tomato production, which were assumed to be comparable to cucumber production (Snyder, 2019).

Starting on 8 Oct. 2018, cucumber fruit were harvested as needed at an average weight of 130 g. Harvesting continued until plants were terminated from the experiment at the specified termination time. Yield was recorded as total marketable fruit count per plot and converted to revenue based on the market price for cucumbers (2.64 USD per kg). Marketable fruit were defined as fruit that measured between 15 and 25 cm and were free of mechanical or insect damage.

Revenue each treatment was analyzed using an analysis of variance (ANOVA) via PROC GLIMMIX. (SAS 9.4, Cary, NC). Block was treated as a random variable. The rate of cucumber production over time was analyzed as a simple linear regression.

Results and Discussion

Revenue from cucumber production increased linearly by approximately $0.23 USD as production cycle length increased (Table 2.2). A 95-day cycle length corresponded to a 63% revenue increase over the 65-day cycle. An 85-day cycle length produced a 36% revenue increase compared to the 65-day cycle. The revenue increase for longer cycle lengths can be attributed primarily to the extended production season for the treatments. The plants had additional time and access to resources for growth and fruit production, thus producing more cucumbers over that time period. The rate of cucumber fruit production over the course of the harvesting season increased slightly. Each plot, consisting of four plants, produced an average of one additional cucumber every 12.5 days after harvesting began, corresponding to an increase in one cucumber per plant every 50 days.

Estimated costs increased as cycle length increased (Table 2.2). Increased costs later in the season can be attributed to liquid propane (LP) used to heat the greenhouse in the colder months. High fuel usage in greenhouse cucumber production was observed in Iran (Mohammadi and Omid, 2010) where the fuel was used primarily for heating purposes and accounted for the highest proportion of input costs for production. Labor costs were estimated at 20 hours per week for the entirety of the production cycle. Labor and LP were the two largest inputs to production costs (Figure 1). As the production season lengthened, daily electricity usage decreased while propane usage increased (Figure 2). Additionally, as the length of the production cycle increased, propane made up more of the total cost of production since heating increased as production cycle length increased, which consequently lowered the proportion of labor cost to total cost.

Feed costs were not considered as a variable cost for cucumber production using AE since feed is required for fish production whether or not the nutrient-rich water that it results in is used for plant production.

The largest profit was found at shorter cycle lengths (Table 2.2). Since LP was being consumed primarily towards the end of the growing season, increased costs for longer cycle-lengths reduced profits as cycle length increased. Cucumber price was held stable at 2.64 USD per kg (1.20 USD/lb) for the purposes of this study; however, seasonal changes in cucumber availability cause the retail price of cucumbers to fluctuate (Figure 3). A sensitivity analysis can determine if price fluctuations are able to further increase profits or aquaponic cucumber production. If price fluctuations are taken into account, greenhouse cucumber production in colder months has the potential to generate an even higher return due to increased retail prices during the colder months, but only if customer demand is high. Most aquaponic operations primarily grow lettuce and other leafy greens (Love et al, 2015; Mchunu et al., 2018). The fast turnaround of these crops compared to the longer maturity time required for fruiting vegetables allows multiple crops to be harvested per year— especially during the cool season when it is less expensive to grow leafy greens due to the lower optimum growth temperature.

Conclusion

While revenue generated from aquaponic cucumber production increased as a linear function of increasing cycle length, profits at shorter cycle lengths were higher. Revenue was highest for the longest cycle length of 95 days; however, the 95-day cycle length also had the highest cost. LP was the main contributor to total cost as cycle length increased. For small aquaponic operations, shorter cycle lengths for fall cucumber production would take advantage of the warmer temperatures earlier in the season to cut back on input costs later in the season. Additionally, switching to a crop that tolerates cooler temperatures in the greenhouse, such as lettuce, during cool season production could be one strategy to optimize production without increasing energy inputs. These practices would increase profits from aquaponic cucumber production, making it a viable option for small-scale aquaponic producers.

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Tables and Figures

Table 2.1 Inputs and multipliers used to determine variable costs of cool-season aquaponic cucumber production (September-December 2018)[z]

Input	Unit	Total amount of input used during experiment	Cost multiplier (USD• unit^{-1})	Area allocated to input
Propane	Liters	2,101	0.36	267m^2
Electricity	kWh	2,851	0.09	
Labor	Hours	336[y]	7.25	
Pesticides	mL	8	0.11	$96\,plants \div \frac{1.32\,plants}{m^2}$

[z] Cost estimate was calculated by dividing the product of the total amount of input per day and the cost multiplier for the input by the area allocated to the input.

[y] Estimate based on greenhouse tomato production (Greenhouse Tomato Handbook: Mississippi State University Extension Publication No. 1828.)

Table 2.2 Aquaponic cucumber revenue and cost (USD per m^2) at 10-day cycle-length intervals

Cycle-Length	Revenue	Cost	Profit
95	17.80a[z]	15.86	1.98a
85	14.89b	12.35	2.54b
75	12.57c	9.26	3.31c
65	10.94c	6.85	4.09c
Significance	L***[y]	N/A	L***

[z] Means with the same letters in a column are not significantly different at ($P<0.05$) as determined by analysis of variance and lsmeans using the GLIMMIX procedure and type III sum of squares in SAS.

[y] Significance established with an analysis of variance using the GLIMMIX procedure and type III sum of squares in SAS. L= linear. ***= significance at alpha=0.0001.

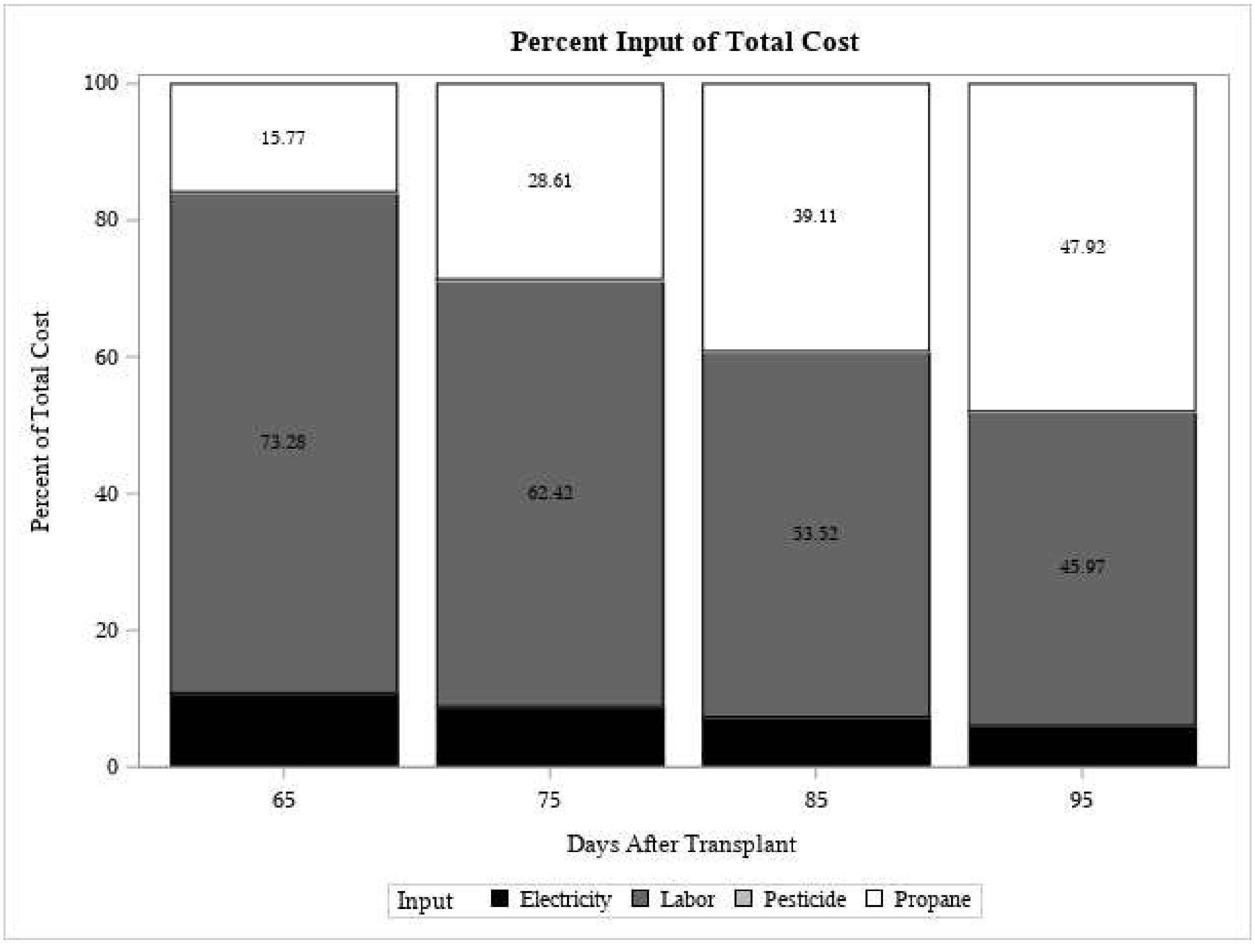

Figure 1. Input contribution to total cost of cool season aquaponic cucumber production.

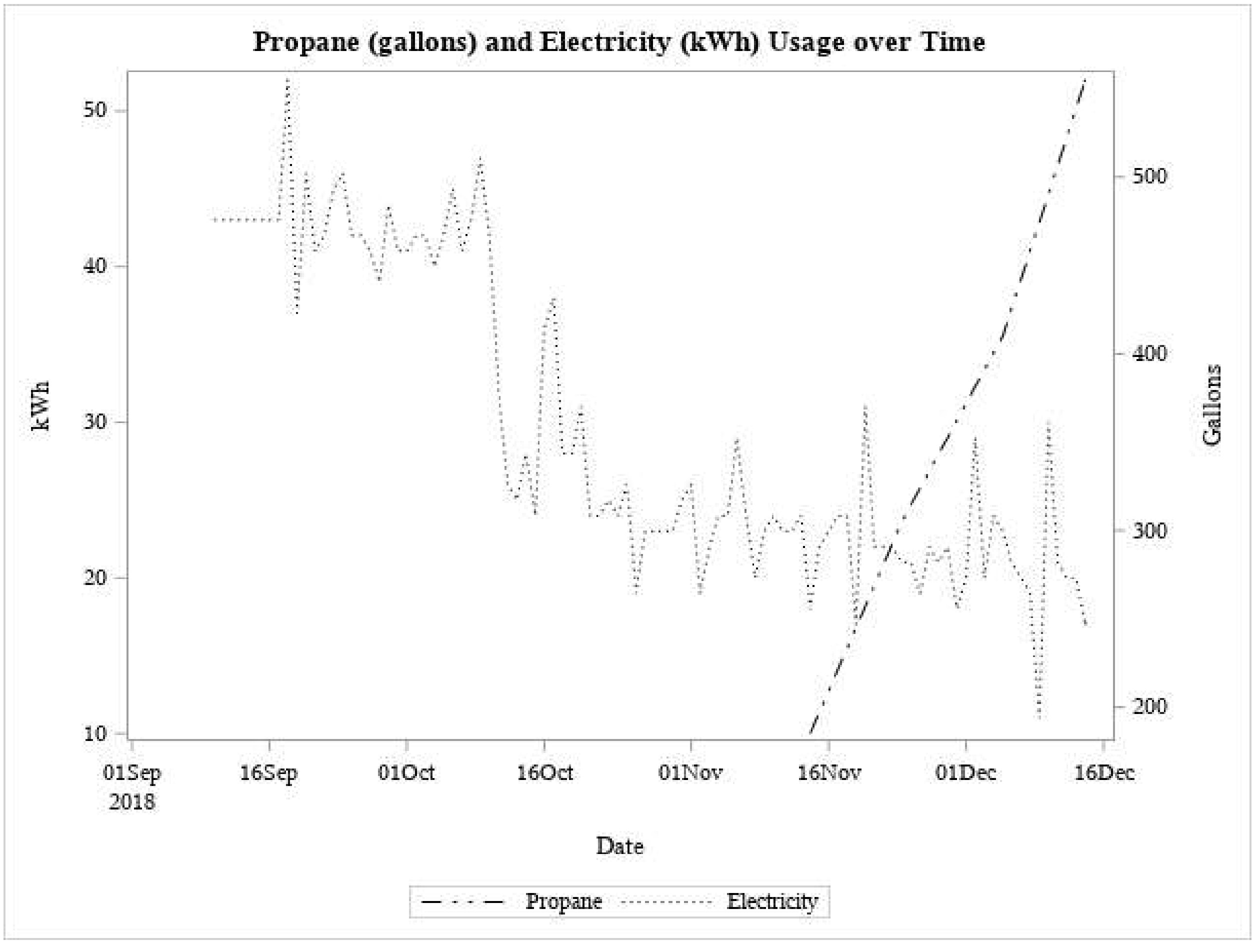

Figure 2. Decrease in electricity usage and increase in propane usage over fall production season for cucumber production using aquaculture effluent as irrigation source.

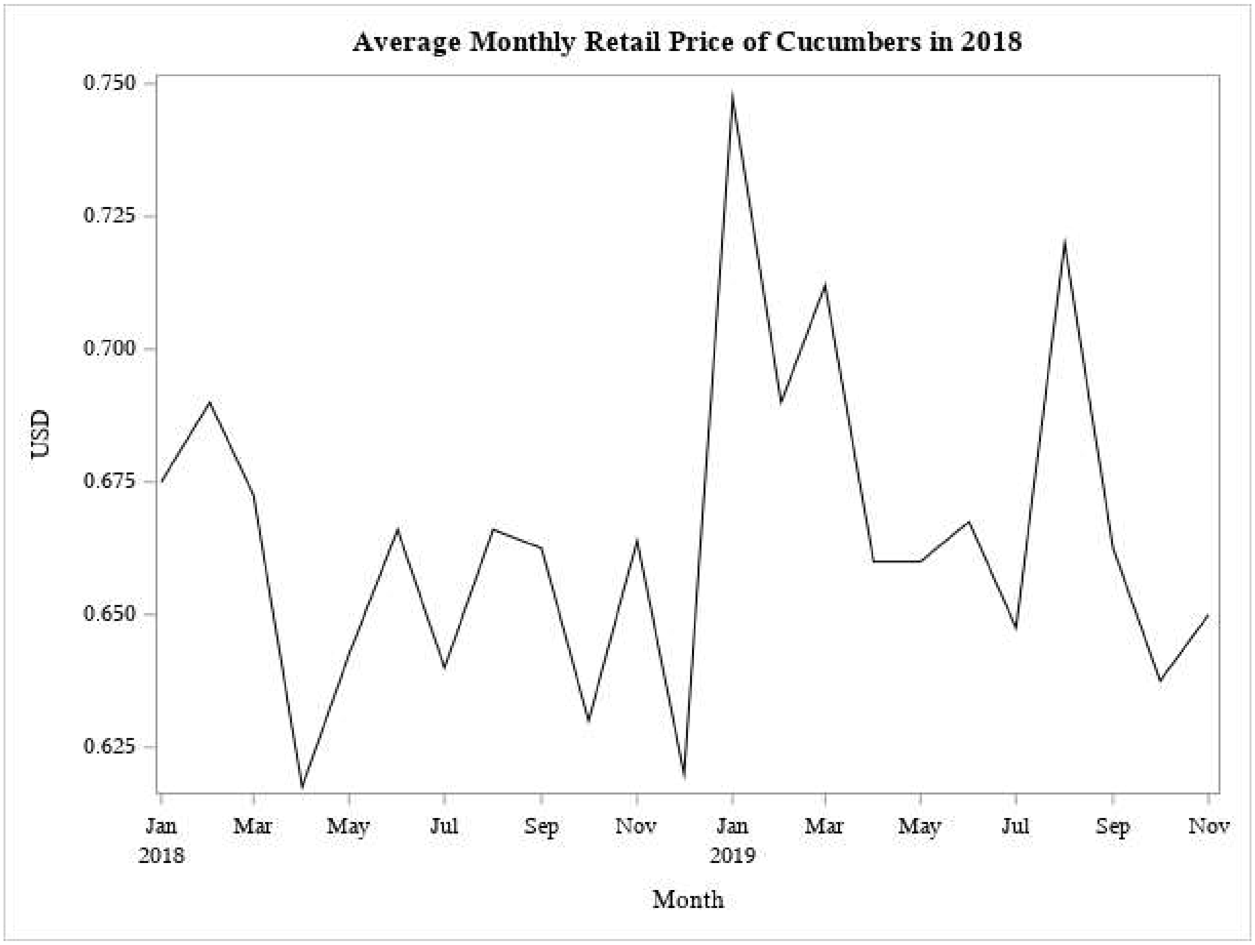

Figure 3. Seasonal fluctuations in retail cucumber prices from January 2018 to November 2019 (USDA-AMS,2019).

Chapter III

Effect of pH on cucumber growth and nutrient availability in a de-coupled aquaponic system with minimal solids removal

Abstract

To determine the effect of pH in a de-coupled aquaponic system, a study was conducted using aquaculture effluent from tilapia culture tanks at 4 pH treatments: 5.0, 5.8, 6.5, and 7.0 to irrigate a cucumber crop. Growth and yield parameters, nutrient content of the irrigation water, and nutrients incorporated into the plant tissue were collected over two growing seasons. pH did not have a practical effect on growth rate, internode length or yield over the two growing seasons.

Availability and uptake of several nutrients were affected by pH, but there was no over-arching effect that would necessitate its use in commercial systems. Nutrient concentrations in the aquaculture effluent would be considered low compared to hydroponic solutions; however elemental analysis of leaf tissue were within the recommended ranges Research into other nutrient sources provided by the system (i.e. solid particles carried with the irrigation water) would provide further information into the nutrient dynamics of this system.

Introduction

Aquaponics combines the nutrient delivery method of hydroponics with aquaculture by taking nutrient-rich wastewater from aquaculture production and using it as a nutrient solution for horticultural crops. Aquaculture production maintains pH levels between 7.0-8.5 to favor nitrification in the biofilters (Timmons, 2012), which allows more of the toxic NH_3 to be converted to NO_3. In regard to plant production, pH plays a vital role in the nutrition of soil-based systems (Foth and Ellis, 1997) as well as hydroponic systems (Hochmuth, 2015; Resh, 2008), with a recommended pH range for plant growth at 5.5-6.5. Different from conventional soil production, aquaponic plant production takes advantage of nutrients dissolved in solution rather than adsorbed to the surface of soil particles. Because aquaponics is linked to aquaculture production in coupled systems, the pH of the irrigation water is typically maintained at a level that favors fish production and nitrification rather than plant production, reconciling both systems at a pH of 7.0 (Tyson, 2007). Newer aquaponic system designs de-couple the aquaculture and horticulture units. These de-coupled systems allow each unit to be optimized separately. Fertilizer supplements can be added, and pH adjusted in the plant unit without affecting the water quality in the fish unit.

Cucumbers are a warm season annual, thriving at temperatures between 26.7 and 29.4°C (80-85°F) with an optimum growing temperature of 27.7°C (82°F) (Hochmuth, 2015; Swaider and Ware, 2002). Cucumbers are a major greenhouse crop primarily produced in hydroponic systems (USDA NASS, 2014). Cucumber production pH is managed at 5.5-6.5, levels commonly used for plant production. However little research has been done to investigate the effect of pH on greenhouse cucumber production in de-coupled biofloc-type aquaponic systems.

Several studies address the differences in pH between hydroponic and aquaponic production by using hydroponic solution at various pH levels to simulate the effects of nitrification in aquaponic solution. (Tyson, 2008; Wortman et al., 2015). Tyson (2008) concluded that a pH of 7.0 would be a suitable compromising pH for aquaponic cucumber production even though early marketable fruit yields were limited. Tyson's and others' results may be best applied to clearwater systems in which organic solids are removed. Monsess et al. (2017), showed that there are nutrients in AE solids, which contain mostly calcium (0.5%) as well as phosphorus (0.3%), a nutrient commonly limited in aquaponics. Sulfur, magnesium, iron, and manganese contributed to solid composition at 0.06, 0.03, 0.03, and 0.002% respectively. These nutrients could be made available to plants, providing adequate nutrition in aquaponic production. Monsees reported aerobic treatment of sludge increased phosphorus (330%) and potassium (31%). Aerobic conditions, such as those available in the perlite media could provide opportunities for nutrients to be made available to plants. Therefore, a study was designed to determine the effects of pH on cucumber growth and yield in a de-coupled, media-based biofloc-type aquaponic system with minimal solids removal.

Materials and Methods

Two experiments occurred in Spring and Summer 2019 using two greenhouse facilities. Cucumber seeds were grown for transplants for 21 days at the Patterson Greenhouse Facility, Auburn University, Alabama, USA. The main experiment was housed in a commercial-sized greenhouse located at E. W. Shell Fisheries Center, Auburn, Alabama, USA. The 30' x 96' greenhouse was covered with a double layer of polyethylene sheet and oriented North to South. The greenhouse was set up for vining crop production with five rows arranged 1.2m apart.

Cucumber (*Cucumis sativus* L. ‘Delta Star’) seeds were sown in 72-count round (58 mL) cells seeding trays (Hydrofarm, Pentaluna, CA). Seedlings were transplanted upon emergence of true leaves into 11-L rectangular Dutch buckets (Crop King Inc., Lodi, Ohio) containing 100% perlite media.

Fish production was conducted in a 9.1m x 29.3m double polyethelene-covered greenhouse which contained two 102,000-L rectangular tanks each holding an average of 6,000 tilapia (*Oreochromis niloticus)*. The aquaculture system was operated as an autotrophic, biofloc system in which an external nutrient source was not applied to promote biofloc production. Approximately 5% of the tank volume (5,100 L) was used for irrigation of plants and replaced with fresh water daily which was sourced from a series of ponds and fed via gravity to the aquaculture system. Tilapia were fed twice daily until satiation with a 36% crude protein commercial aquaculture feed (Cargill, Franklinton, Louisiana). Water quality in the tanks was monitored daily for pH, ammonia, and dissolved oxygen levels. Water pH was maintained at 7.0 for fish production by adding a hydrated lime slurry several times a week as needed to raise pH to the appropriate level. Ammonia and dissolved oxygen levels remained within acceptable levels for fish production for the duration of the experiment. Suspended solids, including uneaten feed, feces, and microbial flocs were settled and removed from aquaculture effluent (AE) using two passive clarifiers connected in a series. The clarifiers were 1,500-L cone-bottom tanks located adjacent to the aquaculture system outside the greenhouse. Aquaculture effluent was continuously pumped into the first clarifier using an air lift and was forced to pass under a solid baffle separating the tank into two halves before being moved to the second clarifier which was used as the irrigation reservoir for the plant greenhouse. The clarifiers removed an average of 50% of suspended solids from AE before it was used to irrigate plants.

Four pH treatments with target levels at 7.0, 6.5, 5.8 and 5.0 were randomly assigned to 16 plots in a randomized complete block experimental design to account for a temperature gradient in the greenhouse. The four blocks were arranged in the greenhouse with each block containing one experimental unit per treatment (Figure 4). Each experimental unit consisted of four Dutch buckets containing one cucumber plant each. Two additional Dutch buckets each containing one cucumber plant were placed on each end of the experimental unit to account for shading effects on the edge plants of the plot and to serve as plants for destructive tissue analysis throughout the season for a total of 96 cucumber plants.

Three Chemilizer chemical injectors (Hydro Systems Company, Cincinnati, Ohio) were installed in the research greenhouse to inject acid (1M citric acid in Spring 2019, and 33% Sulfuric acid in Summer 2019) to lower the irrigation water to the target pH. Citric acid was used to adjust pH in the spring trial, however target levels were not reached. Sulfuric acid was used in the summer trial, and while treatment pH was lower compared to the Spring trial, target pH levels were still not reached. pH is managed in the fish tank by using hydrated lime to increase pH, which creates a highly buffered system. Trials were analyzed separately due to acid type and seasonal changes. Acid was mixed into irrigation water post-injection using an in-line static mixer (Johnson Screens®, St. Paul, Minnesota) to ensure proper mixing of the acid with irrigation water. Acid was not added to one treatment level, target pH of 7.0, to observe the effects of unadjusted aquaculture effluent on plant growth. While pH was not adjusted in the horticulture unit for the 7.0 treatment, pH of the fish tank was monitored and adjusted daily by the addition of hydrated lime to the tank.

Plants received between 6 and 8 liters of AE each day during daylight hours using a Sterling 30 irrigation controller (Superior Controls, Torrance, California) set to water at specific time intervals set by the grower. Irrigation was set higher in the summer to prevent water stress due to increased light intensity and temperatures. Water quality was monitored throughout the duration of the experiment for pH, E.C., and NO_3-N using a HI9813-6 Portable pH/EC/TDS/Temperature Meter (Hanna Instruments, Smithfield, Rhode Island). and L-AQUA twin handheld meters (Horiba, Kyoto, Japan).

Vines were trained up Bato bobbins strung with twine (Crop King, Lodi, Ohio) 2.1 meters to the main trellis cable. Once vines reached the main trellis cable, they were leaned and lowered to continue growing, similar to tomato production practices. Lateral stems were removed as they appeared along the main stem according to Hochmuth (2015).

Fruit was harvested once reaching a marketable size (310 grams in Spring 2019 and 402 grams in Summer 2019). Marketable fruit were defined as fruit that measured between 15 and 25 cm and were free of mechanical or insect damage.

Initial height, final height, and number of nodes (the point of emergence of a new leaf or lateral stem from the main stem) was collected on each plant and averaged across the four plants in each experimental unit to obtain information on growth rate throughout production. Yield was calculated as a sum of marketable fruit at the end of a 60-day cycle length. Tissue samples were collected as a composite sample of the two end plants on each plot for each experimental unit (n=16) from the most-recently matured leaves on 30 DAT and 60 DAT and subjected to elemental analysis by Inductively Coupled Plasma-Emission Spectroscopy (ICP_ES) using AOAC official method 985.01(OMA, 2012) at Waters Agricultural Laboratory in Camilla, Georgia. Water samples from the emitter of each plot (n=16) were taken at 30 DAT and 60 DAT and analyzed for plant-essential nutrient levels at Auburn University's Soil Lab in Auburn, Alabama using Inductively Coupled Atomic Plasma (ICAP) Analysis. AE for solids collection was captured from emitters for each treatment in one block. AE was centrifuged at 4000 rpm for 15 minutes until 2g of wet solids were collected. Solids were dried at 105°C for 24h and analyzed using ICP-ES using AOAC official method 985.01(OMA, 2012) for total nutrient content (Waters Agricultural Laboratory, Camilla, GA).

Data were analyzed using SAS software (SAS Institute, Cary, North Carolina) as an analysis of variance (ANOVA) via PROC GLIMMIX and LSMEANS. (SAS 9.4, Cary, NC). Block was treated as a random variable. Nutrient concentrations in the water samples were compared to standard hydroponic solution standards (Resh, 2008) for cucumber production for each treatment using Dunnett's Test via PROC TTEST in SAS. Element concentrations in plant tissue were compared against standard plant tissue levels (Mills and Jones, 1996) for cucumbers using PROC TTEST in SAS.

Results

A target pH of 7.0 resulted in a 9.5% increase in growth rate compared to the 5.0 treatment in Spring 2019 (Table 3.1), but a similar trend was not observed in Summer 2019. No other measured growth and yield factors, including internode length and total yield, were influenced by target pH treatment in either season (Table 3.1).

Midseason soluble macronutrient ion concentrations in aquaculture effluent (AE) were generally not affected by pH treatment with the exception of potassium (K) in Summer 2019 which decreased only 1.68%, from 175 to 172 mg•L^{-1} K, as target pH decreased from 7.0 to 5.0 (Table 3.2). Macronutrient uptake was not generally influenced by target pH treatment with the exception of foliar P which decreased quadratically by 16% in Spring 2019 and linearly by almost 18% in Summer 2019 (Table 3.3).

By the end of each trial soluble NO_3-N concentrations decreased as target pH decreased by 19% and 6% in Spring and Summer 2019, respectively (Table 3.4). Soluble calcium (Ca) and magnesium (Mg) in AE increased by 7% as target pH decreased from 7.0 to 5.0 each by the end of Summer 2019 (Table 3.4). Similar to midseason results, macronutrient uptake was generally not affected at the end-of-season by target pH treatments with the exception of Ca which decreased linearly by nearly 7% as target pH decreased from 7.0 to 5.0 (Table 3.5). This decrease in Ca uptake correlated seemingly well to the observed decrease in soluble Ca in AE at the same timeframe. However, a similar trend for Mg uptake was not observed.

Soluble micronutrient ions were generally non-detectable or extremely low in AE at each sampling date in both Spring and Summer 2019 (Tables 3.6 and 3.7). At midseason, foliar Mn uptake followed a quadratic trend in relationship with target pH, decreasing by 36% as target pH lowered from 7.0 to 6.5, then increasing by 41% and 29% as target pH decreased to 5.8 and 5.0, respectively (Table 3.8). However, micronutrient uptake was generally not affected by target pH treatments at either sampling dates in Spring and Summer 2019 (Tables 3.8 and 3.9).

Macronutrients in the solids deposited from the emitter contained phosphorus and calcium in the highest amounts (Figure 3.10). There were comparatively low amounts of micronutrients, boron and copper, in particular (Figure 3.11). Of the micronutrients, Manganese and Iron were present in the highest amounts.

Discussion

Nutrients in Aquaculture Effluent

Nitrates in the summer 2019 trial showed a general trend of increasing concentration with increasing pH (Tables 3.2 and 3.4), which supports the trend observed by Zou in 2016.

Soluble K increased 1.68% from pH 5.0 to 7.0 (Table 3.2), which is not practical to consider when using pH for the sole purpose of increasing its availability.

Calcium was added to the fish culture tank in the form of hydrated lime to maintain the pH at 7.0. It is possible that the added calcium in solution reacted with the sulfuric acid used to adjust irrigation pH for the plant greenhouse to form calcium sulfate. Shukla (2008) found that the solubility of calcium sulfate increased with decreasing pH, which could explain the resulting increased calcium observed at lower pH in the summer trial. Magnesium is applied to the system through hydrated lime and magnesium oxide (MgO) in the fish feed. Uneaten feed can become available to the plants as small particles carried by the irrigation water. Solubility of MgO in water occurs more readily under acidic conditions (Lewis, 2007), which could account for the observed increase in Mg under the low-pH treatments in the summer trial.

When compared against nutrient concentrations in standard hydroponic solution for cucumber production, all nutrients, except for Zn and Cu**,** in both mid- and late- season measurements were observed to be statistically lower than concentrations recommended for hydroponic growth (Resh, 2008).

Nutrients Assimilated into Plant Tissue. *S*everal individual nutrients showed changes in incorporation into plant tissue due to pH at the ranges reached for this study.

pH has been shown to have an effect on phosphorus availability in solution (Cerozi and Fitsimmons, 2017); however, this trend was not observed in this study, potentially due to the inability of the acid injection to reach the target pH. pH was observed to have an effect on phosphorus uptake, indicating that pH could have a two-fold effect on phosphorus availability and uptake. Although it has been shown that the pH of the apoplast, the space between the cell wall and the cell membrane, influences phosphorus uptake where high pH (7.0) reduced uptake (Sentenac and Grignon, 1985), this trend was not observed in this study.

Several other macronutrients showed inconsistent changes with pH from mid-season to late-season measurements and across trials. While light and temperature changes occurred due to changing seasons, yield and growth was not statistically different between seasons. Nitrate assimilation into leaf tissue varied significantly with pH during the mid-season measurements of the summer 2019 trial showing highest assimilation at pH 5.8 and the lowest at pH 6.5, a 5% increase between the two treatments (Table 3.3). This was not observed in the late-season measurements nor at all in the spring trial. Calcium uptake showed the highest levels at pH 6.5 in the late-season measurements of the summer 2019 trial, a 9.1% increase from 5.0 to 6.5 (Table 3.5). Differences in mid- to late- season measurements could be due to nutrients being allocated to different plant organs at different rates according to seasonal changes or changes physiological requirements once fruit-set and development occurs (Clark and Boldingh, 1991; Mengel and Kirkby, 2001).

Manganese was the only micronutrient whose assimilation into plant tissues was affected by the AE pH. In soil-based systems, manganese uptake occurs by facilitated diffusion in soil-systems and decreases with the addition of lime to the soil. The decreased uptake at higher pH observed could be due to the addition of lime to the irrigation source to manage pH in the fish culture tank.

When compared against standard tissue concentrations for healthy cucumber tissue (Mills and Jones, 1996), all nutrients- both mid- and late- season for the spring and summer trials were observed to be statistically higher than lower range of recommended tissue concentrations for cucumber leaf tissue.

Preliminary analysis of solids carried in the irrigation water was consistent with observations by Monsees et al. (2017). Phosphorus and calcium constituted the largest proportion of the solids. Additionally, Monsees et al. found that mobilization of these nutrients through aerobic digestion further increases the amount of phosphorus and potassium available. High levels of manganese and iron were observed in the captured solids, compared to low levels of boron and copper. Boron and copper were also observed to be low in solution. Despite low levels of nutrients, both in solution and contained in solids, compared to levels observed in hydroponic solution (Table 3.12), cucumber plants were not deficient in plant essential nutrients. Examination of raw numbers of nutrient levels in both the solid and liquid fraction of aquaculture effluent may not be enough to capture the effects of nutrient flow in the system. It is possible that there are interactions occurring in the media over time that allow for adequate plant nutrition.

Conclusion

Certain nutrients are affected by the pH of AE either by influencing the availability of the nutrient in solution or by influencing the nutrient's uptake and assimilation into plant tissue. However, there is no over-arching increase in plant nutrient availability or uptake to necessitate the use of pH adjustment in these systems. Furthermore, the observed effects of pH on plant essential nutrients did not translate to an increase in yield at the pH range observed in this study.

The acidifying agent used to manage pH should also be taken into further consideration. Phosphoric and nitric acids are commonly used to manage pH in hydroponic systems, however they add important macronutrients, phosphorus and nitrogen, to the system. Citric acid, while not contributing any plant essential elements to solution, is a weak acid and was not able to overcome the buffering capacity of the tank solution. Sulfuric acid was used in the summer trial to try to overcome the buffering capacity and reach target pH. Using sulfuric acid provided unique opportunities in the summer trial for subsequent reactions to take place and make certain nutrients available that were not observed when citric acid was used in the spring trial. Sulfuric acid added sulfur to the system, which not only increased the amount of sulfur available to the plants, but also had the potential to react with other plant nutrients, such as calcium, and precipitating out of solution. Sulfur incorporated into plant tissues was slightly higher in the summer trial compared to the spring trial, but yield was not statistically different.

Another interesting observation was the sufficient levels of plant essential elements incorporated into the plant tissues despite the poor nutrition of the irrigation water observed in relation to the hydroponic solution standard. The cucumber plants were not deficient in any essential plant nutrients even with the irrigation water under-supplying nutrients. This indicates that there are nutrients coming from another source and being made available for plant uptake. This could be from the accumulation of solids in the container over time, allowing plant roots to mine for nutrients in the container, or even from aerobic processes increasing available nutrients in the solid particles, which Monsees et al. (2017) observed in aquacultural sludge. Further research that looks into the nutritional composition of suspended solids carried in the irrigation water would provide additional information on the nutrient dynamics in this system.

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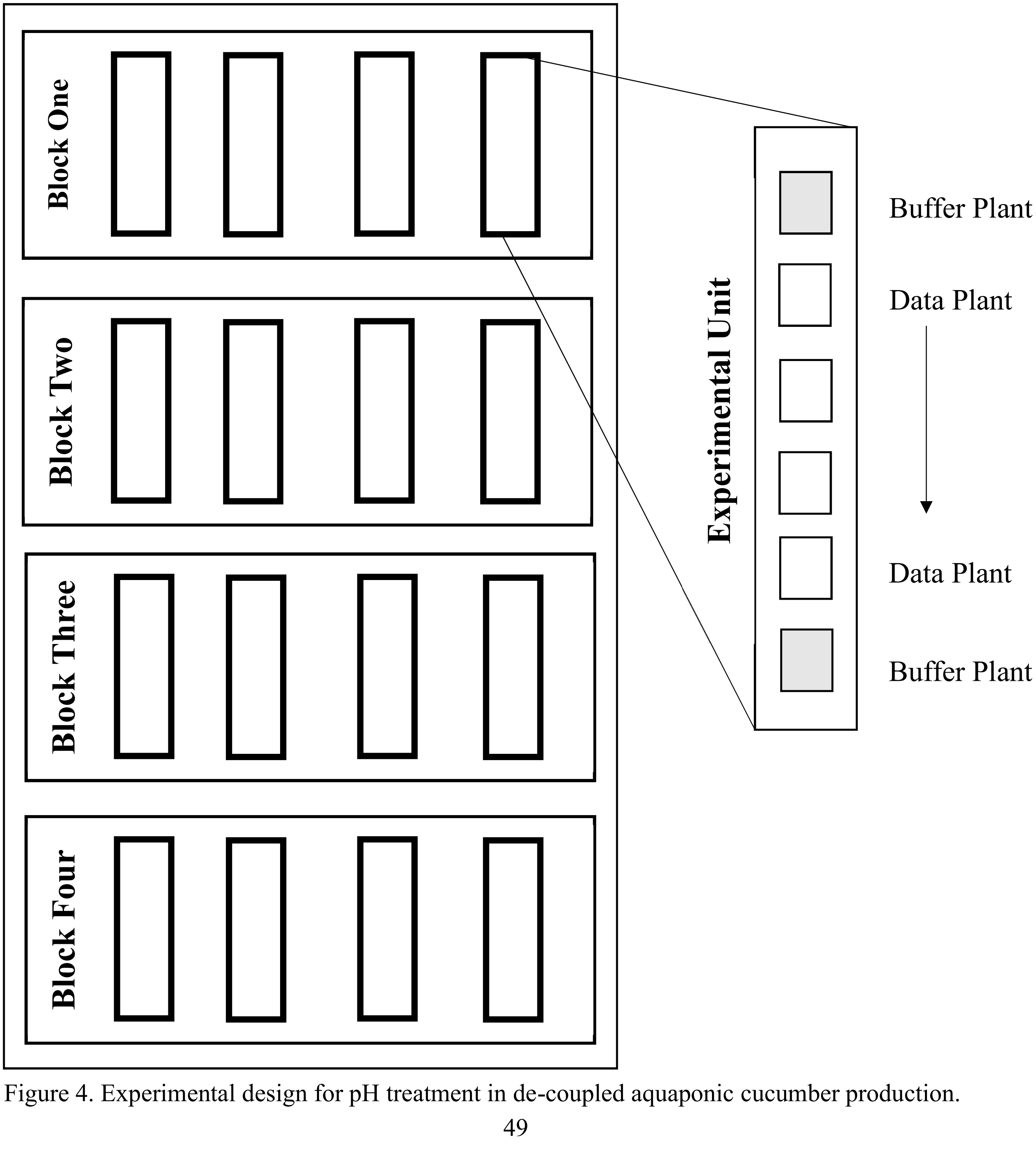

Figure 4. Experimental design for pH treatment in de-coupled aquaponic cucumber production.

Table 3.1 Growth and yield of aquaponic cucumber at four pH treatments

Target pH[z]	Actual pH (±S.E.)	Growth Rate (cm•day^{-1})	Internode Length (cm)	Yield (kg•plant^{-1})
Spring				
7.0	6.9±0.16[y]	4.95a[x]	8.66n.s.	6.59n.s.
6.5	6.7±0.22	4.70ab	8.63	6.01
5.8	6.4±0.30	4.67ab	8.58	6.63
5.0	6.3±0.46	4.75b	8.43	7.01
Summer				
7.0	6.7± 0.22[w]	9.80n.s.	9.50n.s.	8.39n.s.
6.5	6.6±0.22	9.73	9.70	8.24
5.8	6.3± 0.45	9.65	9.65	8.84
5.0	6.1±0.70	9.98	9.70	8.99

[z] Target pH based on conventional RAS (7.0), conventional agriculture (6.5), conventional hydroponics (5.8) and below plant- and fish- recommendation (5.0).

[y] Spring pH levels were managed using citric acid.

[x] Means with the same letters in a column are not significantly different at (P<0.05) as determined by analysis of variance and lsmeans using the GLIMMIX procedure and type III sum of squares in SAS. n.s.=not significant.

[w] Summer pH levels were maintained with 33% sulfuric acid.

Table 3.2 Mid-Season macronutrient analysis of aquaponic irrigation water at four pH treatments[zy]

Target pH	Actual pH (±S.E.)	NO_3-N	P	K	Ca	Mg
Spring[x]						
7.0	6.9±0.16	187n.s.[w]	8n.s.	104n.s.	187n.s.	20n.s.
6.5	6.7±0.22	183	8	104	186	20
5.8	6.4±0.30	184	8	104	188	20
5.0	6.3±0.46	171	8	104	187	20
Significance	N/A	N/A	N/A	N/A	N/A	N/A
Summer[v]						
7.0	6.7± 0.22	93a	11.n.s.	175a	108n.s.	20n.s.
6.5	6.6±0.22	86bc	11	173b	106	19
5.8	6.3± 0.45	88b	12	173b	106	19
5.0	6.1±0.70	85c	12	172b	105	19
Significance	N/A	N/A	N/A	L*[u]	N/A	N/A
Recommended Level[t]	N/A	216[u]	58	286	185	185

[z] All units are in $\text{mg}\cdot\text{L}^{-1}$

[y] When compared against standard hydroponic solution for cucumber production (Resh, 2008), all measured levels of plant-essential elements were lower than the recommended amount for hydroponic production according to t-test results using the TTEST procedure in SAS.

[x] Spring pH levels were managed using citric acid.

[w] Means with the same letters in a column are not significantly different at ($P<0.05$) as determined by analysis of variance and lsmeans using the GLIMMIX procedure and type III sum of squares in SAS. n.s.=not significant.

[v] Summer pH levels were maintained with 33% sulfuric acid.

[u] Significance established using trend analyses in PROC GLIMMIX. L= linear trend. *= significance at alpha=0.05. N/A= no trend test conducted because of non-significant ANOVA.

[t] Sufficiency levels obtained from Jones, 2005.

[u] ppm total nitrogen

Table 3.3 Mid-season foliar analysis of macronutrients as percentage of dry matter for aquaponic cucumbers at four pH treatments[z]

Target pH	Actual pH (±S.E.)	N	P	K	Ca	Mg	S
Spring[y]							
7.0	6.9±0.16	4.98n.s.[x]	0.68a	3.95n.s.	8.36n.s.	0.58n.s.	0.76n.s.
6.5	6.7±0.22	4.96	0.52b	3.65	8.16	0.56	0.74
5.8	6.4±0.30	4.88	0.54b	3.59	7.65	0.54	0.74
5.0	6.3±0.46	4.90	0.57b	3.31	8.28	0.58	0.74
Significance[w]	N/A	N/A	Q*	N/A	N/A	N/A	N/A
Summer[v]							
7.0	6.7± 0.22	5.72ab	0.62a	3.19n.s.	4.56n.s.	0.46n.s.	0.98n.s.
6.5	6.6±0.22	5.57c	0.51b	3.38	4.71	0.51	0.90
5.8	6.3± 0.45	5.85a	0.54b	3.18	4.10	0.51	1.09
5.0	6.1±0.70	5.68bc	0.51b	3.31	4.47	0.46	1.03
Significance	N/A	N/A	L*	N/A	N/A	N/A	N/A
Sufficiency Levels[u]	N/A	4.30	0.30	3.10	2.40	0.35	0.32

[z] When compared against standard percentages in cucumber dry matter (Mills and Jones, 1996), all nutrients were above the lower range of recommended percentage of dry matter for cucumber growth according to t-test results using the TTEST procedure in SAS.

[y] Spring pH levels were managed using citric acid.

[x] Means with the same letters in a column are not significantly different at (P<0.05) as determined by analysis of variance and lsmeans using the GLIMMIX procedure and type III sum of squares in SAS. n.s.=not significant.

[w] Signifiance established using trend analyses in PROC GLIMMIX. L=linear trend, Q=quadratic trend. *= significance at alpha=0.05. N/A= no trend test conducted because of non-significant ANOVA.

[v] Summer pH levels were maintained with 33% sulfuric acid.

[u] Sufficiency levels obtained from Mills and Jones, 1996.

Table 3.4 End-of-season macronutrient analysis of aquaponic irrigation water at four target pH treatments[zy]

Target pH	Actual pH (±S.E.)	NO_3-N	P	K	Ca	Mg
Spring[x]						
7.0	6.9±0.16	110a[w]	20n.s.	171n.s.	139n.s.	23n.s.
6.5	6.7±0.22	103ab	15	167	137	23
5.8	6.4±0.30	98bc	19	154	126	22
5.0	6.3±0.46	89c	13	168	131	23
Significance[v]	N/A	L**	N/A	N/A	N/A	N/A
Summer[u]						
7.0	6.7± 0.22	96a	12n.s.	98n.s.	110b	28c
6.5	6.6±0.22	93ab	13	98	111b	29bc
5.8	6.3± 0.45	93ab	14	98	115a	29b
5.0	6.1±0.70	90b	13	98	118a	30a
Significance	N/A	N/A	N/A	N/A	L***	L***
Recommended Levels[t]	N/A	216[u]	58	286	185	185

[z] All units are in mg·L^{-1}

[y] When compared against standard hydroponic solution for cucumber production (Resh, 2008), all measured levels of plant-essential elements were lower than the recommended amount for hydroponic production according to t-test results using the TTEST procedure in SAS.

[x] Spring pH levels were managed using citric acid.

[w] Means with the same letters in a column are not significantly different at ($P<0.05$) as determined by analysis of variance and lsmeans using the GLIMMIX procedure and type III sum of squares in SAS. n.s.=not significant.

[v] Significance established using trend analyses in PROC GLIMMIX. L=linear trend. **, ***= significance at alpha=0.01 and 0.0001 respectively. N/A= no trend test conducted because of non-significant ANOVA.

[u] Summer pH levels were maintained with 33% sulfuric acid.

[t] Sufficiency levels obtained from Jones, 2005.

[u] ppm total nitrogen.

Table 3.5 End-of-season foliar analysis of macronutrients as percentage of dry matter for aquaponic cucumbers at four target pH treatments[z]

Target pH	Actual pH (±S.E.)	N	P	K	Ca	Mg	S
Spring[y]							
7.0	6.9±0.16	5.26n.s.[x]	0.54n.s.	4.11n.s.	5.46n.s.	0.41n.s.	1.23n.s.
6.5	6.7±0.22	5.16	0.44	4.06	5.61	0.42	1.19
5.8	6.4±0.30	4.84	0.44	3.82	4.92	0.40	1.07
5.0	6.3±0.46	5.23	0.55	4.08	5.52	0.43	1.20
Significance[w]	N/A	N/A	N/A	N/A	N/A	N/A	N/A
Summer[v]							
7.0	6.7± 0.22	3.94n.s.	0.38a	2.70n.s.	8.85ab	0.59n.s.	1.45n.s.
6.5	6.6±0.22	4.48	0.32b	2.50	8.99a	0.63	1.43
5.8	6.3± 0.45	5.10	0.33b	2.52	8.54bc	0.62	1.51
5.0	6.1±0.70	5.16	0.34b	2.66	8.24c	0.62	1.54
Significance	N/A	N/A	N/A	N/A	L*	N/A	N/A
Sufficiency Levels[u]	N/A	4.30	0.30	3.10	2.40	0.35	0.32

[z] When compared against standard percentages in cucumber dry matter (Mills and Jones, 1996), all nutrients were above the lower range of recommended percentage of dry matter for cucumber growth according to t-test results using the TTEST procedure in SAS.

[y] Spring pH levels were managed using citric acid.

[x] Means with the same letters in a column are not significantly different at (P<0.05) as determined by analysis of variance and lsmeans using the GLIMMIX procedure and type III sum of squares in SAS. n.s.=not significant.

[w] Significance established using trend analyses in PROC GLIMMIX. L=linear trend. *= significance at alpha=0.05. N/A= no trend test conducted because of non-significant ANOVA.

[v] Summer pH levels were maintained with 33% sulfuric acid.

[u] Sufficiency levels obtained from Mills and Jones, 1996.

Table 3.6 Mid-Season micronutrient analysis of aquaponic irrigation water at four target pH treatments[z]

Target pH	Actual pH (±S.E.)	Boron	Zinc	Manganese	Iron	Copper
Spring[y]						
7.0	6.9±0.16	0.08	0.08	0.03	0.03	0.03
6.5	6.7±0.22	0.09	0.13	0.04	0.09	0.03
5.8	6.4±0.30	0.09	0.10	0.04	0.03	0.02
5.0	6.3±0.46	0.08	0.08	0.05	0.05	0.02
Summer[x]						
7.0	6.7± 0.22	<0.10	<0.10	0.21	<0.10	<0.10
6.5	6.6±0.22	<0.10	<0.10	0.21	<0.10	<0.10
5.8	6.3± 0.45	<0.10	<0.10	0.21	<0.10	<0.10
5.0	6.1±0.70	<0.10	<0.10	0.22	<0.10	<0.10
Recommended Levels[w]	N/A	0.70	N/A	1.97	6.85	0.07

[z] All units are in $mg \cdot L^{-1}$

[y] Spring pH levels were managed using citric acid.

[x] Summer pH levels were maintained with 33% sulfuric acid.

[w] Sufficiency levels obtained from Jones, 2005.

Table 3.7 End-of-Season micronutrient analysis of aquaponic irrigation water at four target pH treatments[z]

Target pH	Actual pH (±S.E.)	Boron	Zinc	Manganese	Iron	Copper
Spring[y]						
7.0	6.9±0.16	<0.10	<0.10	<0.10	<0.10	0.11
6.5	6.7±0.22	<0.10	<0.10	<0.10	<0.10	0.15
5.8	6.4±0.30	<0.10	<0.10	<0.10	<0.10	0.10
5.0	6.3±0.46	<0.10	<0.10	<0.10	<0.10	0.17
Summer[x]						
7.0	6.7± 0.22	<0.10	<0.10	0.22	<0.10	<0.10
6.5	6.6±0.22	<0.10	<0.10	0.22	0.45	<0.10
5.8	6.3± 0.45	<0.10	0.12	0.25	0.22	<0.10
5.0	6.1±0.70	<0.10	0.13	0.27	1.13	<0.10
Recommended Levels[w]	N/A	0.70	N/A	1.97	6.85	0.07

[z] All units are in $mg \cdot L^{-1}$

[y] Spring pH levels were managed using citric acid.

[x] Summer pH levels were maintained with 33% sulfuric acid.

[w] Sufficiency levels obtained from Jones, 2005.

Table 3.8 Mid-season foliar analysis of micronutrients as dry matter for aquaponic cucumbers at four pH treatments[z]

Target pH	Actual pH (±S.E.)	B	Zn	Mn	Fe	Cu
Spring[y]						
7.0	6.9±0.16	60n.s.[x]	95n.s.	46n.s.	106n.s.	12n.s.
6.5	6.7±0.22	57	84	47	116	12
5.8	6.4±0.30	55	82	46	120	11
5.0	6.3±0.46	59	92	47	119	12
Significance	N/A	N/A	N/A	N/A	N/A	N/A
Summer[w]						
7.0	6.7± 0.22	43n.s.	80n.s.	184ab	144n.s.	11n.s.
6.5	6.6±0.22	38	61	118c	78	9
5.8	6.3± 0.45	40	62	166b	89	10
5.0	6.1±0.70	38	79	214a	86	10
Significance	N/A	N/A	N/A	Q*[v]	N/A	N/A
Sufficiency Levels[u]	N/A	30	25	50	50	8

[z] When compared against standard percentages in cucumber dry matter (Mills and Jones, 1996), all nutrients were above the lower range of recommended percentage of dry matter for cucumber growth according to t-test results using the TTEST procedure in SAS.

[y] Spring pH levels were managed using citric acid.

[x] Means with the same letters in a column are not significantly different at ($P<0.05$) as determined by analysis of variance and lsmeans using the GLIMMIX procedure and type III sum of squares in SAS. n.s.=not significant.

[w] Summer pH levels were maintained with 33% sulfuric acid.

[v] Significance established using trend analyses in PROC GLIMMIX. Q=quadratic trend. *= significance at alpha=0.05. N/A= no trend test conducted because of non-significant ANOVA.

[u] Sufficiency levels obtained from Mills and Jones, 1996.

Table 3.9 End-of-season foliar analysis of micronutrients as dry matter for aquaponic cucumbers at four pH treatments[z]

Target pH	Actual pH (±S.E.)	B	Zn	Mn	Fe	Cu
Spring[y]						
7.0	6.9±0.16	58n.s.[x]	95n.s.	60n.s.	104n.s.	12n.s.
6.5	6.7±0.22	50	95	55	96	11
5.8	6.4±0.30	48	86	54	93	11
5.0	6.3±0.46	53	98	57	94.5	12
Summer[w]						
7.0	6.7± 0.22	59n.s.	69n.s.	391n.s.	107n.s.	11n.s.
6.5	6.6±0.22	53	59	342	100	10
5.8	6.3± 0.45	53	67	285	101	10
5.0	6.1±0.70	54	77	367	109	10
Sufficiency Levels[v]	N/A	30	25	50	50	8

[z] When compared against standard percentages in cucumber dry matter (Mills and Jones, 1996), all nutrients were above the lower range of recommended percentage of dry matter for cucumber growth according to t-test results using the TTEST procedure in SAS.

[y] Spring pH levels were managed using citric acid.

[x] Means with the same letters in a column are not significantly different at (P<0.05) as determined by analysis of variance and lsmeans using the GLIMMIX procedure and type III sum of squares in SAS. n.s.=not significant.

[w] Summer pH levels were maintained with 33% sulfuric acid.

[v] Sufficiency levels obtained from Mills and Jones, 1996.

Table 3.10 Macronutrients (mg • kg dried solids^{-1}) contained in suspended solids of aquaculture effluent[z]

	P		K		S		Ca		Mg	
pH	Pre-Plant	Post-Plant	Pre-Plant	Post-Plant	Pre-Plant	Post-Plant	Pre-Plant	Post-Plant	Pre-Plant	Post-Plant
7.0	2075	1672	757	519	1050	850	1713	2825	300	300
6.5	2628	2392	591	498	1100	875	2625	4188	325	300
5.8	1892	1580	716	540	1150	925	5100	5013	350	400
5.0	1693	2037	747	581	1213	963	3550	3238	350	325

[z] Solids were dried at 105°C for 24h before analysis.

Table 3.11 Micronutrients (mg • kg dried solids^{-1}) contained in suspended solids of aquaculture effluent[z]

	B		Zn		Mn		Fe		Cu	
pH	Pre-Plant	Post-Plant	Pre-Plant	Post-Plant	Pre-Plant	Post-Plant	Pre-Plant	Post-Plant	Pre-Plant	Post-Plant
7.0	5	13	175	150	625	1250	850	938	25	25
6.5	5	1	188	38	338	600	963	488	25	13
5.8	5	1	125	150	450	2238	1038	813	25	13
5.0	1	1	13	150	13	1963	1113	1125	25	13

[z] Solids were dried at 105°C for 24h before analysis.

Table 3.12 Comparison of Daily Nutrient Supply (mg) from Aquaculture Effluent[z] (AE) and Hydroponic Solution[y] based on irrigation rate of 7 L daily.

Nutrient	AE			Hydroponic Solution
	Solid	Liquid	Total	
N	N/A	651	651	1512
P	2.06	98	100	406
K	0.779	686	687.	2002
S	1.25	N/A	N/A	N/A
Ca	5.55	805	811	1295
Mg	0.381	203	203	1295
B	0.005	0.7	0.705	4.9
Zn	0.136	0.84	0.976	N/A
Mn	0.490	1.75	2.24	13.8
Fe	1.13	1.54	2.67	478.0
Cu	0.027	0.7	0.727	0.49

[z] Data for aquaculture effluent (AE) at pH of 5.8.

[y] Values extrapolated from Jones, 2005.

Appendix 1

Auburn University's aquaponics research is housed at the E.W. Shell Fisheries Center in Auburn, Alabama. The goal of the aquaponics project at Auburn University is to improve the profitability and sustainability of aquaponics by developing a system that generates multiple revenue streams from the initial input of fish feed. The system is comprised of one fish production greenhouse, three vegetable production greenhouses, twelve vegetable production raised beds, and two algal turf scrubbers. Water- and nutrient-flow through the system connects all units (Figure 5).

Fish production was conducted in a 9.1m x 29.3m double polyethelene-covered greenhouse which contained two 102,000-L rectangular tanks each holding an average of 6,000 tilapia (*Oreochromis niloticus)*. The aquaculture system was a hybrid RAS-biofloc system in which approximately 5% of the tank volume (5,100 L) was used for irrigation of plants and replaced with fresh water daily which was sourced from a series of ponds and fed via gravity to the aquaculture system. Unlike a true biofloc system, where the tank water is inoculated with bacteria when the fish are initially stocked in the system, the existing "biofloc" was cultivated naturally through bacteria brought in with the pond water and is suspended in the tank's culture water due to the constant aeration supplied by two 1.5 horsepower blowers. A carbon source is not currently being added to the tank culture to encourage heterotrophic fixation of ammonia. Tilapia were fed twice daily until satiation with a 36% crude protein commercial aquaculture feed (Cargill, Franklinton, Louisiana). Water quality in the tanks was monitored daily for pH, ammonia, and dissolved oxygen levels. Water pH was maintained at 6.5 for fish production.

Ammonia and dissolved oxygen levels remained within acceptable levels for fish production for the duration of the experiment. Suspended solids, including uneaten feed, feces, and biofloc were settled and removed from aquaculture effluent (AE) using two passive clarifiers connected in a series. The clarifiers were 1,500-L cone-bottom tanks located adjacent to the aquaculture system outside the greenhouse. Aquaculture effluent was continuously pumped into the first clarifier using an air lift and was forced to pass under a solid baffle separating the tank into two halves before being moved to the second clarifier which was used as the irrigation reservoir for the plant greenhouse. The clarifiers removed an average of 50% of suspended solids from AE before it was used to irrigate plants.

Water is captured from greenhouse production in one of four 378-L sumps and is used in algae production and research. Unlike conventional aquaponic systems, the Auburn system contains a high amount of suspended solids, which places it in a unique position for research.

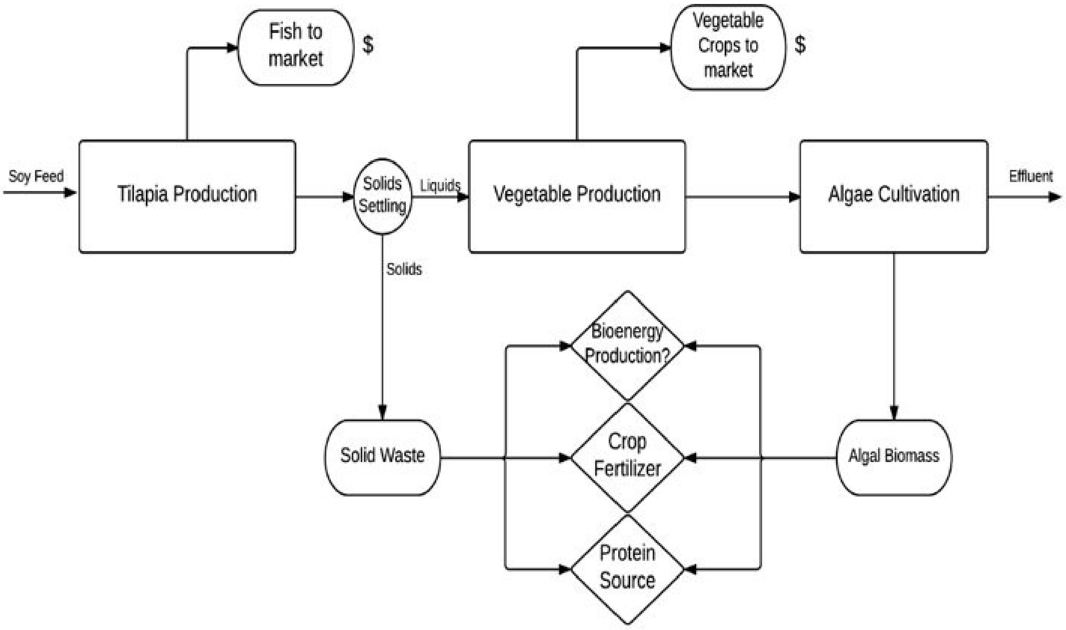

Figure 5. Systems schematic, courtesy of David M. Blersch, of the Auburn University Aquaponics facility.