

**INTEGRATION OF INTENSIVE AQUACULTURE PRODUCTION AND
HORTICULTURE CROP PRODUCTION**

by

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Thesis Abstract

Agricultural sectors can be integrated into mutually beneficial productions systems. Integration provides improved production sustainability, increased ecosystem health, increased human health, and polyculture. Horticulture and aquaculture are two sectors of agriculture readily integrated. Water-reuse, nutrient filtration, decreased environmental loading, decreased production costs, and product diversification are several benefits of irrigating horticulture crop production with aquaculture effluent waters. The objective of studies was to evaluate horticulture crop production in an intensive system utilizing aquaculture effluent water, as compared to standard greenhouse horticulture crop production. Effluent water from an intensive tilapia production facility was utilized as an alternative input to traditional greenhouse crop production. Tilapia production was conducted in a 29.3 x 9.1 m (96 x 30 ft) double layer polyethylene-covered greenhouse, for increased environmental control for year-round production. Fish were stocked at 80 fish·m³, in two 27.4 m x 3.8 m x 1.2 m (90 ft x 12½ x 4 ft) tanks, constructed of wood with steel I-beam and metal cable reinforcements. Due intensive nature of aquaculture production, continuous aeration supplied adequate dissolved oxygen (DO) for fish population. Dissolved oxygen and temperature of fish culture water were recorded with YSI 550A meter (YSI Inc., Yellow Springs, OH). Culture water pH, EC, and salinity were measured with YSI 63 meter. Tank water total ammonia nitrogen (TAN) levels were measured with a test kit (1.0 to 8.0mg·L⁻¹, LaMotte Company,

Chestertown, MD), and NO_3^- -N was measured with ion specific electrode meter (Cardy meter, range 0 to 9,900 $\text{mg}\cdot\text{L}^{-1}$, Spectrum Technologies, Inc., Plainfield, IL).

Water samples were collected weekly and analyzed using ICAP and for NH_4 determination. Water exchange rate was between 4% and 12% daily per tank. Daily feeding rate was between 27 kg to 34 kg (50 lbs to 75 lbs) $\text{feed}\cdot\text{tank}^{-1}\cdot\text{day}^{-1}$ with a 32% crude protein fish feed (Alabama Catfish Feed Mill, Uniontown, AL).

Adjacent to the fish greenhouse, bedding plants and vegetables were irrigated utilizing effluent water provided from the tank directly, or bypassed through a settling tank as experiments required. All experiments were conducted in a 29.3 x 9.1 m (96 x 30 ft) greenhouse, with a double-layer polyethylene cover. The first experiment, conducted between January to March 2009 and July to August, 2009, clear water (CW) and effluent water (EW) irrigation was examined under varying types and rates of fertilizer inputs. Clear water treatments included a soluble 200 $\text{mg N}\cdot\text{L}^{-1}$ application, and a top-dressed controlled release fertilizer applied at 1.58 $\text{kg N}\cdot\text{yd}^{-3}$. Effluent water treatments varied between settled EW and unsettled EW. Results indicate plants grown under effluent water irrigation performed similar to those produced under traditional production methods. In the second experiment, conducted from July to September 2009, irrigation source was again examined between CW and EW applications, while varying rates of a soluble fertilizer were compared. Treatments consisted of 200 $\text{mg N}\cdot\text{L}^{-1}$, 100 $\text{mg N}\cdot\text{L}^{-1}$, and unsettled EW. Results indicate plants grown under effluent water irrigation performed similar to those produced under traditional production methods. Two experiments examined effects of EW irrigation on vegetable production. One study investigated greenhouse production of a 90-day sweetcorn variety, while the second study

examined production of hydroponic cucumbers, in both studies plant received CW and EW irrigation as treatments. Results for the sweetcorn study, conducted January to March 2009, indicate no visual or statistical differences between treatments for plant height, yield, ear weight and length. Results for the hydroponic cucumber study, conducted from September to October 2009, indicate Manar F1 Beit Alpha cucumbers receiving EW performed similarly to plants receiving a specially formulated hydroponic fertilizer for a defined time frame, after which fruit production on plant irrigated with EW was less than the yield of fruit from plants receiving CW. Results indicate intensive aquaculture effluent water to be a viable irrigation source from production of plant species grown.

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I. LITERATURE REVIEW

Integration of Aquaculture and Horticulture

Since the early nineteen seventies, much research and scientific input has occurred regarding the integration of agricultural systems (Holliman, 2008; Rice, 2008). The goal of integration is a more efficient use of overlapping resources for a sustainable outcome. Two areas of interest lie between using aquaculture wastewater and production of horticultural crops via various techniques. Not only has interest peaked in aquaculture and horticulture, but aquaculture is being incorporated into many other facets of agriculture (Perin, 2002, Prinsloo et al., 1998). Agriculture is unique, because many different fields share numerous overlapping concerns, making integration sensible (Girardin and Spiertz, 1993; Perin, 2002). This review will focus on the integration of aquaculture and horticulture, but not before exploring how other agricultural combinations are positively affecting the world around us.

Aquaculture

Fundamentally, aquaculture is an agricultural science involving rearing aquatic life forms, be they plant or animal. The majority of aquaculture inputs are fish and/or shellfish production. Other forms of aquaculture production include the rearing of baitfish, aquarium fish and plant species, and aquatic plant species for consumption as food sources. Furthermore, production of pearls is another form of aquaculture (Rice, 2008).

A contemporary application of aquaculture, deemed ‘intensive aquaculture,’ entails, specifically, fish and shellfish production. Intensive aquaculture is a method of cultivating fish or aquatic life species by way of intensive human input and management practices (Rice, 2008). Various management tasks include calculating stock densities of fish populations per tank or pond, strictly controlling water quality, creating and maintaining a low stress environment, and managing disease levels at all costs. Other characteristics include high capital costs and inputs, and constant vigilance for wellbeing of fish species. Aquaculture producers must have an alternate power source available in case of emergencies, such as a generator in case of power outage (Rice, 2008).

Horticulture

Horticulture is the science and practice of plant cultivation. Economic impacts of horticulture industries (nursery, greenhouse, floriculture, sod, and vegetable production) in Alabama are substantial, accounting for nearly \$291 million in market value in 2007 (ERS, 2007). Within horticulture are many specialized practices many of which can be integrated with intensive aquaculture. In particular, greenhouse crop production is important for year round production and sales opportunities otherwise unavailable in winter months.

Hydroponics is a specialized horticulture method of plant production, in which plants are grown in a soilless media known as a substrate or in a fully liquid media (Jensen, 1997). Substrates are a resource to plants, offering a medium for holding nutrients during and between watering from which the plant secures the nutrients. Liquid media provides nutritional requirements by having nutrients dissolved in media. Environmental control is desirable for fully liquid hydroponic systems due to soilless

conditions and exposed roots. Greenhouses are often used with hydroponic systems for controlling environmental conditions (light, temperature, and water usage) (Jensen, 1997), providing plants with ideal growth conditions.

Agriculture Integrations

Considering the coupling of aquaculture and horticulture is important and valuable. Various agricultural wastes/by-products are being used in integrated systems such as in development of animal feed, biofilters, biofuel, and composted materials for use in plant production substrate. Each will be discussed individually.

Animal Feed Integration

Recycling of waste materials for incorporation into animal feeds often integrates previously separate agricultural systems. Apple pomace (AP), material left over from juicing processes is a readily available and inexpensive byproduct (Hang and Walter, 1989; Smock and Neubert, 1950), having undergone much research for use in animal feeds, and in the production of ethanol (Gupta et al., 1990; Hang et al., 1982; Ngadi and Corrota, 1992). Determining alternative uses of AP due to high content of carbohydrates, acids, fibers, vitamin and mineral levels would bring added value or additional cost recovery to apple processors. Byproducts often leach into environment possibly causing significant loss of valuable natural resources (Hang and Walter, 1989; Joshi and Joshi, 1990). In a study by Joshi and Sandhu (1996), AP was processed via solid-state fermentation for ethanol extraction, and evaluated as a dried powder for nutritional content. AP powder analysis indicated valuable nutrient levels, with three times natural

level of crude proteins, two times fat content, and two times vitamin C content, compared to traditional feeds. Even though AP is lacking in N, nitrogen salts (urea or ammonia) can be used to offset low N. Results concluded AP has excellent potential as a dual-purpose waste; animal feed and ethanol production. Dual use of AP is simple, cheap, and stable with potential to be utilized as a commercial endeavor. Other benefits are reduction of a potentially environmentally harmful waste and added economic value through the reuse of an inexpensive waste product (Joshi and Sandhu, 1996).

Biofilter Integration

Biofiltration is the removal of unwanted materials from a given source that is polluted or impure (Schmidt, 2000). Biofiltration is used in poultry, livestock, wastewater management, and aquaculture industries, as well as many more. Biofilter media may be used to filter polluted gases from production processes, or to filter out chemical pollutants from water sources (Rajeshwarisivaraj et al., 2001; Schmidt, 2000). Benefits and relationships of biofiltration to various agricultural industries will briefly be addressed.

Rajeshwarisivaraj et al. (2001) used activated carbon from cassava peel to reduce presence of dyes and heavy metals from wastewater resources. Cassava is a tropical woody, perennial shrub, and the peel is a byproduct of food processing. Two experiments using the cassava peel were conducted, as an adsorbent for filtering unwanted materials. Carbon was extracted from the cassava physically, by heating the peel to 700°C, and chemically, infusing H₃PO₄ into the peel. Results indicate use of carbon from cassava peel to effectively neutralize dyes and heavy metals from wastewater. Filtration of pollutants was nearly one hundred percent effective under chemical treatment. Chemical extraction proved more efficient over physical release of carbon, while both methods

were effective. Wastes can be employed in valuable ways to reduce environmental pollution. Recycling waste materials, such as waste cassava peel, is economically important due to high prices of activated carbon from commercial providers (Rajeshwarisivaraj et al., 2001).

Hong and Park (2005) investigated filtering of ammonia gas from the composting process. Ammonia-nitrogen (NH_3) release is an environmental problem, associated with livestock houses and livestock feed lots. Biofiltration was adapted to reduce emissions from the composting process. Manure compost and coconut shells were tested as biofilter media, at a ratio of 50:50. The filter dimensions were 500 mm deep and 300 mm wide. Material used for filtering was a mixture of dairy manure, rice hulls, and sawdust. Results indicate with biofilters at a depth of 500 mm, NH_3 released during the composting process was successfully filtered, with 100 percent of NH_3 captured (Hong and Park, 2005). Advantages of reusing agricultural wastes indicate composting to be a valuable and effective means of recycling decomposed organic wastes. Secondly, biofiltration can be managed to successfully eliminate escape of unwanted gases into the environment.

Remediation of heavy metals via biofiltration through the use of agricultural wastes has been investigated due to damaging influences and persistence of heavy metals in waterways and due to availability of byproducts from agricultural industries (Sud et al., 2008). Agricultural wastes are valuable for heavy metal remediation due to affinity of functional groups present in biomass to heavy metal pollutants. Use of non-traditional biofiltration is important because conventional methods are less efficient and harmful chemical sludge byproducts are produced. Heavy metals in water systems such as

chromium, lead, cadmium, and nickel have been effectively removed by using agricultural wastes for remediation. As a modern solution to polluted waters, biofiltration is valuable due to cleansing abilities, and to the available, inexpensive, and renewable nature of byproducts. Results are conclusive that agriculture byproducts are effective for biofiltration (Sud et al., 2008).

Biofuel Integration

Biofuel is important worldwide, as nations pursue alternative fuels to combat rising oil prices and decreasing available supplies. By definition, biofuel is a fuel made from biological materials, usually plants (Kahn, 2007). Plant materials are considered organic materials, thus breaking down biologically with diminutive impact on the environment. Corn, switch grass and biomasses of other feedstocks are currently being tested as potential biofuels. There are other potential resources that have yet to be considered.

As a majority user of all oil supplies (Albukh, 2000), the United States has a growing interest in alternative fuel sources (Um et al., 2003). Chen et al (2007) evaluated bioethanol from agricultural residues consisting of barley hay, barley straw, pearl millet hay, sweet sorghum hay, triticale hay and straw, and wheat straw was conducted. Chemical compositions of feedstocks tested were analyzed, to investigate necessary pretreatments and efficiency of hydrolysis with various enzyme levels, to determine ethanol yield after fermentation. Generally, the chemical qualities that qualify a feedstock as a potential ethanol source are: 40-50% glucan, 15-35% xylan, and 10-20% lignin. The feedstocks tested showed a composition of 28.61-38.58% glucan, 11.19-20.78% xylan, and 22.01-27.57% lignin, qualifying them as candidates for bioethanol production

sources. An important observation to note is the level of glucan, xylan and lignin in the hay feedstocks were significantly lower than levels in the straw feedstocks. Final conclusions obtained signal a need for more research to determine precise levels of chemical pretreatments, enzymatic hydrolysis and fermentation requirements (Chen et al., 2007).

In Northern Ireland and Republic of Ireland, the mushroom industry has grown exponentially over the past decade (Williams et al., 2001), which is both positive and negative. Due to industry growth, growers are now capable of competing with mainland Europe; however, due with industry expansion environmental impacts are beginning to be observed. One problem is excess waste produced, as spent mushroom compost (SMC). Once mushroom growers are finished with a batch, SMC must be replaced with new compost. Due to increased growth of the mushroom industry, SMC production increased. For each kilogram of mushrooms produced, about 5 kg of SMC is produced. Due to large amounts of SMC, governmental bodies are beginning to regulate industry growth due to environmental concerns arising from spent compost. Spent compost leaching has been implicated in the contamination of local water resources (Williams et al., 2001). The prospect of using SMC as a biofuel source is now being investigated. Compost would be combusted in a heat recovery steam generator, in which superheated steam produced would be harnessed and used for energy. Results exhibit inconclusive trends without hard evidence either way, because no firing of SMC has been recorded. However, analyses of SMC on a dry weight basis indicate similar composition of sewage sludge (SS), which has undergone firings for many years (Williams et al., 2001). This is just one example of how environmentally conscious scientists are seeking to better the environment, advance

agriculture, and help economies by way of integrating different aspects of agriculture. If future use of SMC as a biofuel becomes a reality, this will be a useful and sustainable utilization of waste resources. Despite inconclusive reports, studies indicate agricultural wastes, residues, and byproducts can and have multiple uses after the primary use is concluded. Secondary uses of byproducts are environmentally sustainable, aid in advancing agriculture, and help economies.

Plant Media Integration

A further agricultural integration common in practice is recovery of organic wastes and byproducts in horticultural utilization (Carrion et al., 2008). In horticultural integration, use of composted materials is attractive due to the need to find alternatives to *Sphagnum* peat, for reducing peat use (Abad et al., 2001; Fitzpatrick, 2001; Fitzpatrick et al., 1998; Moore, 2005). Waste materials used in plant growing substrates range from traditional materials such as crop residues to nontraditional materials such as sewage sludge, coconut fibers, or composted household garbage (Hernandez-Apaolaza et al., 2005; Hu and Barker, 2004; Lu et al., 2006; Lu, 2008; Perez-Murcia et al., 2006). There is also a growing trend of land application and general landscape use, of agricultural wastes (Hu and Barker, 2004). Extensive studies concerning composts as a viable plant growth substrate have been conducted using a wide range of species (Hu and Barker, 2004).

Various combinations of Pinebark (PB), coconut fiber (CF), and sewage sludge (SS) were tested on *Pinus pinea*, *Cupressus arizonica*, and *C sempervirens*. Substrates combination used were 100% PB, PB with 15% and 30% SS, 100% CF, and CF with 15% and 30% SS compost (Hernandez-Apaolaza et al., 2005). Plants remained healthy

without any indications of nutrient deficiency or toxicity for the yearlong test period. *Cupressus arizonica* and *C. sempervirens* performance was improved with an increasing amount of SS (Hernandez-Apaolaza et al., 2005).

Perez-Murcia et al (2006) evaluated use of composted sewage sludge (CSS) combined with peat on growth of broccoli (*Brassica oleracea* var. *Botryti* cv. Marathon), in response to increasing amounts of CSS from 0%, 15%, 30% and 50% volume for volume. Broccoli showed increased fresh and dry weight with increasing CSS content, however, nearly toxic levels of Na, Cu, Zn, and Cd in tissue were indicated. Highest performance for broccoli was a substrate consisting of 7:3 peat to CSS (Perez-Murcia et al., 2006).

Concerns exist in use of composted materials for potting mixes, often due to high pH of the composted materials, usually above 8.4 (Carrion et al., 2008). Carrion et al (2008) sought to determine effectiveness of acidifying amendments at five different rates on vegetable crop composts. Study consisted of 3 compost combinations: melon (75%), yard trimmings (19%) and almond husk (6%); pepper (75%), almond husk (15%), and yard trimmings (10%); zucchini (70%), cucumber (15%), and pepper (15%) biomass and used elemental sulfur (S) and ferrous sulfate ($\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$) for the acidifying agents. Sulfur and $\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$ were added at five different rates, with rates required to obtain the lowest pH equivalent to 12.4, 8.5, and 9.4 g of S per liter of compost for melon, pepper, and zucchini composts, respectively. For $\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$ the rates for lowest pH were 108.2, 73.9 and 82.3 g for melon, pepper, and zucchini composts, respectively. Results determine potential of controlling pH in growing media composed of composted materials. Evaluation of physical and chemical properties of amended composts showed

proper characteristics for incorporation into commercial production of containerized plants. Although both acidifying agents used were effective, the elemental sulfur demonstrated better pH correction abilities than did the ferrous sulfate (Carrion et al, 2008).

Hu and Baker (2004) evaluated effect of three waste products (agricultural wastes (poultry manure, cranberry presscake), sewage compost (biosolids, woodchips), and yard wastes), mixed with peat moss and soil on growth of tomatoes (*Lycopersicon esculentum*). Four substrate mixes were tested: 100% compost, compost and soil at a ratio of 1:2 by volume (v:v), compost and peat moss at a ratio 1:2 (v:v), and compost, peat, and soil at a ratio 1:1:1 (v:v:v). Tomatoes were grown from a seedling stage to initiation of fruit growth, for six weeks. Each of the four mixes also had a fertilizer (0.15g N, 0.15g P₂O₅, and 0.15g K₂O per kg of media) and non-fertilized treatment, with fertilizer being applied at two week intervals. Results indicated differences among the four compost mixes, with best vegetative growth, and highest in foliar nutrient content in substrates with agricultural wastes, but fruit yield was best in mixes composed of peat moss with supplemental fertilizer (Hu and Barker, 2004).

Reuse of waste products for soil amendment have also been conducted. Pulp mill wastes were tested to determine if various materials effectively altered soil pH (Cabral et al, 2008). Wood ash, dregs, and grits were utilized for amending of soil pH, to a desired level of 6.5. Some materials proved effective as an alternative source to commercial agricultural liming agents. Wood ash effectively adjusted pH, and supplemented other nutrients (P and K). Conclusions indicate pulp mill wastes to be viable for agricultural

liming agents, due to ability to provide necessary degree of control of soil pH (Cabral et al, 2008).

Summary

Agricultural integrations offer potentially long-term positive effects. Assimilating various sectors of agriculture provides increased levels of sustainability, protection of human and environmental health, increased revenues, extended life of materials once thought to be fully used, provision of alternatives to replace limited naturally occurring materials, and providing aid in producing the same quality products as traditional production means. Due to benefits listed, an argument for sustainable and integrated agriculture carries more significance in a changing world. Among such integrated agriculture endeavors is the growing trend of integrating aquaculture and horticulture.

Agriculture Production Intensity Changes

In the last fifty years, various sectors of agriculture have followed the trend of increased production intensities, specifically in dairy, poultry and swine industries (Sheldon, 2000). Reports describe intensification to be a trend in the European Union (EU). Extensive livestock systems in the EU have previously been shown to be economically sensible (Caballero and Gil, 2009). In Henan Province of China, after farmers adapted an intensive “four-crops, four-harvests” technique, yield increases were 2.5 times greater than traditional farming practices (Huajun and van Ranst, 2005). Intensive systems in China indicated land used to be 1.5 times more productive, while labor was nearly 80% more productive, and a ratio of 1 Yuan input resulting in a 1.7 Yuan output. Feeding regimes for animals, whether terrestrial or aquatic, can be

intensified when animals are recruited into a decreased area, so that density (animal·unit area⁻¹) is increased (Watanabe et al., 2002).

Poultry Intensity Changes

Fully mature and complete development of the poultry technology revolution of the last half-century extends only to 15% to 20% of the world population (Sheldon, 2000). Improvements in design and construction of poultry housing have led to increased efficiencies in production, due to better building and insulating materials, more efficient heating and cooling systems, and automated controls. Latter developments have also made possible more efficient farm and business management methods. Ability to control bird environments against extremes of heat and cold has allowed a higher efficiency of feed utilization, better health, and less mortality (Sheldon, 2000). Intensive systems in poultry are not new or novel, but rational in light of cost savings, increased efficiencies in production, use of facilities and labor, as well as increased revenues.

Intensive Aquaculture

Intensive Aquaculture (IA) is a production system of cultivating fish or aquatic life species by way of substantial human input and management practices (Rice, 2008), including exponentially increased stocking densities (fish·area⁻¹), increased feeding regimens, and strict environmental control. The magnitude of the aquaculture industry in the United States (US) can scarcely be estimated, yet projections approximate catfish farming in 2008 to be worth nearly \$400 million to producers (Hanson, 2009). Total value of aquaculture industry in Alabama was estimated to be worth \$100 million during 2007 fiscal year, ranking sixth nationally in total aquaculture production (NASS, 2007a)

National and Alabama Aquaculture Industries

In 2008, US catfish farming produced 231 million kg (510 million lbs) round weight catfish from processing (Hanson, 2009), with Alabama ranked second nationally. Catfish culture in America has an important economic impact, accounting for about \$400 million to producers in 2008, down from previous years, such as 2003 when aquaculture provided approximately \$650 million to producers (Hanson, 2009). Catfish farming is not only important nationally, but locally in Alabama. Aquaculture in Alabama, in 2006, provided producers with \$115 million, with catfish farming accounting for over \$105 million the total (Crews and Chappell, 2007a). Alabama processed catfish accounts for \$200 million in sales across the nation (Crews and Chappell, 2007a).

Decline of National and Local Aquaculture

Despite promising numbers discussed above, both nationally and locally, aquaculture industries, including catfish farming, are struggling among an international market. Various signs of decreasing US markets exist. Locally, in the southeastern US, catfish water acres have dropped by 11 percent from 2007 to 2008, down to nearly 134 thousand acres among Alabama, Arkansas, Louisiana, and Mississippi (Hanson, 2009). In the same states, 75 catfish operations have shutdown, accounting for nearly 11 percent of total operations. From 2003 to 2006, national catfish farm gates sales have decreased by 17 percent (Crews and Chappell, 2007b). US and local markets are shrinking due to a variety of factors.

Foreign Imports

Importing foreign fish products is a major hindrance to domestic markets. Frozen fish products are of particular interest. As of December 2008, US imported 102 million

pounds of frozen catfish products, accounting for nearly 50 percent of catfish market share (Hanson, 2009). Imported catfish products increased by 21 percent from 2007 to 2008.

An increase in readily available information and technology has enabled foreign nations to become leading producers of aquaculture products. China is the top producer all aquaculture products worldwide, for nearly thirty years, accounting for thirty-five percent of global aquaculture products (Barnett and Rose, 2006). China is the world's top exporter of seafood products. Two of the most consumed seafood products in America are shrimp and tilapia, respectively. China produces over half of worldwide tilapia products (Barnett and Rose, 2006). Seventy percent of Chinese tilapia exports are consumed in America, and China is the fourth largest supplier of shrimp to the U.S. (Bean and Xinping, 2006). In country, Chinese aquaculture producers maintain large-scale operations, intended for exporting to America (Engle and Heikes, 2008). This degree of importation is harmful and dangerous to domestic markets, and presents increasing challenges for local fish producers.

Poor Quality of Foreign Seafood Products

Reasons for importing large quantities of fish products are numerous, and concerns arise from poor quality control practices. Due to severely polluted waterways in China, unease has grown over cleanliness and safety of Chinese aquaculture practices and products. Contamination of waterways occurs via municipal runoff, industrial runoff, and mercury emissions from coal power plants (Ellis and Turner, 2007).

Poor quality control is evident from reported cases of food poisoning or banned imports. Eel imports from China in 2003 were barred in Japan due to contamination;

while in 2005, Chinese shrimp imports were brought to a halt in the EU (Ellis and Turner, 2007). Preliminary analyses indicate disease in Chinese aquaculture production accounts for fifteen to twenty percent production losses, equating to losses of five and seven billion Yuan annually (or US \$525-875 million) (Wang, 2001). Other reports of food poisoning include: presence of melamine scrap sickening 14,000 US pets (Barboza and Barrionuevo, 2007), removal of Chinese catfish from Wal-Mart stores when antibiotic contamination of imported fish was discovered by state of Alabama (Nohlgren, 2007), samples of Mandarin fish tested positive for malachite dye in Hong Kong (China CSR, 2006), and in Taiwan when mitten crabs were found with traces of carcinogens (Huang, 2006). Poor quality fish products begin with cultivation practices of farmers. Most common culture mistakes occur due to over application or mishandling antibiotics, pesticides, and fungicides frequently used for water quality control (Food and Water Watch, 2006).

A “Stop-Sale Order” was issued to Mississippi grocery stores in 2007, when imported Chinese catfish products were found to be contaminated with FDA banned antibiotics. Mississippi Commissioner of Agriculture and Commerce, Lester Spell, had sixteen samples tested, of which eleven were positive for contamination (Gallagher, 2007a). Referring to the same incident, Roger Barlow, executive vice president of the Catfish Farmers of America stated, “Frankly, I was not surprised to read about the wheat gluten poisoning incident. We know that Chinese farmers routinely use a variety of chemicals and antibiotics banned in the United States for use in or around human food,” (Gallagher, 2007a). United States FDA has implemented increased security checks on all

farm raised catfish, basa, shrimp, dace, and eel imported from China, holding products until confirming contamination reports to be negative (Gallagher, 2007b).

Contaminated food is problematic due to induced illnesses and death, but often food poisoning is not the only cause of poor quality products, but also mislabeling of products. Paul Raymond of NOAA states, "Substitution in the seafood industry is an unfortunate, but prevalent occurrence, often at the expense of the resource and the consumer," indicating the prevalence of mislabeling of seafood products (Ecosystems, 2008). Predominantly, seafood products are mislabeled in country of origin, for greater economic income for seller. An example from Vietnam exposed farm raised catfish being labeled and marketed as "wild-caught grouper or snapper" (Ecosystems, 2008).

Imports from China are significantly less expensive than locally grown fish products, partly due to lower costs and availability of labor in China and around the world.

Between 1980 and 1998, Chinese aquaculture production increased from 1.68 million metric tons (mmt) to 21.82 mmt, creating a large need for workers. Following the explosion of industry growth, the work force grew by 10 million laborers, and is reported to be growing by 500,000 annually (Wang, 2001). China provides an endless number of laborers as the industry continues to grow, subsequently providing cheaper, though lesser quality, products as trends indicate.

In summary, though focused on China, the importance of recognizing hurdles and concerns facing domestic and local fish producers among a growing global market is comprised of dealing with less expensive products, managing mislabeled products, screening for contaminated products, and lastly competing with cheaper labor rates from overseas providers.

Benefits of Intensive Aquaculture over Traditional Practices

Intensive aquaculture (IA) operations contained within an environmentally regulated system have inherent benefits over traditional aquaculture production practices. Environmentally regulated systems are indoor systems, often in greenhouses, allowing for year round fish production (Holliman, 2006; Masser et al., 1999). Other benefits include reduced land requirement, and possibly a decreased water requirement (Masser et al, 1999; Rakocy, 1989). Year round production of fresh product tends toward increased profitability, as well as being able to operate in closer proximity to local markets (Holliman, 2006; Masser et al., 1999). IA makes use of high stocking densities, disrupting reproductive processes in intensive tilapia culture, producing a marketable fish sooner than in pond culture. Time requirement for feeding and harvesting is decreased in intensive tank culture, which further leads to decreased labor costs (Rakocy, 1989). Due to year round production, trained workers may then remain on staff. All factors considered, IA is an efficient use of resources compared to traditional pond culture, equating to increased revenues.

Pond production is limited by numerous factors, often including available land, functionality of ponds, as well as a need for increased seasonal labor and time inputs. Where climate allows, pond production of tilapia is stocked at a density of 1000-2000 fish·ha⁻¹ (Watanabe et al., 2002) if production is a mixed-sex batch. When ponds are stocked with males only higher stocking densities can be used but supplemental aeration may be needed (Rakocy and McGinty, 1989), with an output range from 300-700kg·ha⁻¹·crop⁻¹ (Watanabe et al., 2002). Additional options include converting existing water

resources into production models of raceways or cage culture. Raceways, or rectangular tanks, are a practical and functional means of rearing fish.

Cage culture of tilapia is common throughout the Americas, with advantages including greater control over feeding, increased ease of harvesting, and interrupted breeding to decrease recruitment (Watanabe et al., 2002). Stocking densities in cage culture are generally $200\text{-}300 \text{ fish}\cdot\text{m}^{-3}$, in 4m^3 cages, with yields reaching up to $150 \text{ kg}\cdot\text{m}^{-3}\cdot\text{crop}^{-1}$ (Watanabe et al, 2002).

Raceway systems, which are commonly used in commercial scale intensive systems, require water exchange and supplemental aeration. In Jamaica, raceway system range in yield from $9000 \text{ kg}\cdot\text{ha}^{-1}\cdot\text{year}^{-1}$ up to $45,000 \text{ kg}\cdot\text{ha}^{-1}\cdot\text{year}^{-1}$ (Watanabe et al., 2002). Small-scale farmers (1-4 ha) produce the lower yield while large-scale farms (21-45 ha) produce the highest outputs.

A progressive increase in $\text{output}\cdot\text{area}^{-1}$ is observed when comparing pond, cage, and raceway systems, indicating intensive cultured tilapia systems outperform pond culture. Other advantages of intensive culture systems include increased output per unit of equipment and per unit of labor (Rakocy, 1989). No additional equipment input is needed for IA systems compared with pond culture, excluding supplemental aeration equipment. Workers can feed a low density of fish in a pond as efficiently as feeding a highly stocked intensive system (Rakocy, 1989). Changes in production intensities aid in meeting demands for tilapia, a tropical fish species, where tilapia culture is now being located in temperate regions due to environmentally controlled intensive systems.

Intensive Aquaculture Concerns

Inherent to IA systems is a need for water exchange to maintain adequate water quality for fish species being grown, meaning a given percentage of water must be removed from the system, while being replenished by clean water. Discharge water is laden with nutrient loads derived from uneaten food particles, feces, phytoplankton, and other organic matter, leading to an increased amount of organic matter via photosynthesis (Boyd and Tucker, 1998). Furthermore, due to increased intensity of production in IA systems, a heavier nutrient load is produced (Boyd, 2004). An increase of fish per area means higher feed input per area and increased waste per area. Excess effluent is not only potentially toxic to production systems, but is drawing the attention of environmental activist groups and governmental agencies (Dierberg and Kiattisimukul, 1996; Goldberg and Triplett, 1997; Naylor et al., 1998, 2000; Palada et al., 1999).

Water Chemistry and System Toxicity

System toxicity is a concern in high-density production models, due to potential lethal accumulation of certain nutrients. Of utmost importance is ammonia in fish water. Total ammonia nitrogen (TAN, comprised of NH_4^+ and NH_3) is the end product of respiration and digestion in fish, excreted through gills and in feces (Durborow et al., 1997b), and is directly proportional to protein concentrations in the feed. Recommended crude protein (CP) levels in feed as a rule-of-thumb follow for catfish: 28-32% CP, tilapia 26-30% CP (Hargreaves and Tucker, 2004; Pompa and Masser, 1999), hybrid striped bass 38-50% CP (Hodson and Hayes, 1989), rainbow trout 34-38% CP, and pacific salmon 30.5-15% CP (Cho et al., 1993). Estimates of nitrogen (N) percentages in

feed are based on protein content, and generally for grow-out feed there is 4.5-5.1% N in CP of feed (Auburn University, 2004). For example, 1 kg of 32% CP feed would contain 16 g N·kg⁻¹ feed. Further, an accepted rule-of-thumb is fish assimilate between 20-30% of N from feed (Auburn University, 2004). Therefore, based on accepted assimilation rates, 11.2-12.8 g N would be excreted as ammonia by-products.

Other sources of TAN are bacterial decomposition of uneaten feed inputs, algae, and other aquatic plant growth (Hargreaves and Tucker, 2004). The two forms of ammonia in aquaculture are ionized ammonia (NH₄⁺, non-toxic) and unionized ammonia (NH₃, toxic) are together considered total ammonia nitrogen (TAN). Unionized ammonia is toxic to fish at certain concentrations, decreasing feed efficiency, as fish cannot obtain energy from food as well (Hargreaves and Tucker, 2004).

Complex production systems inherently have numerous variables with potential for interaction with other factors present. Chemical properties of fish water are ceaselessly changing and subsequently interacting. Predominant factors include pH, carbon dioxide (CO₂), alkalinity and total hardness. Altogether these properties can have confounding effects on dissolved oxygen (DO) and NH₃ concentrations (Wurts and Durborow, 1992).

Due to an average pH of 7.4 of fish blood, water pH is vital for survival, with desirable level between 6.0-9.0 pH (Tucker and D'Abramo, 2008; Wurts and Durborow, 1992). CO₂ and pH interact as plant life and algae blooms in ponds or tanks sequester energy from sun for photosynthesis (Ps) during the day, taking up CO₂ and releasing oxygen. As plants photosynthesize, taking up CO₂, pH will increase which can potentially cause a severe rise in pH where water alkalinity is low, while levels of Ps are

high. During night as P_s decreases and fish respiration continues, CO_2 concentrations increase, resulting in decreasing pH due to CO_2 conversion to carbonic acid ($\text{H}_2\text{O} + \text{CO}_2 = \text{H}_2\text{CO}_3 = \text{H}^+ + \text{HCO}_3^-$). If CO_2 levels are persistently problematic, chemical treatment is possible using hydrated lime ($\text{Ca}(\text{OH})_2$), approximated to remove $1 \text{ mg}\cdot\text{L}^{-1} \text{ CO}_2$ per $1 \text{ mg}\cdot\text{L}^{-1} \text{ Ca}(\text{OH})_2$. Low alkalinity waters should not be treated with $\text{Ca}(\text{OH})_2$ due to low buffering capacity, leading to a threat of lethally high water pH (Wurts and Durborow, 1992).

Alkalinity, measured as $\text{mg}\cdot\text{L}^{-1}$ calcium carbonate (CaCO_3), at adequate concentrations acts as a buffer to prevent pH changes. Chemicals responsible for alkalinity concentrations are carbonates, bicarbonates, hydroxides, phosphates, and berates (Wurts and Durborow, 1992). Recommended levels are $75\text{-}200 \text{ mg}\cdot\text{L}^{-1} \text{ CaCO}_3$. Low alkalinity concentrations will result in pH swings, undesirable in fish production.

Hardness is the amount of dissolved divalent (primarily calcium and magnesium) compounds in culture water, reported as $\text{mg}\cdot\text{L}^{-1} \text{ CaCO}_3$ hardness (Wurts and Durborow, 1992). The difference between alkalinity and hardness of water is the source of alkalinity concentrations. If limestone is source of alkalinity and hardness, result is similar. If bicarbonates or other non-carbonate sources are responsible for alkalinity, it is possible to have low hardness and high alkalinity. Hardness levels are suggested to be $25\text{-}100 \text{ mg}\cdot\text{L}^{-1}$ free calcium, or $63\text{-}250 \text{ mg}\cdot\text{L}^{-1} \text{ CaCO}_3$ hardness. Agricultural lime and agricultural gypsum are two sources for adjusting hardness level (calcium concentrations) in high alkaline waters (Wurts and Durborow, 1992).

Having discussed pH, CO_2 , alkalinity and hardness, it is clear how NH_3 interacts with water quality parameters influencing chemical compounds. Importance of

controlling NH_3 concentrations is due to potentially toxic levels. Other factors influencing NH_3 include water temperature, seasonal variations, and phytoplankton populations (Durborow et al., 1997b). Increasing temperatures cause an increase in NH_3 concentrations, as do increases in pH, and more basic waters (Durborow et al., 1997b; Wurts and Durborow, 1992). Effect of water pH on NH_3 concentration is extremely important, as a single unit increase in pH can cause NH_3 to increase by ten times (Table 1.1). During fall and winter months, due to decreased plankton populations, nitrification slows; resulting in NH_3 concentrations increasing as bacterial activity slows as temperatures cool (Durborow et al., 1997b).

Nitrite (NO_2^-) is another chemical property of culture water to consider. During the nitrification process, TAN is converted, via aerobic nitrifying bacteria (*Nitrosomonas spp.*) from NH_4^+ or NH_3 , to NO_2^- then to nitrate (NO_3^-), which is not toxic to fish. Fixation from NO_2^- to NO_3^- occurs during bacterial nitrification via *Nitrobacter spp.* The ammonia converted to NO_2^- is excreted from fish after feeding, and is in excess in the water column. Concerns about NO_2^- levels arise due to the potential of brown blood disease in fish populations (Durborow et al., 1997a). Toxic NO_2^- levels are rare, triggered by the cooling weather of fall and winter months, which decreases bacterial activity responsible for converting NO_2^- to NO_3^- . High water pH favors decline of NO_2^- elimination. Build up of NO_2^- can occur quickly, as $1 \text{ mg}\cdot\text{L}^{-1}$ TAN converts to three $\text{mg}\cdot\text{L}^{-1}$ NO_2^- . A system overloaded with ammonia exhibits high nitrite levels as nitrification is inhibited. Sodium chloride applications are a common treatment solution (Durborow et al., 1997b).

Nitrification is essential to fish health, and by products of the nitrification process have potential to be further sequestered by applying wastewater to plants for crop production. The preferred form of nitrogen for plant uptake is NO_3^- , as well as NH_4^+ (Mengel and Kirkby, 2001; Mullins and Hansen, 2006; Rakocy et al., 2006).

Environmental Concerns

Mentioned briefly above, pressure from environmental activist groups for increased environmental awareness and effluent management regulation for aquaculture production is steadily increasing. Increased pressure may force governmental bodies to impose regulation, parameters, or best management practices (BMP) (Boyd, 2003).

Activists are concerned due to aquaculture effluent containing elevated levels of dissolved nutrients, plankton bodies, suspended solids and an increased biological oxygen demand (BOD) in relation to watershed bodies into which effluents are discharged (Boyd, 2004; Boyd and Gross, 1999; Schwartz and Boyd, 1994, Tucker, 1998). Further concerns raised against aquaculture exist, including: conversion of agriculture lands to ponds, watershed pollution, excessive use of fresh water resources, introduction of foreign species into natural ecosystems, and flora and fauna community disturbance. Yet while these are concerns, the most notable concern remains water pollution (Boyd and Gautier, 2000; Boyd and Tucker, 2000; Tookwinas, 1996). Aquaculturists agree, enhanced management practices are attainable, but governing bodies should determine which concerns are founded upon fact and those that are not (Boyd and Schmittou, 1999).

Watershed Pollution

Watershed pollution, eutrophication and hypoxia, is the predominant environmental concern among governments and environmental awareness groups. Research indicates excess nitrogen and phosphorus inputs from feed, unutilized by fish for metabolism can result in downstream eutrophication (Ackefors and Enell, 1994; Gowen and Bradbury, 1987; Kelly, 1993). Downstream water sources may also be subject to decreased oxygen concentrations, elevated water pH, increased BOD, and unwanted algal blooms (Aubin, 2006; Bergheim and Siversten, 1981; Boesch et al., 2001; Rennert, 1994; Tervet, 1981). Other influences may include decreased populations of environmentally intolerant fish species, while tolerant species may increase (Carmago, 1992; Doughty and McPhail, 1995; Henderson and Ross, 1995; Loch et al., 1996). Specific factors of concern include nitrate concentrations, excess phosphates, and release of suspended solids. Nitrogen and phosphorus contents of feed inputs are estimated to be utilized by fish at 25%-30%, indicating 70%-75% of N and P inputs are unutilized and potentially discharged (Crab et al., 2007; Sealey et al., 1999).

Nitrate (NO_3^-) is a form of nitrogen converted, in aquaculture, from excess TAN in culture system. NH_4^+ and NH_3 are converted to NO_2^- , which is normally converted to NO_3^- , a non-toxic nitrogen form to fish in culture. However, nitrates introduced into environment raise concern for numerous reasons. One aquaponic study indicated sludge from production contained NO_3^- in concentrations of 2.3-313 $\text{mg}\cdot\text{L}^{-1}$, monitored from 1-29 days of production (Rakocy et al, 2007a). Nitrates released into surrounding environments increase the threat of contaminating drinking water sources, which can lead to blue baby syndrome. Federal regulations have set a maximum nitrate-nitrogen

concentration of $10 \text{ mg}\cdot\text{L}^{-1}$, established on preventing blue baby syndrome (EPA, 2009). Rouse et al, (1999) indicated amphibians, native in North America, are threatened with by lethal NO_3^- concentrations from 13-40 $\text{mg}\cdot\text{L}^{-1}$.

Primary freshwater eutrophication is caused by phosphorus in various forms (Correll, 1998). Increased phosphate loading in receiving water leads to increased populations of algae and cyanobacteria, leading to hypoxia, or low DO concentrations. Increasingly low DO levels result in death of aquatic plants and animals (Correll, 1998). Rakocy et al, (2007a) indicated concentrations of phosphates increased from 6.4-102.7 $\text{mg}\cdot\text{L}^{-1}$, after 29 days of production (Rakocy et al., 2007a).

Water turbidity, or water clarity, is directly affected by the amount of total suspended solids (TSS), or the particulate matter in water. TSS averaged in an aquaponic system at 19,060 $\text{mg}\cdot\text{L}^{-1}$ over twenty-nine-day period (Rakocy et al., 2007a); and in a green-water intensive culture system at 26,230 $\text{mg}\cdot\text{L}^{-1}$ (Rakocy et al., 2004b).

Government Regulation

The Environmental Protection Agency (EPA) in 2003 revised the Clean Water Act (CWA), establishing regulations for concentrated animal feeding operations (CAFO) in regard to permits and effluent discharge regulations (EPA, 2008). Revisions in 2005, called for National Pollutant Discharge Elimination System (NPDES) permitting for CAFOs either discharging effluents or proposing to discharge effluents, as well as submitting nutrient management plans (NMP) to the EPA. Revisions to the CWA also impacted aquaculture producers, due to specific effluent limitation guidelines (ELG) for concentrated aquatic animal production (CAAP) (EPA, 2008). CAAPs are simply CAFOs with aquatic animals, thus where regulation calls for permitting of CAFOs, the same

regulations apply to CAAPs. Producers grossing more than 45,000 kg (100,000 lbs) animal·year⁻¹ are subject to NPDES permits (EPA, 2006).

Some individual states including Alabama, Arizona, Arkansas, Florida, Hawaii, and many others have statewide recommended BMPs. Alabama has developed an NPDES Permit Branch in the Alabama Department of Environmental Management, aiming to provide permits to aquaculture producers, which adhere to both state and federal regulations (Sanderson, 2009). The Federal Water Pollution Control Act (FWPCA) declares any state may enforce a state-level permitting program for discharging into publicly owned waterways, as long as state permit programs are align with NPDES permits. In Alabama, regulations fall under the Alabama Water Pollution Control Act (AWPCA), which is designed as a NPDES based permit system (ADEM, 2009). Criteria contained in permitting regulations include: average monthly discharge limitations, average weekly discharge limitations, implementation of BMPs, amongst others (ADEM, 2009).

Regulations following the CWA have had significant impacts on multiple sectors of aquaculture industry. Trout farming has faced considerable regulation from the early 1990's (MacMillan et al., 2003). Beginning in 1991, trout farms in Idaho under regulation of the CWA, were required to reduce total phosphorus (TP) by 40% and TSS were to be maintained between 3-5 mg·L⁻¹. A concern of such regulation is the ability of farmers to maintain price competitiveness. Often BMP implementations are cost-ineffective, not easily integrating with existing productions systems, causing concern for being out-priced by natural fisheries or already under-priced overseas markets (MacMillan et al., 2003). Two states in particular have faced rather severe consequences

under EPA regulations. Idaho and North Carolina are the top farm-raised trout producing states in US, providing 70-75% and 8% domestic production, respectively (NASS, 2003), yet both states are experiencing setbacks due to regulations (Engle et al., 2005). The EPA supplied several options for effluent treatment, consisting primarily of installation of sediment basins, compliance with BMPs (feed, solids control, drug and chemical, etc), monitoring of TSS, escape prevention, and monitoring of TP amongst others (Engle et al., 2005).

Controlling Environmental Loading

Environmental impact of the aquaculture industry is at a heightened level of awareness with a need for increased environmental-friendliness and sustainability to be addressed. Multiple means of eliminating harmful nutrient loads from effluent before discharging exist, including; flushing, biofiltration, constructed wetlands, nitrification and settling tanks, and plant sequestration of nutrients.

Flushing or water exchange (WE) in aquaculture is discharging heavily tainted water, and replacing it with potentially clear ground water. Crawfish production in Louisiana, flushing is used as means for correcting low DO concentrations (McClain and Romaine, 2008.) WE removes excess nutrients, undesirable phytoplankton blooms, toxic pollutants (NH_3), and aids in salinity concentrations (if salinity is problematic) (Lemonnier and Faninoz, 2006). Due to harmful pollutants potentially entering natural waterways, flushing is not a recommended means of controlling culture water quality.

Aquaculture Effluent Biofiltration

A preferred method for controlling environmental impacts is using biofiltration for removal of unwanted components from a given source that cause pollution or are

impure (Schmidt, 2000). Biofiltration occurs in numerous methods including physical filtration, chemical filtration, and biological filtration (Crab et al., 2007). In aquaculture, biological biofiltration is of supreme importance, because the nitrogen cycle is a biological process. Biofiltration involving nitrification can be conducted in rotating biological contactors, trickling filters, fluidized bed filters, bead filters, and fixed film filters (Ling and Chen, 2005; Malone and Pfeiffer, 2006; van Rijn, 1996). Other techniques include periphyton and bio-floc technologies (Crab et al., 2007). Previous research reports TAN removal rates from various biofilters. Rotating biological contactors removed on average $0.19\text{-}0.79\text{ g TAN}\cdot\text{m}^{-2}\cdot\text{day}^{-1}$, and trickling filters removed $0.24\text{-}0.64\text{ g TAN}\cdot\text{m}^{-2}\cdot\text{day}^{-1}$ (Castro et al., 2006; Edinga et al., 2006; Kamstra et al., 1998; Lyssenko and Wheaton, 2006; Shnel et al., 2002). Bead filters and fluidized sand biofilters on average remove $0.30\text{-}0.60\text{ g TAN}\cdot\text{m}^{-2}\cdot\text{day}^{-1}$ and $0.24\text{ g TAN}\cdot\text{m}^{-2}\cdot\text{day}^{-1}$, respectively (Greiner and Timmons, 1998; Miller and Libey, 1985; Timmons et al., 2006; Timmons and Summerfelt, 1998).

Constructed wetlands are a means of filtration of heavy effluent loads. The use of wetlands is advantageous due to eliminating need for chemical treatment of effluents. Heavy nutrient loads are sequestered by plants, and wetlands may serve as wildlife habitats and aid in balancing hydrological functions within a given ecosystem and waterway (Sealy et al., 1999). According to previous studies, a four-day retention time within the wetland eliminated the most TP and BOD. Grass strips (Bahia) were found to be effective in diminishing suspended solids, BOD, and NH_3 concentrations; however, removal of algae was not effective (Sealy et al., 1999).

As mentioned above, nitrification is the primary biological process required for

effluent management. Often nitrification tanks are used for this process. Settling tanks or basins are often used as low input water quality control methods. Tanks are directly linked to the culturing system, and wastewater is pumped to them, and allowed to sit for a given time, allowing physical, biological, and chemical changes take place (Diab et al., 1992; Hargreaves, 2006).

Worldwide aquaculture growth is expanding into dry, arid environments, even to regions where water resources are limited. Expansion is possible due to recirculating aquaculture systems (RAS) (Chaves et al., 1999). RAS are closed systems, discharging minimal amounts of water, by relying on various means of biofiltration to filter effluent water until filtered sufficiently for reuse in the fish-culturing portion of the system. In addition to various types of biofiltration discussed above, the incorporation of hydroponic crop production is being incorporated as well (Malone et al., 1993). Evidence indicates effluent water contains dissolved nutrients adequate for plant utilization, and subsequent growth. Plants incorporated in RAS sequester excess nutrients, not metabolized by fish in culture. Plants remove and utilize nutrients from the fish culture system, eliminating potentially toxic metabolites, allowing water to be further filtered after other methods of biofiltration, thus recycled in fish-culturing portion of RAS with improved water quality after passing through hydroponic systems, fish may respond with better growth and health (Nair et al., 1985). From a study in Canada, research indicates fish wastewater to be a viable nutrient reservoir for crop production (Savidov, 2004). After four months of aquaponic production, cucumber reached maximum yield at $1.29 \text{ kg}\cdot\text{plant}^{-1}\cdot\text{week}^{-1}$.

Integration of Intensive Aquaculture with Horticulture Crop Production

Mentioned at the beginning of this review, agriculture is a unique industry due to potential level of integration among all sectors. Integrating intensive aquaculture with horticulture crop production is beneficial due to sequestering heavy nutrient loads by plants preventing release into surrounding ecosystems.

Benefits

For both aquaculturists and horticulturists, benefits from integrating exist (Diver, 2006). For horticulture growers, dependent upon individual situations, fish waste may meet USDA standards for organic certification. If crop production meets USDA organic criteria, crops can be advertised in niche markets as locally grown, and as organic crops for increased profit. Further, crop production can serve as a means of biofiltration for recirculating aquaculture systems (RAS), increasing sustainability. Integrated systems are often appealing to consumers who are becoming increasingly environmentally conscious. Using wastewater as an irrigation source for crop production can recycle and use pre-existing byproducts from aquaculture systems, which may prove to be economically sensible, producing two products from one production system's resources (Diver, 2006).

Benefits of integrated aquaculture and horticulture systems (IAHS), however, may have more substantial impacts than listed above. Increased production of both aspects of IAHS is possible. Previous work indicates that with IAHS, production rates for aquaculture rise significantly from 2,720 – 6,800 kg (6,000 – 15,000 lbs) fish·acre⁻¹·year⁻¹ to 136,000 – 363,000 kg (300,000 – 800,000 lbs) fish·acre⁻¹·year⁻¹ (Neori et al., 2004; Rakocy, 2002; Rakocy et al., 2000). An aquaponics system at the University of the Virgin Islands (UVI) generated \$110,000 producing 5,000 kg (11,000 lbs) basil annually and

2,900 kg (6,400 lbs) of okra valued at \$6,400 (Rakocy et al., 2006). In another UVI study, basil was produced via hydroponics (batch and staggered) and in a traditional field setting. Results indicated greater basil yields in hydroponic system over field production: 25.0 kg·m⁻² (batch), 23.4 kg·m⁻² (staggered), and 7.8 kg·m⁻², respectively (Rakocy et al., 2004a). The value of basil produced was US \$550·m⁻²·year⁻¹ (batch), US \$515·m⁻²·year⁻¹ (staggered), and US \$172·m⁻²·year⁻¹. In Alberta Canada, while not operating at full capacity, greenhouse production of tomato, cucumber, and basil was recorded, respectively, at 40 kg·m⁻², 100 kg·m⁻², and 42 kg·m⁻² annually, indicating greater yields than traditional greenhouse production in Canada (Rakocy et al., 2007b).

Historically, data shows an exponential rise in fertilizer costs (NASS, 1990; NASS, 2000; NASS, 2009). Data was collected from farmers in the Southeastern region (AL, FL, GA, SC) and the Delta region (AR, MS, LA), later merged into one region, the Southern region. In 1989, total expenditures of the Southern region for fertilizer inputs was approximately \$330 million (NASS 1990), increasing to \$1.75 billion in 1999 (NASS, 2000), and further increasing to \$4.28 billion in 2008 (NASS, 2009). Over a twenty-year span, fertilizer, lime and soil conditioner costs have risen some 120%. Facing unprecedented cost increase such as expressed here, cost saving measures are necessary for producers. For greenhouse and poultry house heating during winter, a rise in fuel prices has impacted producers, costing some farmers up to 67% more than previous years (Davis, 2003). During 2004, farmers reported fuel and fertilizer costs to be 30% increased from 2003 (Helms, 2004). Such drastic increases in fertilizer costs, as well as fuel cost, encourage use of alternative and supplemental fertilizer additions. Effluent water from aquaculture production is one viable option.

Due to effects of recent droughts, water reuse is strongly recommended. Record drought conditions were observed during August 2007, when Alabama drought status reached “extreme drought” according Palmer Drought Index (PDI) (Ding, 2008). Alabama, though rich in water resources, annually receiving 1,400 mm (55 in) precipitation, is home to 124,000 km (77,000 mi) of rivers and streams, and has 226,600 ha (560,000 ac) of water (ponds, lakes, reservoirs) (Marcus and Kiebzak, 2008), faces limited access to water resources due to governing bodies and laws. The USGS estimated in 2000 that surface water applied for agriculture irrigation in Alabama was 28.7 million gallons daily, or 2% of total surface water use per day. Due to such little surface water use, the question, “Why is water reuse so important?” is posed.

To answer, consider that approximately 96% of Alabama agriculture is rain-fed, indicating that only 4% of agricultural endeavors in state use 28.7 million gallons of water daily (Marcus and Kiebzak, 2008). Thus rain dependent farmers are threatened during seasonal and annual fluctuations in precipitation, so water availability is of great concern. After the drought in 2007, proving to be of the driest summers in Alabama history, farmers and the green industry as a whole suffered much loss of produce. Late June 2007, many Alabama farmers determined that farming in the given conditions was useless and did not plant, or subsequently harvest (Gutzmer et al., 2007). In the nursery and landscape sectors of agriculture, losses observed were extensive. During 2006 California and multiple southeastern states comprised 60% of gross national sales in the green industry (Ding, 2008). Yet, in 2007, the green industry witnessed decreased sales, increased plant mortality, increased watering expenses, and many businesses laid employees off, closed store locations, and even filed for bankruptcy (Ding, 2008).

Concluding from pressing water rights issues, recent environmental trends, and economic volatility, the prospective of integrated systems for the conservation and reuse of water is becoming increasingly favorable. For farmers who are dependent upon rain, integrating horticulture production with pre-existing aquaculture production facilities is a potentially simple solution to limited water resources.

Historical and Modern Examples IAHS

Integrated plant-fish culturing is a centuries old practice, dating back before the time of Christ. Centuries of integrated rice production and fish farming have proved to be priceless polyculture food production systems in Asia. Records indicate production of common carp (*Cyprinus carpio*) in China as early as 1400-1150 BC (Li, 1992). The production of rice and fish in the same plot of land is also traced to originate in China over 1700 years past (Ali, 1992; Coche, 1967). Multiple forms of rice-fish integrated farming exist, including concurrent crops, rotational crops, or alternating culture systems (Halwart and Gupta, 2004). The degree of increased economies between rice monoculture and integrated rice-fish culture varies by region of the world, regional economies, as well as other factors. Rice-fish production in Bangladesh was indicated to be over 50% more profitable than rice monoculture (Gupta et al., 1999). Chinese integrated culture demonstrated an economic advantage between 45% and 270%, where rice-fish culture was three times more profitable than rice production alone (Yan et al., 1995). Increased economies are a result of increased rice yields, lower labor inputs, and decreased materials cost (Li, 1992; Lin et al., 1995).

Research has been conducted evaluating a variety of plants, including seaweed, as

biofilters, and integrating marine aquaculture with seaweed production (Schuenhoff et al., 2003). In freshwater aquaculture, much development has occurred over recent decades involving the integration of hydroponic plant production. Multiple models have been developed, employing nutrient film technique (NFT) or aggregate hydroponics for plant culturing, with several models developed in recent decades (Diver, 2006).

Out of North Carolina State University (NCSU) a model was developed incorporating tilapia production with vegetable culture. Fish tanks are held underground; while vegetables located in ground level, sand-filled hydroponic beds were irrigated with effluent water. NCSU grew tomatoes and cucumbers, with roots serving biofiltration purposes. Results indicate that with integrated aquaponic systems water consumption in integrated models used one percent of the amount of water to produce similar tilapia yields as in pond culture (Diver, 2006).

Developed and modeled loosely upon the NCSU system, is the Speraneo system, developed by Tom and Paula Speraneo, operators of S&S Aqua Farm in Missouri. Tilapia are reared in tanks linked to a solar greenhouse and a full-scale commercial greenhouse for producing a wide variety of vegetables, herbs, and ornamentals. The Speraneo system was developed with nodes, or a single fish tank linked to a single hydroponic bed, allowing each node to be independent of all the others. Yields of produce to fish were calculated to be 20-32 kg (45-70 lbs) produce to every pound of tilapia grown. Basil production was valued at $\$12 \cdot 0.45 \text{ kg}^{-1}$ ($\$12 \cdot \text{lb}^{-1}$) (Diver, 2006).

The Freshwater Institute, of the Conservation Fund, has developed two systems, including a high-tech, indoor RAS as well as a low-tech, outdoor RAS. Due to abundant fresh water stream sources through the Appalachian region, effluent treatment was a

concern. Through collaborative efforts, a hydroponic system was incorporated into the systems. Trial results indicate that nitrogen and phosphorus concentrations can be adequately removed from wastewater (Diver, 2006).

Lastly, and perhaps most famous, is the University of the Virgin Islands (UVI) aquaponic system model. James Rakocy is the designer behind this successful commercial scale aquaponic system. Producing Red and Nile tilapia, effluents are used to irrigate crops located in raft hydroponic beds. The UVI system has been used to produce basil, lettuce, and okra with notably higher yields than traditional field production (Diver, 2006).

Nutrient film technique (NFT) is a model hydroponic system, differing from those discussed above due to several factors. NFT involved plastic troughs, through which a film of effluent water flows continuously over exposed plant roots. As the effluent films passed over plant roots, roots receive water, oxygen, and absorb dissolved nutrients. Advantages include high plant density in troughs while using minimal space and the relative inexpensive nature of NFT components. However, solids must be removed so roots are not suffocated due to accumulation of solids upon them (Rakocy et al., 2006).

Integrated Intensive Aquaculture and Nursery/Greenhouse Crop Production

Alabama Green Industry

In Alabama alone, the Green Industry is not only high-input and high-intensity using up to 20,000 gallons water·acre⁻¹·day⁻¹, but run-off potential is great, impacting surface and ground water resources just as fish effluent discharge can (Avent, 2003; Berghage et al., 1999; Lea-Cox and Ross, 2001). As previously discussed, due to recent

drought threats, decreased water use is a potential for both production aspects. Before addressing potential benefits for the Green Industry particularly, consideration will be given to national and local nursery and greenhouse production levels.

Nursery production in Alabama, including nursery, greenhouse, floriculture and sod production, in 2007, accounted for over \$291 million in sales (ERS, 2007). During 2007, irrigated agriculture land in Alabama was estimated at 751,000 acres (Clark, 2009). During the same year Alabama had 2.33 ha (5.75 ac) dedicated to greenhouse vegetable production, while nationally there were 574 ha (1,400 ac) total, and in 2008 over 688,000 ha (1.7 million ac) of vegetables were harvested. In Alabama, just less than 2,428 ha (6,000 ac) of field grown vegetables were harvested in the same year, valued at \$17.2 million. Considering nursery production, Alabama has 57 ha (141 ac) of nursery stock under various forms of protection, and nearly 3,100 ha (6,758 ac) of open land dedicated to nursery stock production (Clark, 2009). Total area for nursery and greenhouse industry in Alabama is near 3,360 ha (8,300 ac). Considering the industry uses 20,000 gallons water·acre⁻¹·day⁻¹, estimated total water use is 166 million gallons daily.

Alabama has a growing floriculture industry, within the horticulture industry. During 2005-2006, total floriculture producers numbered 221, covering approximately 26 ha (64 ac) statewide (NASS, 2007b). These 221 floriculture producers accounted for \$89 million in sales at wholesale value. Several common annual bedding crops (begonia, impatiens, pansies/violas, petunia, and others) accounted for \$16.2 million in sales of flats, and flats of vegetables is valued at \$7.4 million. Sales of container bedding plants totals \$9.4 million during 2005-2006 (NASS, 2007b). However, due to voluntary reporting and a lack of data collection from many operations (such as Bonnie Plants with

annuals sales estimated in excess of \$172 million) total production is greater than reported (AFC, 2009; J. Sibley, personal communication).

During the last fifteen years a shift in hydroponic greenhouse vegetable production has been observed in Florida, moving from traditional crops (tomatoes and cucumber) to more market specific crops such as peppers, herbs, lettuce, and strawberries (Tyson and Hochmuth, 2009). Cucumber (*Cucumis sativus*) has become an important hydroponically produced greenhouse crop (Tyson et al., 2001), a trend that is continuing, reaching into Alabama.

Vegetable production is an important industry in Alabama. As previously mentioned, during 2008 2,400 ha (6,000 ac) of field grown vegetables were harvested, while only 2.33 ha (5.75 ac) were dedicated to greenhouse vegetable production. Statewide total vegetable production accounted for \$17.2 million in sales (Clark, 2009). Estimate of annual value of fruit, pecan, and vegetable production in Alabama is \$57 million in cash receipts (Higginbotham, 2004). Nationally, Alabama is nationally ranked for production of sweet potato (5th), pecan (7th), blueberry (12th), fresh-market tomatoes (12th), and fresh-market watermelons (16th) (Higginbotham, 2004). As mentioned, greenhouse vegetable production is a growing industry in Florida, and in Mississippi there are 6 ha (15 ac) of greenhouse tomato production (Helms, 2005). During 2005, in Alabama there were between 15 and 20 greenhouse vegetable producers in Alabama, including tomatoes, herbs, microgreens, cucumbers, and lettuce (Helms, 2005).

Traditional field production costs of vegetables and fruits are high (ACES, 2007 and 2008). For slicer cucumbers under irrigation, cost per acre in 2007 was \$4,500 (ACES, 2007). Handpicked, fresh-market sweetcorn production costs reach \$1,300

acre⁻¹. Fresh fruit production costs are exponentially higher. For field grown, irrigated strawberry production costs were estimated at \$16,900·ac⁻¹ in 2008. Nearly \$5,000 of strawberry production costs was fertilizer (ACES, 2008). Greenhouse vegetable production may provide farmers relief from such high production costs. Another area of potential increased profits is off-season greenhouse crop production. Off-season greenhouse production can reduce fuel/energy costs associated with shipping of fresh produce where unavailable and supports local economies (Schonbeck et al., 1991). For fruits such as strawberries, blueberries, peaches, watermelon, blackberries, cantaloupe, and others, winter greenhouse production may provide substantially higher revenues for crops traditionally produced in summer months.

Alabama Aquaculture Industry

Alabama has 25,000 water acres utilized for aquaculture production, yet with the resources available to use 250,000 water acres for aquaculture (Crews and Chappell, 2007a). The potential therefore of statewide aquaculture growth is significant, and thus potentially increasing possibilities of increased aquaculture-horticulture integration.

IAHS, previously discussed, maintains mutual benefits for each production component. Incorporating production of both aspects, on the same farm site, carries as well several benefits. Total land area for fish and crop production will decrease as a means of integrated farms, rather than having separate aquaculture and horticulture production sites (Edwards, 1989, 1998). Overall production costs may decrease due to several factors: intensified production and subsequent income (Costa-Pierce, 2002; Devendra and Thomas, 2002; Edwards, 1998), decreased water costs by recycling irrigation with effluent water (Little and Muir, 1987; Prein, 2002), decreased fertilizer

costs by replacing and/or supplementing traditional fertilizers with dissolved nutrients available from effluent (Al-Jaloud et al., 1993; D'Silva and Maughan, 1995), and decreased fuel costs due to production systems at same location.

Further, integrated systems may be small-scale business endeavors, in close proximity to markets, further reducing fuel costs (Diver, 2006). Other benefits may include the production of season specific crops during non-traditional seasons, such are fresh, locally grown strawberries during winter months. Although winter crop production is limited in Alabama, the outlook remains promising as researchers and growers in Florida are seeking to address barriers. Florida has increased potential for winter production due to mild winters, resulting in minimal heating inputs (Hochmuth, 2006).

Nutrient composition of fish effluent appears to be strictly dependent on a case-by-case evaluation. Intensive systems will produce a heavier nutrient load per water volume than more traditional extensive pond culture (Shireman and Cichra, 1994), feed input will change effluent composition (Tucker and Boyd, 1985), and seasonal differences are reported (Tucker, 1998). However, noted frequently, fish effluent does meet the nutritional demands of horticulture crops, with adequate concentrations of ammonia, nitrate, nitrite, phosphorus, potassium as well as others (Diver, 2006). Further, successful crop production is dependent upon plant species, wherein some species with moderate nutritional requirements may be better adapted for production under fish effluent (Nelson and Pade, 2009). Some plant species will thrive under almost any conditions such as lettuce, spinach, arugula, basil, mint, watercress, chives, and other leafy greens, while plant species such as tomatoes, peppers, cucumbers and other fruiting crops will require a more substantial nutrient regimen, more suited to established, heavily

stocked aquaculture production systems for integration (Nelson and Pade, 2009; Rakocy et al., 2006).

Accepted industry standards for plant nutrient tissue composition is: 1.5% N, 0.2% P, 1.0% K, 0.5% Ca, 0.2% Mg, 100 mg·L⁻¹ Fe, 50 mg·L⁻¹ Mn, 20 mg·L⁻¹ Zn, 6 mg·L⁻¹ Cu, 20 mg·L⁻¹ B, 0.1 mg·L⁻¹ Mo, and 100 mg·L⁻¹ Cl (Table 1.2) (Epstein, 1965). Sufficiency ranges for adequate nutritional composition of greenhouse crops are: 2.5-6% N, 0.30-1.0% P, 2.5-6% K, 0.6-2% Ca, 0.3-1.0% Mg, 0.3-1.0% S, 75-200 mg·L⁻¹ Fe, 50-200 mg·L⁻¹ Mn, 25-100 mg·L⁻¹ Zn, 5-20 mg·L⁻¹ Cu, 30-120 mg·L⁻¹ B, and 1-5 mg·L⁻¹ Mo (Table 1.2) (Argo et al., 2009). Recommended fertilization rates of bedding plants produced under greenhouse conditions are 50-100 mg·L⁻¹ N for plugs, 100-150 mg·L⁻¹ N for slight feeding crops, 150-200 mg·L⁻¹ N for moderate feeding crops, and 200-250 mg·L⁻¹ N for heavy feeders (Table 1.3) (Kessler, 2002). Common greenhouse and nursery production standards for bedding plants, fertilizer at rates between 200 and 500 mg·L⁻¹ N (James and van Iersel, 2001; Nelson, 1994; Rader, 1998).

Mentioned previously, nutritional characteristics of effluent vary case-to-case, however a number of regional averages have been compiled (Tucker, 1998). During a two year period, twenty five catfish farms in Alabama had effluent compositions monitored with averaged results of: 0.073 mL·L⁻¹ settleable solids, 72.63 mg·L⁻¹ TSS, 4.35 mg·L⁻¹ total nitrogen (TN), 1.17 mg·L⁻¹ TAN, 0.24 mg·L⁻¹ TP, and 9.3 mg O₂·L⁻¹ BOD. In Mississippi, effluent from twenty commercial catfish ponds were monitored, averaging 0.078 mL·L⁻¹ settleable solids, 109.63 mg·L⁻¹ TSS, 5.8 mg·L⁻¹ TN, 1.62 mg·L⁻¹ TAN, 0.37 mg·L⁻¹ TP, and 15.57 mg O₂·L⁻¹ BOD. Hybrid striped bass pond production in South Carolina contained effluent characterized with <0.4 mg·L⁻¹ settleable solids, 49.2 mg·L⁻¹

TSS, $0.95 \text{ mg}\cdot\text{L}^{-1}$ TAN, $7.06 \text{ mg}\cdot\text{L}^{-1}$ kjeldahl nitrogen, $0.07 \text{ mg}\cdot\text{L}^{-1} \text{NO}_2^-$, $0.304 \text{ mg}\cdot\text{L}^{-1}$ TP, and $11.6 \text{ mg}\cdot\text{L}^{-1}$ BOD (Table 1.4) (Tucker, 1998). From Brazil, Nile tilapia stocked ponds had a chemical composition of: $1.95 \text{ mg}\cdot\text{L}^{-1} \text{NH}_3\text{-N}$, $0.071 \text{ mg}\cdot\text{L}^{-1} \text{NO}_2\text{-N}$, $0.8 \text{ mg}\cdot\text{L}^{-1} \text{NO}_3\text{-N}$, $0.013 \text{ mg}\cdot\text{L}^{-1} \text{PO}_4^{3-}$, and $109.20 \text{ mg}\cdot\text{L}^{-1} \text{K}^+$ (Castro et al., 2006). In a batch culture system, in which basil was produced, water quality parameters appeared as: $2.2 \text{ mg}\cdot\text{L}^{-1}$ TAN, $0.7 \text{ mg}\cdot\text{L}^{-1} \text{NO}_2^-$ -N, $42.2 \text{ mg}\cdot\text{L}^{-1} \text{NO}_3^-$ -N, $532 \text{ mg}\cdot\text{L}^{-1}$ total dissolved solids (TDS), $11.9 \text{ mg}\cdot\text{L}^{-1}$ Ca, $6.5 \text{ mg}\cdot\text{L}^{-1}$ Mg, $44.9 \text{ mg}\cdot\text{L}^{-1}$ K, $8.2 \text{ mg}\cdot\text{L}^{-1}$ P, $2.5 \text{ mg}\cdot\text{L}^{-1}$ Fe, $0.80 \text{ mg}\cdot\text{L}^{-1}$ Mn, $0.05 \text{ mg}\cdot\text{L}^{-1}$ Cu, $0.44 \text{ mg}\cdot\text{L}^{-1}$ Zn, $0.19 \text{ mg}\cdot\text{L}^{-1}$ B, and $0.01 \text{ mg}\cdot\text{L}^{-1}$ Mo (Rakocy et al, 2004a).

Previous research indicates a specific production rate of N per amount feed fed (Rakocy et al., 2006). Intensive fish culture systems will have a greater feed input than traditional extensive systems, therefore an increased N concentration. Intensive systems produce fish at an approximate stocking density of $59.4 \text{ kg fish}\cdot\text{m}^{-3}$ (Masser et al., 1999). Estimated N concentrations from feed range from $1 \text{ kg TAN}\cdot 45.4 \text{ kg feed fed}^{-1}$ ($2.2 \text{ lbs TAN}\cdot 100 \text{ lbs feed fed}^{-1}$) (Masser et al., 1999). Given a specific feedings rate, N concentration can be determined, and further extrapolated to feed per plant production area. UVI recommendations, for raft hydroponics, are $60\text{-}100 \text{ g fish feed}\cdot\text{m}^{-2}$ of plant production area (Rakocy et al., 2006). Based upon feeding rate listed above, a plant production system consisting of 100 m^2 would need 10 kg (22 lbs) of feed. At given rate, TAN concentrations can be estimated to be 98 g (0.22 lbs). UVI aquaponic system reared tilapia at two different stocking densities, $77 \text{ fish}\cdot\text{m}^{-3}$ (Nile tilapia) and $154 \text{ fish}\cdot\text{m}^{-3}$ (Red tilapia), with feed conversion rates of 1.7 and 1.8 respectively. At this production level, estimated TAN is 8.7 kg (19.2 lbs) for Nile tilapia, and 10.6 kg (23.4 lbs) for Red tilapia.

Knowing the concentrations of various forms of nitrogen in a fish culturing system is desirable for knowing the amount of N available to plants. Calculating N from feed inputs includes several forms of nitrogen, including TAN, NO_3^- , and NO_2^- . A generally accepted rule-of-thumb for on-farm fish population estimation is assuming a 2:1 feeding conversion ratio (FCR), indicating if a farmer were feeding 45.4 kg (100 lbs) feed, estimated weight of fish would be 22.7 kg (50 lbs) fish (B. Daniels, personal communication).

Although impossible to characterize any given aquaculture production systems' effluent based on the nutrient and chemical composition of various systems, sufficient levels of nitrogen appear to be available for plant production (Castro et al., 2006; Clarkson and Lane, 1991; Palada et al., 1999; Rakocy et al., 2006; Rakocy et al., 2004a; Savidov, 2004; Schuenhoff et al., 2003; Seawright et al., 1998; Tucker, 1998). Despite analyses indicating sufficient nutrient levels, a need for supplemental nutrients has been reported. Supplemental inputs counteracting deficient levels of N, P, K, Ca, and Fe may be needed (Al-Hafedh, 2008; Graber and Junge, 2009; Rakocy et al., 2004a). In a tilapia-vegetable aquaponic (AP) and hydroponic systems, tomatoes grown hydroponically and in AP system indicated K concentrations in fruit analysis to be: 40.8 g $\text{K} \cdot \text{kg}^{-1}$ dry matter (hydroponics) and 22.0 g $\text{K} \cdot \text{kg}^{-1}$ dry matter (AP). In another study, though nutrient levels were low compared to hydroponic formulations, nutritional deficiencies symptoms were not observed, due to constant replenishment of nutrients (Al-Hafedh, 2008). During one study investigating an AP system and a greenwater (GW) system, nutrients needing supplementation were Ca, K, and Fe in an AP system, while in a GW system Ca and Cl were supplemented (Rakocy et al., 2007a). During a 29-day

study period, K, NO_3^- N, SO_4^- S, PO_4^- P, Na, Cl, Ca, Mg, B, Mo increased in concentrations, while NH_4^- N and Fe decreased, and Mn, Zn, and Cu fluctuated throughout the study. Sludge from the AP system indicated an increase of NO_3^- N ($\text{mg}\cdot\text{L}^{-1}$) by 13,568% from initial concentration to final concentration (Rakocy et al., 2007a). Further research is needed concerning mineral additions to account for limiting concentrations of given nutrients (N, P, K, Ca, Fe, Cl). A study evaluating productivity of AP system (tilapia with tomatoes, cucumbers, and aubergine production) noted additional KOH to fish culture system for pH stabilization, to be an effective addition to counteract limiting concentrations of K (Graber and Junge, 2009), however this was not a corrective measure for plant production but displayed secondary benefits to plant culture system.

As indicated from recommended greenhouse crop nutrient requirements and chemical analysis of a variety of aquaculture effluents, aquaculture waste effluents appear to have the means to meet the nutritional demand of horticulture crops. Total nitrogen from catfish farms in Alabama and Mississippi ranged from $4.35\text{-}5.8\text{ mg}\cdot\text{L}^{-1}$, while kjeldahl nitrogen from South Carolina HSB farms was $7.06\text{ mg}\cdot\text{L}^{-1}$, which should meet nutritional demands of plant based on recommended plant nutritional composition.

Conclusion

Due to factors such as a growing global market, slack overseas aquaculture regulations, increasing imports, increasing domestic regulations, and an unpredictable economy, fish farmers in America, and Alabama are facing an uncertain industry outlook. Integrating intensive aquaculture production and horticulture crop production is a sensible endeavor, with great potential for increased revenues. Inherent to integrated

systems are benefits such as polyculture from otherwise wasted valuable byproducts, increased environmental protection, and increased competition amongst increasingly competitive markets.

The following chapters will discuss studies conducted during August 2008 and November 2009, evaluating traditional horticulture crop production methods (clear water and fertilizer inputs) and the use of intensive aquaculture effluent water for crop irrigation, fertilization, and production. The research objectives were to determine nutritional suitability of intensive aquaculture effluent water for utilization in production of horticulture crops, as a replacement or supplement to synthetic fertilizer inputs. Determination of nitrogen produced in fish culturing system, portion of nitrogen available for uptake by plants, and plant performance compared to standard nursery production practices.

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Table 1.1. Fraction of toxic, unionized ammonia (NH₃)^z at varying water temperature and pH values.^y

pH	Temperature (°C)												
	6	8	10	12	14	16	18	20	22	24	26	28	30
7.0	0.0013 ^x	0.0016	0.0018	0.0022	0.0025	0.0029	0.0034	0.0039	0.0046	0.0052	0.0060	0.0069	0.0080
7.2	0.0021	0.0025	0.0029	0.0034	0.0040	0.0046	0.0054	0.0062	0.0072	0.0083	0.0096	0.0110	0.0126
7.4	0.0034	0.0040	0.0046	0.0054	0.0063	0.0073	0.0085	0.0098	0.0114	0.0131	0.0150	0.0173	0.0198
7.6	0.0053	0.0063	0.0073	0.0086	0.0100	0.0116	0.0134	0.0155	0.0179	0.0206	0.0236	0.0217	0.0310
7.8	0.0084	0.0099	0.0116	0.0135	0.0157	0.0182	0.0211	0.0244	0.0281	0.0322	0.0370	0.0423	0.0482
8.0	0.0133	0.0156	0.0182	0.0212	0.0247	0.0286	0.0330	0.0381	0.0438	0.0502	0.0574	0.0654	0.0743
8.2	0.0210	0.0245	0.0286	0.0332	0.0385	0.0445	0.0514	0.0590	0.6760	0.0772	0.0880	0.0998	0.1129
8.4	0.0328	0.0383	0.0445	0.0517	0.0597	0.0688	0.0790	0.0904	0.1031	0.1171	0.1326	0.1495	0.1678
8.6	0.0510	0.0593	0.0688	0.0795	0.0914	0.1048	0.1197	0.1361	0.1541	0.1737	0.1950	0.2178	0.2442
8.8	0.0785	0.0909	0.1045	0.1204	0.1376	0.1566	0.1773	0.1998	0.2241	0.2500	0.2774	0.3062	0.3362
9.0	0.1190	0.1368	0.1565	0.1782	0.2018	0.2273	0.2546	0.2836	0.3140	0.3456	0.3783	0.4116	0.4453
9.2	0.1763	0.2008	0.2273	0.2558	0.2861	0.3180	0.3512	0.3855	0.4204	0.4457	0.4909	0.5258	0.5599
9.4	0.2533	0.2847	0.3180	0.3526	0.3884	0.4249	0.4618	0.4985	0.5348	0.5702	0.6045	0.6373	0.6685
9.6	0.3496	0.3868	0.4249	0.4633	0.5016	0.5394	0.5762	0.6117	0.6456	0.6777	0.7078	0.7358	0.7617
9.8	0.4600	0.5000	0.5394	0.5778	0.6147	0.6499	0.6831	0.7140	0.7428	0.7692	0.7933	0.8153	0.8351
10.0	0.5745	0.6131	0.6498	0.6844	0.7166	0.7463	0.7735	0.7983	0.8207	0.8408	0.8588	0.8749	0.8892
10.2	0.6815	0.7152	0.7463	0.7746	0.8003	0.8234	0.8441	0.8625	0.8788	0.8933	0.9060	0.9173	0.9271

^zTo determine NH₃ concentration: based on system pH and temperature, find NH₃ fraction and multiply TAN (total ammonium nitrogen),

NH₄⁺ and NH₃ by fraction given to determine total mg·L⁻¹ NH₃.

^yEmerson, K., R.C. Russo, R.E. Lund, and R.V. Thruston. 1975. Aqueous ammonia equilibrium calculations: Effects of pH and temperature.

Board of Canada. 32:2379-2383.

^xUnit = mg·L⁻¹

Table 1.2. Industry standard foliar nutrient analysis composition.

Nutrient	Unit	General plant nutrition	
		Epstein, 1965 ^Z	Argo et al., 2009 ^Y
Nitrogen	%	1.5	2.5-6.0
Phosphorus	%	0.2	0.3-1.0
Potassium	%	1.0	2.5-6.0
Calcium	%	0.5	0.6-2.0
Magnesium	%	0.2	.0.3-1.0
Sulfur	%	. ^x	0.3-1.0
Iron	ppm	100	75-200
Manganese	ppm	50	50-200
Zinc	ppm	20	25-100
Copper	ppm	6.0	20-May
Boron	ppm	20	30-120
Molybdenum	ppm	0.1	1.0-5.0
Chlorine	ppm	100	.

^Z Epstein, E. 1965. Mineral nutrition. In: J. Bonner and J.E. Varner (eds.) Plant Biochemistry. Academic Press, Inc. Orlando, FL. p. 438-466.

^Y Argo, B., P. Fisher, and K. Santos. 2009. Understanding plant nutrition: Diagnosing problems. GreenhouseGrower.com. 31 Aug. 2009.
<<http://www.greenhousegrower.com/magazine/?storyid=1883>>.

^xNo data provided.

Table 1.3. Recommended fertilization rates for greenhouse production of bedding plants.^z

Nutrient requirement	Rate (mg·L ⁻¹)	Crop
Seedling	50-100	Plug flats
Light	100-150	Africa Violet, Ageratum, Azalea, Begonia, Celosia, Coleus, Cosmos, Dianthus, Gloxinia, Impatiens, Lobelia, Marigold, Melampodium, Pansy, Pentas, Petunia, Portulaca, Primula, Salvia, Snapdragon, Verbena, Vinca
Moderate	150-200	Begonia, Carnation, Coleus, Cyclamen, Dahlia, Dusty Miller, Easter lily, Fuchsia, Geranium, Gerbera Daisy, Holiday Cactus, Hydrangea, Ivy Geranium, Kalanchoe, Lisianthus, Ornamental Cabbage/Kale, Portulaca, Rose, Sunflower, Verbena, Zinnia
Heavy	200-250	Chrysanthemum, Poinsettia

^z Kessler, J.R. 2002. Fertilizing greenhouse crops in Alabama. Cooperative Extension System. ANR-1221.

Table 1.4. Aquaculture effluent nutrient analysis from farms in AL, MS, and SC.^z

	Unit	AL Catfish	MS Catfish	SC Hybrid Striped Bass
Settle Solids	mL·L ⁻¹	0.073	0.078	<0.4
TSS ^y	mg·L ⁻¹	72.63	109.63	49.2
TN ^x	mg·L ⁻¹	4.35	5.8	.
TAN ^w	mg·L ⁻¹	1.17	1.62	0.95
Kjeldahl N	mg·L ⁻¹	. ^t	.	7.06
TP ^v	mg·L ⁻¹	0.24	0.37	0.304
BOD ^u	mg·L ⁻¹	9.3	15.57	11.6

^zTucker, C.S. 1998. Characterization and management of effluents from aquaculture ponds in the southeastern United States. Southern Regional Aquaculture Center. Final Report. 600.

^yTSS = Total suspended solids.

^xTN = Total nitrogen.

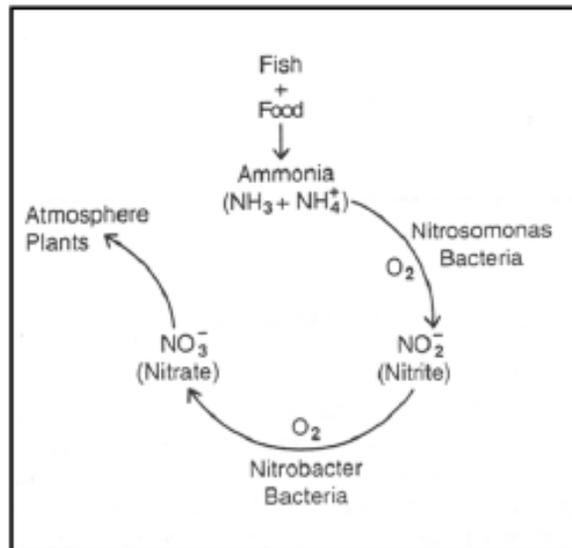
^wTAN = Total ammonium nitrogen.

^vTP = Total phosphorus.

^uBOD = Biological oxygen demand.

^tNo data provided.

Figure 1.1. The nitrogen cycle.^{ZY}



^ZNitrifying bacteria utilize O₂ to convert ammonia (NH₄⁺, NH₃) and nitrite (NO₂⁻) into the nontoxic byproduct, nitrate (NO₃⁻), which is then used by plants or returned to the atmosphere (N₂).

^YFrancis-Floyd, R. and C. Watson. 1996. Ammonia. Institute of Food and Agricultural Services, Cooperative Extension Service. University of Florida. FA-16.

II. Utilization of Intensive Aquaculture Wastewater for Greenhouse Crop Production

Abstract

Irrigation with wastewater from intensive tilapia culture was evaluated as a replacement/supplement to standard greenhouse crop production methods (clear water and fertilizer inputs). Variations of clear water (CW) and effluent water (EW) irrigation were applied with fertilizer inputs consisting of 20-20-20 soluble fertilizer at $200 \text{ mg}\cdot\text{L}^{-1}$, differing rates of 14-14-14 controlled release fertilizer, and incorporation of Micromax[®]. Effluent water applications without any supplemental fertilizer were made as well. In the first experiment, growth index (GI) differences were observed between treatments for *Angelonia angustifolia* and *Petunia x hybrida*, while for *Verbena x hybrida* differences across treatments were not observed. Shoot dry weights (SDW) between treatments for individual species indicate no differences for angelonia and verbena, while petunia exhibited differences. Foliar analyses were conducted and plant tissue nutrient compositions indicated differences between treatments, varying above and below recommended levels. Experiment two had similar results to study one. Trends indicate petunia crops perform best under standard greenhouse production methods compared with EW based upon differences observed in GI and SDW. However, EW did appear to be a viable fertilizer supplement for reducing costs. Vinca results indicate EW was sufficient without supplemental fertilizer when compared with traditional inputs. No differences were observed in GI and SDW for vinca across all treatments.

Index of Words: Horticulture, bedding plants, angelonia, nemesia, petunia, verbena, effluent, fertilizer

Species used in Study: *Angelonia angustifolia*, *Catharanthus roseus* ‘Cooler Rose’, *Nemesia fruticans*, *Petunia x hybrida*, *Verbena x hybrida*

Significance to Industry

Tank effluent from intensive cultured tilapia (*Oreochromis niloticus*) was evaluated as a source of irrigation and fertilization for greenhouse crop production. Plants grown using effluent-laden water from intensive aquaculture performed similarly to plants grown under standard production means. As production costs continue to rise and environmental policies grow more stringent, effluent from intensive fish culture might provide an economical and environmentally sustainable alternative to current water use and fertilizer practices in plant production and intensive aquaculture.

Introduction

Agricultural integrations offer potentially long-term positive effects. Assimilating various sectors of agriculture provides increased levels of sustainability, protection of human and environmental health, increased revenues, extended life of materials once thought to be fully used, provision of alternatives to replace limited naturally occurring materials, and providing aid in producing the same quality products as traditional production means. Due to benefits listed, an argument for sustainable and integrated agriculture carries more significance in a changing world. Among such integrated

agriculture endeavors is the growing trend of integrating aquaculture and horticulture. For horticulture growers, dependent upon individual situations, fish waste from aquaculture may meet USDA standards for organic certification (NOP, 2008). Crops that meet USDA organic criteria can be marketed in niche markets as locally grown as well as organic crops, for increased profit. Further, crop production can serve as a means of biofiltration for recirculating aquaculture systems (RAS), increasing sustainability. Integrated systems are often appealing to consumers who are becoming increasingly environmentally conscious. Using wastewater as an irrigation source for crop production can recycle and use pre-existing byproducts from aquaculture systems (Diver, 2006).

Benefits of integrated aquaculture and horticulture systems (IAHS), however, may have more substantial impacts than listed above. Increased production in both aspects of IAHS is possible. Previous work indicates production rates for IAHS rise significantly from 2,720 – 6,800 kg (6,000 – 15,000 lbs) fish·acre⁻¹·year⁻¹ to 136,000 – 363,000 (300,000 – 800,000 lbs) fish·acre⁻¹·year⁻¹ (Neori et al., 2004; Rakocy, 2002; Rakocy et al., 2000). An aquaponics system at the University of the Virgin Islands (UVI) generated \$110,000 producing 5,000 kg (11,000 lbs) basil annually and 2,900 kg (6,400 lbs) of okra valued at \$6,400 (Rakocy et al., 2006). In another UVI study, basil was produced via hydroponics (batch and staggered) and in a traditional field setting with greater basil yields in hydroponic systems over field production: 25.0 kg·m⁻² (batch), 23.4 kg·m⁻² (staggered), and 7.8 kg·m⁻², respectively (Rakocy et al., 2004). The value of basil produced was US \$550·m⁻²·year⁻¹ (batch), \$515·m⁻²·year⁻¹ (staggered), and \$172·m⁻²·year⁻¹. In Alberta Canada, while not operating at full capacity, greenhouse production of tomato, cucumber, and basil was recorded, respectively, at 40 kg·m⁻², 100 kg·m⁻², and 42

kg·m⁻² annually, indicating greater yields than traditional greenhouse production in Canada (Rakocy et al., 2007).

Due to effects of recent droughts in southeastern US, water reuse is often recommended. Record drought conditions were observed during August 2007, when Alabama drought status reached “extreme drought” according Palmer Drought Index (PDI), further encouraging water conservation production practices (Ding, 2008). Environmental instability has lead to decreased profit margins for many horticulture producers. In nursery and landscape sectors, financial losses observed were extensive. During 2006 California and multiple southeastern states comprised 60% of gross national sales in the Green industry. Yet, in 2007, industry from these regions witnessed decreased sales, increased plant mortality, increased watering expenses, and many businesses laid employees off, closed store locations, or filed for bankruptcy (Ding, 2008).

Fertilizer costs have increased 120% in past two decades (NASS, 2009; NASS, 2000; NASS, 1990), encouraging alternative fertilizer inputs. Fuel costs have risen too. Production costs have risen as a result to increased prices of fertilizers and fuel. The need for alternative fertilizer sources is great, and effluent water from aquaculture is one viable option.

Nursery production in Alabama is comprised of 57 ha (141 ac) of nursery stock under various forms of protection, and nearly 3100 ha (6,758 ac) of open land nursery stock production (Clark, 2009). Total area readily available for the nursery and greenhouse industry in Alabama is near 3360 ha (8,300 ac). Considering the industry uses 20,000 gallons water·acre⁻¹·day⁻¹ (Avent, 2003; Berghage et al., 1999; Lea-Cox and Ross, 2001), estimated total water use is 166 million gallons daily. Integrated systems offer

means to reduce water use dramatically. RAS can use as little as 1-2% of water required a day, meaning 98-99% is reused.

Alabama has a growing floriculture industry, with 221 total floriculture producers, covering approximately 26 ha (64 ac) with \$89 million in sales at wholesale value during 2005-2006 (NASS, 2007). Several common annual bedding crops (begonia, impatiens, pansies/violas, petunia, and others) accounted for \$16.2 million in sales (flats), with sales of vegetable flats valued at \$7.4 million. Sales of container bedding plants were \$9.4 million during same time period (NASS, 2007).

Integrating aquaculture and horticulture systems requires adequate nutrients to be available for plants from effluent water. Noted frequently, fish effluent could meet the nutritional demands of horticulture crops, with adequate concentrations of ammonia, nitrate, nitrite, phosphorus, potassium as well as others (Diver, 2006). Sufficiency ranges for adequate nutritional composition of greenhouse crops are: 2.5-6% N, 0.30-1.0% P, 2.5-6% K, 0.6-2% Ca, 0.3-1.0% Mg, 0.3-1.0% S, 75-200 mg·L⁻¹ Fe, 50-200 mg·L⁻¹ Mn, 25-100 mg·L⁻¹ Zn, 5-20 mg·L⁻¹ Cu, 30-120 mg·L⁻¹ B, and 1-5 mg·L⁻¹ Mo (Table 2.1) (Argo et al., 2009). Recommended fertilization rates of bedding plants produced under greenhouse conditions are 50-100 mg·L⁻¹ N for plugs, 100-150 mg·L⁻¹ N for slight feeding crops, 150-200 mg·L⁻¹ N for moderate feeding crops, and 200-250 mg·L⁻¹ N for heavy feeders (Kessler, 2002).

Nutritional characteristics of effluent vary case-to-case, however a number of regional averages have been compiled (Tucker, 1998). During a two year period, twenty five catfish farms in Alabama had effluent compositions monitored with averaged results of: 0.073 mL·L⁻¹ settleable solids, 72.63 mg·L⁻¹ TSS, 4.35 mg·L⁻¹ total nitrogen (TN), 1.17

mg·L⁻¹ TAN, 0.24 mg·L⁻¹ TP, and 9.3 mg O₂·L⁻¹ BOD. In Mississippi, effluent from twenty commercial catfish ponds were monitored, averaging 0.078 mL·L⁻¹ settleable solids, 109.63 mg·L⁻¹ TSS, 5.8 mg·L⁻¹ TN, 1.62 mg·L⁻¹ TAN, 0.37 mg·L⁻¹ TP, and 15.57 mg O₂·L⁻¹ BOD. Hybrid striped bass pond production in South Carolina contained effluent characterized with <0.4 mg·L⁻¹ settleable solids, 49.2 mg·L⁻¹ TSS, 7.06 mg·L⁻¹ kjeldahl nitrogen, 0.07 mg·L⁻¹ NO₂⁻, 0.95 mg·L⁻¹ TAN, 0.304 mg P·L⁻¹ TP, and 11.6 mg O₂·L⁻¹ BOD (Tucker, 1998). From Brazil, Nile tilapia stocked ponds had a chemical composition of: 1.95 mg·L⁻¹ NH₃-N, 0.071 mg·L⁻¹ NO₂-N, 0.8 mg·L⁻¹ NO₃-N, 0.013 mg·L⁻¹ PO₄³⁻, and 109.20 mg·L⁻¹ K⁺ (Castro et al., 2006).

Previous research indicates a specific production rate of N per amount feed fed (Rakocy et al., 2006). Intensive fish culture systems will have a greater feed input than traditional extensive systems, therefore an increased N concentration. Intensive systems produce fish at an approximate stocking density of 59.4 kg fish·m⁻³ (Masser et al., 1999). Estimated N concentrations from feed range from 1 kg TAN·45.4 kg feed fed⁻¹ (2.2 lbs TAN·100 lbs feed fed⁻¹) (Masser et al., 1999). Given a specific feeding rate, N concentration can be determined, and further extrapolated to feed per plant production area. UVI recommendations, for raft hydroponics, are 60-100 g fish feed·m⁻² of plant production area (Rakocy et al., 2006). Based upon feeding rate listed above, a plant production system consisting of 100 m² would need 10 kg (22 lbs) of feed. At the given rate, TAN concentrations can be estimated to be 98 g (0.22 lbs). UVI aquaponic system reared tilapia at two different stocking densities, 77 fish·m³ (Nile tilapia) and 154 fish·m³ (Red tilapia), with feed conversion rates of 1.7 and 1.8 respectively. At this production level, estimated TAN is 8.7 kg (19.2 lbs) for Nile tilapia, and 10.6 kg (23.4 lbs) for Red

tilapia. Knowing the concentrations of various forms of nitrogen in a fish culturing system is desirable for knowing the amount of N available to plants. Calculating N from feed inputs includes several forms of nitrogen, including TAN, NO_3^- , and NO_2^- . A generally accepted rule-of-thumb for on-farm fish population estimation is assuming a 2:1 feeding conversion ratio (FCR), indicating if a farmer were feeding 45.4 kg (100 lbs) feed, estimated weight of fish would be 22.7 kg (50 lbs) fish (B. Daniels, personal communication).

Although impossible to characterize any given aquaculture production systems' effluent based on the nutrient and chemical composition of various systems, sufficient levels of N appear to be available for plant production (Castro et al., 2006; Clarkson and Lane, 1991; Palada et al., 1999; Rakocy et al., 2006; Rakocy et al, 2004a; Savidov, 2004; Schuenhoff et al., 2003; Seawright et al., 1998; Tucker, 1998). The objective of this study was to evaluate effluent water from intensive tilapia culture as an irrigation replacement, substituting or supplementing fertilizer inputs used in standard greenhouse production methods.

Materials and Methods

Nile tilapia (*Oreochromis niloticus*) was produced in an intensive, bio-floc production system at the E.W. Shell Fisheries Center, North Auburn Unit, in Auburn, AL (Cold Hardiness Zone 8) during 2008 and 2009. Production was conducted in a 29.3 x 9.1 m (96 x 30 ft) double layer, polyethylene covered greenhouse, for environmental control in year-round production. Fish were stocked at $80 \text{ fish} \cdot \text{m}^{-3}$, in two 27.4 x 3.8 m x 1.2 m (90 ft x 12.5 ft x 4 ft) tanks each with a volume of 125 m^3 (33,000 gal), constructed of

plywood with steel I-beam and metal cable reinforcements, and lined with a 12 mm polyethylene liner. Each production tank was partitioned into four netted sections for grading fish by size and for staggered harvest, moving fish forward as size increased, until reaching a marketable weight of 0.45 kg (1 lb). Individual partitions were stocked at different times for a staggered harvest, the most recent stocking on 1 August 2009 of approximately 10,000 50 g tilapia fingerlings. Daily water exchange rates were maintained at low levels, averaging 1-6%, but up to 12% when excess flushing was needed. Tank water was exchanged with water from a local reservoir and with well water. Aeration was provided from two 1.5 hp air-blowers (Sweetwater[®], AES Inc., Apopka, FL).

During winter months, water temperature was maintained within a range suitable for tilapia production ($\geq 22^{\circ}\text{C}$), and recirculation of warm air for aeration provided further warming of water. For heating, a 200,000 BTU corn burner was utilized, cycling reservoir water through the burner at $3.78 \text{ L}\cdot\text{min}^{-1}$ (two $\text{gal}\cdot\text{min}^{-1}$), heated to approximately 48.8°C (120°F). During daylight hours, heated water was provided to the fish production greenhouse, while heating was provided to an adjacent plant greenhouse during the night. Shelled corn, priced at \$2.50-\$3.00 per bushel was used as a fuel, ($\approx 7500 \text{ BTU}\cdot\text{lb}^{-1}$), and pelletized wood was utilized as well.

Tilapia production yields averaged near 9,000-11,000 kg (10-12 tons) of fish for the entire system annually (250 m^3 ; 66,043 gallons water). Estimated production per acre is 136,000-181,400 kg (300,000-400,000 lb) annually.

Dissolved oxygen and temperature of fish culturing water were recorded twice daily (YSI 550A, YSI Inc., Yellow Springs, OH) (Figure 2.1). Water temperature

exhibited an increasing trend between December 2008 and September 2009, however temperatures can be expected to drop during winter months. Culture water DO indicated a general decreasing trend during the same time frame. Culture water pH, EC, and salinity (YSI 63 meter), TAN (1.0 to 8.0 mg·L⁻¹, LaMotte Company, Chestertown, MD), and NO₃⁻-N (Cardy meter, range 0 to 9,900 mg·L⁻¹, Spectrum Technologies, Inc., Plainfield, IL) were measured daily. Water samples were collected directly from fish tanks weekly and analyzed using ICAP and for NH₄ determination (Table 2.2). Tilapia were fed a 32% crude protein feed from Alabama Catfish Feed Mill (Uniontown, AL), fed between 22 kg to 34 kg (50 lbs to 75 lbs) feed·tank⁻¹·day⁻¹. During the production period of December 2008 to September 2009, a general increasing trend in feed fed was observed.

This study consisted of four stages, two preliminary trials and a subsequent experiment, which was repeated. All were conducted at E.W. Shell Fisheries Center, North Auburn Unit, in Auburn, AL 2009.

Trail 1

On January 31, 2009 plugs of *Nemesia Aromatica*TM (*Nemesia fruticans*) and *Petunia Vegetative Suncatcher* (*Petunia x hybrida*) from 162-plug trays (John McBryde, Ball Horticultural Company) were transplanted into common substrate (70% peat moss, 15% perlite, 15% vermiculite), in 15.24 cm (6 in) diameter containers with a volume of 1278 cm³ (6.0 AZ traditional TW, Dillen Products/Meyers Industries, Middlefield OH). The trail was conducted in a 29.3 x 9.1 m (96 x 30 ft) pad and fan greenhouse with a double layer, polyethylene cover. Dependent variables consisted of water source (clear water (CW), effluent water (EW)), top-dressed CRF, liquid fertilizer application, or no

fertilizer application. Each pot was filled level to container rim with substrate, irrigated with CW and allowed to settle, and plugs were stuck following settling. Ten treatments were: 1) CW with 1.6 kg (3.5 lb) CRF·yd⁻³ (low rate), 2) EW with low rate CRF, 3) CW with 3.4 kg (7.5 lb) CRF·yd⁻³ (medium rate), 4) EW with medium rate CRF, 5) CW with 5.4 (12 lb) CRF·y⁻³ (high rate), 6) EW with high rate CRF, 7) CW with 200 mg·L⁻¹ N from soluble fertilizer per watering, 8) EW with incorporated Micromax, 9) CW with incorporated Micromax, and 10) EW alone. The CRF used was 14-14-14 Nutricote (NPK, Florikan E.S.A., Sarasota, FL), the soluble fertilizer used was 20-20-20 TotalGro (NPK, SDT Industries, Winnsboro, LA), while Micromax was applied at 0.45 kg (1 lb)·yd⁻³ to corresponding treatments above.

Each treatment consisted of eleven single pot replications, and set up as a complete randomized block design (CRBD). Units received uniform watering by hand. Initial irrigation applications were 100 ml, increasing to 200 ml as growth determined. Leachates were collected at 11, 18, 25, 35, and 39 days after transplanting (DAT) for petunia, plus two additional dates at 49 and 56 DAT for nemesia, using the Virginia Tech pour-through method (Yeager et al., 2007). Substrate pH and electrical conductivity (EC) were measured (Accumet Excel XL50, Fisher Scientific, Inc., Pittsburgh, PA) at 36 and 56 DAT for petunia, 42 and 58 DAT for nemesia, and shoot fresh weights (SFW) and shoot dry weights (SDW) (80°C (175°F) for 48 hours) were also determined. All data was analyzed using proc GLM, Waller-Duncan K-ratio t test (SAS Version 9.1, SAS Institute, Cary, NC).

Trial 2

On April 24, 2009, a second trial began with plugs of nemesia and petunia (John McBryde, Ball Horticultural Company) transplanted into a common substrate (Fafard 3B, Conrad Fafard Inc., Agawam, MA) in 20.23 cm (8 in.) diameter containers, with a volume of 3063 cm³ (187 in³). Plants were produced in a 29.3 x 9.1 m (96 x 30 ft) pad and fan greenhouse with a double layer, polyethylene cover. Study dependent variables included water source (CW, EW), top-dressed CRF, liquid fertilizer applications, or no fertilizer application. Each pot was filled level to container rim with substrate; plugs were stuck subsequent to irrigating pots, allowing substrate to settle. Five treatments included 1) SF: CW with 200 mg·L⁻¹ N 20-20-20 soluble fertilizer; 2) EW: 100% effluent water, 3) SEW: settled EW, 4) EW and SF: effluent water and one final week of supplemental 200 mg·L⁻¹ N, and 5) CRF: CW with 1.58 kg N·yd⁻³ of controlled release 14-14-14 fertilizer. Each treatment consisted of twelve single pot replications placed on greenhouse benches in a CRBD. Plants received uniform irrigation applications by hand, receiving 200 ml water per irrigation.

Study 1

Based on preliminary trials, an experiment began on June 26, 2009 at E.W. Shell Fisheries Center, North Auburn Unit, in Auburn, AL. *Angelonia AngelMist*TM (*Angelonia angustifolia*), Suncatcher Vegetative Petunia (*Petunia x hybrida*), and Verbena Vegetative AztecTM (*Verbena x hybrida*) plugs (John McBryde, Ball Horticultural Company), were transplanted into a common substrate (Fafard 3B) in 15.24 cm (6 in.) diameter containers, with a volume of 1278 cm³ (78 in³), using the same treatments as those in the second preliminary trial.

Based on preliminary trials, irrigation applications were adjusted by delivery method and by volume. Units received 100 ml, 200 ml, and 300 ml per irrigation event as environmental conditions dictated. Due to summer season, plants were subjected to increased temperatures and increased evaporation rates than experienced in trials, so irrigation rates were increased as needed. Data collected consisted of leachates collected at 18, 42, and 49 DAT, using the Virginia Tech pour-through method (Yeager et al., 2007) measuring pH, EC, and salinity in parts per thousand (ppt) (YSI 63 meter, YSI Inc., Yellow Springs OH). Relative growth indices (RGI) were determined, measuring initial GI_I (cm, (height + widest width + perpendicular width)/3) and final GI_F at 26 DAT and 49 DAT respectively, subtracting GI_I from GI_F . At harvest, SFW and SDW (80°C (175°F) for 48 hours) were weighed and recorded. Foliar nutrient analyses were conducted. All data was analyzed using proc GLM, Waller-Duncan K-ratio t test (SAS Version 9.1, SAS Institute, Cary, NC).

The study was subsequently repeated beginning on August 17, 2009. Container size remained 15.24 cm (6 in.) diameter containers. Plant material used was Celebrity Blue Petunia (*Petunia x hybrida* 'Celebrity Blue') and Cooler Rose Vinca (*Catharanthus roseus* 'Cooler Rose'), potted in common substrate (Fafard 3B). Five treatments included 1) SF: CW with 200 mg·L⁻¹ N 20-20-20 soluble fertilizer; 2) EW: 100% effluent water, 3) SEW: settled EW, 4) EW and SF: effluent water and two final weeks of supplemental 200 mg·L⁻¹ N, and 5) CRF: CW with 1.58 kg N·yd⁻³ of controlled release 14-14-14 fertilizer. Irrigation schedule remained the same, units receiving only 100 ml to 200 ml per watering as needed. Data collected consisted of leachates collected at 23, 30, and 36 DAT, using the Virginia Tech pour-through method (Yeager et al., 2007). GI were

measured 36 DAT. At harvest, SFW and SDW (80°C (175°F) for 48 hours), and nutrient content were determined. Data was analyzed using Proc GLM, Waller-Duncan K-ratio t test (SAS Version 9.1, SAS Institute, Cary, NC).

Results and Discussion

Trial 1

Initial substrate pH was below BMP recommendation level of 4.5-6.5 pH for petunia and nemesia (Tables 2.2, 2.3) (Yeager et al., 2007). Plants receiving CW with low rate CRF and CW with Micromax exhibited pH levels within recommended range for petunia at 11 DAT (Table 2.3). Between 11 DAT and 25 DAT pH levels increased across all treatments to be in recommended range for both species. Nemesia substrate pH displayed a general decreasing trend from 25 DAT to 56 DAT, with exception to plants receiving CW with low rate CRF and CW with Micromax (Table 2.4).

Substrate EC across treatments and species generally exceeded recommended range for EC (Tables 2.5, 2.6) ($0.5\text{-}1.0\text{ mS}\cdot\text{cm}^{-1}$, Yeager et al., 2007), with the exception of plants receiving CW with Micromax, which remained below recommended levels. For petunia and nemesia, all plants receiving EW treatments exceeded recommended range significantly, as did multiple CW treatments. Petunias receiving CW with low rate CRF and CW with soluble N were in range, respectively from 11 DAT-25 DAT and 25 DAT-39 DAT. No trends were observed in nemesia EC levels, except exceeding acceptable range.

At 39 DAT and 56 DAT for petunia and nemesia, respectively, SDW were recorded. Differences in SDW were observed for both species. In petunia, plants

receiving CW soluble N had highest SDW, while plants receiving EW with low rate CRF, CW with high rate CRF, EW with Micromax, and EW indicated no differences (Table 2.7). *Nemesia* SDW displayed fewer differences between treatments, where EW with low rate CRF, CW & EW with medium rate CRF, CW and EW with high rate CRF, and CW with soluble N were similar. EW and CW with low rate CRF treatments showed no differences in *nemesia*. The smallest SDW of plants across all treatments for both species was CW with Micromax (Table 2.7).

Results indicate, while *petunia* under EW irrigation did not perform similarly to plants receiving soluble N inputs, no differences were observed between EW inputs and plants receiving CW with high rate CRF. Further, plants receiving EW with low rate CRF performed similarly to plants receiving EW irrigation alone, a potential cost saving observation; indicating *petunia* crop under EW receive no additional benefit from supplement synthetic fertilizer inputs.

Nemesia is not as heavy a feeder as *petunia*, and subsequently performed similarly across treatment. Plants receiving CW with low rate CRF where out performed by plants receiving EW with low rate CRF, while plants in this treatment performed similarly to industry standard fertilizer regimes of $200 \text{ mg} \cdot \text{L}^{-1}$. Noting no difference between plants receiving high rate CRF, medium rate, or low rate, indicates potential cost savings.

Subsequent experiments were adjusted to reflect more realistic greenhouse and nursery production methods, with an emphasis on soluble fertilizer inputs.

Study 1

Leachates

Leachates recorded for angelonia, at 18, 42, and 49 DAT fluctuated above and below recommended levels for pH (4.5-6.5), EC ($0.5-1.0 \text{ mS}\cdot\text{cm}^{-1}$), and salinity (Table 2.8) (Yeager et al, 2007). Salinity (ppt) was measured after recording substantially increased EC's at 42 DAT compared to 18 DAT. During the study, a single application of 160 kg (350 lbs) sodium chloride was added to tilapia culture system, accounting for increased EC. Substrate pH levels for all treatments remained within recommended levels, except for plants receiving treatments EW, EW and SF, and CRF when pH levels peaked at 42 DAT. Substrate EC levels were generally higher than recommended range. At 18 DAT EC was within range in EW, EW and SF, and SEW, while at 42 DAT the only treatment within range was CRF.

Initial substrate pH for petunia was above recommended range, with the exception of plants receiving SF treatment, which remained within range throughout the duration of the experiment (Table 2.9). However, at 18 DAT statistical analysis indicated no differences across treatments. All remaining treatments exhibited pH levels exceeding BMP range until 49 DAT at which point all treatments were within range, except the CRF treatment. EC levels for petunia displayed similar pattern to that of angelonia EC. No treatment EC remained below, within, or above BMP range for duration of study. At 49 DAT all treatment EC's were below BMP range. Salinity levels were substantial, reaching 1.75 ppt in plant substrates of SEW treatment, but with little effect on overall growth.

Substrate leachate trends in verbena did not resemble those of two previous crops discussed (Table 2.10) Throughout duration of the study, all substrate pH levels remained well within recommended range. Differences however, between treatments were observed. Unlike pH levels, recorded EC's varied. At 18 DAT only treatments EW, EW and SF, and SEW were within range. At 42 DAT due to high EC levels, plant were flushed uniformly, leading to lower than BMP recommended EC levels at 49 DAT. Salinity concentrations remained elevated through study, due to sodium chloride application to tilapia culture system previously mentioned.

Growth Indices and Dry Weights

Growth indices were different across treatments for angelonia, with best performance for SF (38.04 cm), SEW (37.36 cm), and CRF (39.37 cm) treatments. EW growth index was smallest (32.83 cm) with differences indicated compared to all other treatments. However, all SDW across treatments were similar (Table 2.11). Visually, no differences were observed across treatments (Figure 2.2).

Growth indices for petunia were similar between treatments SF (39.24 cm), and CRF (40.57 cm), while both were different from the EW (31.60 cm) treatment. However, between treatments SF and CRF and treatments EW and SF (35.54 cm), and SEW (35.75 cm) no differences were observed. Between EW treatment and treatments EW and SF, and SEW no differences were indicated. SDW were greatest for SF (11.65 g) when compared to all other treatments (Table 2.11).

No differences between treatments for verbena GI and SDW were observed. All treatments performed similarly (Table 2.11).

Foliar Nutrient Analysis

Nutritional requirement ranges used to assess results of foliar analysis are those of Argo et al (2009) for angelonia and Mills and Jones (1996) for petunia and verbena (Table 2.1). Angelonia exhibited sufficient concentrations of N, P, Cu, Mn, Zn, and Ca, however not across all treatments. Plants in the CRF treatment had a deficient range of N, and deficient concentrations of K and Ca were observed in plants in EW and SF, SEW, and CRF treatments. Boron was deficient across all treatments. Excess accumulation of nutrients Mg and Fe across all treatments was observed (Table 2.12).

In petunia, only plants in SF treatment displayed sufficient N concentration, while plants of all other treatments were N-deficient. Other nutrients within sufficiency range include P, Ca, Fe, Mn, and Zn. Plants in SF treatment exhibited excess concentration of P, and all plants displayed excess accumulation of Mg and Cu (Table 2.13).

Plants of all treatments for verbena were below the recommended range for N, P, K, and Zn content, with other nutrients at or exceeding the recommended ranges of sufficiency (Mg, Fe, Cu, B, Na, Al, and Mo). Calcium and Mo (only for treatments EW, EW and SF, and SEW) were within sufficiency range (Table 2.14).

Results of angelonia indicate, despite excessively high EC in plants receiving EW, total growth and overall performance was not hindered, as GI and SDW indicate. A visual assessment indicates as well that plants are similar (Figure 2.2). This study indicates that effluent from this production system could provide adequate nutrition for proper growth and development of petunia. Effluent water used in production of petunia resulted in both statistically and visually similar plants to traditional bedding plant production practices (Figure 2.3). However, it should be noted that using a high rate of N

in a soluble fertilizer produced larger petunia plants, while plants receiving other treatments remained similar. Data for verbena produced indicates this crop to be a potentially highly successful bedding plant for alternative production means, as there was no difference between treatments for GI and SDW (Figure 2.4).

Study 2

Leachates

Substrate pH remained within recommended ranges for petunia from 23-31 DAT, across all treatments (Table 2.15). At 23 DAT, plants of all treatments maintained similar pH levels, except plants in CRF treatment (6.19), which was lower than the others. At 30 DAT, pH for all treatments had declined to be similar to CRF, except plants of SF treatment (6.42). Substrate EC's demonstrated less consistency (Table 2.15). However, at 23 DAT all treatments except CRF ($1.40 \text{ mS}\cdot\text{cm}^{-1}$) were well within BMP range. At 30 DAT, all EC levels had decreased below BMP ranges, except SEW ($0.56 \text{ mS}\cdot\text{cm}^{-1}$). Salinity concentrations remained consistent across treatments, excluding CRF at 23 DAT, which corresponds to high EC level at 23 DAT (Table 2.15).

Vinca substrate pH trends were similar to petunia data, with 23 and 30 DAT falling within recommended range (Table 2.16), with no treatment differences. At 23 DAT all treatments exhibited exceptionally high EC levels, between $1.60 - 2.43 \text{ mS}\cdot\text{cm}^{-1}$, but EC levels decreased to meet BMP recommendations by 30 DAT, with exception of CRF ($1.23 \text{ mS}\cdot\text{cm}^{-1}$) (Table 2.16). Recorded salinity levels correspond to EC levels, initially high at 23 DAT across all treatments, and decreasing to expected range at 30 DAT.

Growth Indices and Dry Weights

There were no differences across treatments for petunia GI (Table 2.17), but SFW and SDW were different. Plants in the CRF treatment had the greatest SFW (112.67 g), which was similar to plants in the SF treatment SFW (102.31 g). Plants of the same two treatments had the greatest SDW, which were different from plants of other treatments (CRF: 9.85 g, SF: 9.80 g). Plants receiving SEW had the smallest SFW (82.25 g), while for SDW plants receiving EW (7.78 g), EW and SF (7.90 g), and SEW (7.59 g) performed similarly (Table 2.17). No visual differences were observed.

Vinca GI, SFW and SDW were similar across treatments, showing no differences (Table 2.17). Further, SFW and SDW were similar for plants across all treatments (Table 2.17). No differences were observed visually.

Results of this study indicate that EW from intensive tilapia production supplies adequate nutrition to produce comparable crops produced under standard methods. Fluctuation in substrate EC is dependent upon effluent water treatments and fish feed inputs. For petunia production, plants receiving EW were different in size and SDW than plants grown under traditional means. During the first study, one treatment received six weeks of EW, replaced by 200 mg·L⁻¹ N for the final week of production, while in the second study, one treatment received five weeks EW and two final weeks of 200 mg·L⁻¹ N; neither of the treatments produced similar sized petunias as standard greenhouse production methods. However, the potential for cost savings exist, where a petunia crop can be fertigated with EW for half or two thirds of production time, and then finished with soluble fertilizers. However the correct time ratio of EW to soluble fertilizer needs

to be determined. For less heavy feeding crops such as angelonia, verbena, and vinca EW did produce similar plants compared to standard greenhouse production methods.

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Table 2.1. Industry accepted foliar nutrient analysis compositions.

Nutrient ^{ZY}	General plant nutrition	<i>Petunia</i> nutrition	<i>Verbena</i> nutrition	<i>Catharanthus</i> nutrition
	Argo et al., 2009 ^X	Mills and Jones, 1996 ^W		
Nitrogen	2.5-6.0	3.87-7.60	3.64-5.84	2.72-6.28
Phosphorus	0.3-1.0	0.47-0.93	0.77-1.19	0.28-0.64
Potassium	2.5-6.0	3.13-6.65	2.79-4.69	1.88-3.48
Calcium	0.6-2.0	1.20-2.81	1.60-2.54	0.93-1.13
Magnesium	.03-1.0	0.36-1.37	0.73-1.58	0.32-0.78
Sulfur	0.3-1.0	0.33-0.80	0.58-0.64	0.22-0.50
Iron	75-200	84-168	56-112	72-277
Manganese	50-200	44-177	55-293	135-302
Zinc	25-100	33-85	65-127	30-51
Copper	5.0-20	3.0-19	3.0-13	6.0-16
Boron	30-120	18-43	43-48	21-49
Molybdenum	1.0-5.0	0.19-0.46	0.14-0.80	0.14-0.46
Sodium	. ^V	3067-10896	257-310	89-1566
Aluminum	.	50-92	41-53	34-136

^ZNutrient: Essential and non-essential nutrients for plant growth and development.

^YUnit: % for N, P, K, Ca, Mg, and S; mg·L⁻¹ for Fe, Mn, Zn, Cu, B, Mo, Na, and Al.

^XArgo, B., P. Fisher, and K. Santos. 2009. Understanding plant nutrition: Diagnosing problems.

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^WMills, H.A. and J.B. Jones. 1996. Plant Analysis Handbook II: A practical sampling, preparation, analysis and interpretation guide. MicroMacro Pub. Inc. Athens, GA.

^VNo data provided.

Table 2.2. Intensive tilapia culture effluent nutrient analysis, during greenhouse plant production from January to September 2009.

Nutrient ^{ZY}	Jan-Mar, 2009	Jul-Aug, 2009	Aug-Sep, 2009
TAN ^X	. ^W	12.20	16.43
NH ₄ -N	19.0	14.90	16.00
NO ₃ ⁻ -N	47.0	44.90	38.88
P	24.0	15.30	19.60
K	40.0	31.50	42.10
Ca	25.0	22.50	36.90
Mg	11.0	40.60	52.10
Fe	<0.1	0.10	0.50
Zn	0.2	0.20	0.20
Mn	0.3	0.40	0.40
Cu	0.1	0.10	0.10
B	0.2	0.10	0.10
Mo	.	0.10	0.10
Na	83	192.70	109.80
pH ^V	5.87	4.65	4.85
EC ^U	2.23	1.21	0.94

^ZNutrient analysis determined by ICAP.

^YAll nutrients measured in mg·L⁻¹, unless noted.

^XTAN: Total ammonia nitrogen (NH₄⁺-N and NH₃-N).

^WData not provided.

^VpH (YSI 63 meter).

^UEC: Electrical conductivity measured in mS·cm⁻¹ (YSI 63 meter).

Table 2.3. *Petunia x hybrida* substrate pH^Z during January-February 2009, potted in common substrate (70% peat, 15% perlite, 15% vermiculite).

Treatment ^Y	11 DAT ^X	18 DAT	25 DAT	35 DAT	39 DAT
CW, Low CRF Rate	5.67a ^W	4.90a	5.69a	5.94a	4.56bc
EW, Low CRF Rate	4.30bc	4.19a	4.97bc	5.51ab	4.36bc
CW, Medium CRF Rate	3.87bc	4.44a	5.16abc	4.90bcd	4.76bc
EW, Medium CRF Rate	3.53c	4.00a	4.94bc	4.32cd	5.21ab
CW, High CRF Rate	4.43bc	4.40a	4.75bc	5.51ab	5.95ab
EW, High CRF Rate	4.07bc	4.75a	6.12a	5.31ab	4.40bc
CW, 200 mg·L ⁻¹ N	3.90bc	4.58a	5.87ab	5.70ab	5.19ab
EW, Micromax [®]	3.83bc	4.26a	4.50c	4.23d	3.97c
CW, Micromax [®]	4.67b	4.70a	5.29abc	4.78bcd	4.29bc
EW	4.27bc	4.98a	5.52abc	5.15abc	4.21c

^ZMedia pH measured with Accumet Excel XL50 meter (Fisher Scientific Inc., Pittsburg, PA).

^YTreatments: CW: Clear Water; EW: Effluent Water (see Table 2.2 for nutrient analysis); CRF: Controlled Release Fertilizer at low (1.58 kg N·yd⁻³), medium (3.40 kg N·yd⁻³), and high (5.44 kg N·yd⁻³) rates (14-14-14, Nutricote); 200 mg·L⁻¹ N (20-20-20, TotalGro); Micromax[®] at 0.45 kg·yd⁻³.

^XDAT = days after transplanting.

^WMeans within columns followed by the same letter are not significantly different (Waller-Duncan K-ratio t test, $P \leq 0.05$, $N = 3$).

Table 2.4. *Nemesia fruticans* substrate pH^z during January-March 2009, potted in common substrate (70% peat, 15% perlite, 15% vermiculite).

Treatment ^y	11 DAT ^x	18 DAT	25 DAT	35 DAT	39 DAT	49 DAT	56 DAT
CW, Low CRF Rate	4.37a ^w	3.96a	6.41a	5.77a	4.52bc	4.72b	4.78b
EW, Low CRF Rate	4.13a	4.92a	5.29abc	4.41bc	3.93c	4.02d	4.14c
CW, Medium CRF Rate	4.20a	4.65a	5.03bc	4.81abc	4.12bc	4.18d	4.27c
EW, Medium CRF Rate	3.50a	4.24a	4.84c	4.71bc	3.90c	4.01d	4.17c
CW, High CRF Rate	3.60a	4.37a	4.76c	4.19c	4.01c	3.95d	4.17c
EW, High CRF Rate	3.97a	4.22a	5.35abc	4.28bc	4.24bc	3.94d	4.17c
CW, 200 mg·L ⁻¹ N	3.90a	4.65a	6.36a	5.21ab	5.18a	5.88a	5.60a
EW, Micromax [®]	3.60a	4.22a	5.60abc	5.01abc	4.42bc	4.12d	4.38c
CW, Micromax [®]	4.03a	4.83a	6.15ab	5.22ab	4.71ab	4.43c	4.47c
EW	4.37a	4.13a	5.31abc	4.89abc	4.49bc	4.12d	4.29c

^zMedia pH measured with Accumet Excel XL50 meter (Fisher Scientific Inc., Pittsburg, PA).

^yTreatments: CW: Clear Water; EW: Effluent Water (see Table 2.2 for nutrient analysis); CRF: Controlled Release Fertilizer at low (1.58 kg N·yd⁻³), medium (3.40 kg N·yd⁻³), and high (5.44 kg N·yd⁻³) rates (14-14-14, Nutricote); 200 mg·L⁻¹ N (20-20-20, TotalGro); Micromax[®] at 0.45 kg·yd⁻³.

^xDAT: days after transplanting.

^wMeans within columns followed by the same letter are not significantly different (Waller-Duncan K-ratio t test, P ≤ 0.05, N = 3).

Table 2.5. *Petunia x hybrida* substrate electrical conductivity^Z during January-March 2009, potted in common substrate (70% peat, 15% perlite, 15% vermiculite).

Treatment ^Y	11 DAT ^X	18 DAT	25 DAT	35 DAT	39 DAT
CW, Low CRF Rate	0.93de ^W	0.60dc	0.73cd	0.30d	0.25d
EW, Low CRF Rate	2.13bc	1.90ab	1.44abc	1.09bc	1.14bc
CW, Medium CRF Rate	1.63cd	1.39abc	0.83cd	0.71cd	0.49d
EW, Medium CRF Rate	3.70a	1.97ab	1.97a	2.01a	1.06cd
CW, High CRF Rate	2.10bc	1.83ab	2.06a	0.85cd	0.35d
EW, High CRF Rate	3.80a	2.39a	1.10bcd	2.03a	2.37a
CW, 200 mg·L ⁻¹ N	1.13cde	1.04bcd	0.70d	0.64cd	0.57d
EW, Micromax [®]	3.03ab	1.91ab	1.74ab	1.59ab	1.80abc
CW, Micromax [®]	0.53e	0.38d	0.53d	0.38d	0.34d
EW	1.87cd	1.34bcd	1.10bcd	1.27bc	2.00ab

^ZMedia EC measured with Accumet Excel XL50 meter (Fisher Scientific Inc., Pittsburg, PA).

^YTreatments: CW: Clear Water; EW: Effluent Water (see Table 2.2 for nutrient analysis); CRF: Controlled Release Fertilizer at low (1.58 kg N·yd⁻³), medium (3.40 kg N·yd⁻³), and high (5.44 kg N·yd⁻³) rates (14-14-14, Nutricote); 200 mg·L⁻¹ N (20-20-20, TotalGro); Micromax[®] at 0.45 kg·yd⁻³.

^XDAT = days after transplanting.

^WMeans within columns followed by the same letter are not significantly different (Waller-Duncan K-ratio t test, P ≤ 0.05, N = 3).

Table 2.6. *Nemesia fruticans* substrate electrical conductivity^z during January-March 2009, potted in common substrate (70% peat, 15% perlite, 15% vermiculite).

Treatment ^y	11 DAT ^x	18 DAT	25 DAT	35 DAT	39 DAT	49 DAT	56 DAT
CW, Low CRF Rate	0.80cd ^w	1.05bc	0.32c	0.45d	0.66c	0.52d	0.45e
EW, Low CRF Rate	2.07ab	1.58abc	1.28ab	2.54ab	2.84b	4.91b	4.32b
CW, Medium CRF Rate	0.87cd	1.02cd	1.09abc	1.15cd	1.74bc	1.98cd	1.94de
EW, Medium CRF Rate	2.93a	2.56a	1.36ab	2.28ab	4.68a	4.75b	4.30b
CW, High CRF Rate	1.63bc	1.32bcd	1.82a	2.40ab	2.18bc	4.64b	3.99bc
EW, High CRF Rate	2.87a	2.08ab	1.69a	3.03a	4.88a	8.12a	7.08a
CW, 200 mg·L ⁻¹ N	1.37bcd	1.14bcd	0.54bc	1.03cd	1.10bc	0.90d	1.19de
EW, Micromax [®]	2.73a	1.17abc	1.07abc	1.72bc	2.82b	3.54bc	2.37cd
CW, Micromax [®]	0.53d	0.43d	0.33c	0.43d	0.41c	0.58d	0.51e
EW	1.60bc	1.58abc	1.44ab	1.92bc	1.94bc	4.10bc	2.90bcd

^zMedia EC measured with Accumet Excel XL50 meter (Fisher Scientific Inc., Pittsburgh, PA).

^yTreatments: CW: Clear Water; EW: Effluent Water (see Table 2.2 for nutrient analysis); CRF: Controlled Release Fertilizer at low (1.58 kg N·yd⁻³), medium (3.40 kg N·yd⁻³), and high (5.44 kg N·yd⁻³) rates (14-14-14, Nutricote); 200 mg·L⁻¹ N (20-20-20, TotalGro);

Micromax[®] at 0.45 kg·yd⁻³.

^xDAT: days after transplanting.

^wMeans within columns followed by the same letter are not significantly different (Waller-Duncan K-ratio t test, $P \leq 0.05$, $N = 3$).

Table 2.7. *Nemesia* and *Petunia* fresh and dry weights during January-March 2009, potted in common substrate (70% peat, 15% perlite, 15% vermiculite).

Treatment ^z	<i>Nemesia</i>			<i>Petunia</i>		
	Shoot fresh weight		Shoot dry weight	Shoot fresh weight		Shoot dry weight
	56 DAT ^y	58 DAT	58 DAT	36 DAT	42 DAT	
CW, Low CRF Rate	20.62c ^x	3.00b	3.00b	34.43c	3.36b	
EW, Low CRF Rate	21.59c	2.37c	2.37c	53.19a	4.31a	
CW, Medium CRF Rate	21.57c	3.15b	3.15b	45.78ab	4.17a	
EW, Medium CRF Rate	15.62d	1.66d	1.66d	51.56a	4.31a	
CW, High CRF Rate	15.58d	2.30c	2.30c	52.75a	4.77a	
EW, High CRF Rate	9.83e	1.13e	1.13e	51.94a	4.45a	
CW, 200 mg·L ⁻¹ N	36.15a	5.78a	5.78a	41.37bc	4.37a	
EW, Micromax [®]	21.09c	2.24c	2.24c	34.86c	2.60c	
CW, Micromax [®]	0.36f	0.05f	0.05f	2.23d	0.14d	
EW	24.89b	2.53c	2.53c	36.52c	2.90bc	

^zTreatments: CW: Clear Water; EW: Effluent Water (see Table 2.2 for nutrient analysis); CRF: Controlled Release Fertilizer at low (1.58 kg N·yd⁻³), medium (3.40 kg N·yd⁻³), and high (5.44 kg N·yd⁻³) rates (14-14-14, Nutricote); 200 mg·L⁻¹ N (20-20-20, TotalGro); Micromax[®] at 0.45 kg·yd⁻³.

^yDAT = days after transplanting.

^xMeans within columns followed by the same letter are not significantly different (Waller-Duncan K-ratio t test, $P \leq 0.05$, N=3).

Table 2.8. *Angelonia angustifolia* leachates during July-August 2009, potted in common substrate (Fafard 3B).

Treatment ^z	pH			Electrical conductivity ^y				Salinity ^x	
	18 DAT ^w	42 DAT	49 DAT	18 DAT	42 DAT	49 DAT	24 DAT	49 DAT	
SF	6.026c ^v	5.70c	6.07b	2.18a	1.44b	0.45a	0.60b	0.36bc	
FW	6.23ab	6.94a	6.21ab	0.82c	1.27b	0.28a	0.55b	1.33ab	
EW & SF	6.33ab	6.58ab	6.25ab	0.74c	1.99b	0.18a	0.90b	0.83bc	
SEW	6.356a	6.09bc	5.94b	0.60c	4.86a	2.44a	2.30a	2.00a	
CRF	6.14bc	6.75ab	6.54a	1.26b	0.50b	0.38a	0.20b	0.167c	

^zTreatment: 1) SF: 200 mg·L⁻¹ N 20-20-20 soluble fertilizer (TotalGro); 2) EW: 100% Effluent water (see table 2.2 for nutrient

analysis of EW); 3) EW & SF: 100% Effluent water for 6 of 7 weeks of study, receiving soluble 200 mg·L⁻¹ N for final week;

4) SEW: Settled effluent water; 5) CRF: 14-14-14 top-dressed controlled release fertilizer at 3.5 lbs·yd⁻³.

^yElectrical conductivity: mS·cm⁻¹ (YSI 63 Meter, YSI Inc., Yellow Springs OH).

^xSalinity: NaCl parts per thousand (YSI 63 Meter, YSI Inc., Yellow Springs OH).

^wDAT = days after transplanting.

^vMeans within columns followed by same letter are not significantly different (Waller-Duncan K-ratio t test, $p \leq 0.05$).

Table 2.9. *Petunia x hybrida* leachates during July-August 2009, potted in common substrate (Fafard 3B).

Treatment ^z	pH			Electrical conductivity ^y			Salinity ^x	
	18 DAT ^w	42 DAT	49 DAT	18 DAT	42 DAT	49 DAT	42 DAT	49 DAT
SF	6.27a ^v	6.09c	6.15b	1.91a	0.67cd	0.48a	0.30cd	0.20c
EW	6.60a	6.53bc	6.41ab	0.49b	2.25ab	0.21b	1.06ab	0.90b
EW & SF	6.67a	6.87ab	6.08b	0.52b	1.52bc	0.21b	0.70bc	0.80b
SEW	6.55a	6.60b	6.19b	0.45b	2.63a	0.36ab	1.26a	1.75a
CRF	6.62a	7.10a	6.75a	0.71b	0.31d	0.32ab	0.13d	0.10c

^zTreatment: 1) SF: 200 mg·L⁻¹ N 20-20-20 soluble fertilizer (TotalGro); 2) EW: 100% Effluent water (see table 2.2 for nutrient analysis of EW); 3) EW & SF: 100% Effluent water for 6 of 7 weeks of study, receiving soluble 200 mg·L⁻¹ N for final week;

4) SEW: Settled effluent water; 5) CRF: 14-14-14 top-dressed controlled release fertilizer at 3.5 lbs·yd⁻³.

^yElectrical conductivity: mS·cm⁻¹ (YSI 63 Meter, YSI Inc., Yellow Springs OH).

^xSalinity: NaCl parts per thousand (YSI 63 Meter, YSI Inc., Yellow Springs OH).

^wDAT = days after transplanting.

^vMeans within columns followed by same letter are not significantly different (Waller-Duncan K-ratio t test, $p \leq 0.05$).

Table 2.10. *Verbena x hybrida* leachates during July-August 2009, potted in common substrates (Fafard 3B).

Treatment ^z	pH		Electrical conductivity ^y				Salinity ^x	
	18 DAT ^w	42 DAT	49 DAT	18 DAT	42 DAT	49 DAT	42 DAT	49 DAT
SF	6.11b ^v	5.56b	5.10b	1.48a	1.21b	0.12a	0.60b	0.56bc
EW	6.32a	6.14ab	5.92a	0.79a	3.36a	1.96a	1.60a	1.80a
EW & SF	6.35a	6.26ab	5.95a	0.74a	3.04a	0.25a	1.46a	1.10b
SEW	6.39a	6.23ab	6.11a	0.74a	3.25a	0.22a	1.56a	1.00b
CRF	6.46a	6.43a	5.63ab	1.08a	0.59b	0.75a	0.26b	0.30c

^zTreatment: 1) SF: 200 mg·L⁻¹ N 20-20-20 soluble fertilizer (TotalGro); 2) EW: 100% Effluent water (see table 2.2 for nutrient analysis of EW); 3) EW & SF: 100% Effluent water for 6 of 7 weeks of study, receiving soluble 200 mg·L⁻¹ N for final week;

4) SEW: Settled effluent water; 5) CRF: 14-14-14 top-dressed controlled release fertilizer at 3.5 lbs·yd⁻³.

^yElectrical conductivity: mS·cm⁻¹ (YSI 63 Meter, YSI Inc., Yellow Springs OH).

^xSalinity: NaCl parts per thousand (YSI 63 Meter, YSI Inc., Yellow Springs OH).

^wDAT = days after transplanting.

^vMeans within columns followed by same letter are not significantly different (Waller-Duncan K-ratio t test, $p \leq 0.05$).

Table 2.11. Growth index and harvest weights of *Angelonia angustifolia*, *Petunia x hybrida*, and *Verbena x hybrida*, during July-August, 2009.

Treatment ^z	<i>Angelonia</i>			<i>Petunia</i>			<i>Verbena</i>		
	Growth index (cm) ^y	Dry weight (g) ^x		Growth index (cm)	Dry weight (g)		Growth index (cm)	Dry weight (g)	
	56 DAT ^w	56 DAT		56 DAT	56 DAT		56 DAT	56 DAT	
SF	38.04abV	20.05a		39.24a	11.65a		31.68a	8.58a	
EW	32.83c	25.12a		31.60b	6.31b		28.24a	6.52a	
EW & SF	34.66bc	13.19a		35.54ab	6.26b		28.54a	6.54a	
SEW	37.36ab	15.61a		35.75ab	5.52b		30.72a	6.37a	
CRF	39.37a	18.36a		40.57a	5.89b		31.88a	7.78a	

^zTreatment: 1) SF: 200 mg·L⁻¹ N 20-20-20 soluble fertilizer (TotalGro); 2) EW: 100% Effluent water (see table 2.2 for nutrient analysis);

3) EW & SF: 100% Effluent water for 6 of 7 weeks of study, receiving soluble 200 mg·L⁻¹ N for final week; 4) SEW: Settled effluent water;

5) CRF: 14-14-14 top-dressed controlled release fertilizer at 3.5 lbs·yd⁻³.

^yGrowth Index = (height + widest width + perpendicular width)/3.

^xDry weight = Plant tissue dried at 80°C (175°F) for 48 hours.

^wDAT = days after transplanting.

^vMeans within columns followed by same letter are not significantly different (Waller-Duncan K-ratio t test, p ≤ 0.05).

Table 2.12. *Angelonia angustifolia* foliar nutrient analysis from study conducted July-August 2009, potted in common substrate (Fafard 3B).

Treatment ^z	%							mg·L ⁻¹						
	N	P	K	Ca	Mg	Fe	Cu	Mn	Zn	B	Na	Al		
SF	3.74a ^y	0.55a	1.25a	0.62b	1.36b	68.74a	20.50a	159.00bc	37.24a	2.50a	2,270b	61.00a		
EW	2.75b	0.35b	1.22a	0.74a	1.57a	40.50b	25.50a	248.25a	36.00a	0.10a	7,999a	32.74a		
EW & SF	3.01b	0.43b	1.22a	0.56b	1.37ab	42.50b	17.24a	207.25ab	29.24a	1.33a	7,023a	31.74a		
SEW	2.35c	0.35b	1.09b	0.57b	1.32b	36.74b	14.50a	170.00bc	33.50a	0.10a	7,388a	31.00a		
CRF	2.10c	0.36b	1.19ab	0.52b	1.20b	41.50b	20.74a	140.00c	24.00a	0.10a	2,546b	35.74a		

^zTreatment: 1) SF: 200 mg·L⁻¹ N soluble 20-20-20 fertilizer (TotalGro); 2) EW: 100% Effluent water (see table 2.2 for nutrient analysis);

3) EW & SF: 100% effluent water for 6 of 7 weeks of study, receiving soluble 200 mg·L⁻¹ N for final week; 4) SEW: Settled effluent water;

5) CRF: 14-14-14 top-dressed controlled release fertilizer at 3.5 lbs·yd⁻³.

^yMeans followed by the same letter are not significantly different (Proc GLM, Tukeys lines, $\alpha = 0.05$, N=4).

Table 2.13. *Petunia x hybrida* foliar nutrient analysis from study conducted July-August 2009, potted in common substrate (Fafard 3B).

Treatment ^z	%					mg·L ⁻¹						
	N	P	K	Ca	Mg	Fe	Cu	Mn	Zn	B	Na	Al
SF	5.30a ^y	1.31a	2.97ab	1.22c	1.90b	163.50a	21.24a	91.74d	49.74ab	5.00c	4,047c	79.75b
EW	3.25b	0.54b	2.72b	1.35bc	2.02ab	158.50a	28.50a	138.50c	52.74ab	0.10c	13,130b	153.25ab
EW & SF	3.35b	0.56b	2.73b	1.62a	2.09a	160.00a	27.74a	186.00a	58.50ab	29.33bc	12,437b	135.25ab
SEW	3.27b	0.63b	3.26a	1.44b	1.93ab	203.25a	34.74a	167.00ab	62.24a	99.50a	15,616a	179.25a
CRF	3.04b	0.64b	3.03ab	1.38b	1.89b	167.50a	20.24a	158.50bc	40.24b	82.75ab	5,542c	134.50ab

^zTreatment: 1) SF: 200 mg·L⁻¹ N soluble 20-20-20 fertilizer (TotalGro); 2) EW: 100% Effluent water (see table 2.2 for nutrient analysis);

3) EW & SF: 100% effluent water for 6 of 7 weeks of study, receiving soluble 200 mg·L⁻¹ N for final week; 4) SEW: Settled effluent water;

5) CRF: 14-14-14 top-dressed controlled release fertilizer at 3.5 lbs·yd⁻³.

^yMeans followed by the same letter are not significantly different (Proc GLM, Tukeys lines, $\alpha = 0.05$, N=4).

Table 2.14. *Verbena x hybrida* foliar nutrient analysis from study conducted July-August 2009, potted in common substrate (Fafard 3B).

Treatment ^z	%					mg·L ⁻¹						
	N	P	K	Ca	Mg	Fe	Cu	Mn	Zn	B	Na	Al
SF	3.46a ^y	0.53a	1.71a	2.12a	2.00a	853.25a	19.76b	385.25a	53.74a	95.24a	3814b	82.25a
EW	2.54b	0.30a	1.06b	2.51a	2.04a	153.75c	31.74a	282.50ab	42.74ab	66.50b	12692b	110.50a
EW & SF	2.60b	0.34a	1.06b	2.55a	2.08a	140.25c	27.00ab	249.00b	39.24ab	58.50b	9980ab	93.75a
SEW	2.54b	0.33a	1.08b	2.38a	2.01a	160.25c	19.00b	220.75b	34.24b	53.56a	12894a	103.50a
CRF	2.64b	0.38a	1.35ab	2.44a	2.01a	478.25b	19.00b	316.75ab	36.24ab	53.00a	3796b	118.75a

^zTreatment: 1) SF: 200 mg·L⁻¹ N soluble 20-20-20 fertilizer (TotalGro); 2) EW: 100% Effluent water (see table 2.2 for nutrient analysis);

3) EW & SF: 100% effluent water for 6 of 7 weeks of study, receiving soluble 200 mg·L⁻¹ N for final week; 4) SEW: Settled effluent water;

5) CRF: 14-14-14 top-dressed controlled release fertilizer at 3.5 lbs·yd⁻³.

^yMeans followed by the same letter are not significantly different (Proc GLM, Tukeys lines, $\alpha = 0.05$, N=4).

Table 2.15. *Petunia x hybrida* 'Celebrity Blue' leachates during July–August 2009, potted in common substrates (Fafard 3B).

Treatments ^z	pH			Electrical conductivity ^y			Salinity ^x		
	23 DAT ^w	30 DAT	35 DAT ^u	23 DAT	30 DAT	35 DAT ^u	23 DAT	30 DAT	35 DAT ^u
SF	6.23ab ^v	6.42a	6.06a	0.76b	0.37a	0.44a	0.33b	0.16a	0.16a
EW	6.38ab	6.25b	6.05a	0.63b	0.39a	0.39a	0.26b	0.16a	0.16a
EW & SF	6.34ab	6.24b	6.04a	0.83b	0.42a	0.27a	0.36b	0.16a	0.16a
SEW	6.45a	6.16b	6.09a	0.63b	0.56a	0.36a	0.30b	0.26a	0.26a
CRF	6.19b	6.17b	5.93a	1.40a	0.47a	0.28a	0.60a	0.23a	0.23a

^zTreatment: 1) SF: 200 mg·L⁻¹ N 20-20-20 soluble fertilizer (TotalGro); 2) EW: 100% Effluent water (see table 2.2 for nutrient analysis of EW); 3) EW & SF: 100% Effluent water for 5 of 7 weeks of study, receiving soluble 200 mg·L⁻¹ N for final week;

4) SEW: Settled effluent water; 5) CRF: 14-14-14 top-dressed controlled release fertilizer at 3.5 lbs·yd⁻³.

^yElectrical conductivity: mS·cm⁻¹ (YSI 63 Meter, YSI Inc., Yellow Springs OH).

^xSalinity: NaCl parts per thousand (YSI 63 Meter, YSI Inc., Yellow Springs OH).

^wDAT = days after transplanting.

^vMeans within columns followed by same letter are not significantly different (Waller-Duncan K-ratio t test, $p \leq 0.05$, N=3).

^u35 DAT leachates measured with Fisher Scientific Accumet Excel XL50 pH and EC Meter.

Table 2.16. *Catharanthus roseus* 'Cooler Rose' leachates during July-August 2009, potted in common substrate (Fafard 3B).

Treatments ^z	pH			Electrical conductivity ^y			Salinity ^x	
	23 DAT ^w	30 DAT	35 DAT ^u	23 DAT	30 DAT	35 DAT ^u	23 DAT	30 DAT
SF	6.02a ^v	6.27a	6.28ab	1.86ab	0.75a	0.35b	0.80ab	0.33b
EW	6.13a	6.24a	6.09b	1.73ab	0.80a	0.72ab	0.80ab	0.26b
EW & SF	6.13a	6.22a	6.36a	1.60b	0.96a	0.36b	0.67b	0.46ab
SEW	6.24a	6.20a	6.16ab	1.60b	0.87a	0.76a	0.60b	0.43ab
CRF	6.06a	6.07a	6.16ab	2.43a	1.23a	1.04a	1.00a	0.63a

^zTreatment: 1) SF: 200 mg·L⁻¹ N 20-20-20 soluble fertilizer (TotalGro); 2) EW: 100% Effluent water (see table 2.2 for nutrient analysis of EW); 3) EW & SF: 100% Effluent water for 5 of 7 weeks of study, receiving soluble 200 mg·L⁻¹ N for final week;

4) SEW: Settled effluent water; 5) CRF: 14-14-14 top-dressed controlled release fertilizer at 3.5 lbs·yd³.

^yElectrical conductivity: mS·cm⁻¹ (YSI 63 Meter, YSI Inc., Yellow Springs OH).

^xSalinity: NaCl parts per thousand (YSI 63 Meter, YSI Inc., Yellow Springs OH).

^wDAT = days after transplanting.

^vMeans within columns followed by same letter are not significantly different (Waller-Duncan K-ratio t test, $p \leq 0.05$, N=3).

^u35 DAT leachates measured with Fisher Scientific Accumet Excel XL50 pH and EC Meter.

Table 2.17. Growth index and shoot weights at harvest for *Petunia* and *Catharanthus*, during September 2009, potted in common substrate (Fafarb 3B).

Treatments ^z	<i>Petunia</i>			<i>Catharanthus</i>		
	Growth index (cm) ^y	Fresh weight (g)	Dry weight (g) ^x	Growth index (cm)	Fresh weight (g)	Dry weight (g)
SF	37.63a ^v	102.31ab	9.80a	31.00a	56.30a	6.58a
EW	35.71a	87.13c	7.78b	28.87a	49.35a	5.99a
EW & SF	34.57a	91.64bc	7.90b	28.50a	49.16a	5.88a
SEW	34.07a	82.25c	7.59b	29.00a	47.84a	5.64a
CRF	37.74a	112.67a	9.85a	27.87a	52.30a	6.28a

^zTreatment: 1) SF: 200 mg·L⁻¹ N 20-20-20 soluble fertilizer (TotalGro); 2) EW: 100% Effluent water (see table 2.2 for nutrient analysis of EW);

3) EW & SF: 100% Effluent water for 5 of 7 weeks of study, receiving soluble 200 mg·L⁻¹ N for final two weeks; 4) SEW: Settled effluent water;

5) CRF: 14-14-14 top-dressed controlled release fertilizer at 3.5 lbs·yd⁻³.

^yGrowth Index = (height + widest width + perpendicular width)/3.

^xDry weight = Plant tissue dried at 80°C (175°F) for 48 hours.

^wDAT = days after transplanting.

^vMeans within columns followed by same letter are not significantly different (Waller-Duncan K-ratio t test, P ≤ 0.05, N=3).

Figure 2.1. Intensive tilapia production feeding, water temperature and dissolved oxygen trends from December 2008 to October 2009.

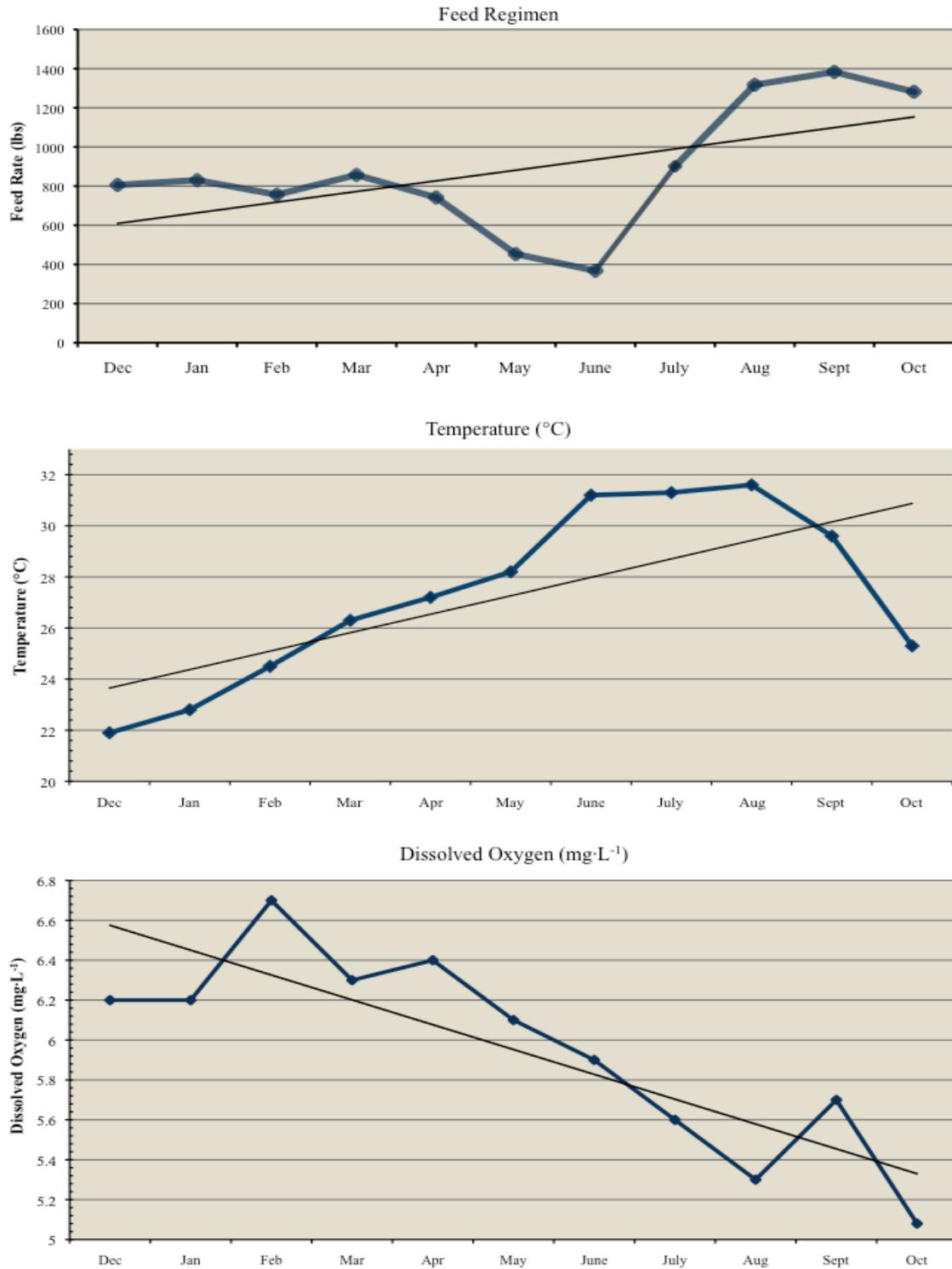
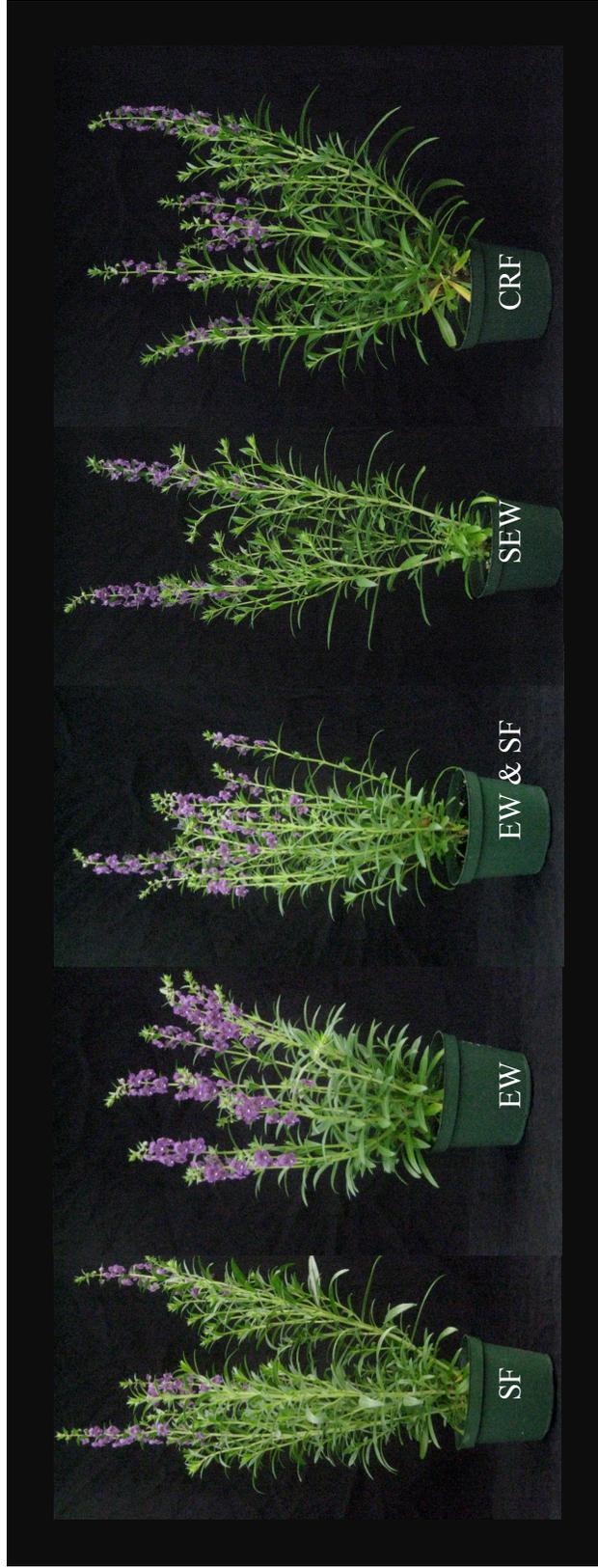
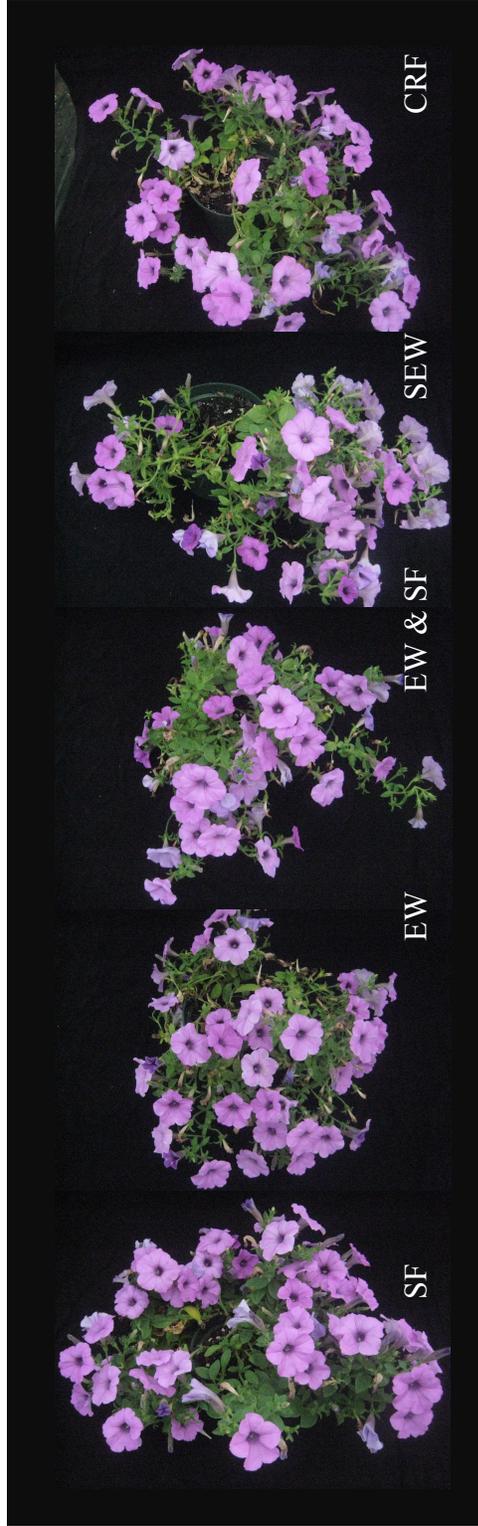


Figure 2.2. Comparison of *Angelonia angustifolia* under various irrigation and fertilizer regimes.



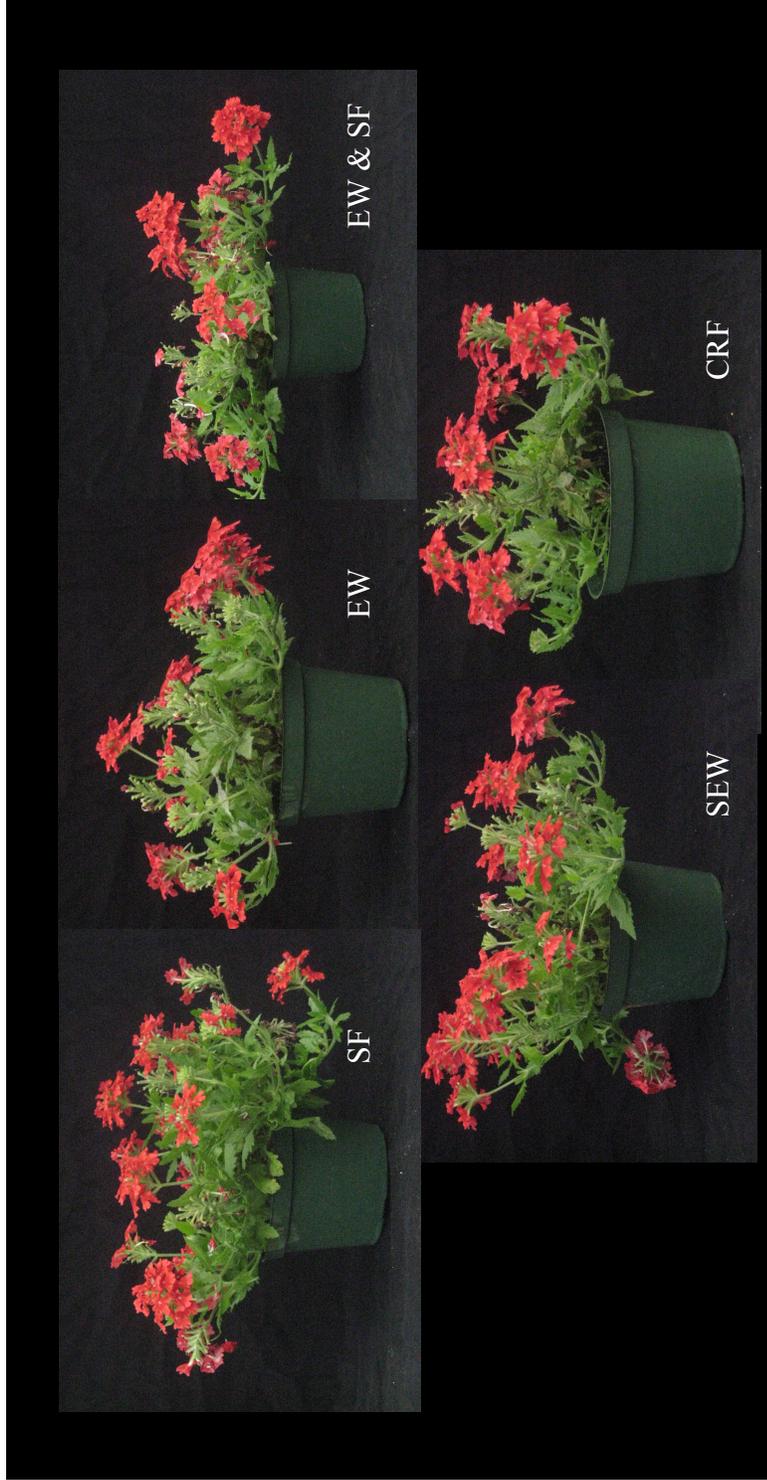
SF: 200 mg·L⁻¹ N 20-20-20 soluble fertilizer; 2) EW: 100% Effluent water; 3) EW & SF: 100% Effluent water for 6 of 7 weeks of study, receiving soluble 200 mg·L⁻¹ N for final week; 4) SEW: Settled effluent water; 5) CRF: 14-14-14 top-dressed controlled release fertilizer at 3.5 lbs·yd⁻³.

Figure 2.3. Comparison of *Petunia x hybrida* under various irrigation and fertilizer regimes.



SF: 200 mg·L⁻¹ N 20-20-20 soluble fertilizer; 2) EW: 100% Effluent water; 3) EW & SF: 100% Effluent water for 6 of 7 weeks of study, receiving soluble 200 mg·L⁻¹ N for final week; 4) SEW: Settled effluent water; 5) CRF: 14-14-14 top-dressed controlled release fertilizer at 3.5 lbs·yd⁻³.

Figure 2.4. Comparison of *Verbena x hybrida* under various irrigation and fertilizer regimes.



SF: 200 mg·L⁻¹ N 20-20-20 soluble fertilizer; 2) EW: 100% Effluent water; 3) EW & SF: 100% Effluent water for 6 of 7 weeks of study, receiving soluble 200 mg·L⁻¹ N for final week; 4) SEW: Settled effluent water; 5) CRF: 14-14-14 top-dressed controlled release fertilizer at 3.5 lbs·yd⁻³.

III. Greenhouse Crop Production Utilizing Aquaculture Wastewater

Abstract

Irrigation application of intensive tilapia culture effluent was evaluated as a replacement/supplement to standard nursery greenhouse production methods (clear water and fertilizer inputs). Irrigation was applied with effluent water (EW) or clear water (CW) at two different rates of 20-20-20 soluble fertilizer. In the first experiment, growth index (GI) differences were observed between treatments for *Petunia x hybrida*, while for *Angelonia angustifolia* and *Verbena x hybrida* no differences across treatments were observed. Shoot fresh weights (SFW) and dry weights (SDW) between treatments for individual species were determined. Differences were observed in angelonia and petunia, while all treatments for verbena were similar. This study was replicated and results follow as reported for study one. Petunia displayed differences in GI, SFW, and SDW between soluble fertilizer treatments and effluent irrigated treatment. *Catharanthus roseus* results were similar across all treatments for GI, SFW, and SDW.

Index of Words: Horticulture, intensive fish production, bedding plants, angelonia, calibrachoa, nemesia, petunia, verbena, vinca, effluent, fertilizer recommendations

Species used in Study: *Angelonia angustifolia*, *Calibrachoa x hybrida*, *Catharanthus roseus*, *Nemesia fruticans*, *Petunia x hybrida*, *Verbena x hybrida*

Significance to Industry

Tank effluent from intensive cultured tilapia (*Oreochromis niloticus*) was evaluated as a source of irrigation and fertilization for greenhouse crop production. Results indicate several plants grown using effluent-laden water from intensive aquaculture performed similarly to plants grown with clear water and soluble fertilizer application. As greenhouse production costs continue to rise and environmental policies grow more stringent, effluent from intensive fish culture might provide an economical and environmentally sustainable alternative to current water use and fertilizer practices in plant production and intensive aquaculture.

Introduction

Agricultural integrations offer potentially long-term positive effects. Assimilating various sectors of agriculture provides increased levels of sustainability, protection of human and environmental health, increased revenues, extended life of materials once thought to be fully used, provision of alternatives to replace limited naturally occurring materials, and providing aid in producing the same quality products as traditional production means. Due to benefits listed, an argument for sustainable and integrated agriculture carries more significance in a changing world. Among such integrated agriculture endeavors is the growing trend of integrating aquaculture and horticulture. For horticulture growers, dependent upon individual situations, fish waste from aquaculture may meet USDA standards for organic certification (NOP, 2008). Crops that meet USDA organic criteria can be marketed in niche markets as locally grown as well as organic crops, for increased profit. Further, crop production can serve as a means of biofiltration

for recirculating aquaculture systems (RAS), increasing sustainability. Integrated systems are often appealing to consumers who are becoming increasingly environmentally conscious. Using wastewater as an irrigation source for crop production can recycle and use pre-existing byproducts from aquaculture systems (Diver, 2006).

Benefits of integrated aquaculture and horticulture systems (IAHS), however, may have more substantial impacts than listed above. Increased production in both aspects of IAHS is possible. Previous work indicates production rates for IAHS rise significantly from 2,720 – 6,800 kg (6,000 – 15,000 lbs) fish·acre⁻¹·year⁻¹ to 136,000 – 363,000 (300,000 – 800,000 lbs) fish·acre⁻¹·year⁻¹ (Neori et al., 2004; Rakocy, 2002; Rakocy et al., 2000). An aquaponics system at the University of the Virgin Islands (UVI) generated \$110,000 producing 5,000 kg (11,000 lbs) basil annually and 2,900 kg (6,400 lbs) of okra valued at \$6,400 (Rakocy et al., 2006). In another UVI study, basil was produced via hydroponics (batch and staggered) and in a traditional field setting with greater basil yields in hydroponic systems over field production: 25.0 kg·m⁻² (batch), 23.4 kg·m⁻² (staggered), and 7.8 kg·m⁻², respectively (Rakocy et al., 2004). The value of basil produced was US \$550·m⁻²·year⁻¹ (batch), \$515·m⁻²·year⁻¹ (staggered), and \$172·m⁻²·year⁻¹. In Alberta Canada, while not operating at full capacity, greenhouse production of tomato, cucumber, and basil was recorded, respectively, at 40 kg·m⁻², 100 kg·m⁻², and 42 kg·m⁻² annually, indicating greater yields than traditional greenhouse production in Canada (Rakocy et al., 2007).

Due to effects of recent droughts in southeastern US, water reuse is often recommended. Record drought conditions were observed during August 2007, when Alabama drought status reached “extreme drought” according Palmer Drought Index

(PDI), further encouraging water conservation production practices (Ding, 2008).

Environmental instability has led to decreased profit margins for many horticulture producers. In nursery and landscape sectors, financial losses observed were extensive.

During 2006 California and multiple southeastern states comprised 60% of gross national sales in the Green industry. Yet, in 2007, industry from these regions witnessed decreased sales, increased plant mortality, increased watering expenses, and many businesses laid employees off, closed store locations, or filed for bankruptcy (Ding, 2008).

Fertilizer costs have increased 120% in past two decades (NASS, 2009; NASS, 2000; NASS, 1990), encouraging alternative fertilizer inputs. Fuel costs have risen too. Production costs have risen as a result to increased prices of fertilizers and fuel. The need for alternative fertilizer sources is great, and effluent water from aquaculture is one viable option.

Nursery production in Alabama is comprised of 57 ha (141 ac) of nursery stock under various forms of protection, and nearly 3100 ha (6,758 ac) of open land nursery stock production (Clark, 2009). Total area readily available for the nursery and greenhouse industry in Alabama is near 3360 ha (8,300 ac). Considering the industry uses 20,000 gallons water·acre⁻¹·day⁻¹ (Avent, 2003; Berghage et al., 1999; Lea-Cox and Ross, 2001), estimated total water use is 166 million gallons daily. Integrated systems offer means to reduce water use dramatically. RAS can use as little as 1-2% of water required a day, meaning 98-99% is reused.

Alabama has a growing floriculture industry, with 221 total floriculture producers, covering approximately 26 ha (64 ac) with \$89 million in sales at wholesale value during 2005-2006 (NASS, 2007). Several common annual bedding crops (begonia, impatiens,

pansies/violas, petunia, and others) accounted for \$16.2 million in sales (flats), with sales of vegetable flats valued at \$7.4 million. Sales of container bedding plants were \$9.4 million during same time period (NASS, 2007).

Integrating aquaculture and horticulture systems requires adequate nutrients to be available for plants from effluent water. Noted frequently, fish effluent could meet the nutritional demands of horticulture crops, with adequate concentrations of ammonia, nitrate, nitrite, phosphorus, potassium as well as others (Diver, 2006). Sufficiency ranges for adequate nutritional composition of greenhouse crops are: 2.5-6% N, 0.30-1.0% P, 2.5-6% K, 0.6-2% Ca, 0.3-1.0% Mg, 0.3-1.0% S, 75-200 mg·L⁻¹ Fe, 50-200 mg·L⁻¹ Mn, 25-100 mg·L⁻¹ Zn, 5-20 mg·L⁻¹ Cu, 30-120 mg·L⁻¹ B, and 1-5 mg·L⁻¹ Mo (Table 3.1) (Argo et al., 2009). Recommended fertilization rates of bedding plants produced under greenhouse conditions are 50-100 mg·L⁻¹ N for plugs, 100-150 mg·L⁻¹ N for slight feeding crops, 150-200 mg·L⁻¹ N for moderate feeding crops, and 200-250 mg·L⁻¹ N for heavy feeders (Kessler, 2002).

Nutritional characteristics of effluent vary case-to-case, however a number of regional averages have been compiled (Tucker, 1998). During a two year period, twenty five catfish farms in Alabama had effluent compositions monitored with averaged results of: 0.073 mL·L⁻¹ settleable solids, 72.63 mg·L⁻¹ TSS, 4.35 mg·L⁻¹ total nitrogen (TN), 1.17 mg·L⁻¹ TAN, 0.24 mg·L⁻¹ TP, and 9.3 mg O₂·L⁻¹ BOD. In Mississippi, effluent from twenty commercial catfish ponds were monitored, averaging 0.078 mL·L⁻¹ settleable solids, 109.63 mg·L⁻¹ TSS, 5.8 mg·L⁻¹ TN, 1.62 mg·L⁻¹ TAN, 0.37 mg·L⁻¹ TP, and 15.57 mg O₂·L⁻¹ BOD. Hybrid striped bass pond production in South Carolina contained effluent characterized with <0.4 mg·L⁻¹ settleable solids, 49.2 mg·L⁻¹ TSS, 7.06 mg·L⁻¹

kjeldahl nitrogen, $0.07 \text{ mg}\cdot\text{L}^{-1} \text{ NO}_2^-$, $0.95 \text{ mg}\cdot\text{L}^{-1} \text{ TAN}$, $0.304 \text{ mg P}\cdot\text{L}^{-1} \text{ TP}$, and $11.6 \text{ mg O}_2\cdot\text{L}^{-1} \text{ BOD}$ (Tucker, 1998). From Brazil, Nile tilapia stocked ponds had a chemical composition of: $1.95 \text{ mg}\cdot\text{L}^{-1} \text{ NH}_3\text{-N}$, $0.071 \text{ mg}\cdot\text{L}^{-1} \text{ NO}_2\text{-N}$, $0.8 \text{ mg}\cdot\text{L}^{-1} \text{ NO}_3\text{-N}$, $0.013 \text{ mg}\cdot\text{L}^{-1} \text{ PO}_4^3$, and $109.20 \text{ mg}\cdot\text{L}^{-1} \text{ K}^+$ (Castro et al., 2006).

Previous research indicates a specific production rate of N per amount feed fed (Rakocy et al., 2006). Intensive fish culture systems will have a greater feed input than traditional extensive systems, therefore an increased N concentration. Intensive systems produce fish at an approximate stocking density of $59.4 \text{ kg fish}\cdot\text{m}^{-3}$ (Masser et al., 1999). Estimated N concentrations from feed range from $1 \text{ kg TAN}\cdot 45.4 \text{ kg feed fed}^{-1}$ ($2.2 \text{ lbs TAN}\cdot 100 \text{ lbs feed fed}^{-1}$) (Masser et al., 1999). Given a specific feeding rate, N concentration can be determined, and further extrapolated to feed per plant production area. UVI recommendations, for raft hydroponics, are $60\text{-}100 \text{ g fish feed}\cdot\text{m}^{-2}$ of plant production area (Rakocy et al., 2006). Based upon feeding rate listed above, a plant production system consisting of 100 m^2 would need 10 kg (22 lbs) of feed. At the given rate, TAN concentrations can be estimated to be 98 g (0.22 lbs). UVI aquaponic system reared tilapia at two different stocking densities, $77 \text{ fish}\cdot\text{m}^{-3}$ (Nile tilapia) and $154 \text{ fish}\cdot\text{m}^{-3}$ (Red tilapia), with feed conversion rates of 1.7 and 1.8 respectively. At this production level, estimated TAN is 8.7 kg (19.2 lbs) for Nile tilapia, and 10.6 kg (23.4 lbs) for Red tilapia. Knowing the concentrations of various forms of nitrogen in a fish culturing system is desirable for knowing the amount of N available to plants. Calculating N from feed inputs includes several forms of nitrogen, including TAN, NO_3^- , and NO_2^- . A generally accepted rule-of-thumb for on-farm fish population estimation is assuming a 2:1 feeding conversion ratio (FCR), indicating if a farmer were feeding 45.4 kg (100 lbs)

feed, estimated weight of fish would be 22.7 kg (50 lbs) fish (B. Daniels, personal communication).

Although impossible to characterize any given aquaculture production systems' effluent based on the nutrient and chemical composition of various systems, sufficient levels of N appear to be available for plant production (Castro et al., 2006; Clarkson and Lane, 1991; Palada et al., 1999; Rakocy et al., 2006; Rakocy et al, 2004a; Savidov, 2004; Schuenhoff et al., 2003; Seawright et al., 1998; Tucker, 1998). The objective of this study was to evaluate effluent water from intensive tilapia culture as an irrigation replacement, substituting or supplementing fertilizer inputs used in standard greenhouse production methods.

Material and Methods

Nile tilapia (*Oreochromis niloticus*) was produced in an intensive, bio-floc production system at the E.W. Shell Fisheries Center, North Auburn Unit, in Auburn, AL (Cold Hardiness Zone 8) during 2008 and 2009. Production was conducted in a 29.3 x 9.1 m (96 x 30 ft) double layer, polyethylene covered greenhouse, for environmental control in year-round production. Fish were stocked at 80 fish·m⁻³, in two 27.4 x 3.8 m x 1.2 m (90 ft x 12.5 ft x 4 ft) tanks each with a volume of 125 m³ (33,000 gal), constructed of plywood with steel I-beam and metal cable reinforcements, and lined with a 12 mm polyethylene liner. Each production tank was partitioned into four netted sections for grading fish by size and for staggered harvest, moving fish forward as size increased, until reaching a marketable weight of 0.45 kg (1 lb). Individual partitions were stocked at different times for a staggered harvest, the most recent stocking on 1 August 2009 of

approximately 10,000 50 g tilapia fingerlings. Daily water exchange rates were maintained at low levels, averaging 1-6%, but up to 12% when excess flushing was needed. Tank water was exchanged with water from a local reservoir and with well water. Aeration was provided from two 1.5 hp air-blowers (Sweetwater[®], AES Inc., Apopka, FL).

During winter months, water temperature was maintained within a range suitable for tilapia production ($\geq 22^{\circ}\text{C}$), and recirculation of warm air for aeration provided further warming of water. For heating, a 200,000 BTU corn burner was utilized, cycling reservoir water through the burner at $3.78 \text{ L}\cdot\text{min}^{-1}$ (two $\text{gal}\cdot\text{min}^{-1}$), heated to approximately 48.8°C (120°F). During daylight hours, heated water was provided to the fish production greenhouse, while heating was provided to an adjacent plant greenhouse during the night. Shelled corn, priced at \$2.50-\$3.00 per bushel was used as a fuel, ($\approx 7500 \text{ BTU}\cdot\text{lb}^{-1}$), and pelletized wood was utilized as well.

Tilapia production yields averaged near 9,000-11,000 kg (10-12 tons) of fish for the entire system annually (250 m^3 ; 66,043 gallons water). Estimated production per acre is 136,000-181,400 kg (300,000-400,000 lb) annually.

Dissolved oxygen and temperature of fish culturing water were recorded twice daily (YSI 550A, YSI Inc., Yellow Springs, OH) (Figure A1). Water temperature exhibited an increasing trend between December 2008 and September 2009, however temperatures can be expected to drop during winter months. Culture water DO indicated a general decreasing trend during the same time frame. Culture water pH, EC, and salinity (YSI 63 meter), TAN (1.0 to $8.0 \text{ mg}\cdot\text{L}^{-1}$, LaMotte Company, Chestertown, MD), and $\text{NO}_3^{-}\text{-N}$ (Cardy meter, range 0 to $9,900 \text{ mg}\cdot\text{L}^{-1}$, Spectrum Technologies, Inc.,

Plainfield, IL) were measured daily. Water samples were collected directly from fish tanks weekly and analyzed using ICAP and for NH_4 determination (Tables A1). Tilapia were fed a 32% crude protein feed from Alabama Catfish Feed Mill (Uniontown, AL), fed between 22 kg to 34 kg (50 lbs to 75 lbs) feed·tank⁻¹·day⁻¹. During the production period of December 2008 to September 2009, a general increasing trend in feed fed was observed.

Trial

On September 3, 2008 cuttings of Deep Blue Improved *Nemesia Aromatica*TM (*Nemesia fruticans*) and Calibrachoa CabaretTM Red (*Calibrachoa x hybrida*), (John McBryde, Ball Horticultural Company), were stuck into 72 cell-pack trays filled with common starter substrate (Germinating Mix, Conrad Fafard Inc., Agawam, MA) at Paterson Greenhouse Complex, Auburn University (USDA Cold Hardiness Zone 8). Cuttings were placed under mist irrigation at ten seconds every ten minutes, for 2 weeks. Upon rooting, plugs were transported to a 30.5 x 9.0 m (96 x 30 ft) pad and fan greenhouse with polyethylene cover at E.W. Shell Fisheries Center. Plugs were transplanted into 15.24 cm (6 in.) diameter containers, with a volume of 1278 cm³ (78 in³) (6.0 AZ traditional TW, Dillen Products/Myers Industries, Middlefield, OH), into a common greenhouse substrate (70% peat, 15% perlite, and 15% vermiculite). Dependent variables included water source (CW or EW, or a 50:50 blend) and incorporated fertilizer at 1.5 lbs N·yd⁻³ using 12-6-6 Gro and Sho Nursery Special (The State Plant Food Inc., Dothan, AL). Each pot was filled level to rim with substrate, irrigated to settled substrate, and plugs were placed following settling. Six treatments included 1) 100% CW, no fertilizer; 2) 100% EW, no fertilizer; 3) 50:50 CW:EW, no fertilizer; 4) 100% CW,

fertilizer; 5) 100% EW, fertilizer; and 6) 50:50 CW:EW, fertilizer. All treatments received 1.4 kg (3 lbs) lime·yd⁻³. Each treatment consisted of twelve single pot replications for both nemesia and calibrachoa, and organized into a complete randomized block design (CRBD). Plants received uniform irrigation applications, receiving 100 ml to 200 ml water per irrigation as needed.

Leachates were collected using Virginia Tech pour-through method measuring pH and electrical conductivity (EC) at 20, 27, 34, and 41 days after transplanting (DAT) (Yeager et al., 2007). Leaf chlorophyll content was measured 17 DAT using a SPAD-502 Chlorophyll Meter (Minolta Camera Co., Ramsey NJ). Growth indices were measured 33 DAT, and SFW and SDW (48 hours at 155^oF) were recorded, foliar nutrient analyses were conducted, and a visual root rating based on root density was taken (1= lowest density, 5 = highest density). All data was analyzed using proc GLM, Waller-Duncan K-ratio t-test using SAS (Version 9.1; SAS Institute, Cary, NC).

Study 1

Based on preliminary trials (data not shown), an experiment began on July 28, 2009. Plugs of Angelonia AngelMistTM (*Angelonia angustifolia*) Suncatcher Vegetative Petunia (*Petunia x hybrida*), and Vegetative AztecTM Verbena (*Verbena x hybrida*) (John McBryde, Ball Horticultural Company) were transplanted from 72-plug trays into a common substrate (Fafard 3B) in 15.24 cm (6 in) diameter containers, with a volume of 1278 cm³ (78 in³). This study was conducted in a 30.5 x 9.15 m (96 x 30 ft) pad and fan greenhouse with polyethylene cover. Dependent variables consisted of water source (CW or EW) and application of soluble fertilizer. Plugs were transplanted following settling of substrate, previously irrigated for settling. There were a total of two treatments consisting

of treatments receiving CW with 200 mg·L⁻¹ N 20-20-20 TotalGro (SDT Industries, Winnsboro, LA) or 100% EW, with ten single pot replications, placed on greenhouse benches in a CRD. Units received uniform watering of 200 ml throughout duration of study.

Data collected included substrate pH, EC, and salinity, and relative growth indices (RGI), shoot fresh weight (SFW), and shoot dry weight (SDW). Petunia crop was grown for 31 days, verbena for 38 days, and angelonia for 44 days. Leachates were collected at 21 and 31 DAT for petunia, at 21, 31, and 37 DAT for verbena, and 21, 31, 37, and 44 DAT for angelonia. Leachates were collected using the Virginia Tech pour-through method (Yeager et al., 2007) and evaluated for pH, EC, and salinity (YSI 63 meter, YSI Inc., Yellow Spring, OH). RGI were recorded, initial GI_I measured at 17 DAT for all species grown. Final GI_F was measured for petunia at 31 DAT, for verbena at 37 DAT, and angelonia at 44 DAT. SFW weights were recorded on the same date as GI_F, followed by SDW measurements. Data was analyzed using proc GLM, Tukey's Standardized Range Test (SAS Version 9.1, SAS Institute, Cary, NC).

Study 2

The study was subsequently repeated, beginning on August 20, 2009. Containers remained 15.24 cm (6 in.). Plant material used was Celebrity Blue Petunia (*Petunia x hybrida* 'Celebrity Blue'), and Cooler Rose Vinca (*Catharanthus roseus* 'Cooler Rose'), transplanted from 288-plug trays into a common substrate (Fafard 3B). The study was conducted in a 30.5 x 9.15 m (96 x 30 ft) pad and fan greenhouse with a double layer polyethylene cover. Dependent variables were the same as those in the first study. Treatments included 1) CW with 200 mg·L⁻¹ N (20-20-20 TotalGro), 2) 100 mg·L⁻¹ N

(20-20-20 TotalGro), and 3) 100% EW. Each treatment consisted of nine single pot replications, placed on greenhouse benches in a CRD, receiving uniform irrigation throughout duration of study at 200 ml per irrigation.

Data collected included substrate pH, EC, and salinity, as well as GI, SFW, and SDW. Both crops were grown for a total of 45 days. Leachates were collected at 18 and 45 DAT. Leachates were collected using the Virginia Tech pour-through method (Yeager et al., 2007) and evaluated for pH, EC, and salinity (YSI 63 meter). GI and SFW were recorded at 45 DAT. SDW were recorded after drying plant at 80°C (175°F) for 48 hours. Data was analyzed using proc GLM, Waller-Duncan K-ratio t test (SAS Version 9.1, SAS Institute, Cary, NC).

Results and Discussion

Trail

Leachates

Substrate pH of nemesia remained within recommended range (4.5-6.5) across all treatments throughout the study (Table 3.2) (Yeager et al., 2007). Highest pH levels for CW (5.62) and EW (5.49) treatments were recorded at 34 DAT, while both CW with fertilizer (6.15) and 50%CW: 50% EW (6.16) had highest pH levels at 20 DAT. Effluent water treatment had an increasing pH throughout duration of study. Electrical conductivity, recommended at 0.5-1.0 mS·L⁻¹, levels peaked at 27 DAT for EW (1.16), CW with fertilizer (0.88), 50% CW: 50% EW with no fertilizer (0.47), and 50% CW: 50% EW (1.92), while 50% CW:50% EW and CW exhibited a general decreasing EC

trend from 20 DAT to 41 DAT. Treatment EW with fertilizer displayed EC levels within BMP range for duration of study (Table 3.2).

Calibrachoa substrate pH levels from 20 DAT to 41 DAT remained within BMP recommended range across all treatments (Table 3.3.). Substrate pH levels for CW with fertilizer, EW with fertilizer, and 50% CW: 50% EW with fertilizer treatments were highest at 20 DAT. Generally, substrate EC fluctuated above and below recommended range ($0.5\text{-}1.0\text{ mS}\cdot\text{cm}^{-1}$). All treatments exhibited EC's below BMP range from 34 DAT to 41 DAT, except 50% CW: 50% EW with fertilizer treatment which was within range at 34 DAT, declining below range by 41 DAT.

Growth Index and Dry Weights

Nemesia GI was greatest for treatment EW with fertilizer (29.74 cm) which was different from other treatments, but followed by 50% CW: 50% EW with fertilizer (27.49 cm) (Table 3.4). Plants grown with CW exhibited the smallest GI (13.74 cm). Shoot dry weights correspond directly to GI with differences across treatments, with treatment EW with fertilizer having the greatest SDW (11.68 g). Plants in the clear water treatment had the smallest SDW (1.28 g).

Growth indices for Calibrachoa indicated differences across treatments, with EW with fertilizer performing best (31.04 cm), and with the greatest SDW (8.07 g), different from other treatments (Table 3.5). Trends for GI and SDW for calibrachoa had similar trends as those of nemesia, with plant in the CW treatment exhibiting the poorest performing plants (GI = 14.88 cm; SDW = 1.35 g).

Results indicate that substrate pH can be maintained in an adequate range using EW. Electrical conductivity was generally out of range, but with little if any negative

effects on over all plant growth. Results of GI for both species indicates EW with a medium rate CRF performed best under production conditions, out performing all other treatments for both species. Further, results suggest that EW and CRF may have had a symbiotic interaction, producing better plant than plants produced in other treatments.

Study 1

Leachates

Substrate pH for angelonia remained within recommended range (4.5-6.5) from 21 DAT to 44 DAT, with no difference between the plant in the two treatments except at 31 DAT (Table 3.6). Although no differences were observed between treatments for EC, EW treatment EC remained higher than BMP range (0.5-1.0) from 21 DAT to 44 DAT, treatment SF remained within range, except at 31 DAT where is temporarily rose above range. No differences were observed for salinity between plants in the two treatments.

Petunia substrate pH from 21DAT to 31 DAT indicated no differences between plants in the treatments, while pH levels remained within range for duration of the study (Table 3.7). Differences between plants in the treatments were observed from both EC and salinity. Substrate EC levels were above BMP range for plants in EW treatment for duration of study, while plant in the SF treatment EC's remained with in range (Table 3.7).

Leachates for verbena resemble recorded levels for angelonia and petunia. Substrate pH levels remained within recommended range (4.5-6.5) from 21 DAT to 37 DAT, both treatments' pH levels were similar for the same time frame (Table 3.8). Substrate EC's were different at 21 DAT, plants in the SF treatment ($0.93 \text{ mg}\cdot\text{L}^{-1}$) were within range ($0.5\text{-}1.0 \text{ mS}\cdot\text{cm}^{-1}$) while EW ($2.83 \text{ mS}\cdot\text{cm}^{-1}$) was above BMP range. From 31

DAT to 37 DAT, plants in the treatment EC levels were similar. Salinity reflects trends of EC, while at 21 DAT treatments were different, and from 31 DAT to 37 DAT treatments were similar (Table 3.8).

Growth Indices and Shoot Weights

Angelonia GI displayed no differences between treatments (SF = 40.29 cm; EW = 40.43 cm). Shoot weights were different between treatments. Plants in the SF treatment exhibited a greater SFW and SDW (117.28 g, 21.87 g respectively) than EW treatment (88.54 g, 17.41 g) (3.9).

Petunia GI, SFW, and SDW trends followed trends of previously reported petunia results. Differences were observed between treatments for GI (SF = 25.90 cm; EW = 20.87 cm), SFW (SF = 60.56 g; EW = 31.77 g), and SDW (SF = 5.22 g; EW = 2.83 g) (Table 3.10).

Contrary to results of angelonia and petunia, verbena exhibited no differences between treatments for GI, SFW, or SDW (Table 3.11). Growth index for SF was 21.67 cm and EW was 23.13 cm. For SF treatment SFW was 25.32 g and for EW was 31.83 g, while SDW for SF was 4.09 g and for EW was 4.25 g. Plants receiving EW displayed delayed blooming compared to SF treatments.

Results support previous results where effluent water from intensive aquaculture provided insufficient nutrients to produce a similar plant to units receiving $200 \text{ mg} \cdot \text{L}^{-1} \text{ N}$ of soluble fertilizer for angelonia and petunia. Verbena however, grew similarly under both treatments. Heavy feeding plants appear to not receive adequate nutrients to grow as expected. However, for less heavy feeding plant, such as verbena, sufficient nutrients were provided producing a similar crop. Visual differences are slight for angelonia and

petunia; however, plants from both treatments are aesthetically pleasing, while EW water verbena plants are pictured with increased flower number.

Study 2

Leachates

Substrate pH for petunia remained within recommended range (4.5-6.5) for duration of study, with exception of EW treatment at 45 DAT (Table 3.12) (Yeager et al., 2007). At 18 DAT, all pH levels are within range, though differences were observed. At 45 DAT, EW treatment pH (7.37) was difference than 200 and 100 mg·L⁻¹ N soluble 20-20-20 fertilizer treatments (6.05, 6.24 respectively). Electrical conductivity for substrates of petunia was generally outside of recommended range (0.5-1.5 mS·cm⁻¹, Yeager et al., 2007). At 18 DAT all treatment EC levels were above recommended range, while at 45 DAT only treatment 200 mg·L⁻¹ (0.82 mS·cm⁻¹) was within BMP range, other treatments were below range (Table 3.12). Substrate salinities remained at lower concentrations than recorded in previous studies, all recorded at 18 DAT and 45 DAT under 1.0 ppt.

Substrate pH for vinca indicated similar trends to substrate pH of petunia. All pH levels were within BMP range, excluding treatment receiving 100 mg·L⁻¹ N soluble 20-20-20 fertilizer rates, which was slightly outside recommended range (6.54) (Table 3.13). Initial substrate EC for all treatments was higher than BMP range, while at 45 DAT all treatment EC levels dropped within range (Table 3.13). Salinity concentrations of vinca substrates were below 1.0 ppt, and declined from 18 DAT to 45 DAT. No differences were observed among all treatments at 18 DAT and 45 DAT for substrate pH, EC, and salinity.

Growth Index and Shoot Weight

There were no differences in GI for petunia at 45 DAT across all treatments (Table 3.14). Shoot fresh weights indicated differences among 100 mg·L⁻¹ and EW treatments (94.45 g, 78.15 g respectively), while plants treated with 200 mg·L⁻¹ were similar to plant in other treatments (87.94 g). Shoot dry weights were not similar to trends of SFW, no differences were observed across all treatments, indicating the differences observed in SFW was due to water content (Table 3.14).

Vinca growth indices exhibited no differences across treatments, nor were differences observed across treatments for SFW and SDW (Table 3.14). Visual assessment of treatments between vinca indicated an increased uniformity among plants irrigated with EW.

Results support prior experiments where petunia receiving standard nursery inputs exhibited increased growth compared to plants receiving EW. Due to heavy nutrient requirement of petunia, EW from the intensive aquaculture production system utilized could not meet the nutritional demand of petunia. More research should be conducted to determine the ratio of supplemented EW to standard fertilizer inputs for highest cost reduction, if petunia is to be produced. Vinca has lower nutritional requirement than petunia, and grew without differences between all treatments, indicating adequate nutrition was supplied to crop from EW as well as soluble fertilizer inputs. Results from vinca conclude that EW from the intensive tilapia production results in comparable crop production to standard production practices.

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Table 3.1. Industry accepted foliar nutrient analysis compositions.

Nutrient ^{ZY}	General plant nutrition	<i>Petunia</i> nutrition	<i>Verbena</i> nutrition	<i>Catharanthus</i> nutrition
	Argo et al., 2009 ^X	Mills and Jones, 1996 ^W		
Nitrogen	2.5-6.0	3.87-7.60	3.64-5.84	2.72-6.28
Phosphorus	0.3-1.0	0.47-0.93	0.77-1.19	0.28-0.64
Potassium	2.5-6.0	3.13-6.65	2.79-4.69	1.88-3.48
Calcium	0.6-2.0	1.20-2.81	1.60-2.54	0.93-1.13
Magnesium	.03-1.0	0.36-1.37	0.73-1.58	0.32-0.78
Sulfur	0.3-1.0	0.33-0.80	0.58-0.64	0.22-0.50
Iron	75-200	84-168	56-112	72-277
Manganese	50-200	44-177	55-293	135-302
Zinc	25-100	33-85	65-127	30-51
Copper	5.0-20	3.0-19	3.0-13	6.0-16
Boron	30-120	18-43	43-48	21-49
Molybdenum	1.0-5.0	0.19-0.46	0.14-0.80	0.14-0.46
Sodium	. ^V	3067-10896	257-310	89-1566
Aluminum	.	50-92	41-53	34-136

^ZNutrient: Essential and non-essential nutrients for plant growth and development.

^YUnit: % for N, P, K, Ca, Mg, and S; mg·L⁻¹ for Fe, Mn, Zn, Cu, B, Mo, Na, and Al.

^XArgo, B., P. Fisher, and K. Santos. 2009. Understanding plant nutrition: Diagnosing problems.

Greenhousegrower.com. 31 Aug. 2009. <<http://www.greenhousegrower.com/magazine/?storyid=1833>>.

^WMills, H.A. and J.B. Jones. 1996. Plant Analysis Handbook II: A practical sampling, preparation, analysis and interpretation guide. MicroMacro Pub. Inc. Athens, GA.

^VNo data provided.

Table 3.2. Intensive tilapia culture effluent nutrient analysis, during greenhouse plant production from September 2008 to September 2009.

Nutrient ^{ZY}	Sep-Oct, 2008	Jul-Aug, 2009	Aug-Sep, 2009
TAN ^X	. ^W	12.20	16.43
NH ₄ -N	11.10	14.90	16.00
NO ₃ ⁻ -N	26.70	44.90	38.88
P	14.00	15.30	19.60
K	23.10	31.50	42.10
Ca	18.50	22.50	36.90
Mg	.	40.60	52.10
Fe	.	0.10	0.50
Zn	.	0.20	0.20
Mn	.	0.40	0.40
Cu	.	0.10	0.10
B	.	0.10	0.10
Mo	.	0.10	0.10
Na	128.00	192.70	109.80
pH ^V	5.90	4.65	4.85
EC ^U	0.40	1.21	0.94

^ZNutrient analysis determined by ICAP.

^YAll nutrients measured in mg·L⁻¹, unless noted.

^XTAN: Total ammonia nitrogen (NH₄⁺-N and NH₃-N).

^WData not provided.

^VpH (YSI 63 meter).

^UEC: Electrical conductivity measured in mS·cm⁻¹ (YSI 63 meter).

Table 3.3. *Nemesia fruticans* leachates during September-October 2008, potted in common substrate (70% peat, 15% perlite, 15% vermiculite).

Treatment ^z	pH					Electrical conductivity ^y						
	20 DAT ^x	27 DAT	34 DAT	41 DAT	21 DAT	27 DAT	34 DAT	41 DAT	21 DAT	27 DAT	34 DAT	41 DAT
CW	5.33b ^v	5.33b	5.62a	5.43b	0.53ab	0.46b	0.40a	0.25b				
EW	5.43b	5.49ab	5.73a	5.45b	0.59ab	1.16ab	0.80a	0.38b				
50% CW:50% EW	5.95ab	5.80a	5.85a	6.10a	0.45ab	0.47b	0.45a	0.31b				
CW, 0.23 kg N·yd ⁻³	6.15a	5.65ab	5.63a	5.55ab	0.35b	0.88ab	0.58a	0.47ab				
EW, 0.23 kg N·yd ⁻³	5.78ab	5.81a	5.87a	5.59ab	0.91a	0.86ab	0.64a	0.81a				
50% CW:50% EW, 0.23 kg N·yd ⁻³	6.16a	5.85a	5.52a	5.64ab	0.58ab	1.92a	0.55a	0.37b				

^zTreatment: CW = Clear Water; EW = Effluent water (see table 3.2 for nutrient analysis); Fertilization rate at 0.23 kg N·yd⁻³ (1/2 lb N·yd⁻³).

^yElectrical conductivity (mS·cm⁻¹) measured with Accumet Excel XL50 Meter (Fisher Scientific).

^xDAT = days after transplanting

^vMeans within columns followed by same letter are not significantly different (Waller-Duncan K-ratio t test, $p \leq 0.05$).

Table 3.4. *Calitbrochoa x hybrida* leachates during September–October 2008, potted in common substrate (70% peat, 15% perlite, 15% vermiculite).

Treatment ^z	pH				Electrical conductivity ^y			
	20 DAT ^x	27 DAT	34 DAT	41 DAT	20 DAT	27 DAT	34 DAT	41 DAT
CW	5.94a	6.05a	6.02a	5.92a	0.31c	0.42a	0.22b	0.25a
EW	5.94a	5.69b	5.63ab	5.84a	0.62c	0.54a	0.21b	0.25a
50% CW:50% EW	6.04a	6.24a	5.89ab	5.85a	0.32c	0.65a	0.15b	0.29a
CW, 0.23 kg N·yd ⁻³	6.01a	5.64b	5.63ab	5.73a	4.75a	0.72a	0.44ab	0.31a
EW, 0.23 kg N·yd ⁻³	6.04a	5.54b	6.05a	5.73a	2.08b	0.70a	0.42ab	0.34a
50% CW:50% EW, 0.23 kg N·yd ⁻³	5.98a	5.71b	5.36b	5.64a	2.09b	0.91a	0.61a	0.38a

^zTreatment: CW = Clear Water; EW = Effluent water (see table 3.2 for nutrient analysis); Fertilization rate at 0.23 kg N·yd⁻³ (1/2 lb N·yd⁻³).

^yElectrical conductivity (mS·cm⁻¹) measured with Accumet Excel XL 50 (Fisher Scientific).

^xDAT = days after transplanting

^wMeans within columns followed by same letter are not significantly different (Waller-Duncan K-ratio t test, $P \leq 0.05$, $N=3$).

Table 3.5. Effect of irrigation source on growth of *Nemesia fruticans* during September-October 2008, potted in common substrate (70% peat, 15% perlite, 15% vermiculite).

Treatment ^z	Leaf chlorophyll content ^y		Growth index (cm) ^w	Shoot dry weight (g)		Root rating ^u
	17 DAT ^x	33 DAT		50 DAT	na	
CW	34.57b ^y	13.74e	1.28d	1.16c		
EW	36.75ab	25.03c	9.23b	4.00a		
50% CW:50% EW	31.52b	21.19d	4.82c	3.50ab		
CW, 0.23 kg N·yd ⁻³	30.92b	25.11c	5.76c	3.16b		
EW, 0.23 kg N·yd ⁻³	41.45a	29.74a	11.68a	3.75ab		
50% CW:50% EW, 0.23 kg N·yd ⁻³	36.27ab	27.49b	8.83b	3.92a		

^zTreatment: CW = Clear Water; EW = Effluent water (see table 3.2 for nutrient analysis); Fertilization rate at 0.23 kg N·yd⁻³ (1/2 lb N·yd⁻³).

^yLeaf chlorophyll content quantified using a SPAD-502 Chlorophyll Meter (Minolta Camera Co., Ramsey, NJ).

^xDAT = days after transplanting.

^wGrowth Index = (Height + Widest Width + Perpendicular Width)/3.

^vMeans within columns followed by same letter are not significantly different (Waller-Duncan K-ratio t test, $p \leq 0.05$).

^uRoot ratings were rated by visual assessment of density of roots systems (1 = Lowest Density, 5 = Highest Density).

Table 3.6. Effect of irrigation source on growth of *Calibrachoa hybrida* during September-October 2008, in common substrate (70% peat, 15% perlite, 15% vermiculite).

Treatment ^z	Leaf chlorophyll content ^y		Growth index (cm) ^w		Shoot dry weight (g)		Root rating ^u
	17 DAT ^x	33 DAT	33 DAT	50 DAT	50 DAT	na	
CW	37.23a	14.88f ^v	14.88f ^v	1.35e	1.35e	2.83ab	
EW	44.83a	24.19d	24.19d	4.85bc	4.85bc	2.25b	
50% CW:50% EW	40.94a	21.61e	21.61e	3.34d	3.34d	2.07b	
CW, 0.23 kg N·yd ⁻³	38.97a	26.24c	26.24c	4.41c	4.41c	3.33a	
EW, 0.23 kg N·yd ⁻³	45.33a	31.04a	31.04a	8.07a	8.07a	3.33a	
50% CW:50% EW, 0.23 kg N·yd ⁻³	41.63a	28.45b	28.45b	5.71b	5.71b	2.42ab	

^zTreatment: CW = Clear Water; EW = Effluent water (see table 3.2 for nutrient analysis); Fertilization rate at 0.23 kg N·yd⁻³ (1/2 lb N·L⁻¹).

^yLeaf chlorophyll content quantified using a SPAD-502 Chlorophyll Meter (Minolta Camera Co., Ramsey, NJ).

^xDAT = days after transplanting.

^wGrowth Index = (Height + Widest Width + Perpendicular Width)/3.

^vMeans within columns followed by same letter are not significantly different (Waller-Duncan K-ratio t test, $p \leq 0.05$).

^uRoot ratings were rated by visual assessment of density of roots systems (1 = Lowest Density, 5 = Highest Density).

Table 3.7. *Angelonia angustifolia* leachates during July-September 2009, potted in common substrate (Fafard 3B).

Treatment ^z	pH				Electrical conductivity ^y				Salinity ^x			
	21 DAT ^w	31 DAT	37 DAT	44 DAT	21 DAT	31 DAT	37 DAT	44 DAT	21 DAT	31 DAT	37 DAT	44 DAT
SF	6.28a ^v	6.10b	6.05a	5.98a	0.63a	1.03a	1.02a	0.86b	0.26a	0.46a	0.43a	0.36b
EW	6.25a	6.95a	6.44a	6.32a	2.30a	1.42a	2.02a	1.30a	1.03a	0.50a	0.96a	0.60a

^zTreatment: 1) SF: 200 mg·L⁻¹ N 20-20-20 soluble fertilizer (TotalGro); 2) EW: 100% Effluent water (see table 3.2 for nutrient analysis of EW).

^yElectrical conductivity: mS/cm⁻¹ (YSI 63 Meter, YSI Inc., Yellow Springs OH).

^xSalinity: NaCl parts per thousand (YSI 63 Meter, YSI Inc., Yellow Springs OH).

^wDAT = days after transplanting.

^vMeans within columns followed by same letter are not significantly different (Tukey's standardized range test, $\alpha = 0.05$, N =3).

Table 3.8. *Petunia x hybrida* leachates during July-August 2009, potted in common substrate (Fafard 3B).

Treatment ^z	pH		Electrical conductivity ^y		Salinity ^x	
	21 DAT ^w	31 DAT	21 DAT	31 DAT	21 DAT	31 DAT
SF	5.47a ^v	6.13a	0.63b	0.93b	0.30b	0.40b
EW	6.01a	6.28a	2.70a	1.83a	1.26a	0.83a

^zTreatment: 1) SF: 200 mg·L⁻¹ N 20-20-20 soluble fertilizer (TotalGro); 2) EW: 100% Effluent water (see table

3.2 for nutrient analysis of EW).

^yElectrical conductivity: mS/cm⁻¹ (YSI 63 Meter, YSI Inc., Yellow Springs OH).

^xSalinity: NaCl parts per thousand (YSI 63 Meter, YSI Inc., Yellow Springs OH).

^wDAT = days after transplanting.

^vMeans within columns followed by same letter are not significantly different (Tukey's standardized range test,

$\alpha = 0.05$, N =3).

Table 3.9. *Verbena x hybrida* leachates during July-August 2009, potted in common substrate (Fafard 3B).

Treatment ^z	pH			Electrical conductivity ^y			Salinity ^x		
	21 DAT ^w	31 DAT	37 DAT	21 DAT	31 DAT	37 DAT	21 DAT	31 DAT	37 DAT
SF	5.90a ^v	5.58a	6.12a	0.93b	1.75a	1.28a	0.40b	0.67a	0.63a
EW	5.77a	5.98a	6.65a	2.83a	3.10a	1.03a	1.33a	1.40a	0.50a

^zTreatment: 1) SF: 200 mg·L⁻¹ N 20-20-20 soluble fertilizer (TotalGro); 2) EW: 100% Effluent water (see table 3.2 for nutrient analysis of EW).

^yElectrical conductivity: mS/cm⁻¹ (YSI 63 Meter, YSI Inc., Yellow Springs OH).

^xSalinity: NaCl parts per thousand (YSI 63 Meter, YSI Inc., Yellow Springs OH).

^wDAT = days after transplanting.

^vMeans within columns followed by same letter are not significantly different (Tukey's standardized range test, $\alpha = 0.05$, N =3).

Table 3.10. Growth index and harvest weights of *Angelonia angustifolia* during September 2009, potted in common substrate (Fafard 3B).

Treatment ^z	Growth index (cm) ^y	Fresh weights (g)	Dry weights (g) ^x
	44 DAT ^w	44 DAT	
SF	40.29a ^v	117.28a	21.87a
EW	40.43a	88.54b	17.41b

^zTreatments: 1) SF: 200 mg·L⁻¹ N 20-20-20 soluble fertilizer (TotalGro); 2) EW: 100% Effluent water (see table 3.2 for nutrient analysis of EW).

^y Growth index = [(Widest width+Perpendicular width+Height)/3].

^x Dry Weight = Plant tissue dried at 80°C (175°F) for 48 hours.

^wDAT = days after transplanting.

^vMeans within columns followed by same letter are not significantly different (Tukey's standardized range test, $\alpha = 0.05$, N =3).

Table 3.11. Growth index and harvest weights of *Petunia x hybrid* during September 2009, potted in common substrate (Fafard 3B).

Treatment ^z	Growth index (cm) ^y	Fresh weights (g)	Dry weights (g) ^x
	31 DAT ^w	31 DAT	
SF	25.90a ^y	60.56a	5.22a
EW	20.87b	31.77b	2.83b

^zTreatments: 1) SF: 200 mg·L⁻¹ N 20-20-20 soluble fertilizer (TotalGro); 2) EW: 100% Effluent water (see table 3.2 for nutrient analysis of EW).

^y Growth indices = [(Width₁+Width₂+Height)/3].

^x Dry Weight = Plant tissue dried at 80°C (175°F) for 48 hours.

^wDAT = days after transplanting.

^vMeans within columns followed by same letter are not significantly different (Tukey's standardized range test, $\alpha = 0.05$, N =3).

Table 3.12. Growth index and harvest weights of *Verbena x hybrida* during September 2009, potted in common substrate (Fafard 3B).

Treatment ^z	Growth Index (cm) ^y	Fresh weights (g)	Dry weights (g) ^x
	38 DAT ^w	38 DAT	
SF	21.67a	25.32a	4.09a
EW	23.13a	31.83a	4.25a

^zTreatments: 1) SF: 200 mg·L⁻¹ N 20-20-20 soluble fertilizer (TotalGro); 2) EW: 100% Effluent water (see table 3.2 for nutrient analysis of EW).

^yGrowth indices = [(Width₁+Width₂+Height)/3].

^xDry Weight = Plant tissue dried at 80°C (175°F) for 48 hours.

^wDAT = days after transplanting.

^vMeans within columns followed by same letter are not significantly different (Tukey's standardized range test, $\alpha = 0.05$, N =3).

Table 3.13. *Petunia* substrate leachates during July-August 2009, potted in common substrate (Fafard 3B).

Treatment ^z	pH		Electrical conductivity ^y		Salinity ^x	
	18 DAT ^w	45 DAT	18 DAT	45 DAT	18 DAT	45 DAT
200 mg·L ⁻¹ N	6.14 ^b ^v	6.05 ^b	1.84 ^a	0.82 ^a	0.83 ^a	0.36 ^a
100 mg·L ⁻¹ N	6.33 ^{ab}	6.24 ^b	1.25 ^b	0.40 ^a	0.56 ^b	0.16 ^a
EW	6.34 ^a	7.37 ^a	1.41 ^{ab}	0.42 ^a	0.63 ^{ab}	0.20 ^a

^zTreatment: 1) 200 mg·L⁻¹ N 20-20-20 soluble fertilizer (TotalGro); 2) 100 mg·L⁻¹ N 20-20-20; 3) EW: 100% Effluent water (see table 3.2 for nutrient analysis of effluent water).

^yElectrical conductivity: mS/cm⁻¹ (YSI 63 Meter, YSI Inc., Yellow Springs OH).

^xSalinity: ppt NaCl (YSI 63 Meter, YSI Inc., Yellow Springs OH).

^wDAT = days after transplanting.

^vMeans within columns followed by same letter are not significantly different (Waller-Duncan K-ratio t test, $P \leq 0.05$, $N = 3$).

Table 3.14. *Catharanthus* substrate leachates during July-August 2009, potted in common substrate (Fafard 3B).

Treatment ^z	pH		Electrical conductivity ^y			Salinity ^x	
	18 DAT ^w	45 DAT	18 DAT	45 DAT	18 DAT	45 DAT	
200 mg·L ⁻¹ N	6.44a ^v	6.29a	1.47a	0.99a	0.66a	0.46a	
100 mg·L ⁻¹ N	6.27a	6.54a	1.27a	0.94a	0.60a	0.46a	
EW	6.25a	6.48a	1.36a	0.54a	0.63a	0.26a	

^zTreatment: 1) 200 mg·L⁻¹ N 20-20-20 soluble fertilizer (TotalGro); 2) 100 mg·L⁻¹ N 20-20-20; 3) EW: 100% Effluent water (see table 3.2 for nutrient analysis of effluent water).

^yElectrical conductivity: mS/cm⁻¹ (YSI 63 Meter, YSI Inc., Yellow Springs OH).

^xSalinity: ppt NaCl (YSI 63 Meter, YSI Inc., Yellow Springs OH).

^wDAT = days after transplanting.

^vMeans within columns followed by same letter are not significantly different (Waller-Duncan K-ratio t test, P ≤ 0.05, N = 3).

Table 3.15. Growth index and harvest weights for *Petunia* and *Catharanthus* during September, 2009 potted in common substrate (Fafard 3B).

Treatment ^z	<i>Petunia</i>			<i>Catharanthus</i>		
	Growth index (cm) ^y	Fresh weight (g)	Dry weight (g) ^x	Growth index (cm)	Fresh weight (g)	Dry weight (g)
	45 DAT ^w			45 DAT		
200 mg·L ⁻¹ N	32.94a ^y	87.94ab	8.35a	28.56a	52.54a	7.69a
100 mg·L ⁻¹ N	35.91a	94.45a	9.35a	27.64a	50.75a	7.16a
EW	33.90a	78.15b	8.07a	29.63a	55.54a	8.15a

^zTreatment: 1) 200 mg·L⁻¹ N 20-20-20 soluble fertilizer (TotalGro); 2) 100 mg·L⁻¹ N 20-20-20; 3) EW: 100% Effluent water (see table 3.2 for nutrient analysis of effluent water).

^yGrowth Index = (height + widest width + perpendicular width)/3

^xDry Weight = plant tissue dried at 80°C (175°F) for 48 hours.

^wDAT = days after transplanting.

^vMeans within columns followed by same letter are not significantly different (Waller-Duncan K-ratio t test, $P \leq 0.05$, $N = 10$).

Figure 3.1. Intensive tilapia production feeding, water temperature and dissolved oxygen trends from December 2008 to October 2009.

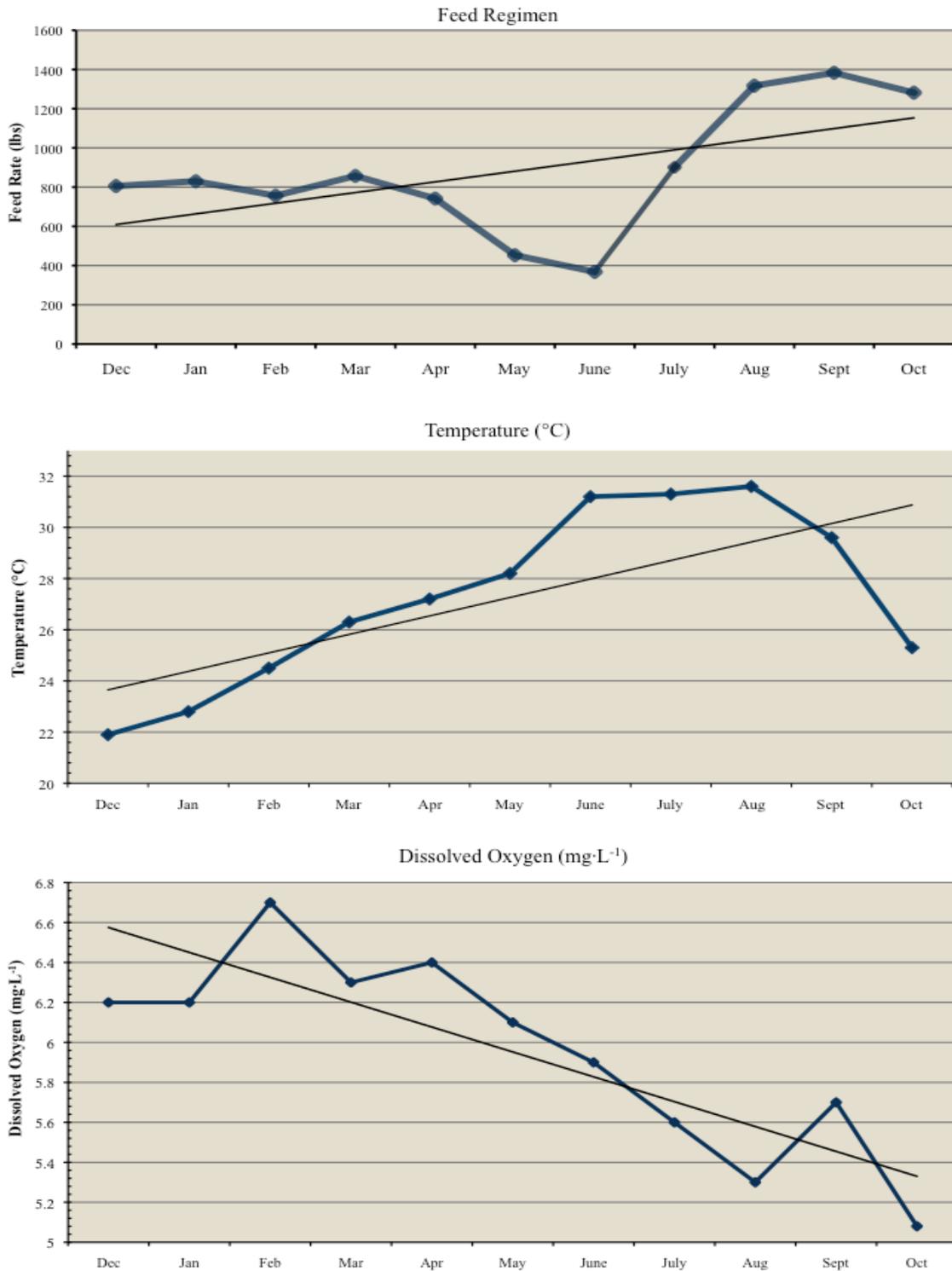


Figure 3.2. Lateral and overhead comparison of *Verbena x hybrida* receiving EW and SF treatments.^Z



^ZEW: 100% Effluent water as irrigation and fertilizer source; SF: 200 mg·L⁻¹ N from 20-20-20 soluble fertilizer (TotalGro).

IV. Greenhouse Vegetable Production Utilizing Intensive Aquaculture Effluent for Irrigation

Abstract

Irrigation application of intensive tilapia culture effluent was evaluated as a replacement/supplement to standard nursery greenhouse production methods (clear water and fertilizer inputs) of vegetable crops. Production of sweet corn and cucumber was evaluated using clear water (CW) with standard soluble fertilizer inputs and effluent water (EW) from an intensive aquaculture production system. In the first experiment, height of corn plants were recorded after 3 weeks growth, and at harvest ear length, weight per plot, and yield per plot were recorded. Differences were not observed among height, or corn weight, length, and yield across treatments. In the second experiment, cucumbers were produced hydroponically in perlite filled bags. Treatments consisted of irrigation with EW or CW and a specially formulated hydroponic fertilizer. Cucumber plant heights were recorded weekly starting at 15 days after planting (DAT), fruit harvest began at 29 DAT and continued throughout study, recording total yield. Cucumbers were rated for quality into three categories, established from previous research. Results of study one indicate EW from intensive aquaculture can produce a similar crop of corn compared to standard greenhouse production methods. Production of cucumber irrigated with a hydroponic fertilizer yielded an increased fruit number compared to EW irrigated cucumber plants; however no differences were observed among weight and length of fruit.

Index of Words: Fish production, intensive tilapia culture, horticulture, beet alpha cucumber, sweet corn

Significance to Industry

Tank effluent from intensive cultured tilapia (*Oreochromis niloticus*) was evaluated as a source of irrigation and fertilization for greenhouse vegetable crop production. Studies indicate sweet corn and cucumbers grown using effluent-laden water from intensive aquaculture performed similarly to plants grown under standard production means. As production costs continue to rise, environmental policies grow more stringent, and interest in greenhouse vegetable production increases, effluent from intensive fish culture might provide an economical and environmentally sustainable alternative to current water use and fertilizer practices in plant production and intensive aquaculture.

Introduction

Agricultural integrations offer potentially long-term positive effects. Assimilating various sectors of agriculture provides increased levels of sustainability, protection of human and environmental health, increased revenues, extended life of materials once thought to be fully used, provision of alternatives to replace limited naturally occurring materials, and providing aid in producing the same quality products as traditional production means (Diver, 2006). Due to benefits listed, an argument for sustainable and integrated agriculture carries more significance in a changing world. Among such integrated agriculture endeavors is the growing trend of integrating aquaculture and

horticulture. This integration is valuable due to multiple benefits. Integrating intensive aquaculture with horticulture crop production is beneficial due to sequestering heavy nutrient loads by plants preventing release into surrounding ecosystems.

Intensive aquaculture (IA) operations contained within an environmentally regulated system have inherent benefits over traditional aquaculture production practices. Environmentally regulated systems are indoor systems, often in greenhouses, allowing for year round fish production (Holliman, 2006; Masser et al., 1999). Other benefits include reduced land requirement, and possibly a decreased water requirement (Masser et al, 1999; Rakocy, 1989). Year round production of fresh product tends toward increased profitability, as well as being able to operate in closer proximity to local markets (Holliman, 2006; Masser et al 1999). IA makes use of high stocking densities, disrupting reproductive processes in intensive tilapia culture, producing a marketable fish sooner than in pond culture. Time requirement for feeding and harvesting is decreased in intensive tank culture, which further leads to decreased labor costs (Rakocy, 1989). Due to year round production, trained workers may then remain on staff. All factors considered, IA is an efficient use of resources compared to traditional pond culture, equating to increased revenues.

For both aquaculturists and horticulturists, benefits from integrating exist (Diver, 2006). For horticulture growers, dependent upon individual situations, fish waste from aquaculture may meet USDA standards for organic certification. If crop production meets USDA organic criteria, crops can not only be advertised in niche markets as locally grown, but as organic crops, for increased profit. Further, crop production can serve as a means of biofiltration for recirculating aquaculture systems (RAS), increasing

sustainability. Integrated systems are often appealing to consumers who are becoming increasingly environmentally conscious. Using wastewater as an irrigation source for crop production can recycle and use pre-existing byproducts from aquaculture systems, which may prove to be economically sensible, producing two products from one production system's resources (Diver, 2006).

Benefits of integrated aquaculture and horticulture systems (IAHS), however, may have more substantial impacts than listed above. Increased production of both aspects of IAHS is possible. Previous work indicates that with IAHS, production rates for aquaculture rise significantly from 2,720 – 6,800 kg (6,000 – 15,000 lbs) fish·acre⁻¹·year⁻¹ to 136,000 – 363,000 kg (300,000 – 800,000 lbs) fish·acre⁻¹·year⁻¹ (Neori et al., 2004; Rakocy, 2002; Rakocy et al., 2000). An aquaponics system at the University of the Virgin Islands (UVI) generated \$110,000 producing 5,000 kg (11,000 lbs) basil annually and 2,900 kg (6,400 lbs) of okra valued at \$6,400 (Rakocy et al., 2006). In another UVI study, basil was produced via hydroponics (batch and staggered) and in a traditional field setting. Results indicated greater basil yields in hydroponic system over field production: 25.0 kg·m⁻² (batch), 23.4 kg·m⁻² (staggered), and 7.8 kg·m⁻², respectively (Rakocy et al., 2004). The value of basil produced was US \$550·m⁻²·year⁻¹ (batch), US \$515·m⁻²·year⁻¹ (staggered), and US \$172·m⁻²·year⁻¹. In Alberta Canada, while not operating at full capacity, greenhouse production of tomato, cucumber, and basil was recorded, respectively, at 40 kg·m⁻², 100 kg·m⁻², and 42 kg·m⁻² annually, indicating greater yields than traditional greenhouse production in Canada (Rakocy et al., 2007).

Due to effects of recent droughts in southeastern US, water reuse is recommended. Record drought conditions were observed during August 2007, when

Alabama drought status reached “extreme drought” according Palmer Drought Index (PDI), further encouraging water conservation production practices (Ding, 2008).

Environmental instability has lead to decreased profit margins for many horticulture producers. In nursery and landscape sectors, financial losses observed were extensive.

During 2006 California and multiple southeastern states comprised 60% of gross national sales in the Green industry. Yet, in 2007, industry from these regions witnessed decreased sales, increased plant mortality, increased watering expenses, and many businesses laid employees off, closed store locations, or filed for bankruptcy (Ding, 2008).

Fertilizer costs have increased 120% in past two decades (NASS, 2009; NASS, 2000; NASS, 1990), encouraging alternative fertilizer inputs. Fuel costs have risen too. Production costs have risen as a result to increased prices of fertilizers and fuel. The need for alternative fertilizer sources is great, and effluent water from aquaculture is one viable option. Concluding from mutual benefits, pressing environmental trends, and economic volatility, the prospective of integrated systems for the conservation and reuse of water is becoming increasingly favorable.

Nursery production in Alabama is comprised of 57 ha (141 ac) of nursery stock under various forms of protection, and nearly 3100 ha (6758 ac) of open land nursery stock production (Clark, 2009). Total area readily available for the nursery and greenhouse industry in Alabama is near 3360 ha (8300 ac). Considering the industry uses 20,000 gallons water·acre⁻¹·day⁻¹ (Avent, 2003; Berghage et al., 1999; Lea-Cox and Ross, 2001), estimated total water use is 166 million gallons daily. Integrated systems offer means to reduce water use dramatically. Recirculating aquaculture systems (RAS) can use as little as 1-2% of water required a day indicating 98-99% is reused.

During the last fifteen years a shift in hydroponic greenhouse vegetable production has been observed in Florida, moving from traditional crops (tomatoes and cucumber) to more market specific crops such as peppers, herbs, lettuce, and strawberries (Tyson and Hochmuth, 2009). Cucumber (*Cucumis sativus*) has become an important hydroponically produced greenhouse crop (Tyson et al., 2001), a trend that is continuing, reaching into Alabama.

Vegetable production is an important industry in Alabama. During 2008, 2400 ha (6000 ac) of field grown vegetables were harvested, while only 2.33 ha (5.75 ac) were dedicated to greenhouse vegetable production. Statewide total vegetable production accounted for \$17.2 million in sales (Clark, 2009). Estimate of annual value of fruit, pecan, and vegetable production in Alabama is \$57 million in cash receipts (Higginbotham, 2004). Alabama is nationally ranked for production of sweet potato (5th), pecan (7th), blueberry (12th), fresh-market tomatoes (12th), and fresh-market watermelons (16th) (Higginbotham, 2004). As mentioned, greenhouse vegetable production is a growing, in Mississippi there are 6 ha (15 ac) of greenhouse tomato production (Helms, 2005). During 2005, in Alabama there were between 15 and 20 greenhouse vegetable producers in Alabama, producing crops such as tomatoes, herbs, microgreens, cucumbers, and lettuce (Helms, 2005).

Traditional field production costs of vegetables and fruits are high (ACES, 2007; 2008). For slicer cucumbers under irrigation, cost per acre in 2007 was \$4500 (ACES, 2007). Handpicked, fresh-market sweet corn production costs reach \$1300·acre⁻¹. Fresh fruit production costs are exponentially higher. For field grown, irrigated strawberry production costs were estimated at \$16,900·ac⁻¹ in 2008. Nearly \$5000 of strawberry

production costs was fertilizer (ACES, 2008). Greenhouse vegetable production may provide farmers relief from such high production costs. Another area of potential increased profits is off-season greenhouse crop production. Off-season greenhouse production can reduce fuel/energy costs associated with shipping of fresh produce where unavailable and supports local economies (Schonbeck et al., 1991). For fruits such as strawberries, blueberries, peaches, watermelon, blackberries, cantaloupe, and others, winter month greenhouse production may provide substantially higher revenues for crops traditionally produced in summer months.

The market for increased greenhouse production of vegetables is great in Alabama. The objective of this study was to evaluate nutritional suitability effluent water from intensive tilapia culture as an irrigation replacement, and substitute or supplement fertilizer inputs used in standard greenhouse production methods.

Material and Methods

Nile tilapia (*Oreochromis niloticus*) was produced in an intensive, bio-floc production system at the E.W. Shell Fisheries Center, North Auburn Unit, in Auburn, AL (Cold Hardiness Zone 8) during 2008 and 2009. Production was conducted in a 29.3 x 9.1 m (96 x 30 ft) double layer, polyethylene covered greenhouse, for environmental control in year-round production. Fish were stocked at 80 fish·m⁻³, in two 27.4 x 3.8 m x 1.2 m (90 ft x 12.5 ft x 4 ft) tanks each with a volume of 125 m³ (33,000 gal), constructed of plywood with steel I-beam and metal cable reinforcements, and lined with a 12 mm polyethylene liner. Each production tank was partitioned into four netted sections for grading fish by size and for staggered harvest, moving fish forward as size increased,

until reaching a marketable weight of 0.45 kg (1 lb). Individual partitions were stocked at different times for a staggered harvest, the most recent stocking on 1 August 2009 of approximately 10,000 50 g tilapia fingerlings. Daily water exchange rates were maintained at low levels, averaging 1-6%, but up to 12% when excess flushing was needed. Tank water was exchanged with water from a local reservoir and with well water. Aeration was provided from two 1.5 hp air-blowers (Sweetwater[®], AES Inc., Apopka, FL).

During winter months, water temperature was maintained within a range suitable for tilapia production ($\geq 22^{\circ}\text{C}$), and recirculation of warm air for aeration provided further warming of water. For heating, a 200,000 BTU corn burner was utilized, cycling reservoir water through the burner at $3.78 \text{ L}\cdot\text{min}^{-1}$ (two $\text{gal}\cdot\text{min}^{-1}$), heated to approximately 48.8°C (120°F). During daylight hours, heated water was provided to the fish production greenhouse, while heating was provided to an adjacent plant greenhouse during the night. Shelled corn, priced at \$2.50-\$3.00 per bushel was used as a fuel, ($\approx 7500 \text{ BTU}\cdot\text{lb}^{-1}$), and pelletized wood was utilized as well.

Tilapia production yields averaged near 9,000-11,000 kg (10-12 tons) of fish for the entire system annually (250 m^3 ; 66,043 gallons water). Estimated production per acre is 136,000-181,400 kg (300,000-400,000 lb) annually.

Dissolved oxygen and temperature of fish culturing water were recorded twice daily (YSI 550A, YSI Inc., Yellow Springs, OH) (Figure 4.1). Water temperature exhibited an increasing trend between December 2008 and September 2009, however temperatures can be expected to drop during winter months. Culture water DO indicated a general decreasing trend during the same time frame. Culture water pH, EC, and

salinity (YSI 63 meter), TAN (1.0 to 8.0 mg·L⁻¹, LaMotte Company, Chestertown, MD), and NO₃⁻-N (Cardy meter, range 0 to 9,900 mg·L⁻¹, Spectrum Technologies, Inc., Plainfield, IL) were measured daily. Water samples were collected directly from fish tanks weekly and analyzed using ICAP and for NH₄ determination (Tables 4.1). Tilapia were fed a 32% crude protein feed from Alabama Catfish Feed Mill (Uniontown, AL), fed between 22 kg to 34 kg (50 lbs to 75 lbs) feed·tank⁻¹·day⁻¹. During the production period of December 2008 to September 2009, a general increasing trend in feed fed was observed.

Seneca Arrowhead sweet corn (*Zea mays var. rugosa* 'Seneca Arrowhead') was produced January through March 2009, at the E.W. Shell Fisheries Center, North Auburn Unit in Auburn, Alabama. Three-inch transplants were transplanted on January 27, 2009 (zero days after transplant, DAT) in a 29.3 x 9.1 m (96 x 30 ft) pad and fan greenhouse with a double layer polyethylene cover. Transplants were planted in twin rows along black plastic (Pliant Corporation, Trussville, AL) covered, 6.1 m (20 ft) rows, along north-south axis of greenhouse. This study was designed as a CRBD, consisting of two treatments replicated five times. Each replication is a plot, each plot containing sixty individual plants (Figure 4.2, 4.3). Treatments consisted of two different irrigation sources: clear water (CW) and effluent water (EW). CW irrigated corn was fertilizer with 60 mg·L⁻¹ N (120 lbs·ac⁻¹) (20-10-20 TotalGro, SDT Industries Inc., Winnsboro, LA), injected via a Dosatron fertilizer injector (Dosatron, Clearwater, FL). Irrigation was delivered via ¼" T-Tape[®] at 20 psi (0.45 gpm/100 ft, T-Systems International, San Diego, CA). Pollination was provided by physically tapping flower heads to induce pollination, and with the addition of wind from electric fans stationed in the greenhouse.

At 21 DAT corn heights were measure. Seneca Arrowhead corn is a 90-day crop, 91 DAT ears were harvested. Data collected at harvest (91 DAT) includes ear weight per plot (kg), yield per plot, and ear length (cm).

A second study was conducted at the E.W. Shell Fisheries Center, North Auburn Unit in Auburn, AL in 2009. On September 8, 2009 seedlings of Manar F1 (Beit Alpha, European seedless) cucumbers (*Cucumis sativus*, De Ruiters Seeds, Inc., Lakewood, CO) were transplanted into drip-irrigated perlite-filled, lay-flat hydroponic bags in a 29.3 x 9.1 m (96 x 30 ft) pad and fan greenhouse with a double layer polyethylene cover. Irrigation of CW with an injected hydroponic fertilizer was compared to irrigation with intensive tilapia culture EW. Beit Alpha cucumber seeds were sown into four-inch cell packs on August 17, 2009. A total of 180 seeds were sown, and were watered twice daily with clear water for 10 day after germination (DAG), followed by twelve days of watering with fish effluent.

Hydroponic bags, approximately three feet long by six inches wide, were filled with perlite and twist and zip-tie sealed (CropKing Inc., Lodi, OH). Bags were positioned length-wise along north-south axis, on greenhouse floor atop raised areas, with access troughs on either side (Figure 4.4). Treatments were irrigation with CW with an injected hydroponic-specific formulated fertilizer solution (HS) and irrigation with fish EW. Along each replication (row) were fourteen bags (seven/treatment), with five replications (rows), thus seventy lay-flat bags. A single bag served as an experimental unit, with two cucumber plants per bag.

Irrigation was designed so that each row received CW and EW, delivered by timer-controlled solenoid valves placed at the beginning of ¾" polypipe lines. Each line, CW

and EW, branched along the replications (rows) with drip emitters placed at each lay-flat bag. Each bag contained two cucumber plants, thus two emitters per bag. A single slit was made on the sidewall of each bag at a forty-five degree angle, one inch above ground, on sidewall facing trough for drainage and leachate collection.

Nutrient compositions of formulated hydroponic fertilizer can be from pre-mixed commercial fertilizers or formulated from individual ingredients (Hochmuth and Hochmuth, 2006). In this experiment treatment one fertilizer solutions were derived from a tomato special (3-13-29) soluble fertilizer (SDT Industries Inc., Winnsboro, LA). Hydroponic fertilizer nutrient solution comprised $30 \text{ mg}\cdot\text{L}^{-1}$ N delivered via injection for length of study. Fertilizer formula consisted of 3% NO_3^- -N, 13% P_2O_5 , 29% K_2O , 5.4% Mg, 0.34% Fe, 10% S, 0.05% Zn, 0.10% Mn, 0.10% Cu, 0.10% b, and 0.01% Mo (Table 4.1). Fertilizer derived from potassium nitrate, potassium sulfate, potassium phosphate, iron EDTA chelate, magnesium EDTA chelate, copper EDTA chelate, zinc EDTA chelate, sodium borate, and sodium molybdate. Calcium nitrate (15.5% N, 19.9% Ca) was supplemented to account for $70 \text{ mg}\cdot\text{L}^{-1}$ N (0 DAT-21 DAT) and $150 \text{ mg}\cdot\text{L}^{-1}$ N (21 DAT-49 DAT) (Table 4.1, 4.2). Nutrient parameters of tilapia culture water consisted of temperature and DO measured twice-daily (YSI Model 550 meter, YSI Inc. Yellow Springs, OH). Water pH and EC was measured daily using YSI Model 63 meter, total ammonium nitrogen (TAN, $0.0\text{-}8.0 \text{ mg}\cdot\text{L}^{-1}$; LaMotte Company, Chestertown, MD) and nitrate-nitrogen (NO_3^- -N, Cardy meter, $0\text{-}9999 \text{ mg}\cdot\text{L}^{-1}$; Spectrum Technologies, Inc. Plainfield Il.) were measured daily as well. Full elemental analysis, was conducted weekly using ICAP. Nutrient concentrations from fish effluent consist of $20.18 \text{ mg}\cdot\text{L}^{-1}$ TAN, $16.50 \text{ mg}\cdot\text{L}^{-1}$ NH_4^+ -N, $32.84 \text{ mg}\cdot\text{L}^{-1}$ NO_3^- -N, $20.00 \text{ mg}\cdot\text{L}^{-1}$ P, $50.00 \text{ mg}\cdot\text{L}^{-1}$ K,

45.20 mg·L⁻¹ Ca, 56.80 mg·L⁻¹ Mg, 0.80 mg·L⁻¹ Fe, 0.20 mg·L⁻¹ Zn, 0.40 mg·L⁻¹ Mn, 0.20 mg·L⁻¹ Cu, 0.10 mg·L⁻¹ B, and 0.10 mg·L⁻¹ Mo, 5.01 pH and 0.65 EC (mS·cm⁻¹) (Table 4.1).

All units received uniform watering throughout study. Irrigation was scheduled to meet increasing water demand with increase in plant biomass, as well as increased water demand when fruiting began. At transplantation into lay-flat hydroponic bags, plants received two waterings·day⁻¹ of 20 ml at seven am and four pm to 14 DAT From 15 DAT to 30 DAT, cucumbers received three waterings·day⁻¹ of 20 ml at seven am, twelve pm, and four pm. From 31 DAT to 49 DAT, cucumber were irrigated at the same rate at seven am, ten am, one pm, and four pm. Pressure on both clear water and fish effluent lines was regulated at 20 psi. Eight plastic two-cup containers were placed throughout greenhouse with spare emitters to allow for spot-checking of uniform flow rate between water sources, as well as checking for clogged emitters. (Hochmuth and Hochmuth, 2009).

Cucumbers were trellised from floor to ceiling to floor as growth permitted. Prior to initiation of study, plastic string was secured to greenhouse ceiling cable (8ft high) and staked into floor next to hydroponic bags, repeated for each individual plant. As plant growth required, plastics clips were used to trellis plants to plastic strings (Figure 4.4) (Hochmuth et al, 2003; Shaw and Cantliffe, 2003). Once growth reached cable, plants were trained for six inches, and then allowed to grow to floor. Lateral growth was removed up to the eighth node, while remaining lateral growth was allowed to grow to second node and pruned (Shaw and Cantliffe, 2003). Fruit was harvested beginning 7 September 2009 through 27 October 2009. Biet Alpha type cucumbers are parthenocarpic, so measures were taken to avoid pollination of flowers. Based on

previous research, cucumbers were graded into three categories: No.1: Fruit of 1½-inch diameter or smaller, good shape; No.2: Diameter over 1½-inch, semi-curved shape; and Cull: generally poor (misshapen, obvious defects), recording number of fruit per rating per treatment (Hochmuth et al. 2003). Plant heights were measured weekly at 15, 22, 29, 36, and 43 DAT. Total harvest yield, weight (kg), and fruit length were determined from harvests at 29, 31, 34, 36, 41, 43, 47, and 49 DAT.

Results and Discussion

Study One

Production of sweet corn early in the season under protection of a greenhouse, comparing soluble fertilizer input with EW resulted in the production of a similar crop across both treatments. Heights of corn, by treatments were similar for CW (17.83 cm) and soluble fertilizer and for EW (17.73 cm) treatments (Table 4.3). Total weight of ears of corn per treatment was similar between CW and soluble fertilizer (8.93 kg) and EW (9.87 kg) treatments. Average number of fruit per plot was similar between treatments, averaging 39.4 ears for plant irrigated with CW and soluble fertilizer and 42.8 ears for plant receiving EW. Length of ears was recorded, and no differences were observed between treatments, CW ears averaged 27.19 cm and EW ears averaged 26.22 cm (Table 4.3).

Results of sweetcorn study indicate EW meets nutritional demand for production of corn for a substantial harvest. Greenhouse production of a short, 90-day corn crop is a promising endeavor, potentially increasing profits for sales of early season produce. However, research should be conducted on a larger scale to further assess functionality

and profitability of greenhouse produced corn. Early or late season production of sweetcorn may be a profitable endeavor, if direct marketed to special niche markets, sold at a premium price (\$6/dozen ears), and as a portion of an overall production system sold at special niche markets.

Study Two

Height measurements of Manar F1 cucumbers produced via hydroponics in a greenhouse indicate initial differences between treatments at 15 DAT (HS: 53.53 cm; EW: 36.85 cm) and 22 DAT (HS: 84.73 cm; EW: 57.21 cm); however plant heights were similar at 29, 36, 43, and 56 DAT (HS: 151.82 cm; EW: 145.64 cm) (Table 4.4) (Figure 4.5).

Fruit harvest began at 29 DAT. Harvests were subsequently conducted at 31, 34, 36, 41, 43, 47, 49, and 52 DAT. No differences in fruit yield·bag⁻¹ were observed from 29 to 47 DAT (HS: 11.68 cucumber·bag⁻¹; EW: 11.36 cucumber·bag⁻¹); however differences were observed from 29 to 52 DAT (HS: 26.80 cucumber·bag⁻¹; EW: 15.54 cucumber·bag⁻¹) (Table 4.5) (Figure 4.6).

Following the trend of fruit harvest, fruit weight per bag exhibited no difference from 29 to 47 DAT (HS: 1.39 kg ; EW: 1.54 kg); however from 29 to 52 DAT differences were observed (HS: 2.74 kg; EW: 1.71 kg) (Table 4.6) Fruit length remained similar for duration of the study (HS: 14.73 cm; EW: 14.02 cm) (Table 4.6). Fruit quality ratings indicated differences between treatments (Table 4.7).

Initial results indicate aquaponically produced cucumbers perform similarly to cucumbers produced hydroponically using specialty formulated hydroponic fertilizers. Due to a higher rate of N in the hydroponic special fertilizer, vegetative growth of plants

receiving the hydroponic fertilizer was greater than vegetative growth of plants receiving EW. However, plants of both treatments exhibited similar heights and growth rate from 29 to 43 DAT, after which plants of the HS treatment again exceeded growth rate of EW irrigated plants. Fruit production was similar in all regards (yield, length, weight) across plants of both treatments after seven weeks of production. Following after seven weeks, plants receiving EW were outperformed in all regards by plants receiving HS treatment.

Conclusions indicate that EW provides insufficient nutrient levels for cucumber production compared to plants irrigated with the HS treatment. In this experiment, EW was sufficient for producing a similar yield to plants receiving HS treatment until 47 DAT. After 47 DAT, plants irrigated with EW produced minimal yields, whereas plants receiving HS produced fruit at a much greater rate. Calcium deficiencies were observed in plants receiving EW. Calcium chloride was supplemented to the fish culturing system to alleviate CA deficiencies.

Further research is needed to determine if increased feeding rates to fish culture, variations in irrigation scheduling, or other environmental factors may increase suitability of EW for production of hydroponic cucumber production.

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Table 4.1. Nutrient analysis of irrigation inputs for greenhouse grown *Zea mays* var. *rugosa* 'Seneca Arrowhead' and *Cucumis sativus*.

Nutrient	Hydroponic solution ^z	Calcium nitrate ^y	Effluent water ^x		
			Jan-Mar, 2009	Aug-Sep, 2009	Sep-Oct, 2009
TAN ^w	. ^u	.	.	16.43	19.54
NH ₄ -H	.	.	19.0	16.00	16.50
NO ₃ ⁻ -N ^v	3	15.5	47.0	38.88	35.48
P	13	.	24.0	19.60	20.00
K	29	.	40.0	42.10	50.00
Ca	.	19.9	25.0	36.90	45.20
Mg	5.4	.	11.0	52.10	56.80
S	10
Fe	0.34	.	<0.1	0.50	0.80
Zn	0.05	.	0.2	0.20	0.20
Mn	0.10	.	0.3	0.40	0.40
Cu	0.10	.	0.1	0.10	0.20
B	0.10	.	0.2	0.10	0.10
Mo	0.01	.	.	0.10	0.10
Na	.	.	83	109.80	63.80
pH	.	.	5.87	4.85	5.78
EC	.	.	2.23	0.94	0.58

^zHydroponic Solution: Hydroponic tomato special formulation, injected at 30 mg·L⁻¹ (3-13-29, TotalGro). for *Cucumis sativus* only (units = %).

^yCalcium-Nitrate: Injected at 70 mg·L⁻¹ for 0 DAT - 21 DAT; injected at 150 mg·L⁻¹ from 22 DAT - 49 DAT, combined with hydroponic solution for *Cucumis sativus* only (units = %).

^xEffluent water: nutrient analysis determined by ICAP (units = mg·L⁻¹).

^wTAN: Total Ammonia Nitrogen (NH₄⁺-N and NH₃-N) (0.0-8.0 mg·L⁻¹, LaMotte Company, Chestertwon, MD).

^vNO₃⁻-N: Nitrate-nitrogen (Cardy meter, 0-9999 mg·L⁻¹; Spectrum Technologies Inc., Plainfield IL).

^uNo data provided.

Table 4.2. Nutrient solution regimen at various stages of growth for hydroponically produced cucumber.^Z

Nutrient	Unit	Growth stage		
		Seeding to transplant	Transplant to 3 weeks	3 Weeks to termination
N	mg·L ⁻¹	80	100	180
P	mg·L ⁻¹	50	50	50
K	mg·L ⁻¹	120	150	200
Ca	mg·L ⁻¹	150	150	150
Mg	mg·L ⁻¹	40	40	50
S	mg·L ⁻¹	50	50	60
Fe	mg·L ⁻¹	2.8	2.8	2.8
Cu	mg·L ⁻¹	0.2	0.2	0.2
Mn	mg·L ⁻¹	0.8	0.8	0.8
Zn	mg·L ⁻¹	0.3	0.3	0.3
B	mg·L ⁻¹	0.7	0.7	0.7
Mo	mg·L ⁻¹	0.05	0.05	0.05

^ZHochmuth, R.C., L.L.L. Davis, W.L. Laughlin, E.H. Simonne, S.A. Sargent, and A. Berry. 2003. Evaluation of twelve greenhouse mini cucumber (Beit Alpha) cultivars and two growing systems during the 2002-2003 winter season in Florida. University of Florida, Institute of Food and Agricultural Sciences. North Florida Research and Education Center, Suwannee Florida. Research Report 2003-04.

^YFor duration of study hydroponic tomato special fertilizer (3-13-29, TotalGro) supplied 30 mg·L⁻¹ N.

^XCalcium-nitrate supplied 70 mg·L⁻¹ N for 0 DAT - 21 DAT, and 150 mg·L⁻¹ N from 21 DAT to termination.

Table 4.3. Evaluation of greenhouse grown sweetcorn under different irrigation sources, during January - March, 2009.

Treatment ^z	Height (cm)	Yield (kg)	Fruit·Plot ⁻¹	Length (cm)
	21 DAT ^y		91 DAT	
CW	17.83a ^x	8.93a	39.40a	27.19a
EW	17.73a	9.87a	42.80a	26.22a

^zCW: Clear water with soluble 20-10-20 fertilizer at a rate of 120 lbs/ac N; EW = Effluent water (see table 4.1 for nutrient analysis).

^yDAT = days after transplanting.

^xMeans followed by the same letter are not significantly different Tukey's Standardized Range Test ($\alpha = 0.05$, N = 30).

Table 4.4. Height measurements for greenhouse grown *Cucumis sativus*^z Septemeber - October, 2009 in perlite-filled hydroponic bags.

Treatment ^y	Height ^x					
	15 DAT ^w	22 DAT	29 DAT	36 DAT	43 DAT	56 DAT
HS	53.53a ^v	84.73a	94.77a	117.16a	137.37a	151.82a
EW	36.85b	57.21b	94.59a	116.73a	134.80a	145.64a

^z*Cucumis sativus* 'Manar F1' (Biet Alpha type).

^yTreatment: HS: Hydroponic solution at 30 mg·L⁻¹ (3-13-29, TotalGro)

(see table 4.1 for nutrient analysis of HS); EW: 100% Effluent water (see table A1 for nutrient analysis).

^xHeight: measured in cm.

^wDAT: days after transplanting.

^vMeans followed by same letter are not significantly different (Proc GLM, Tukey's Standardized

Range Test, $\alpha = 0.05$, N = 35).

Table 4.5 Effect of irrigation source on fruit yield of greenhouse grown, hydroponically produced *Cucumis sativus*.^Z

Treatment ^Y	Total number of fruit ^X	
	29 - 47 DAT ^W	29 - 52 DAT
Hydroponic Solution	11.68a	26.80a
Effluent Water	11.36a	15.54b

^Z*Cucumis sativus* 'Manar F1' (Biet Alpha type).

^YTreatments: Hydroponic solution injected at 30 mg·L⁻¹ (3-13-29, TotalGro), and calcium-nitrate injected at 70 mg·L⁻¹ for 0 DAT - 21 DAT; injected at 150 mg·L⁻¹ from 22 DAT - 56 DAT; Effluent water (see Table 4.1 for nutrient analysis).

^XTotal number of fruit: Average number of fruit per bag.

^W DAT: days after transplanting

^UMeans followed by same letter are not significantly different (Proc GLM, Tukey's Standardized Range Test, $\alpha = 0.05$, N = 35).

Table 4.6. Effect of irrigation source on fruit growth of *Cucumis sativus*^z produced hydroponically in a greenhouse.

Treatment ^y	Total yield ^x		Fruit Length ^w
	29 - 47 DAT ^v	29 - 52 DAT	
Hydroponic Solution	1.39a	2.74a	14.73a
Effluent Water	1.54a	1.71b	14.02a

^z*Cucumis sativus* 'Manar F1' (Biet Alpha type).

^yTreatments: Hydroponic solution injected at 30 mg·L⁻¹ (3-13-29, TotalGro), and calcium-nitrate injected at 70 mg·L⁻¹ for 0 DAT - 21 DAT; injected at 150 mg·L⁻¹ from 22 DAT - 56 DAT; Effluent Water (see Table 4.1 for nutrient analysis).

^xTotal yield: Average weight (kg) of fruit per bag.

^wFruit Length: Average fruit length (cm).

^vDAT: days after transplanting.

^wMeans followed by same letter are not significantly different (Proc GLM, Tukey's Standardized Range Test, $\alpha = 0.05$, N = 35).

Table 4.7 Effect of irrigation source on fruit quality of *Cucumis sativus* produced hydroponically in a greenhouse.

Rating ^z	Treatment	
	Hydroponic solution	Effluent water
Total No. 1 Wt (kg)	84.13a ^y	51.47b
Total No. 1 Fruit	804a	468b
Total No. 2 Wt (kg)	17.04a	12.04a
Total No. 2 Fruit	108a	90a
Total Cull Wt (kg)	1.14a	0.71a
Total Cull Fruit	7a	7a
Total Marketable (kg)	101.17a	63.51b
Total Marketable Fruit	921a	558b

^zRating: #1) Fruit with diameter under 3.81 cm (1.5 in), good overall shape; #2) Fruit with diameter > 3.81 cm (1.5 in), fair shape; Cull) Fruit with poor shape or obvious defects.

^yMeans followed by same letter are not significantly different (Proc GLM, Tukey's Standardized Range Test, $\alpha = 0.05$).

Figure 4.1. Intensive tilapia production feeding, water temperature and dissolved oxygen trends from December 2008 to October 2009.

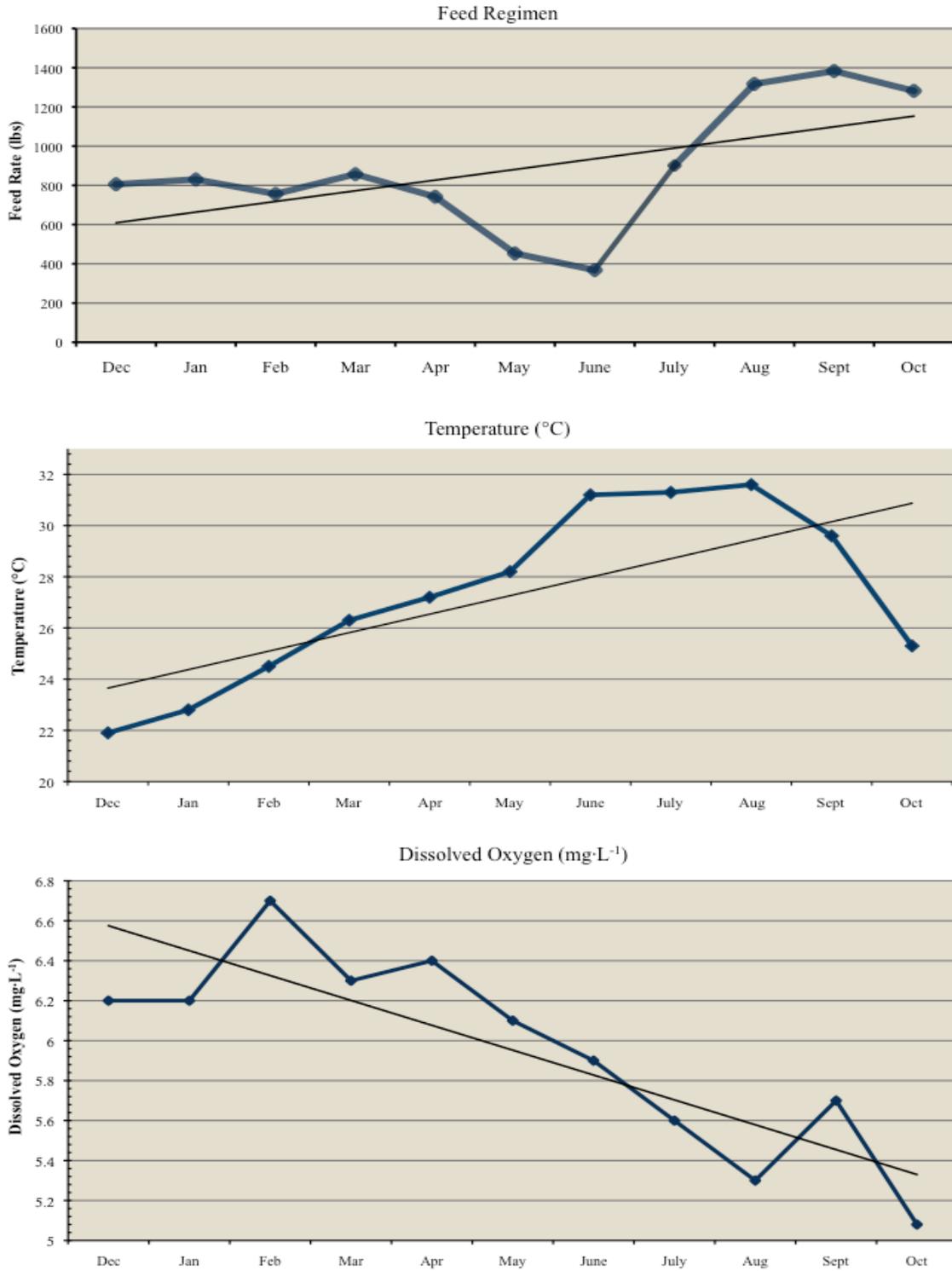
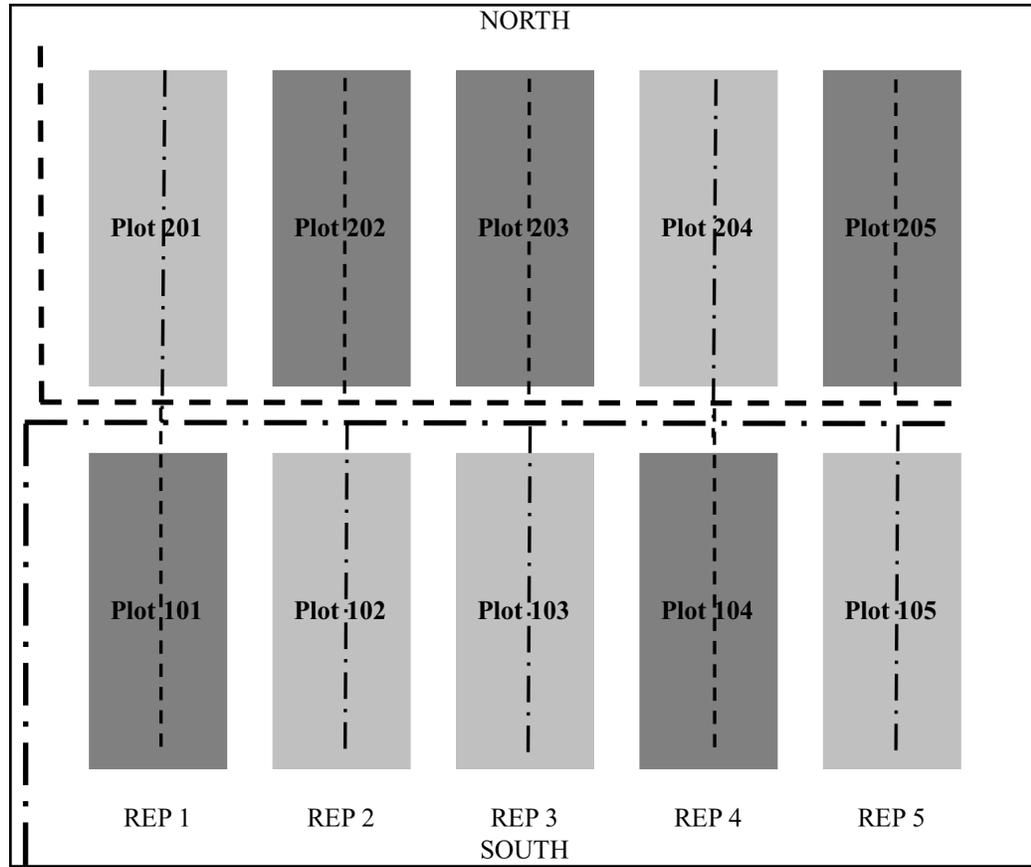


Figure 4.2. Diagram of Seneca Arrowhead sweetcorn greenhouse production plots.^Z



Plot	Treatment
	Clear Water
	Effluent Water

Notes:

6.1 m (20 ft) plot length

60 plants per plot

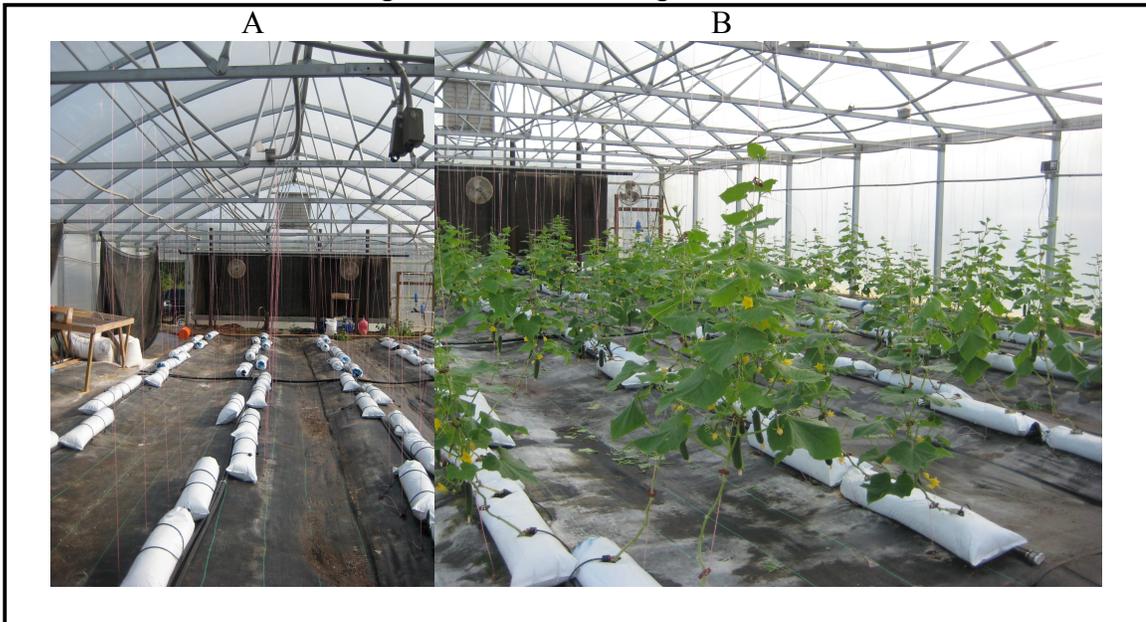
^ZCorn produced in a double layer, polyethylene-covered greenhouse during January through March 2009.

Figure 4.3. Seneca Arrowhead sweetcorn produced January through March 2009, in a greenhouse.^Z



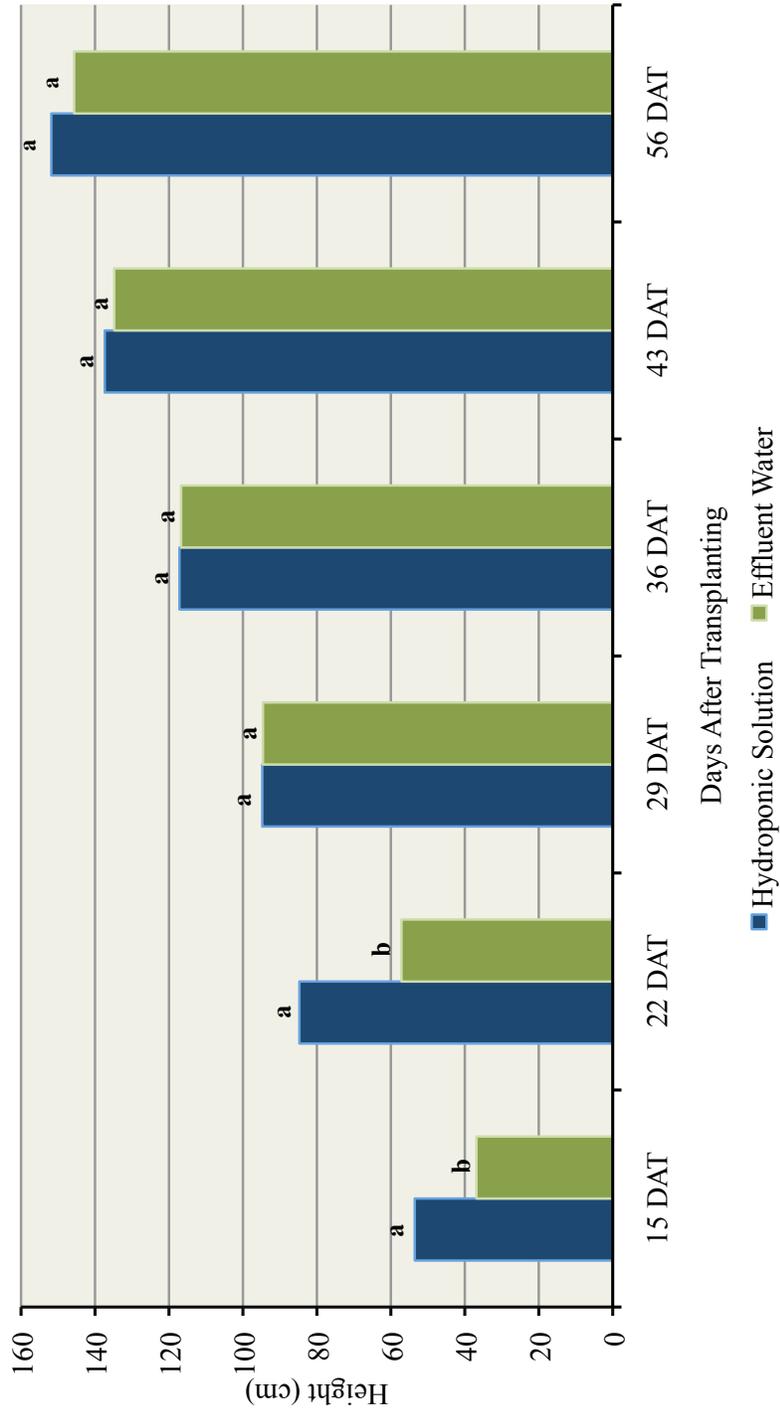
^ZCorn produced in a double layer, polyethylene-covered greenhouse during January through March 2009.

Figure 4.4. Experimental layout for hydroponically produced *Cucumis sativus*, and demonstration of cucumber growth on trellis strings.^Z



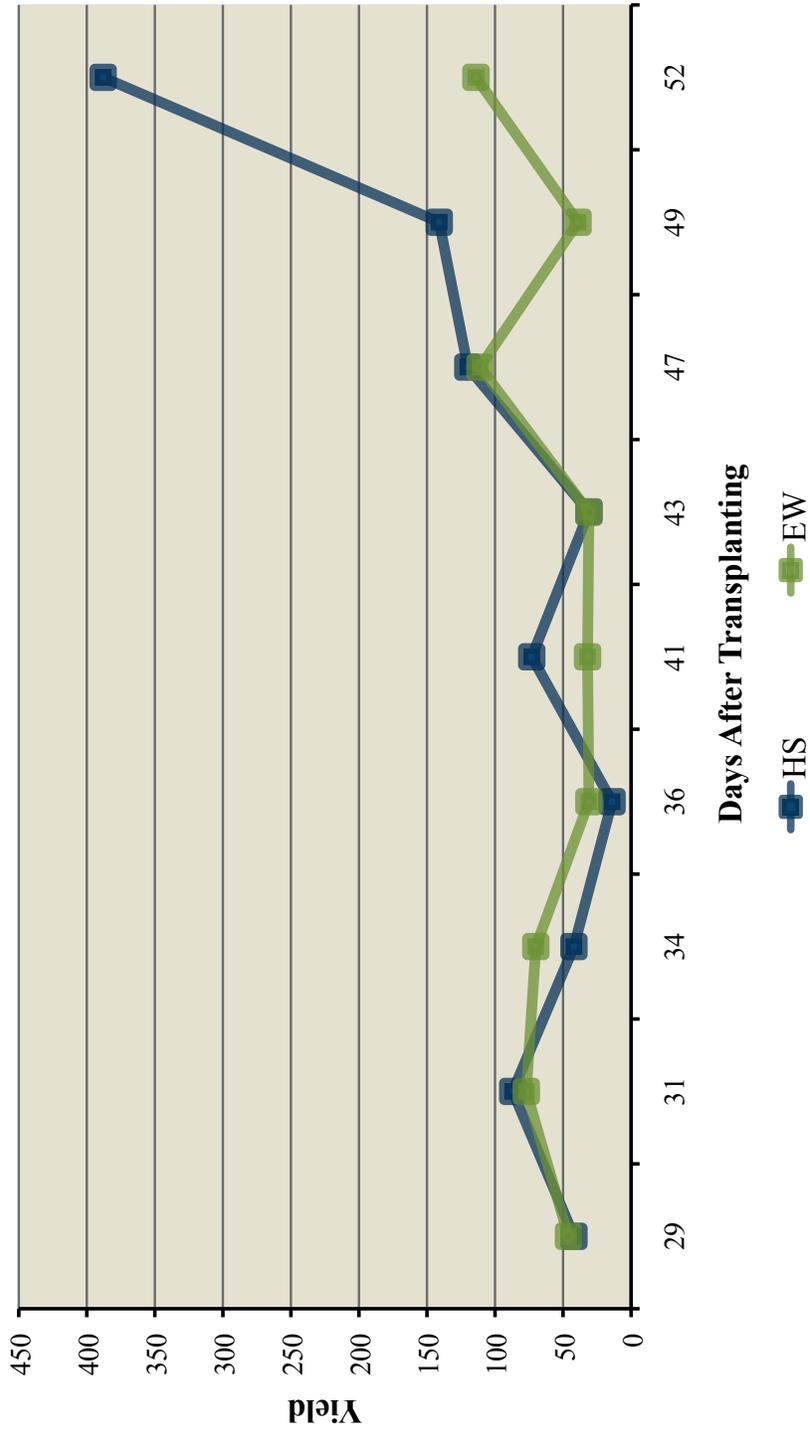
^Z *Cucumis sativus* produced in perlite filled hydroponic bags utilizing a hydroponic solution and effluent water from intensive tilapia production.
A) Peak and trough of greenhouse floor for irrigation drainage from hydroponic bags.
B) *Cucumis sativus* trellised via twine to greenhouse ceiling (8 ft).

Figure 4.5. Height of *Cucumis sativus* produced hydroponically and aquaponically in a greenhouse, August – October, 2009.^{ZY}



^Z See table A1 for nutrient analysis of effluent water used for aquaponically produced cucumbers.
^Y Columns labels with same letter are not significantly different (Proc GLM, Tukey's Standardized Range test, $\alpha = 0.05$. N = 35)

Figure 4.6. Fruit yields of *Cucumis sativus* produced hydroponically in a greenhouse.^{ZY}



^Z *Cucumis sativus* 'Manar F1' (Beit Alpha type).

^Y HS: Hydroponic solution injected at 30 mg·L⁻¹ N (3-13-19 TotalGro); EW: 100% Effluent water.

V. Final Discussion

Rising production costs are a concern in the Alabama horticulture industry due to rising fuel and fertilizer costs (NASS 1990, 2000, 2009), recent environmental instability (Ding, 2008) pressing water rights issues (Marcus and Kiebzak, 2008), and economical volatility (Ding, 2008). Further, due to pressure from environmental activists and pending government regulations, both the horticulture and intensive aquaculture industries are facing pressure to decrease water use and to control environmental loading from discharge of nutrients (Boyd, 2003).

Intensive aquaculture produces a heavy nutrient-laden wastewater (Boyd, 2004; Boyd and Tucker 1998), containing nutrients valuable for plant production (Malone et al., 1993; Savidov, 2004). Intensive aquaculture utilizes commercially produced fish feed for increased growth rate of fish populations. Commercial feed contains a known amount of nitrogen (N), allowing amount of N assimilated into the fish and the amount passed through the fish as waste to be determined. Knowing the N concentrations in wastewater is valuable, if effluent is to be utilized for horticulture crop production.

At the E.W. Shell Fisheries Center, North Auburn Unit in Auburn Alabama, effluent from intensively cultured tilapia was utilized for greenhouse crop production, during August 2008 to October 2009. Studies conducted evaluated the effectiveness of aquaculture effluent as a viable alternative to standard greenhouse crop production methods.

Bedding Plant Production

Throughout the studies, *Petunia x hybrida* was grown. Performance of petunias grown utilizing EW varied from study to study with petunias grown utilizing standard greenhouse production methods, generally performing better than those grown with EW.

Other annual bedding plant species (*Calibrachoa x hybrida*, *Catharanthus roseus*, *Nemesia fruticans*, *Verbena x hybrida*) were produced utilizing EW and standard production methods. Results indicate that adequate nutrient levels were present in the intensive tilapia production system for production of similar crops, to those grown under standard production practices.

Research is limited concerning bedding plant production and production of other ornamental crops utilizing EW. Aquaponic production has focused primarily on growing leafy plant species (lettuce, spinach, basil, mint, duckweed, water fern, and others) and vegetable crops (tomato, cucumber, peppers and others) (Nelson and Pade, 2009; Rakocy et al., 2006). Markets for bedding plants are extensive, attested to by the retail nursery industry, and the potential savings of producing bedding plants by irrigating with EW from aquaculture is large, due to replacing or supplementing traditional fertilizer inputs. Further research is needed to evaluate the production of bedding plants fertigated with EW. Ornamental species such as *Nelumbo*, *Iris*, *Canna*, *Crinum*, *Colocasia*, and others should be evaluated for production receiving EW as irrigation, as well as the production of cut flowers and foliage, such as leatherleaf fern. Research should investigate nutrient sequestering to aid in addressing environmental issues facing both industries, examine market potential and feasibility of EW produced bedding plants, and an economic analysis, determining total savings potential due to decreased fertilizer inputs.

Vegetable Crops

During research sweetcorn was produced in a greenhouse early in the season, with no differences in yield observed between plants irrigated with soluble fertilizer or EW. Production of corn in a protected environment, either early or late in the season, is a means of season extension which is a principle easily applied to many other fruit or vegetable crops. A crop of particular interest is strawberry. Strawberries are continuously in high demand and the winter production of strawberries and similar high demand crops irrigated with EW needs to be investigated. Cost reduction due to decreased fertilizer use coupled with premium prices of fresh locally grown, off-season strawberries and other crops appear have the potential for substantial revenues.

Beit Alpha ‘Manar F1’ cucumbers were produced hydroponically and aquaponically utilizing EW, and no differences were observed in yield and fruit quality, while differences were observed in plant height between plants of the two treatments. Due to the design and components used in this study, costs were substantial and unrealistic for commercial production. However, further research is needed utilizing components requiring less capital investment. Such components may consist of permanent troughs lined with a polyethylene liner, partitioned into sub-compartments, filled with a less expensive, and readily available substrate. One such substrate evaluated could be light-weight expanded clay aggregates (LECA), which can be reused with a lengthy lifespan (Pickens, 2008).

General Observations

Early in our research we noticed a layer of sludge build-up upon the substrate surface in which plants were grown when irrigated with EW directly from the tilapia

production tank. Sludge from EW has several disadvantages including attracting fungus gnats, decreasing percolation rate of water into substrate, creating anaerobic conditions in root ball leading to root rot, and potentially decreasing overall plant performance.

Trials consisting of production of *Hermocallis* spp. and *Rhondodendron* spp. revealed visually noticeable differences in plant appearance, size, and health due to sludge build-up. In native azaleas, potential rot root was observed, leading to leaf burn and necrosis (Figure 5.1). During production of bedding plants, the sludge build-up was observed, but no differences were evident between plants irrigated with settled EW as compared to unsettled EW, indicating sludge build up was not the cause of differences observed between plants produced with EW and those produced under standard methods.

During production of tomatoes and cucumbers, calcium deficiencies were observed in plants irrigated with EW. In tomatoes, the first fruit set displayed blossom end rot (BER) symptomology, and upon addition of calcium to EW located in a small tank (no fish present), the second fruit set did not exhibit BER. In cucumber production, severe calcium deficiencies were observed in leaf tissue, but did not affect fruit production. However, calcium chloride was added to fish production to aid in alleviation of calcium deficiency symptoms.

Demonstrations

Multiple, small-scale potential research projects were examined, simply as demonstrations of the many facets of aquaponics. Tomatoes were produced in 2.44 x 0.45 x 0.61 m (0.67 m³ volume) (8 x 2 x 1.5 ft) metal troughs filled with LECA. Irrigation was provided from EW pumped into a holding tank beneath troughs, continuously recirculated via ebb and flow through the use of a bell siphon (Figure 5.2). Troughs were

filled with EW and drained at ten minute intervals, pulling fresh air to plant roots as water was quickly siphoned into the holding tank. Four troughs with this design were planted with five tomato plants each. Initially, BER was detected in the first fruit set, but with addition of supplemental calcium, the deficiency was alleviated. After four weeks of fruiting, a total of 23.54 kg (96 lbs) of tomatoes were harvested from the twenty plants.

Okra was produced on a small scale, however due to no provision made for pollination vegetative growth was excellent, however fruit yield data was not taken. Greenhouse production of okra is a valuable research endeavor and merits further study.

A small foam tray of basil plugs was placed directly into the fish production tanks, with roots protected from being eaten by the fish (Figure 5.3). The basil grew exponentially in the fish tanks, eventually leaning over to be eaten by fish. Further research is needed investigating the production of herbs and aquatic plants grown directly in fish tanks. Consideration would need to be given to separating the fish from the plants' root systems.

Other potential research for consideration may be the production of crawfish utilizing the EW from fish production. The empty space provided underneath greenhouse benches holds potential for utilization by placing small tanks underneath for the production of crawfish. If this proves to be a viable means of production, utilization of space will be potentially doubled, and three different products may be produced from the resources of a single intensive tilapia production system.

Aquaponic Potential

Rakocy et al. (2006) have determined through previous research that 60 to 100 g fish feed·m⁻² plant growing area achieves optimal output in integrated aquaculture and

hydroponic systems. Research is needed to determine production rates of plants and fish in a maxed out system. The intensive tilapia and plant production system utilized during our research is estimated to be able to produce 136,000 to 181,000 kg fish·acre⁻¹ annually. However, most revenue in integrated systems is not from fish production, but rather plant production. Tomato yield from our intensive system is estimated at 9,000-10,900 kg (10-12 tons) annually, or 136-163 metric tons (mt) (150-180 tons)·ac⁻¹·year⁻¹.

Sweet corn irrigated with EW yielded an average of 10 kg ears·plot⁻¹ or 200 kg·greenhouse⁻¹. Approximately twenty-two 29.3 x 9.1 m greenhouses will fit on an acre, which would potentially provide 4400 kg (5 tons) sweet corn, if feeding approximately 27.2 kg (60 lbs) feed·day⁻¹ to tilapia stocked at 80 fish·m⁻³ water in 125 m³ tanks.

An acre of trade gallon pots generally requires 51,400 L (13,577 gal) of water a day or 9.1 million L (2.4 million gal) of water per season (assuming a 180 day growing season). If this water were to be obtained from municipal water supplies, the approximate cost of water (in Auburn AL) would be \$5,500. Fertilizer inputs would be another costs to consider. One acre can hold 174,240 trade gallon pots, which equates to 493 m³ (645 yd³) per acre. If a grower were to fertilizer at 1.36 kg (3 lbs)·ac⁻¹, with an 18-6-12 fertilizer, approximately 5000 kg (11,000 lbs) of fertilizer would be needed. If a grower purchased fertilizer in 22.67 kg (50 lbs) bags, it would cost nearly \$12,000 to fertilizer. The total cost of water and fertilizer alone is about \$17,500·acre⁻¹·season⁻¹. Integrated intensive aquaculture and horticulture production systems have the potential to alleviate or eliminate municipal water and fertilizer costs. Our system can provide 15 tilapia production greenhouses to the acre, which would provide 3,750 m³ water (990,645 gal) at any given time. This amount of water would meet the watering demands of a full scale

nursery production system, while providing dissolved nutrient via fish effluent.

Depending on the crop, the cost of water, fertilizer expenses and many other facets the cost of nursery production may increase or decrease, however it is important to note, that in the example given, there is a potential cost savings of \$17,500.

Fish species for production in intensive systems need to be tolerant of increased levels of toxic nutrients and generally poor water quality. Other species for production may include catfish species, largemouth bass, sea bass, rainbow trout, and carp species. However, as noted in literature, most common intensive aquaculture systems in the production of tilapia, due to the species high tolerance of poor water quality.

Recapturing solids removed from intensive systems have potential to be reused. Dried solids from intensive aquaculture may be a valuable resource for use as landscaping soil amendments, or for incorporation into nursery substrates for plant production. Other potential uses are sequestering methane release for energy or heating purposes for production systems in non-tropical regions.

The prospect of proximity to urban centers for marketing and selling of aquaponic produce is an issue to be addressed. Utilization of old, empty warehouses is a potential solution to locating commercial scale aquaponic systems in close proximity to cities. Fish do not respond to daylight or darkness, and therefore indoor production is not problematic. Further, old warehouses are spacious enough to hold numerous rows of stacked fish production tanks. For plant production, greenhouses or various forms of protection could be assembled outside of the warehouse. This would be a relatively cheap alternative to starting an aquaponic system from nothing, and would allow a grower to

produce fish and fresh produce within an urban center, subsequently reducing transportation costs, marketing costs, and increasing market access.

Conclusion

Research conducted indicates that EW as a means of supplementation and replacement of standard fertilizer inputs is a viable means of producing greenhouse crops. More research is needed for defining areas where further innovation is possible, for defining the limitations and potential solutions to those limitations. Developing practical, on-farm applications methods for integration are needed.

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Figure 5.1. Sludge build-up upon substrate surface of plants irrigated with EW.^Z



^ZEW: Effluent water.

A) Sludge layer immediately after irrigation application.

B) Sludge layer after drying, and necrotic leaf on plant.

Figure 5.2. Tomato demonstration, grown in LECA in an EW^Z irrigated ebb and flow recirculating culture system.



^ZEW: Effluent water

A) Five tomato plants grown in ebb and flow EW system, controlled by a bell siphon, cycled at 10 minute intervals; B) Outer wall of bell siphon; C) Standpipe inside of bell siphon, level of standpipe controls level to which water rises in tank before being siphoned out.

Figure 5.3. Basil grown directly in fish tanks, floating on a foam tray, with roots protected from tilapia fish.

