Response of Hydroponic Bibb Lettuce (*Lactuca sativa*) to Chloride Additives in Integrated Aquaculture Systems

by

William Robert Beauchamp

A thesis submitted to the Graduate Faculty of
Auburn University
in partial fulfillment of the
requirements for the Degree of
Master of Science

Auburn, Alabama
May 9, 2010

Keywords: aquaponics, lettuce, salt, NaCl, $CaCl_2$

Approved by

Jeff L. Sibley, Chair, Bohmann Professor of Horticulture
Jesse A. Chappell, Associate Professor of Fisheries and Allied Aquaculture
N. Rob Martin, Professor Emeritus, Agricultural Economics and Rural Sociology

Abstract

Agricultural managers are continually looking for ways to increase production while at the same time reduce costly inputs. One such way is an integration of two or more crops or production systems. Sustainability is a key word in our world today and one component of sustainability is the reduction of waste and the reuse of all available resources and refuse. Aquaponics is the combination of plant production and fish production whereby the byproducts of one system are used as an input for the other, thereby creating a symbiotic effect. One such system that has been used and shows promise is the combination of intensive production of tilapia (*Oreochromis spp*) and greenhouse production of lettuce (*Lactuca sativa* L.). Monoculture systems have traditionally relied on proven methods and inputs. However, in cross-cultural systems, what has proven to be "standard practice" for one production system may actually be harmful to the other production system. In intensive tank culture of fish, it is a common practice to add salt in the form of NaCl or $CaCl_2$ to the tank in order to ease stress on fish. Fish tend to have a high tolerance for salt and benefit in many ways from it. However, plants typically do not have such a tolerance. This study was conducted at facilities at Auburn University, Alabama, to determine the tolerance of hydroponic Bibb lettuce to various chloride concentrations resulting from the addition of NaCl or $CaCl_2$ to the hydroponic solution. In a 29.3 x 9.1 m (96 x 30 ft) double insulated greenhouse located at the E.W. Shell Fisheries Center, North Auburn Unit lettuce seeds (*Lactuca sativa* L. 'Charles') were sown in one inch (1") oasis blocks in July, 2010. In August, three weeks after sowing, the blocks were placed in a hole cut in 20 cm x 20 cm (8" x 8") ½" thick styrofoam squares. The squares were placed on top of 6 liter (1 gal.) buckets containing a standard hydroponic fertilizer and concentrations of NaCl or $CaCl_2$ measured in ppm chloride ranging from 0 to 500 ppm. A one (1) inch airstone was put in each pot powered by an 80 watt compressor. Salinity, measured in parts-per-thousand (ppt), electro-conductivity (EC) and pH of the treatment solutions were monitored using a YSI Model

63 meter (YSI Inc., Yellow Springs OH). Chlorophyll level in the foliage was measured with a SPAD-502 meter (Minolta, Spectrum Industries Inc.). Water loss from plant uptake and evaporation was replaced with fresh water throughout the experiment. Approximately 30 days after transplanting, plants were harvested and fresh shoot weight (FSW), fresh root weight (FRW), and a Growth Index were taken. The Growth Index was calculated by taking the average of the height and two perpendicular widths of each plant. Plant tissue was then dried in a forced-air oven for nine (9) days at 45^{o} C (113^{o} F) and dry shoot weight (DSW) and dry root weight (DRW) measurements were taken. Results from these experiment showed that Bibb lettuce (*Lactuca sativa* L. 'Charles') is not adversely affected by either NaCl or $CaCl_2$ at levels ranging from 0 to 500 ppm (0.5 ppt) chloride. A second set of experiments was begun in December 2010 with increased concentrations of NaCl or $CaCl_2$ in order to determine salt concentrations needed to observe adverse effects on Bibb lettuce. The second experiment was conducted at the Paterson Greenhouse Complex located on the main campus of Auburn University in a 14.6 x 6.1 m (48 x 20 ft) gas-heated greenhouse where the air temperature was maintained between 21^{o} and 35^{o}C (70 to 95^{o}F). Concentrations of NaCl or $CaCl_2$ were from 0 ppm to 20,000 ppm (20 ppt) chloride. Experimental setup was the same. Water quality parameters were again monitored and growth parameters taken 35 days later at harvest. Results showed that SPAD readings and pH ranges were poor indicators for measuring the effects of chloride concentrations on Bibb lettuce. Treatments of NaCl or $CaCl_2$ over 5000 ppm (5 ppt) chloride were lethal to Bibb lettuce. Significant adverse affects were seen at concentrations above 2000 ppm (2 ppt) chloride for both salts. Regression lines of the Growth Index, FSW, FRW, DSW, and DRW suggests that the decline begins prior to where differences are significant. Our research suggests that managers not view the 2000 ppm (2 ppt) chloride level as a tipping point for lettuce but as the point at which significant adverse physiological root-mediated plant responses occur. More research is needed to determine a more precise point at which physiological health and growth is significantly affected. Perhaps more importantly, additional research is needed to determine a more specific salt level at which plant health and growth begins to be adversely affected in hydroponic Bibb lettuce.

Acknowledgements

Concerning all the advice, aid, and assistance received during this project, the author extends his deepest gratitude to all who helped. Namely, I would like to first thank Dr. Jeff Sibley. His friendship, wisdom, and encouragement have been a great help and resource. His willingness to help the author finish a degree started a long time ago speaks volumes of the depth of his character. To Dr. Jesse Chappell, whose research is the catalyst for this project, thank you. What you do is making an impact on the lives of people throughout the world. To Dr. Rob Martin, who continues to inspire and lead, thank you. Part of the author's personal success in business throughout the years can be traced back to lessons you taught me long ago. To Jeremy Pickens for the ideas, help, and assistance in not only putting the experiments together but also in the analysis of the data, the author extends special thanks. To my wife, Robbi, I have always loved you. To my children, Bobby, Tara, and Andee: you continue to bring me the greatest happiness. And finally, I wish to thank Jesus Christ, *King of Kings and Lord of Lords, The Rider on the White Horse*, my Master, my Lord, and my God.

Table of Contents

Abstract ii

Acknowledgments iv

List of Tables vi

List of Figures vii

Introduction 1

Literature Review 6

Materials and Methods 18

Results and Discussion 20

Final Discussion 26

Literature Cited 29

Appendix 38

List of Tables

Table 1: Treatments used in NaCl or $CaCl_2$ evaluation on hydroponic Bibb lettuce. Experiment 1, August/September, 2010. 38

Table 2: Treatments used in NaCl or $CaCl_2$ evaluation on hydroponic Bibb lettuce. Experiment 2, December 2010/January 2011..... 39

Table 3: Final results from $CaCl_2$ treatments of hydroponic lettuce (*Lactuca sativa* L. 'Charles') from Experiment 1, August/September 2010. 40

Table 4: Final results from NaCl treatments of hydroponic lettuce (*Lactuca sativa* L. 'Charles') from Experiment 1, August/September 2010. 41

Table 5: Final results from $CaCl_2$ treatments of hydroponic lettuce (*Lactuca sativa* L. 'Charles') from Experiment 2, December2010/January 2011..... 42

Table 6: Final results from NaCl treatments of hydroponic lettuce (*Lactuca sativa* L. 'Charles') from Experiment 2, December2010/January 2011..... 43

List of Figures

Figure 1: Plot of EC over chloride concentration for $CaCl_2$ treatments in lettuce experiments conducted in December 2010/January 2011. 44

Figure 2: Plot of Growth Index over chloride concentration for $CaCl_2$ treatments in lettuce experiments conducted in December 2010/January 2011. 45

Figure 3: Plants from $CaCl_2$ treatments in lettuce experiments conducted in December 2010/January 2011. Concentrations of chloride shown are in ppm. First photo is from above. The second photo is the same plants shot at an angle. 46

Figure 4: Plot of EC over chloride concentration for NaCl treatments in lettuce experiments conducted in December 2010/January 2011. 47

Figure 5: Plot of Growth Index over chloride concentration for NaCl treatments in lettuce experiments conducted in December 2010/January 2011. 48

Figure 6: Plants from NaCl treatments in lettuce experiments conducted in December 2010/January 2011. Concentrations of chloride shown are in ppm. First photo is from above. The second photo is the same plants shot at an angle. 49

Figure 7: Plot of Fresh Shoot Weight over chloride concentration for NaCl treatments in lettuce experiments conducted in December 2010/January 2011. 50

Figure 8: Plot of Fresh Root Weight over chloride concentration for NaCl treatments in lettuce experiments conducted in December 2010/January 2011. 51

Figure 9: Plot of Dry Shoot Weight over chloride concentration for NaCl treatments in lettuce experiments conducted in December 2010/January 2011. 52

Figure 10: Plot of Dry Root Weight over chloride concentration for NaCl treatments in lettuce experiments conducted in December 2010/January 2011. 53

INTRODUCTION

With the ever-increasing earth population and the resulting decreasing supply of natural resources, two of the key phrases used in today's culture are "Sustainability" and "Go Green". The overriding concepts of these two phrases are three-fold. The first concept is an effort on the part of the manager or producer to optimize the use of any resources that are used in producing a product, which encompasses using as small an amount of an input as needed while at the same time making as big an impact as possible. The second concept follows directly behind the first which is that the first objective is usually best achieved by concentrating the inputs or production area into as small a physical footprint as possible. Intense, highly concentrated inputs are applied only in the area where that input is directly needed. An example in the field of horticulture would be irrigation. A grower can water an entire field of crops using sprinkler or overhead irrigation or put water directly at individual plants using drip irrigation. Many variables, such as equipment costs and water availability determine levels of intensity. The third concept in "Sustainability" and the "Go Green" movement is the need to determine ways to use, sell, or recycle any byproducts of the production cycle itself. Managers need to explore and know ways that leftovers can be sold, recycled, or even used by another system.

According to the Food and Agricultural Organization of the United Nations (FAO, 2007), total world fishery production in 2004 was 142 metric tons (156 million tons) produced in three basic systems. The largest system by far is the harvest of natural fish populations. Of the 142 metric tons, approximately 94 metric tons comes from the harvest of natural freshwater and oceanic stocks (FAO, 2007). Even though harvesting natural resources is the largest source of production, the per unit area production on a weight of fish per hectare basis is very low, 3.08 kilograms per surface hectare of ocean water (2.75 pounds per surface acre). The second source of fish production comes from the science of aquaculture. Aquaculture is defined as the culture of aquatic organisms

under controlled or semi-controlled conditions (Stickney, 1996). Aquaculture, in its conventional sense, is the science of building production ponds, stocking a large number of a single fish species, and then feeding the fish until they reach a harvestable size. Ponds are either drained and fish caught in catch basins, or the fish are harvested by seining. Aquaculture is much more intense than natural systems with production levels typically running in the 8000 to 15,000 kilogram of fish per hectare (7137 to 13,383 lbs/acre) basis or 2500 to 5000 times that of natural systems (Popma and Lovshin, 1996). A third system that is being used to produce fish is an even more intense level, and that is rearing fish in an artificial tank system where the fish are stocked at extremely high densities and the manager controls the input of all variables into the system. Production on a per hectare basis of tank culture systems can be in excess of 100,000 kilograms per hectare (89,218 lbs/acre) or 33,000 times the ability of natural biological systems (Popma and Lovshin, 1996).

Natural systems tend to be stable and, in many cases, seemingly self-sustaining. However, with the increase in demand for fish protein, natural systems have, over the last 50 years, been subject to overfishing when demand has exceeded supply (University of Michigan, 2006). Pond aquaculture systems can supplement the supply of fresh fish; however, with pond culture, capital requirements, land footprint, water availability, labor requirements for maintenance, and harvest are issues that become a concern. For tank culture systems, if the manager controls all inputs, then high levels of maintenance and monitoring become important and the culturist must stay on top of the entire process and literally control it all. In tank culture, crowding, disease, infections, and water quality become a major concern.

For human food plant culture (vegetables, grains, etc.), three levels of production also exist. Natural plant systems, even at their highest level of production, simply do not produce sufficient amounts of harvestable food per hectare to sustain large human populations. A second system, and the one responsible for the majority of the world food supply, is in field cultivation of food crops. Worldwide, production levels range from several hundred to 60,000 kilograms per hectare (53,531 lbs/acre). The third system of intensity is greenhouse production of food. Greenhouse production encompasses total control of vegetable and food production. Greenhouse production has been on an

increase for the last 30 years with levels of greenhouse production as high as 650,000 kilograms per hectare (579,916 lbs/acre) (Heyden, 2009).

For plant culture, there are pluses and minuses of the three levels of production. It goes without saying that natural production will not supply the demand for food crops worldwide. For field culture, weather affects production and harvest. Insects and pest control, as well as labor for harvesting, are also major concerns. Many areas of the world have soils not suitable for food production or have an insufficient water supply. In greenhouse production, monitoring, maintaining, and supplying all the basic nutrients and water needed for healthy plants is of upmost importance.

In intense integration production systems, the first two concepts of "Sustainability" and "Go Green" are accomplished. Smaller amounts of "purchased" inputs are needed because some of the necessary inputs may be provided as byproducts of another production system. Maximum profit is accomplished when there is optimal use of resources. In intense agricultural production systems, the plants or animals being grown are typically crowded together or confined in high densities to a small footprint or production area. The third concept, recycling or reuse of byproducts becomes a chief concern.

Much work has been done in studying the integration of many intense culture systems over the last few years. One such integration is the cross between raising fish in an intensely managed tank culture while raising plants in a nearby greenhouse system.

In intense fish culture, byproducts include large percentages of the food and other nutrients supplied to the fish which are not converted into fish flesh. On the average, 1.8 to 2.0 kilograms of feed is required to produce one kilogram of fish flesh (Popma and Lovshin, 1996). Additional byproducts of intense fish production are in the form of feces, urine, and ammonia. Fish naturally excrete large amounts of ammonia from their bodies through their gills. For most fish, high amounts of nitrogen-based byproducts such as ammonia are toxic and can result in poor growth, disease, and death. Brown-blood disease, methemoglobin anemia, is a lethal fish disease caused by high levels of nitrites. High ammonia, rather than low dissolved oxygen, is often the most limiting water quality constraint in intense fish culture (Popma and Lovshin, 1996). In intense culture, excess nutrients have to be flushed from the system or remain in the tank until

broken down by bacteria. Flushing requires water. Supplies of fresh water are not always available and, in the United States, many aquifers and water resources are now being regulated and restricted by government agencies. If left in the tank, nutrient byproducts are eventually broken down by bacteria. *Nitrosomonas* and *Nitrosospira* bacteria take fish production byproducts and break them down into nitrite (NO_2^-) (Francis-Floyd et al., 2009). Nitrites are readily absorbed by fish and are toxic to most fish. If left in this condition, water quality conditions quickly deteriorate to toxic levels. Other bacteria, *Nitrobacter* and *Nitrospira*, take nitrites and break them into nitrate (NO_3). Nitrates are not as toxic as nitrites to fish (typically 10 times less).

Plants need nitrogen. Of the major plant nutrients, nitrogen (N) is typically the element of greatest demand by plants and is necessary for almost every biological function. In plants, nitrate is often a preferential source of N for crop growth (Mengel and Kirkby, 1979).

Much research has demonstrated how water from an intense fish tank system can be filtered to remove fecal and other solids, and redirected through an adjacent greenhouse containing vegetables or other plants (Holliman, 2006; Rahman, 2010; Rakocy, 2002; Sleeper, 2009). After plants filter out many of the nitrates, the water can be recycled back into the fish production tanks. The plants benefit from byproducts of the fish and the fish benefit from filtered water via the plants, a symbiotic relationship.

One such fish-plant production system that has been shown to work successfully is integration of the fish culture of tilapia (*Oreochromis spp*) and the greenhouse plant culture of lettuce (*Lactuca sativa*) (Diver 2006; Rahman, 2010; Rakocy, et al., 1992). Just as there may be benefits of combining fish culture and plant production, there are problems as well.

Because of the intense nature of the fish systems, it is not uncommon for fish to become stressed in some way. Stress can arise from many different causes such as diseases, parasites, bacteria, crowding, or poor water quality. In fish culture, managers often add salt to a tank of fish in the form of NaCl or $CaCl_2$ at the first sign of stress. As a general rule in fish culture, the common "cure-all" is to add salt to distressed fish.

Salt provides many benefits for fish. Brown-blood disease is caused when excess ammonia, primarily from fish respiration, builds up in fish tanks. In fish, the buildup of

ammonia causes a condition called methemoglobin anemia in which the ammonia prevents the absorption of oxygen needed for respiration by fish, quite often resulting in death. Salts help prevent this from occurring. Also, when stressed, fish naturally excrete high levels of ions from their bodies into the water. Additions of salts to the water provide a concentration gradient against which the flow of ions is reduced or stopped. Salts also resupply ions and results in an almost immediate "calming down" of the fish. Fish parasites do not do well in saline water. The salts irritate them and, in many cases, cause them to fall off their host and/or die. Bacteria, as well, do not do well in saline water, and as a result, many bacterial infections clear up with the addition of salts. The commonly used treatment for stress, brown-blood disease, parasites, and bacterial infections is the addition of salt to the water in the form of NaCl or $CaCl_2$ (Brunson, 2009).

Currently, there is a lack of information regarding response of hydroponic Bibb lettuce to varying salt levels, NaCl or $CaCl_2$, present in effluent from intensive tilapia production systems. This is in contrast to the abundance of soil salinization studies in arid areas of the western United States, Australia, and the Middle East where salts have built up over time from high rates of evaporation, heavy fertilization and irrigation, or the use of brackish waters as a water source. The purpose of this study was to determine threshold concentrations of NaCl or $CaCl_2$, measured in parts-per-million (ppm) chloride, to a crop of hydroponic lettuce before seeing a decline in growth.

LITERATURE REVIEW

Aquaculture

Aquaculture is defined as the agricultural science of rearing aquatic life forms, be they plant or animal (Sleeper, 2009). The majority of aquaculture crops are fish and/or shellfish. Aquaculture is big business. In Alabama alone, in 2006, aquaculture provided producers with a farm-gate income value of $105 million dollars (Crews and Chappell, 2007).

Different aquaculture systems are segregated based on their levels of intensity, ranging from extensive systems where human input is minimal, followed by semi-intensive, and then intensive systems where man controls both water quality and available food resources (Tave, 1986). Intensive aquaculture has been defined as a production system of cultivating fish or aquatic life species by way of substantial human input and management practices (Rice, 2008).

Within the aquaculture industry, production systems can be divided into open and closed systems. Open systems take water in from one point and discharge it at another, thereby using the water one time. Closed systems involve reusing the same water and only adding new or fresh water as water is lost. Closed systems often have a series of filtration and processing components that help maintain water quality appropriate for fish (Berghage, et al., 1999). One of the key issues is the management of effluents produced by either system. Effluents are of a concern because they are sometimes discharged into rivers and creeks which, in turn, can lead to environmental issues. Effluents are also a concern because they represent unused value. Feed conversion rates for tilapia in intense systems are typically 1.8 to 2.0 grams of feed for every gram of fish flesh assimilated (Popma and Lovshin, 1996). This equates to a "waste" of 40% to 50% of the feed value in the form of unused nutrients. Extracting some or all of this value would be a significant recovery from the environment. Currently the industry is developing guidelines and technological innovations to tackle environmental issues by implementing

practices to lessen pollution loads and to make use of aquacultural waste in an economical and beneficial way (EPA, 1980). Under the National Pollutant Discharge Elimination System (NPDES) permitting process, regulations place limitations on the discharge of animal wastes. In order to reduce runoff of effluents into navigable waters, minimum standards and a nutrient management plan are required before a permit is granted.

Tank Culture

Tank culture of fish is typically intense and can result in production levels of 100 to 600 metric tons per hectare (89,218 to 535,307 pounds per acre) (Popma and Lovshin, 1996). Tank culture systems can be more efficient than earthen pond culture, in part, because of their lower requirements of land, labor, and water resources (Rakocy, 1989; Rakocy and McGinty, 1989). Commercial tank culture of Nile tilapia (*Oreochromis niloticus*) or other species offers several advantages to other types of culture. Dense stocking rates disrupt breeding which allows both sexes to reach a more marketable size in a shorter period of time due to lack of energy expenditure on reproduction (Popma and Lovshin, 1996). Such systems can be managed to a high degree to achieve optimum production levels (Shaw and Cantliffe, 2003). Indoor tank culture, in thermally controlled systems, also allows many species of fish to be grown and produced in geographical ranges where they would not normally survive. For example, tropical fish, endemic to South America are grown successfully in tanks in the very northern regions of North America. Tilapia are cultured commercially in Wisconsin and the Dakotas, both of which are well out of the normal temperature ranges where tilapia can survive in surface pond systems.

Fish Feeds

Nitrogen (N) content of feed is important. Nitrogen levels in feed are reflected in crude protein where % Crude Protein = 6.25 X %N. Recommended crude protein levels in food varies between fish species; however, for tilapia, the protein range is generally as low as 26% crude protein to 38% crude protein in modern production diets (Hargreaves and Tucker, 2004; Popma and Masser, 1999), which corresponds to four (4%) to six (6%)

percent N in each kilogram of food. Of the N supplied in fish feed, approximately 20% to 30% is recovered in the fish flesh (Auburn University, 2004), with the remainder ultimately entering the culture system as ammonia nitrogen (NH_4) (Graber and Junge, 2008). For example, 1 kg of 32% crude protein feed would produce 40 g NH_4/kg feed.

The effluents from intensive systems tend to be high in nutrients and organic matter. Discharge of nutrient-rich effluents into natural bodies of water can cause eutrophication or excessive plankton and algae growth (Boyd and Tucker, 1998). Excess nutrients in effluent from fish production is not only potentially toxic to production systems, but is drawing the attention of environmental activist groups and governmental agencies (Dierberg and Kiattisimukul, 1996; Goldburg and Triplett, 1997; Naylor et al., 2000; Palada et al., 1999) and many countries have established water quality regulations to protect against high organic discharges. In the United States, effluent laden discharges fall under the Clean Water Act and require a NPDES permit.

Biological filtration is one of the most important elements by which water treatment is accomplished in many aquaculture production systems. In intense tank culture, the N cycle is the primary filtration method used to breakdown ammonia. In the N cycle, ammonia is changed to nitrite by *Nitrosospira* and *Nitrosomonas* bacteria. Other bacteria, *Nitrospira* and *Nitrobacter* convert the nitrite into nitrate (Timmons and Losordo, 1994).

Horticulture and Hydroponics

Horticulture is defined as that branch of agriculture concerned with intensively cultivating plants that are used by people for food, for medicinal purposes, and for aesthetic gratification (USDA, 2011a). Production methods used in horticulture include field production, greenhouse production, outdoor container-plant production as well as others. Olericulture is the study of horticulture as it relates to the production of vegetables for human consumption. The US Department of Agriculture valued the 2010 U.S. vegetable crop at $11.2 billion dollars (USDA, 2011b). With an estimated 1000 acres in hydroponic production in the United States, this equates to an estimated value of $6.6 million dollars of vegetables grown hydroponically.

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). Hydroponics has developed by determining what substrates work when combined with the appropriate nutrient solution for a particular plant (Resh, 2004). Widespread commercial use of hydroponics began over the past 60 years in Europe, Australia, and Israel beginning in the 1950's (Resh, 2004). Greenhouses are often used with hydroponic systems for controlling environmental conditions (light, temperature, and water usage) (Jensen, 1997).

Hydroponics provides some clear advantages over traditional field-grown production systems (Alleman, 1985). Some of these are:

- Reduced water waste.
- Increased water-use efficiency.
- Standardization of culture.
- Sterile root-zone substrates.
- Complete control of nutrients.
- Spacing of plants is only limited by light availability.
- Expanded growing seasons (multi-cropping) and locations.
- Higher production yields.
- Weeds and soil diseases can be reduced or eliminated.
- Potentially reduced insect and pest problems.
- Water stress can be eliminated.

In reference to production output, hydroponics has been shown to increase total production significantly in some crops. Field-grown lettuce will produce an average yield ranging between 20,000 kg/ha and 30,000 kg/ha (17,843 lbs/ac to 26,765 lbs/ac); on the other hand, top growing hydroponics facilities can produce yields that are over 10 times larger than that of field production (Resh, 2004). For tomatoes, hydroponic production can be up to 30 times greater than field production (Sleeper, 2009). For cucumbers, field production is approximately 10,000 kg/ha (8922 lbs/ac) versus hydroponic production of 200,000 kg/ac (178,436 lbs/ac), a factor twenty (20) times

larger (Sleeper, 2009). Australian Hydroponic & Greenhouse Association (AHGA) produced 585,000 kg/ha of tomatoes hydroponically as compared to 69,231 kg/ha of tomatoes grown in the field (Smith, 2007). Wolverton (1987) reported increased wheat production of 143% per acre when grown hydroponically versus in the field. He further demonstrated that potatoes, cabbage, tomatoes, and lettuce production was 87%, 138%, 54%, and 233% higher, respectively, when grown hydroponically. Resh (2004) stated that, under the same greenhouse conditions, where the only difference was soil versus soilless media, the increased production of tomatoes hydroponically was typically 20 to 25 percent, which was still a 400% to 1000% increase over conventional field-grown conditions due to several possible factors such as lack of nutrients in field soil, possible poor soil structure, and the presence of pest or disease in the soil.

Hydroponic culture is possibly the most intensive method of crop production in today's agricultural industry (Hamdy et al., 2009). The future of hydroponics, as it continues to grow, will depend greatly on the development of production systems that are competitive in terms of costs with open field agriculture (Hamdy 1993, 1996). Control of variables such as the nutrient content of the media, pest management, temperature fluctuations, as well as water quality and quantity, will allow agricultural managers to direct specific inputs in specific amounts, when and where needed, thus resulting in reduced costs and increased production.

Tilapia

Tilapia (*Oreochromis spp*) belong to the Cichlidae family and are endemic to Africa, but interest in their aquacultural potential has led to nearly worldwide distribution within the past fifty years. Most tilapia species are herbivorous, readily reproduce in small ponds, and are highly tolerant of poor water quality. For commercial production, the Nile tilapia has been the preferred species (Popma and Lovshin, 1996; Watanabe et al., 2002). Tilapia are more tolerant than most commonly cultured fish to salinity, high water temperature, low dissolved oxygen, and high ammonia concentrations. Concerning salt tolerance, most tilapia species used in commercial culture are freshwater species, but all are euryhaline (tolerant to brackish water). Some tilapia species, *Oreochromis mossambicus* and *O. spilurus*, can grow and even reproduce in seawater. Quite often,

tilapia producers will add salt to their tanks to provide a number of benefits to their fish (Brunson, 2009). A common problem with intense culture of tilapia is the buildup of high nitrite levels due to the breakdown of ammonia nitrogen from fish respiration, fecal matter, and unused feed. High levels of nitrite can result in methemoglobin anemia, brown-blood disease, which is toxic to tilapia. The chloride portion of the salt competes with nitrite and helps prevent the nitrite from being absorption through the gills. Any form of fish stress can causes tilapia to excrete ions from their bodies. High salt content in water creates an ionic gradient that prevents tilapia from losing these ions. Inability of tilapia to tolerate low temperatures is a serious constraint for traditional commercial culture in temperate regions. The lethal low temperature for most tilapia species is 10° or 11°C (50^{o} to 52^{o} F), so the majority of the tilapia grown in the United States and Europe require some form of winter thermal protection (Popma and Lovshin, 1996). Because of their ability to withstand water quality extremes, tilapia have become an excellent fish species to be raised in intense cultures. In intensive tank culture, it is possible to produce tilapia at a level as high as 600,000 kg/hectare/crop (535,307 lbs/acre/crop) (Popma and Lovshin, 1996). Large-scale commercial tilapia production is almost exclusively done with "all-male" fish (>95% male). Recently hatched fry are fed a 3 to 4 week diet containing the male steroid, 17-a-methyltestosterone, that makes genetic females develop testicular tissue, producing fish that function and grow as males. Percentage of phenotypic males after treatment usually exceeds 95% (Popma and Lovshin, 1996).

Salinity and Lettuce

Afzal et al., 2006; Allakhverdiev et al., 2000; Madidi et al., 2004; Munns, 2002; Munns and Tester, 2008; and Parida and Das, 2005 all stated that salinity is the most serious threat to agriculture today and the major environmental factor that limits crop production and growth. Soil salinization, salinity stress, and plant responses to high salinity have been discussed for over four decades (Ehret and Plant, 1999; Flowers et al., 1977; Greenway and Munns, 1980; Hasegawa et al., 2000; Zhu, 2002). Salt has been shown to affect plants in a variety of ways. For the vast majority of terrestrial plants, the electrolytes sodium (a cation) and chloride (an anion) are extremely toxic to most plants at relatively low concentrations due to the adverse effect both have on cellular

metabolism, as well as cell structure. Only a few plants, called halophytes, particularly those found along the coast near bodies of saltwater and inland within high evaporative areas, saline lakes, or desert regions have adapted to tolerate high salt levels (> 0.5% NaCl) (Choukr-Allah et al., 1996).

The literature reveals that a myriad of experimental procedures have been used for determining salt tolerance (Maas and Hoffman, 1977; Takemura et al., 2000). Likewise, plant response to salinity has been measured in several ways and at various stages of growth (Chartzoulakis and Klapaki, 2000; Chartzoulakis and Loupassaki, 1997; Chartzoulakis et al., 2002). Many studies were concerned with the effects of salinity on horticultural crops and the mechanisms caused by salinity gradients or concentrations (Debouba et al., 2006; Grattan and Grieve, 1999; Meiri et al., 1981; Munns, 2002; Shannon and Grieve, 1999; Sonneveld, 1988, 2000).

Salinity increases, depending on the species of plant, are known to retard growth in most terrestrial plants by influencing several factors of plant behavior like osmotic adjustments, ion uptake, protein and nucleic acid synthesis, photosynthesis, enzyme activities and hormonal balance (El-Gamal, 2000). The extent to which salinity stress affects plant growth and development is also dependent on various factors, including plant species, cultivar, phenological stage, soluble salt composition, stress intensity and duration, and edaphoclimatic conditions (Cramer et al., 1994). Dependent on these factors, adverse effects of increased salinity on plant growth and yield have been attributed to simultaneous reduction in leaf area and root growth which affects photosynthesis and water and mineral uptake (Shannon and Grieve, 1999).

During the onset of salt stress within a plant, all the major processes such as photosynthesis, protein synthesis, and energy metabolism are affected (Parida and Das, 2005). Mass and Hoffman (1977) reported that most plants adversely affected by salt usually appear normal but may have dark green leaves which, in some cases, are thicker and more succulent.

Salinity is typically measured in terms of electrical conductivity (EC). The EC value of a solution is linearly related to the equivalent salt concentration in solution. While EC is indeed a measure of salt levels, EC is a measurement of all salts present (Bagley et al., 1997). Which salts are present and their respective concentrations cannot

be differentiated based on EC values alone. Cuartero and Fernandez-Munos (1999) and Li and Stanguellini, (2001) concluded that high EC's are not always the result of NaCl salt and that analysis of salts should be taken into consideration when growing horticultural crops.

Lettuce (*Lactuca spp*) belongs to the family of Asteraceae. Lettuce is one of the most important vegetable plants worldwide (Ayers et al., 1951; Mass and Hoffman, 1977). In 2010, in terms of production value, lettuce was the second leading vegetable crop in the United States, accounting for $2.25 billion dollars in sales (USDA, 2011b).

Bolarin et al. (2001) stated that when salinity is due to NaCl, its effects on lettuce are associated with accumulation of Na^+ and Cl^- in cells and to ionic imbalance. Al-Maskri et al., (2010) and Karam et al., (2005), reported a decreasing pattern in growth of lettuce was observed with increasing salinity. Gokce et al., (2005) studied the effect of NaCl or $CaCl_2$ on lettuce (*Lactuca sativa* L. var. *longifolia*) in a closed hydroponic system. Their results concluded that the effects of salinity on lettuce are primarily sodium specific and that chloride did not seem to impact the growth suppression of lettuce. Growth suppression was observed only when elevated salinity was caused by adding NaCl to the standard solution, whereas $CaCl_2$ salinity was not harmful to lettuce at EC levels up to 5.5 dS/m (Gokce et al., 2005).

Reports are varied regarding the effect of salts on lettuce. Yuichi (2005) tested the salt tolerance of lettuce with NaCl levels ranging from 0 to 11,688 ppm (0 to 200 mM) and found that growth parameters such as total leaf area and dry weight decreased gradually with the increasing concentrations of NaCl. Salinities up to 5844 ppm (100 mM) did not affect the survival percentage, but salinity levels over 8764 ppm (150 mM) reduced survival to less than 40%. Studies at the U.S. Salinity Laboratory in Riverside, CA showed that the threshold EC of lettuce grown in soil was 1.3 dS/m, with lettuce being classified as moderately salt sensitive (Ayers et al., 1951). Ünlükara et al., (2008) reported that lettuce has a threshold value of 1.1 dS/m and the relative yield decrease in slope after this threshold is 9.3%. Andriolo et al., (2005), reported that salinity levels above 2.0 and 2.6 dS/m reduced lettuce yield and plant growth. Pasternak et al. (1986) reported that quality and yield of field-grown lettuce was not affected by sprinkling with water at EC level of 4.4 dS/m. Other studies have shown a wide range of salt tolerance

across lettuce cultivars (Coons et al., 1990; De Pascale and Barbieri, 1995; Odegbaro and Smith, 1969; Pasternak et al., 1986; Shannon and McCreight, 1984; Shannon et al., 1983; Tzortzakis, 2009). Most studies have concluded that lettuce is considered moderately sensitive to salinity (Ayers et al., 1951; Maas, 1990; Maas and Hoffman, 1977; Shannon et al., 1983) when compared to other vegetable crops.

Sustainability

Sustainability has social, environmental, and economic aspects which should be aligned to seek the best benefit from the production system. One of the major issues in intense fish culture is that high fish population densities can be a major factor in producing pollution (Bloom, 1995) in terms of strength of nutrient effluents. Consumers have shown an increasing demand for more responsible, sustainable, and environmentally friendly forms of food production which has led to the locovore and "eco-friendly" movements. This, in turn, has lead to the innovation of other sources of protein such as processed soybeans to replace fish meal in fish diets. Another innovation is the manipulation of more specific fish diets that are dependent on current environmental factors such as temperature, location, and time of year, as well as, fish species, age, and growth stage. Still another innovation has been improved feeding strategies in a production facility such as multiple small feedings versus single heavy feeding per day. Such improvements have resulted in improved feed conversions and reduced waste (Lyon et al., 2008). Another innovation has been the integration of aquaculture with other animal and plant systems where byproducts of each system are used by the other system resulting in better efficiencies of each system and reduced costs of production. Using such innovations will enable the systems to adapt to increasing product demands leading to an advantage over traditional production practices (Wurts, 2000).

Integration

Throughout history and continuing still today, farming has been integrated. Food, fuel, fiber, and other essentials were grown on the same plot of land. Waste from one crop was used in production of another. Two examples of this were animal manure being used as fuel or fertilizer and crop byproducts, not consumed by humans, being used as

animal feed. In the past, diversification was the key to making use of what was produced in as many ways a possible. In recent decades however, farmers have gotten away from these principles. Crops have come to be grown in monoculture with very specific purposes and little concern, if any, for handling wastes and byproducts. Typically, either maximization or optimization of production is the driving force. Monoculture systems seem more efficient in the short run in terms of income generated, but they present a potentially high risk long-term (Edwards, 1993).

Agriculture is unique, because many different fields share numerous overlapping concerns, making integration sensible (Girardin and Spiertz, 1993). Integrating other systems with the aquaculture industry could provide valuable resources to help reduce production costs, improve productivity, and help reduce or eliminate negative consequences such as water pollution. Since the early 1970's, much research and scientific input has occurred regarding the integration of agricultural systems (Holliman, 2006; Rice, 2008). Edwards (1993) stated that "an output from one sub-system in an integrated farming system which may otherwise have been wasted becomes an input to another sub-system resulting in a greater efficiency of output of desired products from the land/water area under the farmer's control". In the past, the most common integration has been the use of on-farm nutrients as sources of fertilizer for fish ponds. Commercial feeds and fertilizers are valuable and the byproducts of production allow fish foods to develop in the form of phytoplankton, zooplankton, and other benthic organisms, which are utilized in the food chain and ultimately work their way up to the primary consumers, the fish (Colman and Edwards, 1987; Knud-Hansen, 1998; Kwei Lin et al., 1997). Other examples of production integration has been seen between fish and livestock slaughterhouses, poultry farms, and feed lots. Integrations must be in the proper sequence in order to be successful. Many early integrations failed because of this. Old models used manures and effluents to fertilize fish ponds, which ultimately destroyed the water quality. Fish needed to get first use of the inputs. Modern systems take this approach.

One of the most appealing forms of agricultural integration is a fish to plant integration whereby both systems benefit from the other (Zweig, 1986). Aquaponics is the combination of closed, recirculating aquaculture and hydroponics where effluent-rich

water that results from raising fish provides a source of nutrients for growing plants (Neori et al., 2004). The primary input from the farm manager is in the form of pelleted feed which is fed to the fish. Bacterial breakdown of unused feed, along with the breakdown of ammonia from fish respiration produces nitrites and nitrates in the water. This nutrient-rich effluent is the major waste of intense aquaculture systems. The solids (another usable byproduct) are removed from the effluent and the nitrogen-rich water used to irrigate plants, which filter out the nitrogen. When in balance with hydroponic plants, the plants can provide the fish with "clean" water. In turn, the closed-loop system improves the water quality for fish production by removing otherwise harmful nutrients through assimilation into plant biomass (Rakocy, 2002).

In some cases integration of aquaculture and hydroponics may be the result of environmental stewardship or even necessity due to water conservation concerns. Advantages include:

- Reducing the cost and expertise required in mixing hydroponic nutrient solutions.
- Providing the plants with an inexpensive or recycled source of natural food.
- Reduced water consumption and waste.
- Potential to increase the value of the plants through "organic" certification.

Some of the disadvantages of integrating hydroponic and intense aquaculture include:

- Some conventional pesticides used in plant production are toxic to fish.
- Some chemicals used in treating fish are toxic to plants.
- Compromising the best pH (6.5 to 7.0) to fit both the plants and fish physiology.
- Initial capital investments in terms of facilities, piping, etc..
- The need for development of simultaneous marketing.

To reap the benefits of integrated systems, several trials have been conducted to show what plant-fish integration can provide. In Alberta, Canada an integrated tilapia crop system was developed by Savidov et al., (2007) to evaluate the commercial feasibility of an integrated system in the northernmost regions of North America. In the Canadian system, yields of tomatoes and mini-cucumbers reached 20.7 kg/plant/year (45.5 lbs/plant/year) and 33.4 kg/plant/year (73.5 lbs/plant/year), respectively, exceeding

the average yields produced in commercial hydroponic systems in the same region which produced 16.8 kg/plant/year (37 lbs/plant/year) and 28.1 kg/plant/year (61.8 lbs/plant/year) of tomatoes and mini-cucumbers, respectively. Takeda et al., (1993), studied the growth rates of strawberries grown in aquacultural effluents from trout when compared to conventional hydroponic systems.

In experiments at the University of the Virgin Islands, Rakocy et al., (2004) studied a batch versus staggered system of tilapia and basil. When connected to a 500 m^2 (5382 ft^2) hydroponic bed, average yield of basil was 1.9 kg/m^2 (0.39 lbs/ft^2) using the batch and staggered production of tilapia as compared to 0.6 kg/m^2 (0.12 lbs/ft^2) in regular field production. The primary attractions, when compared to field production, for such integrated systems can be both increased productivity as well as increased financial return to the farm manager in the form of savings. Nutrient recovery from aquaculture effluents reduces hydroponic chemical costs. High quality water is repeatedly used to support the growth of both fish and vegetables which reduces costs even further.

Integration of hydroponic lettuce and intensive tilapia culture is an appealing combination and several farmers throughout the southeastern United States have begun such integration. As a result, a more thorough understanding of the relationship between the two species is needed. Currently, there is a lack of information regarding hydroponic lettuce response to variable levels and types of salts used in tilapia production and present in effluent from intensive systems, specifically NaCl or $CaCl_2$. The purpose of this study was to determine threshold concentrations of NaCl or $CaCl_2$, measured in parts-per-million (ppm) chloride, to a crop of hydroponic Bibb lettuce before seeing a decline in growth performance.

MATERIALS AND METHODS

Experiments were conducted at two greenhouses located at Auburn University, Alabama. The first set of experiments was conducted at the E.W. Shell Fisheries Center, North Auburn Unit, Auburn, AL (32°36'14"N - 85°29'30"W) in the fall of 2010. In a 29.3 x 9.1 m (96 x 30 ft) pad and fan-cooled greenhouse with a double layer, polyethylene cover seeds of *Lactuca sativa* L. 'Charles' were sown in one inch (1") oasis blocks, watered, maintained, and kept inside the greenhouse for three weeks prior to transplanting. Plants were then transplanted into a hole cut in a one-half inch (½") thick styrofoam square measuring approximately 20 cm x 20 cm (8" x 8") which was placed on top of a 6-liter (1 gallon) water bucket fitted with an air line to provide aeration. A municipal water source was used. Each bucket contained a one inch (1") airstone powered by an 80 watt compressor pump. The buckets were arranged in a complete randomized block design on top of elevated benches.

A standard hydroponic fertilizer solution (8-15-36) was used in all replicates. Calcium nitrate ($CaNO_3$ - 15.5-0-0 plus 19% Ca) and Epsom salt ($MgSO_4$) was also added to supplement the nutrient solution in all replications (Jeremy Pickens, personal communication). Treatments ranging from 0 ppm to 500 ppm chloride from NaCl or 0 ppm to 500 ppm chloride from $CaCl_2$ were prepared using commercially available bagged material and calculated amounts were added to the nutrient solution (Table 1). There were five replicates of each treatment with NaCl and $CaCl_2$ treatments analyzed separately.

The first trial involved 75 buckets with 5 replicates each of treatments 1 through 15. The buckets were topped off with clean water every three days to insure constant volume and to compensate for water loss due to evaporation.

The lettuce was transplanted on August 21, 2010 into the foam bucket lids. After transplanting, plants were assessed for growth periodically where several indices were collected. Chlorophyll level in the foliage was measured with a SPAD-502 meter

(Minolta, Spectrum Industries Inc.). Salinity, measured in parts-per-thousand, (ppt), electro-conductivity (EC) and pH of the treatment solutions were measured using a YSI Model 63 meter (YSI Inc., Yellow Springs OH).

Plants were harvested at maturity on September 13, 2010 (23 days after transplanting). At harvest, a Growth Index was calculated by taking the average of the height and two perpendicular width measurements for each plant. At harvest, shoot and root fresh weights were recorded. Shoot and root tissues were then dried in a forced air oven drier at 45^{o} C (113^{o} F) for nine (9) days and dry weights were recorded.

Based on the first experiment, the study was subsequently repeated beginning in December, 2010 with some modifications. The second experiment was conducted at the Paterson Greenhouse Complex located on the main campus of Auburn University. The trial was conducted in a 14.6 x 6.1 m (48 x 20 ft) pad and fan-cooled greenhouse with a double layer, polyethylene cover. Air temperature was maintained in this greenhouse between 21^{o} and 35^{o}C (70 to 95^{o}F) with supplemental gas heat. Treatments ranging from 0 ppm to 20,000 ppm chloride from NaCl or 0 ppm to 20,000 ppm chloride from $CaCl_2$ were prepared. Again, while the second study was comprised of twenty-eight (28) total treatments with five (5) replicates of each treatment (Table 2), NaCl and $CaCl_2$ treatments were analyzed separately.

Lettuce seeds were sown in one inch (1") oasis blocks three weeks prior to transplanting. Hydroponic and treatment solutions were prepared as discussed above. Lettuce seedlings were transplanted on December 16, 2010 and harvested on January 20, 2011, thirty-five (35) days after transplanting. Water quality and growth indices were taken periodically and at harvest as described above. Data collected was analyzed using SAS statistical software (SAS, Institute, Inc., Cary, NC).

RESULTS AND DISCUSSION

Experiment 1 - $CaCl_2$ Treatments -- August/September 2010.

For pH values, the 400 ppm $CaCl_2$ treatment was different than the 0 ppm and 50 ppm treatments but similar to all others (Table 3). Likewise, the 0 ppm and 50 ppm $CaCl_2$ treatments were similar to all others except the 400 ppm treatment. For EC values, the 400 ppm and 500 ppm $CaCl_2$ treatments were similar to each other but different than the others. The 200 ppm and 300 ppm $CaCl_2$ levels were also similar to each other but different than all others. The remaining treatments were similar to each other. A similar pattern was seen in the salinity values, the only difference being that the 200 ppm and 300 ppm $CaCl_2$ treatments were not similar to each other. Again, as expected the salinity and EC values increased in direct relationship to the amount of $CaCl_2$ in the treatments. SPAD readings showed no difference between treatments. SPAD readings ranged from 28 to 32. Fresh shoot weight (FSW), dry root weight (DRW), and dry shoot weight (DSW) showed no difference between $CaCl_2$ treatments. The highest FSW was the control at 164.6 grams and the lowest was 103.2 grams at 500 ppm; however, differences were not significant. Fresh root weight (FRW) $CaCl_2$ treatments were similar to each other except the 500 ppm $CaCl_2$ treatment. The 500 ppm was similar to the 50 ppm, 75 ppm, and 100 ppm $CaCl_2$ treatment but different than all the others. There was no difference between treatments for Growth Index.

Experiment 1 - NaCl Treatments -- August/September 2010.

There was no difference in pH values across all NaCl treatments; pH ranged from 7.0 to 7.3 (Table 4). Electro-conductivity readings (EC) were all different except the control (0 ppm), which was similar to both the 50 ppm NaCl and 75 ppm NaCl treatment. Salinity values, likewise, were all different with the exception of the control which was, again, similar to the 50 ppm and 75 ppm NaCl treatments. In both cases, the actual salinity and EC value at the 50 ppm was slightly lower than the control, possibly due to

plant uptake of the NaCl provided in the treatment, whereas the control provided no NaCl. As expected, both EC values and salinity values generally increased in direct proportion to the increase in NaCl treatment. SPAD readings were all similar except at the 50 ppm and 300 ppm NaCl treatments. FSW, FRW, and DSW were not different between NaCl treatments. The highest FSW was 172 grams at 100 ppm and the lowest was 130 grams at 400 ppm, but were not significant. For DRW, the 200 ppm treatment was similar to the 400 ppm and 500 ppm NaCl treatments but different than all others. All others, including the 400 ppm and 500 ppm NaCl treatments were similar. There were no differences for Growth Index between any treatments.

Conclusions of Experiment 1.

The first series of experiments indicated that Bibb lettuce (*Lactuca sativa* L. 'Charles') was not adversely affected by either NaCl or $CaCl_2$ at levels ranging from 0 ppm to 500 ppm chloride. In this study, 500 ppm $CaCl_2$ produced an EC value of 2300µS/cm. This is a higher EC than those reported to cause injury by Ayers et al., (1951) and Ünlükara et al., (2008) of 1300 µS/cm. These results are also slightly higher than the low range of 2000 µS/cm but below the high range of 2600 µS/cm reported by Andriolo et al., (2005). However, these results are considerable lower than those reported by Pasternak et al. (1986) of 4400 µS/cm and Gokce et al., (2005) at EC levels up to 5500 µS/cm.

Observationally, pH and SPAD readings do not appear to be descriptive measurements of salt effect at the concentrations evaluated in this study. As a result of this study, a second set of experiments was deemed necessary with increased salt concentrations in order to determine the salt concentrations needed to observe adverse effects on Bibb lettuce.

Experiment 2 - $CaCl_2$ Treatments -- December 2010/January 2011.

Three of the $CaCl_2$ treatments (75 ppm, 200 ppm, and 400 ppm) from the first experiment were repeated in December, 2010. All plants at concentrations of 10,000 ppm and 20,000 ppm chloride died within 10 days of transplanting. For pH, no $CaCl_2$ treatment appeared exclusively different than any other; however, the lower treatments (0

ppm to 1500 ppm) were mostly similar to each other and the higher treatments (2000 ppm to 20K ppm) were similar to each other (Table 5). The five highest treatments (2000, 3000, 5000, 10K, and 20K ppm) had the lowest pH. For EC values, the two highest $CaCl_2$ treatments were different than all other treatments. A graph of the EC values shows an almost linear relationship with treatment concentrations (Figure 1). Similarly, salinity generally increased as chloride concentration increased. For SPAD readings, the only treatment that was different was the $CaCl_2$ treatment at 5000 ppm chloride; all others were similar to each other. The 5000 ppm chloride treatment had a SPAD reading of 21; all others were in the 30's. FRW's were lowest at 3000 ppm and 5000 ppm, which were similar to each other but different than the others. Other treatments were similar in various combinations. For DRW, the only treatment that was different was the 5000 ppm treatment which also had the lowest value. FSW, DSW, and the Growth Index gave similar results to each other. Significantly differences and the lowest values were seen at 3000 ppm and 5000 ppm treatments for FSW, DSW, and Growth Index. The high for FSW was 142 grams at 600 ppm and the low was 4 grams at 5000 ppm. A plot of the Growth Index (Figure 2) shows a distinct pattern; as $CaCl_2$ concentrations increase beyond 2000 ppm, Growth Index decrease rapidly. Visually, laying the plants side-by-side, a dramatic change is observed between the 2000 ppm and 3000 ppm treatment (Figure 3).

Experiment 2 - NaCl Treatments -- December 2010/January 2011.

Three of the NaCl treatments (75 ppm, 200 ppm, and 400 ppm) from the first experiment were repeated in December, 2010. All plants at 10,000 ppm and 20,000 ppm NaCl died within 10 days of transplanting. For pH, no treatments stood out as different than all others. Each treatment was similar to at least seven (7) other treatments (Table 6). The range for pH values was a high of 8.2 at 75 ppm to 7.9 at 3000 ppm. For EC values, treatments over 2000 ppm were different than all others, with treatments at 2000 ppm and below similar to other treatments. As expected, EC values were directly related to the concentration level of NaCl; as NaCl level increased, so did the EC value (Figure 4). Likewise, for salinity, as NaCl increased, so did the salinity value. For salinity measurements, the only two treatments that were different than all others were the 10,000

ppm and 20,000 ppm treatments. All others levels had similarities to others; however, similarities were with adjacent treatments only. As seen in the earlier experiments, the lowest salinity was not the control, but was the lowest treatment (75 ppm). Again, this suggests that the plants absorb and utilize some of the NaCl present in the treatments. SPAD readings were scattered with similarities between treatments showing no distinguishable pattern. The mean values of all SPAD readings were within five (5) points of each other (29.5 to 34.7). For FSW, similarities were between adjacent treatments. There was a significant difference between all NaCl treatments below 1200 ppm and all NaCl treatments over 2000 ppm, suggesting that chloride values over 2000 ppm have an adverse effect on the plants. The smallest difference between these two groupings of FSW was a 30% difference (75 grams vs. 106 grams); all other differences were greater. For DSW, all treatments below 1500 ppm were similar and were also significantly different than all treatments at 2000 ppm and above. For FRW, 3000 ppm and 5000 ppm were similar to each other, but different than all others. Treatments below 3000 ppm were randomly similar to each other. A similar phenomenon was observed with the DRW. The analysis of the Growth Index showed similar results as the FSW analysis. NaCl treatments over 2000 ppm were different than all others. A graph of the Growth Index (Figure 5) shows a very similar polynomial curve to the $CaCl_2$ treatments in Figure 2.

Visually, a significant change can be seen between plants at 2000 ppm and 3000 ppm (Figure 6).

Conclusions.

SPAD readings and pH ranges do not appear to be helpful indicators or instruments for measuring the effects of chloride concentrations on Bibb lettuce.

Yuichi (2005) tested the salt tolerance of lettuce with NaCl levels ranging from 0 to 11,688 ppm (0 to 200 mM). Concentrations up to 5844 ppm (100 mM) did not affect the survival percentage, but salinity levels more than 8764 ppm (150 mM) significantly reduced survival to less than 40%. The current study shows chloride concentrations above 5000 ppm chloride were lethal to Bibb lettuce.

Significant differences in terms of growth and water measurements begin to appear above 2000 ppm chloride using either NaCl or $CaCl_2$. For NaCl, the significant EC level in this study (> 4332 µS/cm) corresponds to a chloride concentration of 2000 ppm which is higher than those reported to cause injury by Ayers et al., (1951) and Ünlükara et al., (2008) of 1300 µS/cm and those reported as 2000 µS/cm to 2600 µS/cm by Andriolo et al., (2005); however, they are below those reported by Pasternak et al. (1986) of 4400 µS/cm and Gokce et al., (2005) at EC levels up to 5500 µS/cm. This affirms the finding of other studies showing that there is wide range of salt tolerance between lettuce cultivars (Coons et al., 1990; Odegbaro and Smith, 1969; Pasternak et al., 1986; Shannon and McCreight, 1984; Shannon et al., 1983).

The Waller-Duncan analysis ($P \leq 0.05$) and the graphic representation of the Growth Index (Figures 2 and 5) in both NaCl and $CaCl_2$ treatments shows significance at values over 2000 ppm chloride; however, the regression lines suggest that the decline begins prior to where differences are significant. Growth, measured as fresh shoot weight (FSW), fresh root weight (FRW), dry shoot weight (DSW), and dry root weight (DRW) showed a similar curve and began to decrease prior to 2000 ppm chloride (Figures 7, 8, 9, and 10). Andriolo et al., (2005), reported that EC above 2000 µS/cm reduced lettuce yield, which corresponds to approximately 1000 ppm chloride using either NaCl or $CaCl_2$, supporting the idea of the decline beginning at some point slightly below 2000 ppm chloride. Visually, Figure 3 and Figure 6 show a decline in size at 2000 ppm chloride, a dramatic decline at 3000 ppm and 5000 ppm, and death at 10,000 ppm and 20,000 ppm chloride. These results suggest there to be only minor differences between the effect of $CaCl_2$ and NaCl on Bibb lettuce.

For integrated systems of hydroponic Bibb lettuce and intensive fish culture, managers need to monitor and test the effluent coming out of the fish tanks to insure that chloride concentrations of the effluent are no more than 2000 ppm. Our research suggests that managers not view the 2000 ppm chloride level as a tipping point for lettuce but as the point at which significant adverse effects occur. More research is needed to determine a more precise point at which growth is significantly affected. Also, and perhaps more importantly, more research is needed to determine a more specific salt level at which growth begins to be adversely affected in hydroponic Bibb lettuce.

Furthermore, as integrated aquaculture/horticulture systems increase in popularity, growth responses to elevated salt levels will need to be determined for other vegetable crops commonly grown in combination with fish effluents such as bell peppers, pak choi, cucumbers, tomatoes, chard, and basil. As integration increases as a whole, managers must understand the dynamics of these cross-cultural systems. Managers will need to have a thorough understanding of the effect one input in one system has on all other systems. What may be seen as a traditional, proper, and beneficial treatment in one system could very well have an equally or even more destructive affect on another. Further studies on integration are needed in order to understand the dynamics of these combined systems.

FINAL DISCUSSION

Over the past 50 years many issues have been a major concern for agricultural managers. Ever-increasing production costs, water rights, fertilizer and fuel costs, government regulations, as well as, environmental concerns have been issues that managers have had to deal with in the last half century. In the current economical, political, and environmental climate, these issues are all even more at the forefront of concerns facing the agricultural industry. Currently, new legislative regulations, as well as the activities of environmentalists, are putting pressure on the horticulture and aquaculture industries to decrease or limit water use and control nutrient effluents discharged into natural systems (Boyd, 2003), while at the same time, economic concerns and world food needs demand that managers push production systems to their very upper limits.

Aquaponics is the combination of intensive tank fish culture and hydroponic plant production. Both systems are able to utilize byproducts of the other as inputs, thereby potentially reducing costs and nutrient-laden discharges. For fish species, one of the most common species used in tank culture is tilapia. For plant species, aquaponics has typically focused on vegetable crops (lettuce, basil, tomatoes, cucumbers, peppers and others) (Nelson and Pade, 2009; Rakocy et al., 1992).

In aquaponic systems however, research is limited concerning the effect a "standard practice" in one system has on the other system(s) and vice versa. A commonly used practice in intensive fish culture is the frequent use of salt, usually in the form of NaCl, as a calming agent for distressed fish. This study was designed to determine the level of salt, NaCl or $CaCl_2$ that hydroponic Bibb lettuce can withstand before adversely affecting production. Our results showed that chloride concentrations over 2000 ppm were significantly detrimental to hydroponic Bibb lettuce.

Studies continue to be conducted on the effects of both NaCl and $CaCl_2$ on other vegetable crops (Wu et al., 2004). Additional research is needed to examine the effect of salt from intensive fish culture has on not only other vegetable crops, but all horticultural crops that can be grown in conjunction with fish (ornamentals, floral, bedding plants, etc.).

General Experimental Observations

Even though chloride concentrations over 2000 ppm can be seen as producing significant adverse effects, graphs of the various growth indexes showed that the trend downward in terms of growth began somewhere before 2000 ppm. Further research is needed to evaluate where this downward tipping point begins. Experimentally, our results would suggest that more treatments in the 500 ppm to 3000 ppm range, combined with regression analysis, need to be done in order to more precisely determine this tipping point. Also, our study showed that salt levels somewhere between 5000 ppm and 10,000 ppm chloride caused death of Bibb lettuce. Further research is needed to determine a more precise level that causes death. One final observation would be the need to design an experimental setup that helps retain the salts within the hydroponic buckets. Because of the bubbling action of the airstones, the bottom of the styrofoam blocks as well as the edges of the buckets, both inside and out, showed salt precipitation and buildup. Perhaps sealing the bucket top with cellophane or some other plastic material, except for a slit for the plant roots, would help alleviate this design problem so that only the salts taken up by the plants are removed. We did not statistically examine the difference between the NaCl and $CaCl_2$ treatments; however, we observed no significant difference. Because of cost and availability, we believe managers will almost always be using NaCl as the source of salt and that future experiments should concentrate on this salt.

Conclusion

Research conducted indicates that chloride levels over 2000 ppm are harmful to hydroponic Bibb lettuce. More research is needed for defining the exact level at which growth begins to decline, as well as, research to define a more precise point at which salt

concentrations kill Bibb lettuce. Finally, further research is needed to examine the effect of salt on other horticultural crops used in aquaponics, and the effect of salt on plant attributes other than just growth (i.e. flavor, leaf area, mineral composition).

LITERATURE CITED

Afzal, I., S.M.A. Basra, M. Farooq, and A. Nawaz. 2006. Alleviation of salinity stress in spring wheat by hormonal priming with ABA, salicylic acid and ascorbic acid. Int. J. Agric. Biol. 8:23–28.

Al-Maskri, A., L. Al-Kharusi, and H. Al-Miqbali. 2010. Effects of salinity stress on growth of lettuce (*Lactuca sativa*) under closed-recycle nutrient film technique, Int. J. Agric. Biol. 12:377-380.

Allakhverdiev, S.I., A. Sakamoto, Y. Nishiyama, M. Inaba, and N. Murata. 2000. Ionic and osmotic effects of NaCl-induced inactivation of photosystems I and II in *Synechococcus sp.* Plant Physiol. 123:1047-1056.

Alleman, J.E. 1985. Elevated nitrate occurrence in biological waste-water treatment systems. Water Science and Tech. 17:407-419.

Andriolo, J. L., G. L. Da Luz, M.H. Witter, R. dos S. Godoi, G.T. Barros, and O.C. Bortolotto. 2005. Growth and yield of lettuce plants under salinity. Hortic. Bras. 23:931-934.

Auburn University. 2004. Alabama Aquaculture Best Management Practice (BMP): Feed Management. Auburn University. United States Department of Agriculture, Natural Resources Conservation Service. BMP No. 7. 1 Feb. 2011. <http://www.aces.edu/dept/fisheries/aquaculture/BMP.php>.

Ayers, A.D., C.H. Wadleigh, and L. Bernstein. 1951. Salt tolerance of six varieties of lettuce. Proc. Am. Soc. Hort. Sci. 57:237-242.

Bagley, C.V., J. Kotuby-Amacher, K. Fareel-Poe. 1997. Analysis of Water Quality For Livestock, Utah State University Cooperative Extension.

Berghage, R.D., S.F. MacNeal, T.F. Wheeler, and W.H. Zachritzt. 1999. "Green" water treatment for green industry: opportunities for bio-filtration of greenhouse and nursery irrigation water and runoff with constructed wetlands. HortScience 34:50-54.

Bloom, T.T. 1995. Reducing the potential environmental impact of tank aquaculture effluents through intensification and recirculation. Aquaculture 226:35-44.

Bolarin, M.C., M.T. Estan, M. Caro, R. Romero-Aranda, and J. Cuartero. 2001. Relationship between tomato fruit growth and fruit osmotic potential under salinity. Plant Science 160:1153-1159.

Boyd, C.E. 2003. Guidelines for aquaculture effluent management at the farm-level. Aquaculture 226:101-112.

Boyd, C. E., and C. S. Tucker. 1998. Pond Aquaculture Water Quality Management. Kluwer Academic Publishers, Boston, Massachusetts, USA.

Brunson, M.W. 2009. Brown Blood Disease. Mississippi State University Extension System Information sheet 1390. 1 Feb. 2011. <http://www.msucares.com/pubs/infosheets/is1390.htm>.

Chartzoulakis, K. and G. Klapaki. 2000. Response of two greenhouse pepper hybrids to NaCl salinity during different growth stages. Sci. Hort. 86:247-260.

Chartzoulakis, K. and M. Loupassaki. 1997. Effects of NaCl salinity on germination, growth, gas exchange and yield of greenhouse eggplant. Agric. Water Manage. 32:215-225.

Chartzoulakis, K., M. Loupassaki, M. Bertaki, and I. Androulakis. 2002. Effects of NaCl salinity on growth, ion content and CO_2 assimilation rate of six olive cultivars. Sci. Hort. 96:235-247.

Choukr-Allah, R., C.V. Malcolm, and A. Hamdy. 1996. Halophytes and biosaline agriculture, Marcel Dekker Inc., New York, pp. 401.

Colman, J.A., and P. Edwards. 1987. Feeding pathways and environmental constraints in waste-fed aquaculture: Balance and optimization. In Detritus and Microbial Ecology in Aquaculture, v-vii, eds. D.J.W. Moriarty, and R.S.V. Pullin. Manila, Philippines: International Center for Living Aquatic Resources Management 240-281.

Coons, J.M., R.O. Kuehl, and N.R. Simon. 1990. Tolerance of ten lettuce cultivars to high temperature combined with NaCl during germination. J. Am. Soc. Hort. Sci. 115:1004-1007.

Cramer, G.R., G.J. Alberico, and C. Schmidt. 1994. Salt tolerance is not associated with the sodium accumulation of two maize hybrids. Austral. J. Plant Physiol. 21:675-692.

Crews, J.R., and J.A. Chappell. 2007. 2006 Alabama aquaculture factsheet. Timely Information. Alabama Cooperative Extension System. 1 Feb. 2011. <http://www.aces.edu/timelyinfo/Fisheries/2006/April/05aqua_factsheet.pdf>.

Cuartero, J., and R. Fernandez-Munos. 1999. Tomato and salinity. Scientia Horticulturae 78:83-125.

De Pascale, S. and G. Barbieri. 1995. Effects of soil salinity from long-term irrigation with saline-sodic water on yield and quality of winter vegetable crops. Sci. Hort. 64:145–157.

Debouba, M., G. Houdea, S. Akira, and M.H. Ghorbel. 2006. NaCl stress effects on enzyme involved in nitrogen assimilation pathway in tomato "*Lycopersicon esculentum*" seedlings. J. Plant Physiology 163:1247-1258.

Dierberg, F.E., and W. Kiattisimukul. 1996. Issues, impacts, and implications of shrimp aquaculture in Thailand. Environmental Management 20:649-666.

Diver, S. 2006. Aquaponics-Integration of hydroponics with aquaculture. ATTRA National Sustainable Agriculture Information Service. 1 Feb. 2011. <www.attrancat.org/attra-pub/PDF /aquaponics.pdf>.

Edwards, P. 1993. Environmental issues in integrated agriculture-aquaculture and wastewater-fed fish culture systems. In: Pullin, R.S.V., Rosenthal, H., Maclean, J.L. (Eds.), Environment and Aquaculture in Developing Countries. ICLARM Conference Proceedings 31. ICLARM, Manila, Philippines, pp. 139-170.

Ehret, D.L. and A.L. Plant. 1999. Salt tolerance in crop plants. In: Dhaliwal, G.S., Arora, R. (Eds.), Environmental Stress in Crop Plants, Commonwealth Publishers, New Delhi, India, pp. 69-120 (Chapter 5).

El-Gamal, I.M. 2000. Interaction between cobalt and salinity on the plant growth, ICEHM2000, Cairo University, Egypt. September 2000:525-533.

Environmental Protection Agency (EPA). 1980. Design manual: onsite wastewater treatment and disposal systems. EPA 625/1-80-012:103-297.

Food and Agriculture Organization of the United Nations (FAO). 2007. The state of the world fisheries and aquaculture 2006. ISSN 1020-5489. 1 Feb. 2011. <http://www.fao.org/docrep/009/a0699e/A0699E04.htm>.

Flowers, T.J., P.F. Troke, and A.R. Yeo. 1977. The mechanism of salt tolerance in halophytes. Annu. Rev. Plant Physiol. 28:89-121.

Francis-Floyd, R., C. Watson, D. Petty, and D.B. Pouder. 2009. Ammonia in aquatic systems. University of Florida IFAS Extension Publication #FA-16.

Girardin, P., and J.H.J. Spiertz. 1993. Integrated agriculture in Western Europe: Researchers' Experience and Limitations. J. Sustainable Agric. Vol. 3/4:155-170.

Gokce, T., N. Papadandonakis, and D. Savvas. 2005. Responses of lettuce (*Lactuca sativa* L. var. *longifolia*) grown in a closed hydroponic system to NaCl- or $CaCl_2$-salinity. J. Applied Bot. and Food Quality 79:136-140.

Goldburg, R., and T. Triplett. 1997. Murky waters: Environmental effects of aquaculture in the United States. Environmental Defense Fund. Washington, DC. 198pp.

Graber A. and R. Junge. 2008. Aquaponic Systems: Nutrient recycling from fish wastewater by vegetable production. Desalination 246:1-14.

Grattan, S.R. and C.M. Grieve. 1999. Salinity-mineral nutrient relations in horticultural crops. Sci. Hort. 78:127-157.

Greenway, H. and R. Munns. 1980. Mechanism of salt tolerance in non-halophytes. Annu. Rev. Plant Physiol. 31:149-190.

Hamdy, A. 1993. Workshop on: "Environmentally Sound Water Management of Protected Agriculture under Mediterranean and Arid Climates", CHIEAM/MAI - Bari, Valenzano (Bari), Italy 16-18 July. 472.

Hamdy, A. 1996. Protected agriculture. In: Irrigation fertilizer management and soilless culture under protected agriculture (Advanced short course). Agadir, 10-17 Oct 1993. 1-35.

Hamdy, A., R. Kaf Alghazal, G. Pacucci, and C. Troccoli. 2009. Green Salad Production in soilless culture under saline irrigation practice. Acta Hort. (ISHS) 843:103-110.

Heyden, L. 2009. Hydroponics in commercial food production. 1 Feb. 2011. <http://www.besthydroponicsystems.com/hydroponic-systems-in-commercial-food-production>.

Hargreaves, J.A. and C.S. Tucker. 2004. Managing ammonia in fishponds. Southern Regional Aquaculture Center 4603.

Hasegawa, P.M., R.A. Bressan, J.K. Zhu, and H.J. Bohnert. 2000. Plant cellular and molecular response to high salinity. Annu. Rev. Plant Physiol. Plant Mol. Biol. 51:463-499.

Holliman, J.B. 2006. An economic analysis of integrating hydroponic tomato production into an indoor recirculating aquacultural production system. Auburn Univ. Auburn, MS Thesis.

Jensen, M.H. 1997. Hydroponics. HortScience 32:1018-1021.

Karam, N.S. and B.H. Al-Daood. 2005. Response of asiatic lily to nutrient solution recycling in a closed soilless culture. Acta Hort. 697:199–212.

Kwei Lin, T.F., R.D.L. Hamilton, P.A. Sandifers, C.L. Browdy, and A.D. Stokes. 1997. Effect of water exchange rate on production, water quality, effluent characteristics and nitrogen budgets of intensive shrimp ponds. J. World Aqua. Soc. 24:304-320.

Knud-Hansen, D.L. 1998. The role of phosphorus in the eutrophication of receiving waters: A Review. J. Environ. Qual. 27: 261-266.

Li, Y.L. and C. Stanguellini. 2001. Analysis of the effects of EC and potential transpiration on vegetative growth of tomato. Scientia Horticulturae 89:9-21.

Lyon, D.D., J. Paret, A. Barkoh, and E. Nunez. 2008. Effect of floating feed ring on advanced channel catfish fingerling production. Tex. Dept. Inland Fisheries Div. ii, 10 pp.

Maas, E.V. 1990. Crop salt tolerance. In: Tanji, K.K. (Ed.), Agricultural Salinity Assessment and Management. ASCE Manuals and Reports on Engineering No. 71, ASCE, New York, pp. 262-304.

Maas, E.V. and G.J. Hoffman. 1977. Crop salt tolerance - current assessment. J. Irrig. Drainage Div. ASCE 103(IR2) 115-134.

Madidi, S.E., B.E. Baroudi, and F.B. Aameur. 2004. Effects of salinity on germination and early growth of barley (*Hordeum vulgare* L.) cultivars. Int. J. Agric. Biol. 6:67–70.

Meiri, A., Z. Plaut, and L. Pinkas. 1981. Salt tolerance of glasshouse grown muskmelon (cv. Galia). Soil Sci. 131:189-193.

Mengel, K., and E.A. Kirkby. 1979. Principles of Plant Nutrition. International Potash Institute, Berne, Switzerland.

Munns, R. 2002. Comparative physiology of salt and water stress. Plant Cell Environ. 25:239-250.

Munns, R. and M. Tester. 2008. Mechanism of salinity tolerance. Annl. Rev. Plant Biol. 59:651–681.

Naylor, R.L., R.J. Goldburg, J.H. Primavera, N. Kausky, M.C.M. Beveridge, J. Clay, C. Folke, J. Lubchenco, Mooney, J, and M. Troell. 2000. Effect of aquaculture on world fish supplies. Nature 405:1017-1024.

Nelson, R. and J. Pade. 2009. Recommended plant and fish in aquaponics. Aquaponics.com. March 2011. <http://www.aquaponics.com/InfoPlantsAndFish.htm>.

Neori, A., T. Chopin, M. Troell, A. H. Buschmann, G. P. Kraemer, C. Halling, M. Shpigel, and C. Yarish. 2004. Integrated aquaculture: Rationale, evolution and state of the art emphasizing seaweed biofiltration in modern mariculture. Aquaculture 231:361-391.

Odegbaro, O.A., and O.E. Smith. 1969. Effects of kinetin, salt concentration, and temperature on germination and early seedling growth of *Lactuca sativa* L. J. Amer. Soc. Hort. Sci. 94:167-170.

Palada, M.C., W.M. Cole, and S.M.A. Crossman. 1999. Influence of effluents from intensive aquaculture and sludge on growth and yield of bell peppers. J. Sustainable Agriculture 14:85-103.

Parida, A.K., and A.B. Das. 2005. Salt tolerance and salinity effects on plants: a review. Ecotoxicology and Environmental Safety 205 (60):324-349

Pasternak, D., Y. de Malach, I. Borovic, M. Shram, and C. Aviram. 1986. Irrigation with brackish water under desert conditions. IV. Salt tolerance studies with lettuce (*Lactuca sativa* L.) Agric. Water Manage. 11:303-311.

Popma, T. and M. Masser. 1999. Tilapia: Life history and biology. Southern Regional Aquaculture Center 283.

Popma, T.J. and L.L. Lovshin. 1996. World prospects for commercial production of tilapia. In: Research and Development Series 41 International Center for Aquatic Environment, Department of Fisheries and Allied Aquaculture, Auburn University, Alabama 24 pp.

Rahman, S.S. 2010. Effluent water characterization of intensive tilapia culture units and its application in an integrated lettuce aquaponic production facility. Auburn Univ. Auburn, MS Thesis.

Rakocy, J.E. 1989. Tank culture of tilapia. Southern Regional Aquaculture Center 282.

Rakocy, J.E. 2002. An integrated fish and field crop system for arid areas. In: Ecological Aquaculture: Evolution of the Blue Revolution. Blackwell Science. Costs-Pierce, B.A. (Ed.), Oxford p. 263-285.

Rakocy, J.E., T.M. Losordo, and M.P. Masser. 1992. Recirculating aquaculture tank production systems: Integrating fish and plant culture. Southern Regional Aquaculture Center, No. 454.

Rakocy, J.E., and A.S. McGinty. 1989. Pond culture of tilapia. Southern Regional Aquaculture Center 280.

Rakocy, J.E., R.C. Shultz, D.S. Bailey, and E.S. Thoman. 2004. Aquaponic production of tilapia and basil: comparing batch and staggered cropping system. Acta Hort. 648:63-69.

Resh, H.M. 2004. Hydroponic Food Production, 6th ed. New Concept Press, Inc., Mahwah, NJ. 221-238.

Rice, M. 2008. Rhode Island Sea Grant Fact Sheet: Aquaculture. 1 Feb. 2011. <http://seagrant.gso. uri.edu/factsheets/aquaculture.html>.

Savidov, N.A., E. Hutchings, and J.E. Rakocy. 2007. Fish and plant production in a recirculating aquaponic system: a new approach to sustainable agriculture in Canada. Acta Horticulturae 742:209-221.

Shannon, M.C., and C.M. Grieve. 1999. Tolerance of vegetable crops to salinity. Scientia Hort. 78:5-38.

Shannon, M.C., and J.D. McCreight. 1984. Salt tolerance of lettuce introductions. HortScience 19:673-675.

Shannon, M.C., J.D. McCreight, and J.H. Draper. 1983. Screening tests for salt tolerance in lettuce. HortScience 18:225-230.

Shaw, N.L., and D.J. Cantliffe. 2003. Hydroponically produced mini-cucumber with improved powdery mildew resistance. Proc. FL State Hort. Soc. 116:58-62.

Sleeper, A.M. 2009. Integration of intensive aquaculture production and horticulture crop production. Auburn Univ. Auburn, MS Thesis.

Smith, G. 2007. Field vs glasshouse tomatoes. Practical Hydroponics and Greenhouses May/June (Issue 94). <http://www.hydroponics.com.au/free-articles/issue-94-field-vs-glasshouse-tomatoes/>.

Sonneveld, C. 1988. The salt tolerance of greenhouse crops. Neth. J. Agric. Sci. 36:63-73.

Sonneveld, C. 2000. Effects of salinity on substrate grown vegetables and ornamentals in greenhouse horticulture. Ph.D. Thesis, University of Wageningen, The Netherlands.

Stickney, R.R. 1996. Aquaculture in the United States. New York: John Wiley & Sons.

Takeda, F., P. Adler, and D. Glenn. 1993. Growing greenhouse strawberries with aquaculture effluent. Acta Hort. 348:264-267.

Takemura, T., N. Hanagata, K. Sugihara, S. Baba, I. Karube, and Z. Dubinsky. 2000. Physiological and biochemical responses to salt stress in the mangrove, *Bruguiera gymnorrhiza*. Aquat. Bot. 68:15-28.

Tave, D. 1986. Genetics for fish hatchery managers. Westport, CN: AVI Pub Co.

Timmons, M. and T. Losordo. 1994. Aquaculture water reuse systems: engineering design and management. New York, NY: Elsevier 18-27.

Tzortzakis, N.G. 2009. Influence of NaCl and Calcium Nitrate on lettuce and endive growth using nutrient film technique. International J. of Veg. Science. 1931-5279. 15 (1):44-56.

United States Department of Agriculture (USDA). 2011a. USDA definition of specialty crops. 1 Feb. 2011. <http://www.nifa.usda.gov/funding/pdfs/definition_of_specialty_crops.pdf>.

United States Department of Agriculture (USDA). 2011b. Vegetables 2010 Summary. ISSN 0884-6413. 1 Feb. 2011. <http://usda.mannlib.cornell.edu/usda/current/VegeSumm/VegeSumm-01-27-2011_new_format.txt>.

University of Michigan. 2006. World fisheries: declines, potential and human reliance. 1 Feb. 2011. <http://www.globalchange.umich.edu/globalchange2/current/lectures/fisheries/fisheries.html>.

Ünlükara, A., B. Cemek, S. Karaman, and S. Ersahin. 2008. Response of lettuce (*Lactuca sativa* var. *crispa*) to salinity of irrigation water. New Zealand J. Crop Hort. Sci. 36:265–273.

Watanabe, W., T. Losordo, T. Fitzsimmons, and F. Hanley. 2002. Tilapia production systems in the Americas: Technological advances, trends, and challenges. Rev. in Fish. Sci. 10(3&4):465-498.

Wolverton, B.C. 1987. Aquatic plants for wastewater treatment: An overview. In Aquatic Plants for Water Treatment and Resource Recovery, 3-15, eds. K.R. Reddy, and W.H. Smith. Orlando, FL. Magnolia Publishing, Inc.

Wu, M., J.S. Buck, and C. Kubota, 2004. Effects of nutrient solution EC, plant microclimate and cultivars on fruit quality and yield of hydroponic tomatoes (Lycopersicon esculentum). Acta Hort. 659:541-547.

Wurts, W. 2000. Sustainable aquaculture in the twenty-first century. Reviews in Fisheries Science 8:141-150.

Yuichi, U. 2005. Genetic improvement of lettuce for salt tolerance by Arabidopsis DREB gene transfer. 1 Feb. 2011. <http://www.saltscience.or.jp/kenkyu/jyoseilist/ENGsum/0513-E.pdf>.

Zhu, J.K. 2002. Salt and drought stress signal transduction in plants. Annu. Rev. Plant Biol. 53:247-273.

Zweig, R. 1986. An integrated fish culture hydroponic vegetable production system. Aquacult. Mag. May/June 34-40.

APPENDIX

Table 1. Treatments used in NaCl or $CaCl_2$ evaluation on hydroponic Bibb lettuce. Experiment 1, August/September, 2010[z].

Treatment	Chloride from NaCl	Treatment	Chloride from $CaCl_2$
Trmt 1	0 ppm		
Trmt 2	50 ppm	Trmt 9	50 ppm
Trmt 3	75 ppm	Trmt 10	75 ppm
Trmt 4	100 ppm	Trmt 11	100 ppm
Trmt 5	200 ppm	Trmt 12	200 ppm
Trmt 6	300 ppm	Trmt 13	300 ppm
Trmt 7	400 ppm	Trmt 14	400 ppm
Trmt 8	500 ppm	Trmt 15	500 ppm

[z] 37.9 L (10 gal.) quantities of each treatment were prepared and mixed with hydroponic fertilizers (8-15-36), calcium nitrate ($CaNO_3$ - 15.5-0-0 plus 19% Ca) and Epsom salt ($MgSO_4$).

Table 2. Treatments used in NaCl or $CaCl_2$ evaluation on hydroponic Bibb lettuce. Experiment 2, December 2010/January 2011[z].

Treatment	Chloride from NaCl	Treatment	Chloride from $CaCl_2$
Trmt 1	0 ppm		
Trmt 2	75 ppm	Trmt 9	75 ppm
Trmt 3	200 ppm	Trmt 10	200 ppm
Trmt 4	400 ppm	Trmt 11	400 ppm
Trmt 5	600 ppm	Trmt 12	600 ppm
Trmt 6	800 ppm	Trmt 13	800 ppm
Trmt 7	1000 ppm	Trmt 14	1000 ppm
Trmt 8	1200 ppm	Trmt 15	1200 ppm
Trmt 17	1500 ppm	Trmt 23	1500 ppm
Trmt 18	2000 ppm	Trmt 24	2000 ppm
Trmt 19	3000 ppm	Trmt 25	3000 ppm
Trmt 20	5000 ppm	Trmt 26	5000 ppm
Trmt 21	10,000 ppm	Trmt 27	10,000 ppm
Trmt 22	20,000 ppm	Trmt 28	20,000 ppm

[z] 37.9 L (10 gal.) quantities of each treatment were prepared and mixed with hydroponic fertilizers (8-15-36), calcium nitrate ($CaNO_3$ - 15.5-0-0 plus 19% Ca) and Epsom salt ($MgSO_4$).

Table 3. Final results from $CaCl_2$ treaments of hydroponic lettuce (*Lactuca sativa* L. 'Charles') from Experiment 1 August-September 2010[Z].

Treatment[Y]	Growth Index[XQ]		Fresh Shoot Weight[WQ]		Dry Shoot Weight[VQ]		Fresh Root Weight[WQ]		Dry Root Weight[VQ]		Salinity[UP]		EC[TP]		SPAD[SP]		pH[RP]	
0 ppm	20.2	A[O]	164.6	A	17.0	A	49.0	A	9.0	A	0.5	D	967	C	30.1	A	7.2	A
50 ppm	18.0	A	123.4	A	14.8	A	42.2	AB	8.7	A	0.6	D	1157	C	29.5	A	7.2	A
75 ppm	19.5	A	147.8	A	16.4	A	45.4	AB	8.9	A	0.6	D	1125	C	27.5	A	7.1	AB
100 ppm	20.7	A	163.0	A	17.3	A	46.0	AB	8.9	A	0.6	D	1156	C	31.8	A	7.1	AB
200 ppm	19.5	A	154.6	A	16.6	A	47.0	A	9.0	A	0.8	C	1542	B	32.1	A	6.9	AB
300 ppm	19.9	A	152.6	A	16.4	A	48.8	A	8.9	A	0.9	B	1785	B	30.1	A	6.9	AB
400 ppm	20.1	A	158.4	A	16.8	A	53.2	A	9.2	A	1.1	A	2174	A	27.8	A	6.7	B
500 ppm	15.1	A	103.2	A	12.2	A	34.0	B	7.0	A	1.1	A	2301	A	29.2	A	6.8	AB

[Z] Analysis performed using Waller-Duncan K-ratio t test - $P \leq 0.05$ - SAS statistical software (SAS, Institute, Inc., Cary, NC).

[Y] ppm = parts-per-million measured as Chloride.

[T] Electroconductivity (EC) measured in µS/cm.

[X] Calculated using the average of the height and two perpendicular widths.

[S] Measurements taken from new leaf growth.

[W] Measured in grams at harvest.

[R] Measured with a YSI Model 63 meter (YSI Inc., Yellow Springs OH).

[V] Measured in grams after oven drying for nine (9) days at 45° C (113°F).

[Q] Number of replicates = 5.

[U] Measured in parts-per-thousand (ppt).

[P] Number of replicates = 3.

[O] Means with different letters within columns are significantly different.

Table 4. Final results from NaCl treaments of hydroponic lettuce (*Lactuca sativa* L. 'Charles') from Experiment 1 August-September 2010[Z].

Treatment[Y]	Growth Index[XQ]		Fresh Shoot Weight[WQ]		Dry Shoot Weight[VQ]		Fresh Root Weight[WQ]		Dry Root Weight[VQ]		Salinity[UP]		EC[TP]		SPAD[SP]		pH[RP]	
0 ppm	20.2	A[O]	164.6	A	17.0	A	49.0	A	9.0	A	0.5	CD	967	DE	30.1	AB	7.2	A
50 ppm	20.1	A	151.2	A	16.3	A	47.8	A	8.9	A	0.4	D	892	E	31.1	A	7.2	A
75 ppm	19.5	A	147.0	A	16.0	A	48.2	A	9.0	A	0.6	C	1206	CD	30.7	AB	7.1	A
100 ppm	19.7	A	172.0	A	17.1	A	50.2	A	9.0	A	0.6	C	1262	C	29.2	AB	7.3	A
200 ppm	18.4	A	132.4	A	15.1	A	43.0	A	8.5	B	0.8	B	1696	B	27.9	AB	7.0	A
300 ppm	20.0	A	163.0	A	16.8	A	49.2	A	9.1	A	1.1	A	2185	A	26.5	B	7.1	A
400 ppm	18.1	A	129.6	A	14.8	A	43.8	A	8.8	AB	1.2	A	2317	A	28.1	AB	7.1	A
500 ppm	18.7	A	139.2	A	15.6	A	48.0	A	8.9	AB	1.2	A	2384	A	28.2	AB	7.1	A

[Z] Analysis performed using Waller-Duncan K-ratio t test P ≤ 0.05 - SAS statistical software (SAS, Institute, Inc., Cary, NC).

[Y] ppm = parts-per-million measured as Chloride.

[X] Calculated using the average of the height and two perpendicular widths.

[W] Measured in grams at harvest.

[V] Measured in grams after oven drying for nine (9) days at 45° C (113°F).

[U] Measured in parts-per-thousand (ppt).

[T] Electroconductivity (EC) measured in µS/cm.

[S] Measurements taken from new leaf growth.

[R] Measured with a YSI Model 63 meter (YSI Inc., Yellow Springs OH).

[Q] Number of replicates = 5.

[P] Number of replicates = 3.

[O] Means with different letters within columns are significantly different.

Table 5. Final results from $CaCl_2$ treaments of hydroponic lettuce (*Lactuca sativa* L. 'Charles') from Experiment 2 December 2010 - January 2011[Z].

Treatment[Y]	Growth Index[XQ]		Fresh Shoot Weight[WQ]		Dry Shoot Weight[VQ]		Fresh Root Weight[WQ]		Dry Root Weight[VQ]		Salinity[UQ]		EC[TQ]		SPAD[SQ]		pH[RQ]	
0 ppm	20.7	A[P]	131.5	AB	6.4	ABC	14.7	BC	2.4	AB	0.3	D	676	H	30.8	A	8.1	A
75 ppm	19.1	ABC	104.4	BCD	6.1	BC	12.6	C	2.4	AB	0.3	D	650	H	31.2	A	8.2	A
200 ppm	19.7	ABC	115.8	ABC	6.4	ABC	13.8	C	2.3	AB	0.4	D	939	GH	30.9	A	8.2	A
400 ppm	20.8	A	128.6	AB	7.8	A	19.1	A	2.7	A	0.5	D	1074	GH	31.7	A	8.1	A
600 ppm	20.8	A	142.3	A	7.5	AB	19.9	A	2.4	AB	0.9	CD	1730	FGH	30.7	A	7.9	B
800 ppm	20.2	AB	111.5	BC	6.1	BC	19.9	A	2.4	AB	1.1	CD	2189	EFG	31.0	A	7.9	B
1000 ppm	17.4	C	76.1	D	5.0	C	18.7	AB	2.7	A	1.1	CD	2101	EFG	33.4	A	7.9	B
1200 ppm	19.0	ABC	93.4	CD	6.0	BC	19.6	A	2.6	A	1.3	CD	2448	EF	32.6	A	7.8	BC
1500 ppm	20.3	AB	115.8	ABC	7.0	AB	18.9	A	2.5	A	2.0	CD	3183	DE	33.1	A	7.9	B
2000 ppm	17.9	BC	87.3	CD	5.2	C	12.5	C	2.0	B	2.9	C	4656	C	31.5	A	7.6	DE
3000 ppm	13.7	D	32.0	E	2.9	D	8.3	D	2.0	B	2.2	CD	3835	CD	32.5	A	7.7	CD
5000 ppm	8.6	E	4.5	F	1.0	E	4.3	D	1.4	C	2.9	C	4812	C	21.4	B	7.7	BCD
10,000 ppm	0.0	F[O]	0.0	F[O]	0.0	E[O]	0.0	E[O]	0.0	D[O]	9.8	B	14,925	B	0.0	C[O]	7.5	EF
20,000 ppm	0.0	F[O]	0.0	F[O]	0.0	E[O]	0.0	E[O]	0.0	D[O]	21.2	A	33,133	A	0.0	C[O]	7.3	F

[Z] Analysis performed using Waller-Duncan K-ratio t test P ≤ 0.05 - SAS statistical software (SAS, Institute, Inc., Cary, NC).

[Y] ppm = parts-per-million measured as Chloride.

[X] Calculated by taking the average of the height and two perpendicular widths.

[W] Measured in grams at harvest.

[V] Measured in grams after oven drying for nine (9) days at 45° C (113°F).

[U] Measured in parts-per-thousand (ppt).

[T] Electroconductivity (EC) measured in µS/cm.

[S] Measurements taken from new leaf growth.

[R] pH Measured with a YSI Model 63 meter (YSI Inc., Yellow Springs OH).

[Q] Number of replicates = 5.

[P] Means with different letters within columns are significantly different.

[O] Dead plant.

Table 6. Final results from NaCl treaments of hydroponic lettuce (*Lactuca sativa* L. 'Charles') from Experiment 2 December 2010 - January 2011[Z].

Treatment[Y]	Growth Index[XQ]		Fresh Shoot Weight[WQ]		Dry Shoot Weight[VQ]		Fresh Root Weight[WQ]		Dry Root Weight[VQ]		Salinity[UQ]		EC[TQ]		SPAD[SQ]		pH[RQ]	
0 ppm	20.7	A[P]	131.5	A	6.4	A	14.7	AB	2.4	A	0.3	H	676	H	30.8	CD	8.1	AB
75 ppm	19.1	ABC	111.4	ABC	6.6	A	10.2	C	2.1	ABC	0.3	H	696	H	31.2	CD	8.2	A
200 ppm	19.5	ABC	112.1	AB	6.2	A	13.5	ABC	2.4	A	0.5	GH	918	GH	32.0	BCD	8.1	ABC
400 ppm	19.3	ABC	109.5	ABC	5.7	A	16.1	AB	2.3	AB	0.6	GH	1156	GH	31.7	BCD	8.2	AB
600 ppm	19.8	ABC	106.7	BC	5.9	A	15.2	AB	2.3	AB	0.9	FGH	1756	FGH	29.9	D	8.0	BC
800 ppm	19.8	ABC	105.8	BC	5.7	A	14.3	AB	2.2	AB	1.0	FGH	1989	FGH	33.0	ABC	8.1	AB
1000 ppm	20.5	AB	121.4	AB	6.1	A	16.1	A	2.1	AB	1.4	EFGH	2673	EFG	31.1	CD	8.2	AB
1200 ppm	20.0	ABC	113.3	AB	5.7	A	14.7	AB	2.2	AB	1.6	EFG	3113	EF	29.5	D	8.0	ABC
1500 ppm	18.7	C	88.4	CD	5.5	A	12.2	BC	2.1	BC	2.0	EF	3863	E	30.4	CD	8.0	BC
2000 ppm	19.8	BC	75.3	D	3.7	B	12.2	ABC	2.1	ABC	2.3	DE	4332	E	30.1	D	8.1	ABC
3000 ppm	14.9	D	32.6	E	2.3	C	4.9	D	1.8	CD	3.4	CD	6399	D	34.1	AB	7.9	C
5000 ppm	12.1	E	12.3	EF	1.2	CD	2.7	DE	1.7	D	4.4	C	8585	C	34.7	A	7.9	C
10,000 ppm	0.0	F[O]	0.0	F[O]	0.0	D[O]	0.0	E[O]	0.0	E[O]	9.0	B	15,500	B	0.0	E[O]	7.9	C
20,000 ppm	0.0	F[O]	0.0	F[O]	0.0	D[O]	0.0	E[O]	0.0	E[O]	13.1	A	20,145	A	0.0	E[O]	7.9	C

[Z] Analysis performed using Waller-Duncan K-ratio t test P ≤ 0.05 - SAS statistical software (SAS, Institute, Inc., Cary, NC).

[Y] ppm = parts-per-million measured as Chloride.

[X] Calculated by taking the average of the height and two perpendicular widths.

[W] Measured in grams at harvest.

[V] Measured in grams after oven drying for nine (9) days at 45° C (113°F).

[U] Measured in parts-per-thousand (ppt).

[T] Electroconductivity (EC) measured in µS/cm.

[S] Measurements taken from new leaf growth.

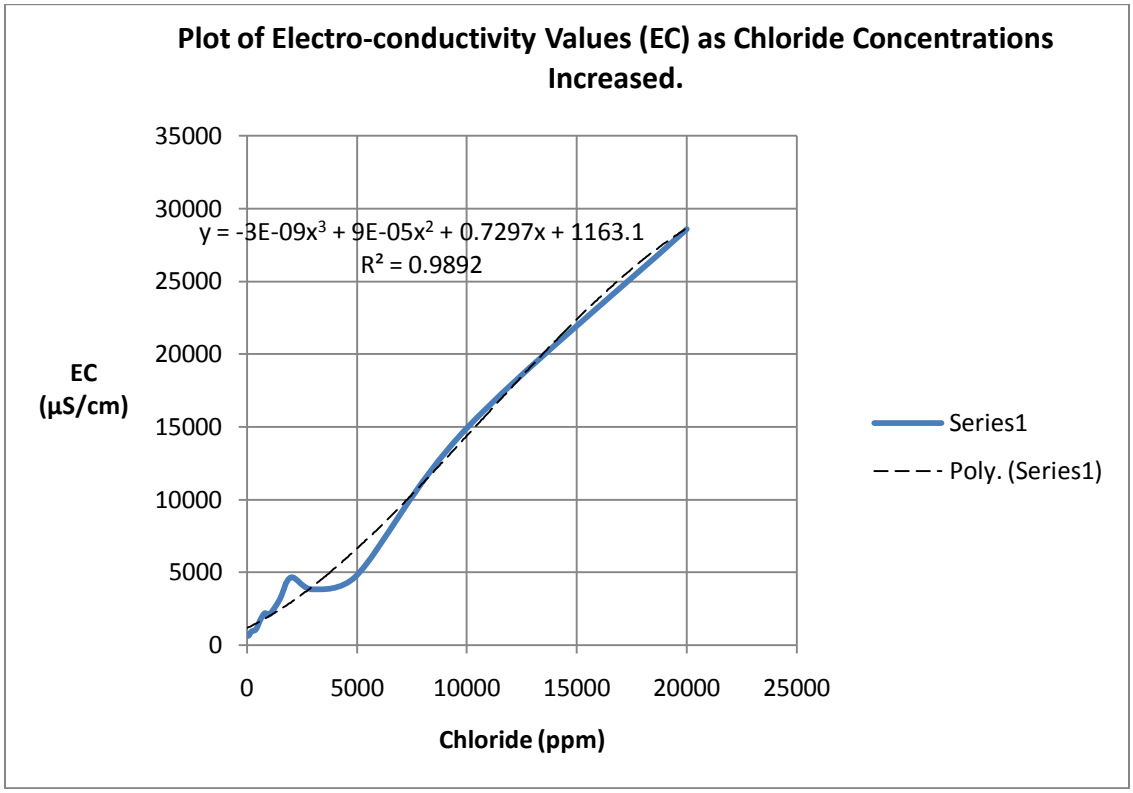
[R] pH Measured with a YSI Model 63 meter (YSI Inc., Yellow Springs OH).

[Q] Number of replicates = 5.

[P] Means with different letters within columns are significantly different.

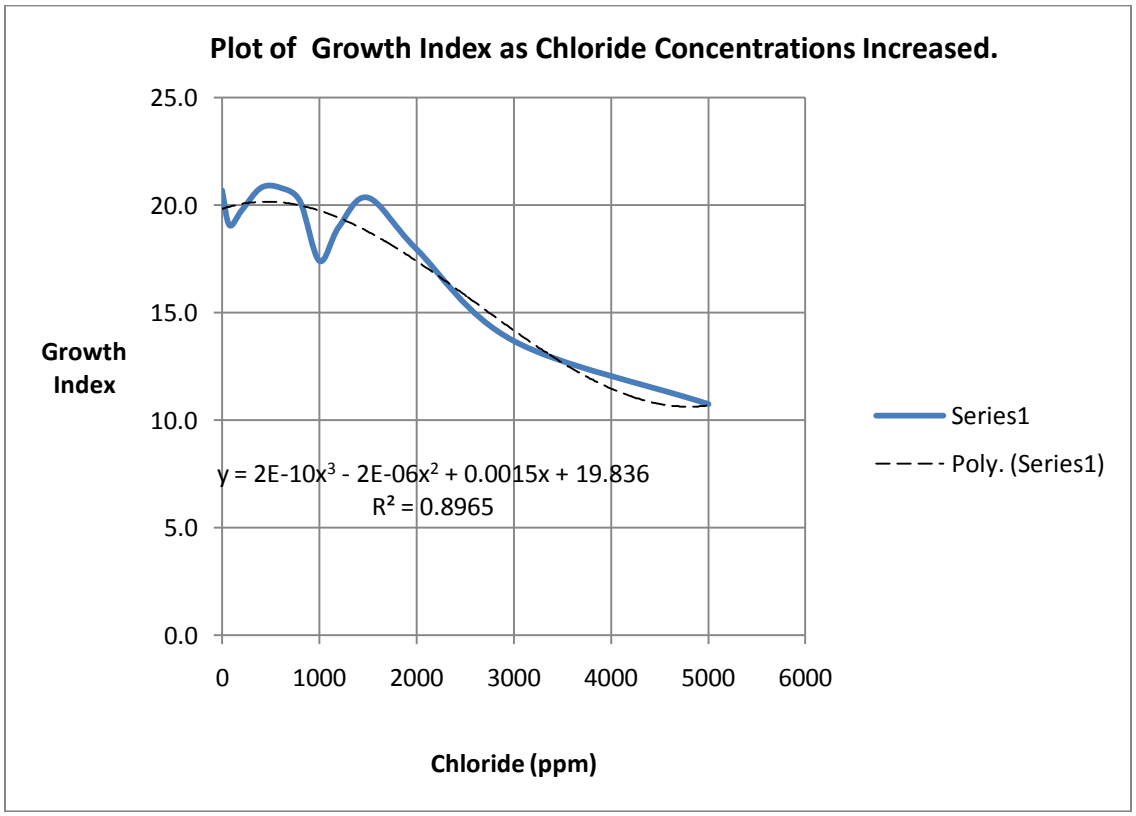
[O] Dead plant.

Figure 1. Plot of EC[Z] over chloride concentration for $CaCl_2$ treatments in lettuce experiments conducted in December 2010/January 2011[Y].



[Z] Electro-conductivity (EC) measured in µS/cm.
[Y] Prepared with Excel graph.

Figure 2. Plot of Growth Index[Z] over chloride concentration for $CaCl_2$ treatments in lettuce experiments conducted in December 2010/January 2011[Y].



[Z] Growth Index calculated by taking the average of the height and two perpendicular widths of the plants.
[Y] Prepared with Excel graph.

Figure 3. Plants from $CaCl_2$ treatments in lettuce experiments conducted in December 2010/January 2011. Concentrations of chloride shown are in ppm. First photo is from above. The second photo is the same plants shot at an angle.

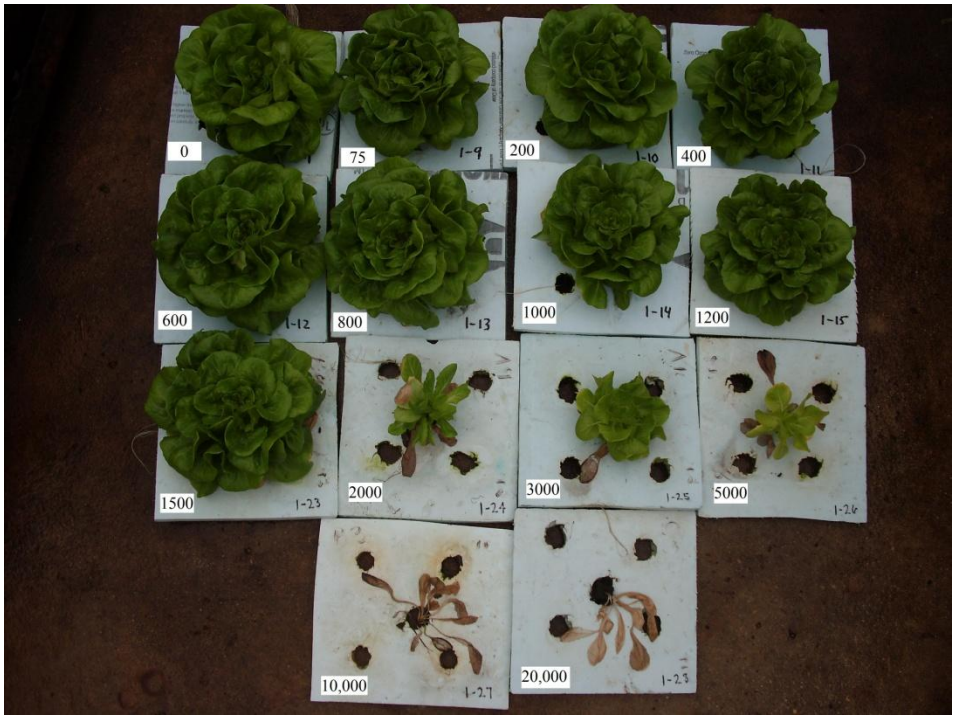
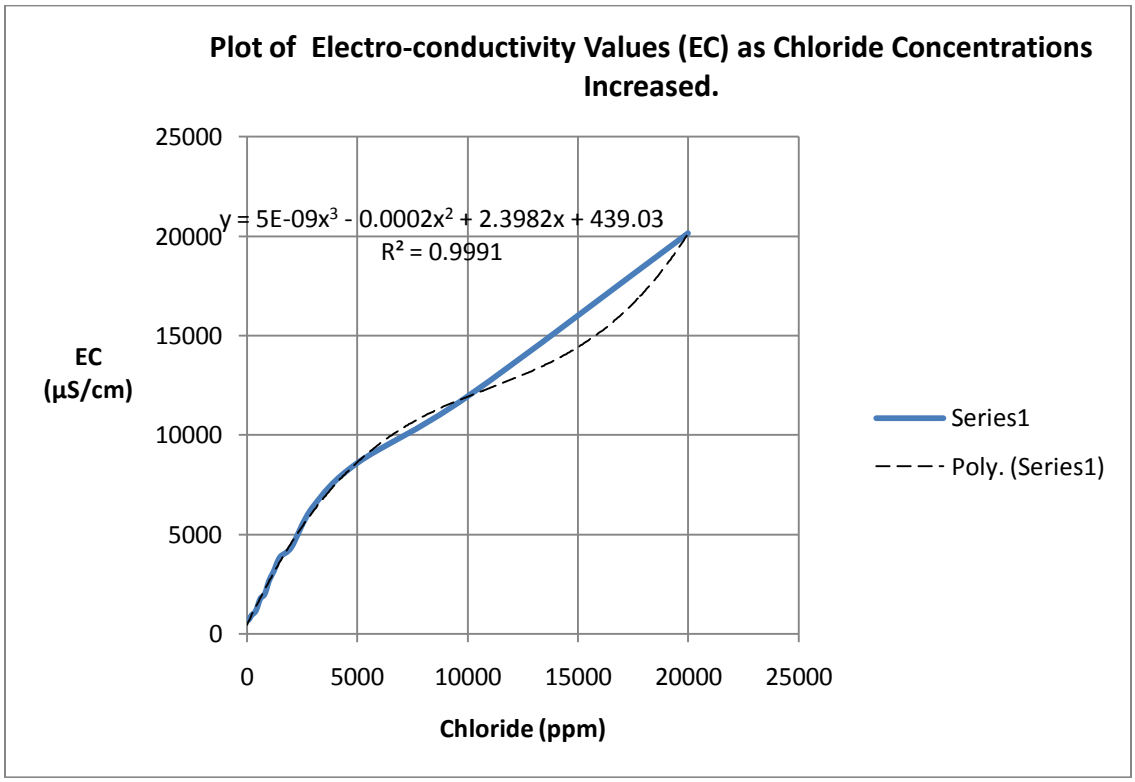
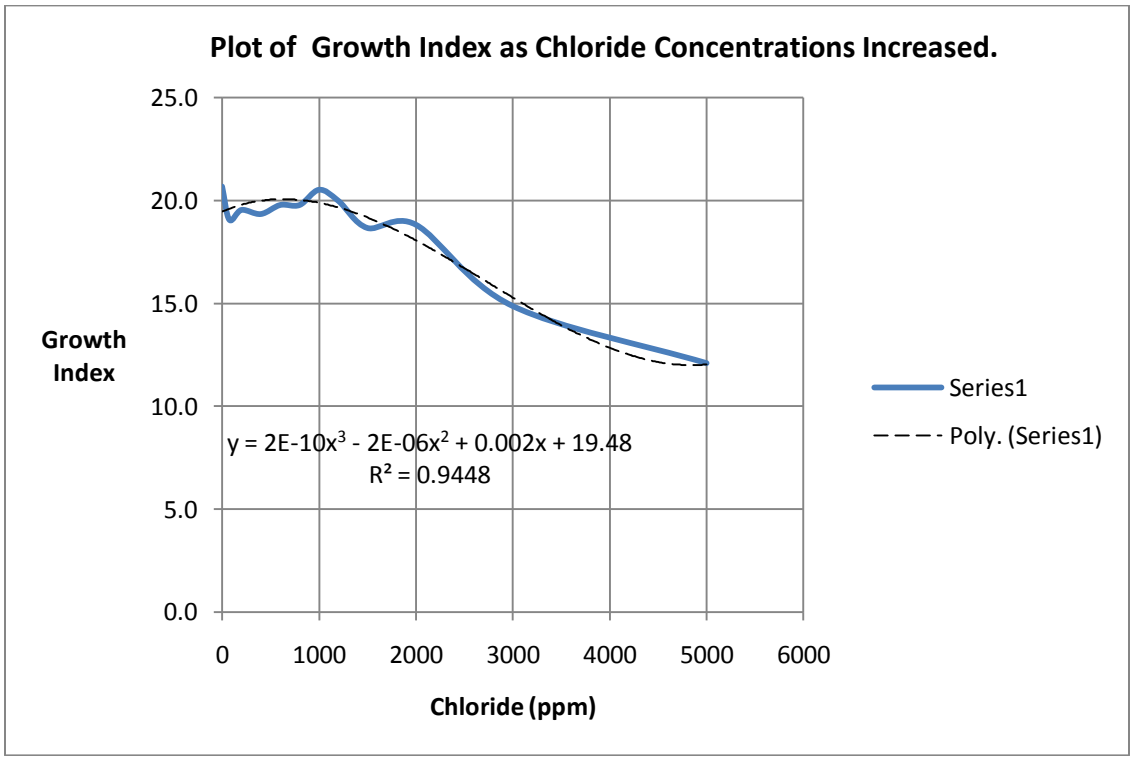


Figure 4. Plot of EC[Z] over chloride concentration for NaCl treatments in lettuce experiments conducted in December 2010/January 2011[Y].



[Z] Electro-conductivity (EC) measured in µS/cm.
[Y] Prepared with Excel graph.

Figure 5. Plot of Growth Index[Z] over chloride concentration for NaCl treatments in lettuce experiments conducted in December 2010/January 2011[Y].



[Z] Growth Index calculated by taking the average of the height and two perpendicular widths of the plants.
[Y] Prepared with Excel graph.

Figure 6. Plants from NaCl treatments in lettuce experiments conducted in December 2010/January 2011. Concentrations of chloride shown are in ppm. First photo is from above. The second photo is the same plants shot at an angle.

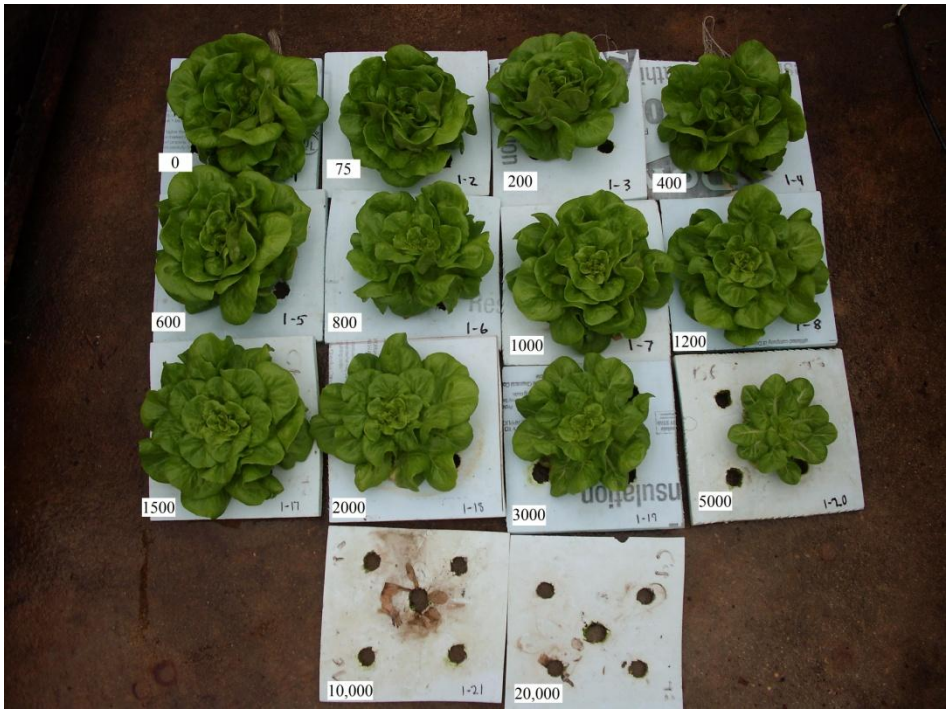
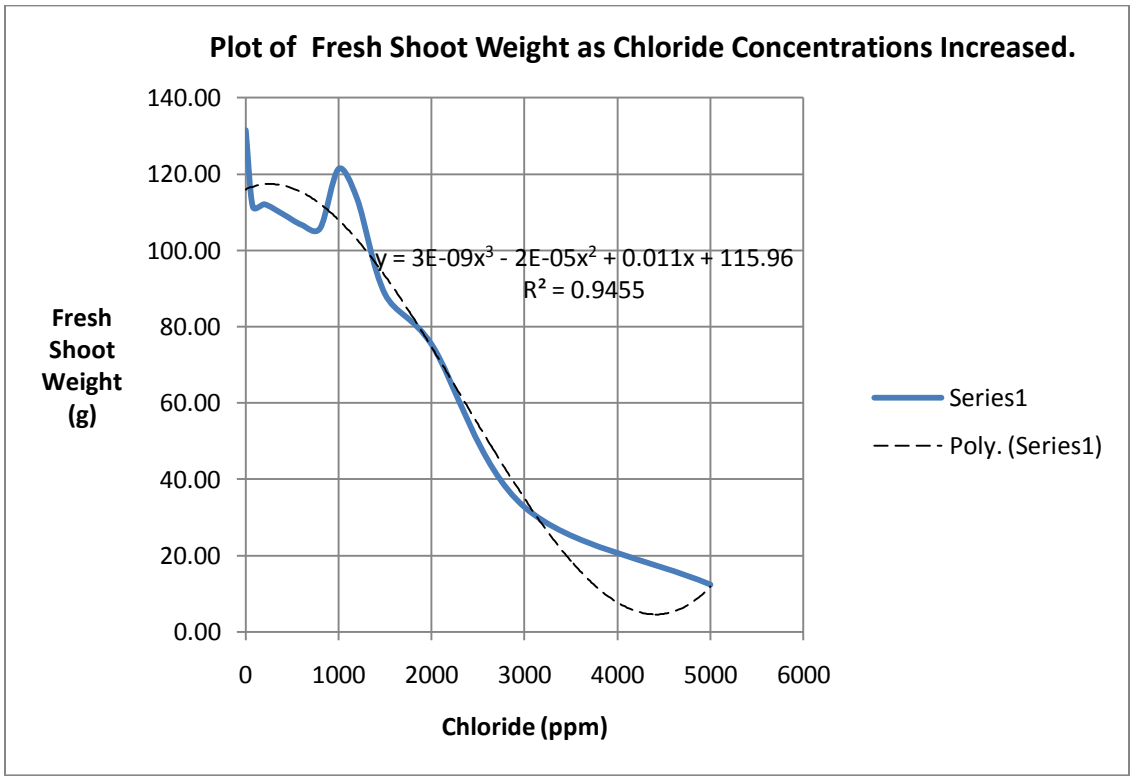
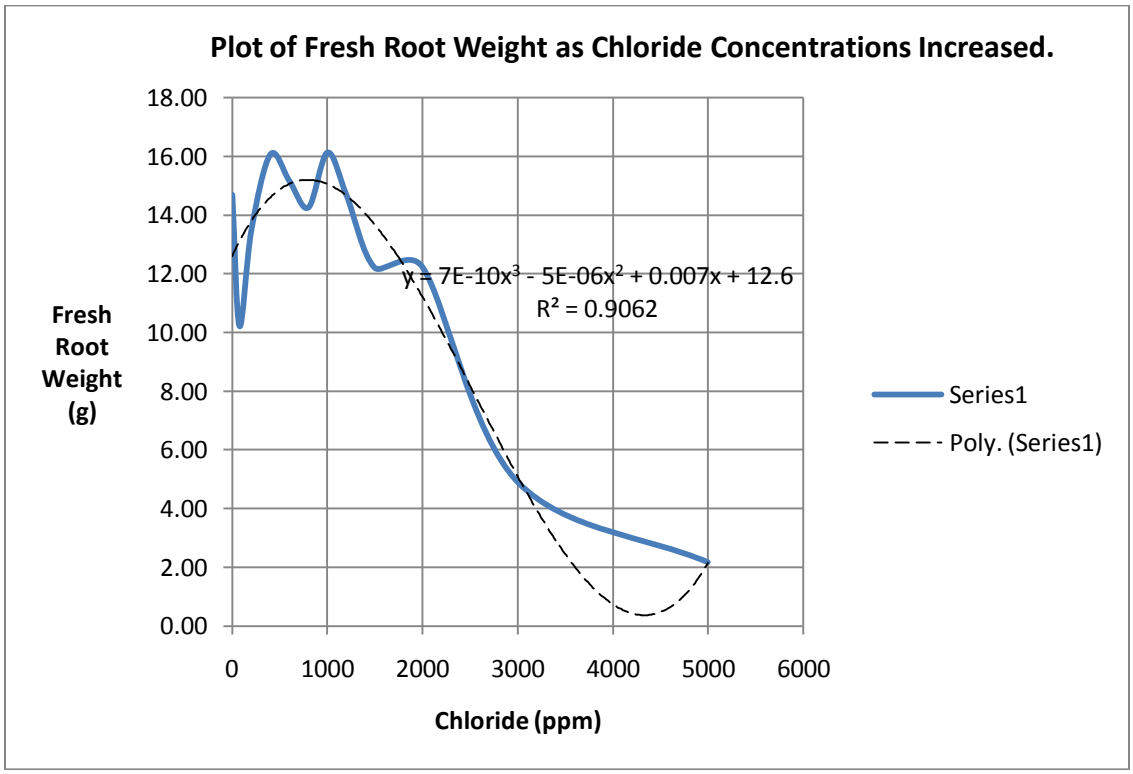


Figure 7. Plot of Fresh Shoot Weight[Z] over chloride concentration for NaCl treatments in lettuce experiments conducted in December 2010/January 2011[Y].



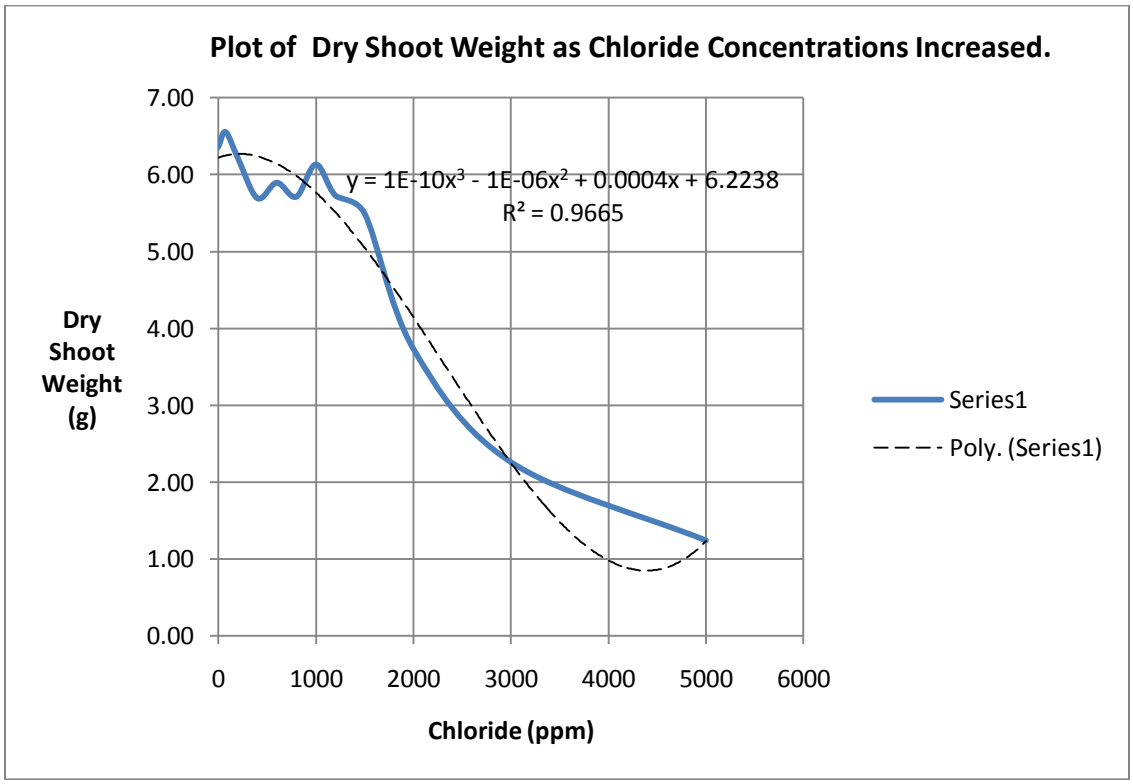
[Z] Measured in grams at harvest.
[Y] Prepared with Excel graph.

Figure 8. Plot of Fresh Root Weight[Z] over chloride concentration for NaCl treatments in lettuce experiments conducted in December 2010/January 2011[Y].



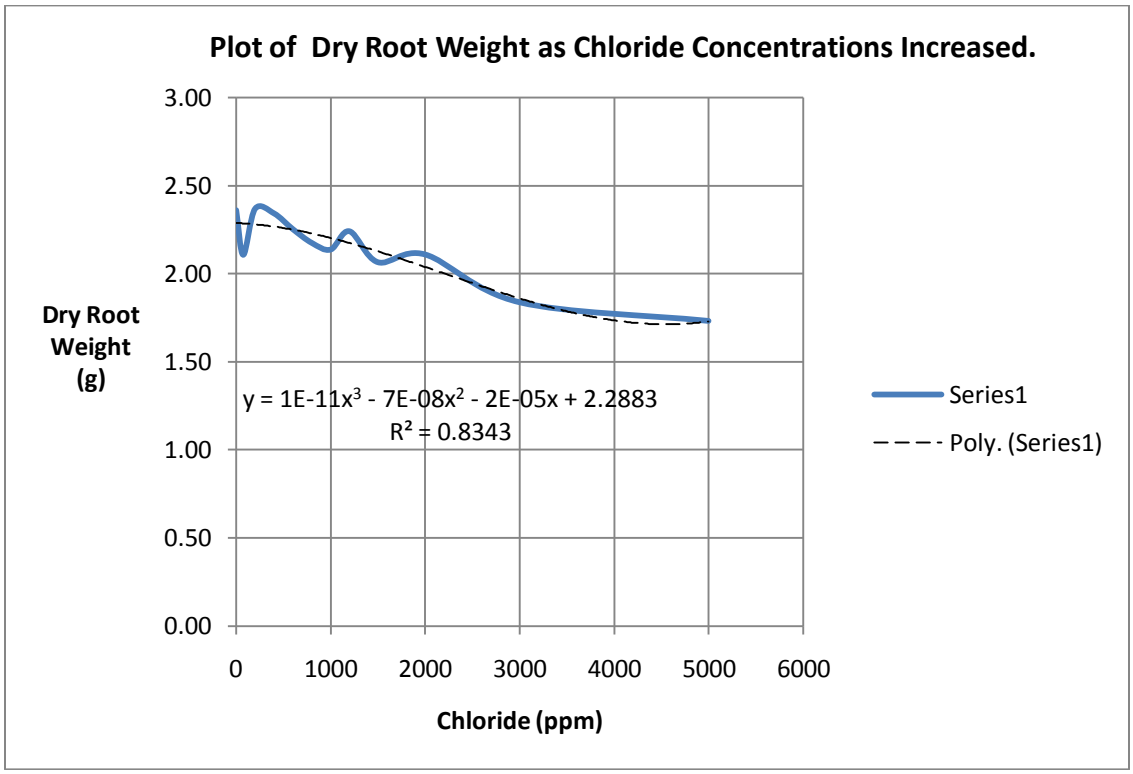
[Z] Measured in grams at harvest.
[Y] Prepared with Excel graph.

Figure 9. Plot of Dry Shoot Weight[Z] over chloride concentration for NaCl treatments in lettuce experiments conducted in December 2010/January 2011[Y].



[Z] Measured in grams after oven drying for nine (9) days at 45° C (113°F).
[Y] Prepared with Excel graph.

Figure 10. Plot of Dry Root Weight[Z] over chloride concentration for NaCl treatments in lettuce experiments conducted in December 2010/January 2011[Y].



[Z] Measured in grams after oven drying for nine (9) days at 45° C (113°F).
[Y] Prepared with Excel graph.