

Saline Irrigation of Five Diverse Landscape Plant Species for the Southeastern United States

by

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Abstract

Research was conducted to determine salt tolerance of five common landscape species, *Illicium parviflorum*, *Itea virginica* ‘Henry’s Garnet’, *Muhlenbergia capillaris*, *Begonia* × *semperflorens-cultorum* and *Portulaca oleracea*. Two experiments were performed, one with high NaCl concentrations and one with low NaCl concentrations. Plants received daily irrigation of tap water containing one of the following NaCl concentrations: 0 (tap water), 2000, 4000, 6000, 8000, or 10000 ppm ($\text{mg}\cdot\text{L}^{-1}$) (high NaCl conc. exp.) or 0 (tap water), 250, 500, or 1000 ppm ($\text{mg}\cdot\text{L}^{-1}$) (low NaCl conc. exp.). *Illicium parviflorum*, *M. capillaris*, *B. × semperflorens-cultorum*, and *P. oleracea* were tolerant of saline irrigation that could be expected from greywater. *Itea virginica* showed signs of salt stress at the lowest NaCl concentration [250 ppm ($\text{mg}\cdot\text{L}^{-1}$)] and should not be irrigated with saline greywater or other saline irrigation sources.

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Chapter I

Saline Irrigation Sources: A Review

Introduction

Water reuse has become increasingly important due to population increase and drought (Jordan et al., 2001). Alternative water sources provide an opportunity to reduce the demand for potable water. Greywater is defined as wastewater without any input from toilets. This includes water produced from bathtubs, showers, hand basins, laundry machines, and kitchen sinks. Greywater is less polluted than combined wastewater due to the absence of feces, urine, and toilet paper (Eriksson et al., 2002; Valee et al., 2010). The reuse of greywater shows the most promise when used for landscape irrigation or toilet flushing. Greywater irrigation can provide homeowners and municipalities with an alternative use for this resource instead of being wasted. However, few landscape species, commonly used in the southeastern United States, have been studied to determine the potential side effects of greywater.

Internationally, use of greywater has gained most support in areas that are under extreme drought. Research and application have been conducted in Sweden, Germany, Canada, Cyprus, Saudi Arabia, Jordan, and the United Kingdom. In the United States and Australia, some legislation has been established regulating greywater use as landscape irrigation (Valee et al., 2010). Each country has different reasons for promoting greywater use. Japan's greywater initiative was prompted by rising population density and small land area. The United States, Australia, Saudi Arabia, Cyprus, and Jordan have been searching for potable water alternatives for irrigation in response to prolonged drought. Most of the countries participating in greywater

research use the same model of application for toilet flushing and irrigation of crops or landscapes (Al-Jayyousi, 2003; Valee et al., 2010). Lu and Leung (2003) found that in Hong Kong, implementing greywater reuse would help reduce the city's reliance on water from the dwindling supply of the Dongjiang River. In Australia, water used while gardening accounted for approximately 34% of the total household water use budget, while toilet flushing accounted for approximately 20% (Christova-Boal et al., 1996). This presents a significant opportunity for recycled and alternative irrigation implementation.

Chemical characteristics of greywater

Chemical characteristics of greywater vary greatly depending on the quality of the greywater, the network of pipes that the water has traveled through (leaching from the pipes and biofilm on the inside of the pipes), and the activities of the household that produce the greywater. One common chemical characteristic of greywater is high sodium content, usually in the form of sodium chloride (NaCl). Reviews of several greywater studies found that sodium concentrations of greywater range from 7.4-480 mg·L⁻¹ while chlorine ranged from 9-88 mg·L⁻¹. (Christova-Boal et al., 1996; Eriksson et al., 2002). High concentrations of salinity in irrigation water can cause soil structure deterioration, decrease in soil permeability, and reduction of crop yield due to toxicity and osmotic effects (Al-Hamaiedeh and Bino, 2010). Christova-Boal et al. (1996) found that separating greywater produced from showers, baths, hand basins, and kitchen sinks from the more saline laundry waste water provided a greywater that is less saline. Laundry waste water is more saline due to the high salt content found in laundry detergents. By separating the greywater streams, it would be possible to use greywater from showers, baths, hand basins, and kitchen sinks for irrigation, while laundry greywater could then be used for toilet flushing.

Physiology of salt stress

High sodium content in greywater can produce salt stress when used to irrigate plants not adapted to saline environments, a characteristic of many plants used in southeastern U.S. landscapes. In a comparison of plant physiological responses of salt and water stress, plant responses to salt and water stress were similar (Munns, 2002). High salinity in irrigation water reduces a plant's ability to take up water and results in other metabolic changes in the plant. Including: reductions in leaf and root elongation, reduced final leaf size, reduced number of lateral shoots, altered flower time, and reduction in seed production. Salt taken up through the roots is usually accumulated in the leaves. In salt sensitive plants, salt can reach toxic levels causing premature senescence that eventually reduces the photosynthetic leaf area of the plant to a point that it cannot survive. Reductions in growth from salinity happen rapidly. If this reduction is only temporary, it could be followed by a gradual recovery at a reduced growth rate. Saline irrigation applied to wheat and barley caused premature floral initiation and reduced seed production (Munns and Rawson, 1999) Symptoms of salt injury in plants often resemble drought. Symptoms include wilting, reduced growth, stunting, and tissue death (Blaylock, 1994).

Boland (2008) identified four primary categories of salinity stress. These were osmotic effects, toxicity, nutritional imbalance, and altered soil structure. Osmotic effects result from salts increasing the osmotic potential of soil solution limiting the plants ability to take up water. Toxicity occurs when too much NaCl is in the soil, causing leaf burn, necrosis, and defoliation after NaCl ions are taken up by the plant. Severity of injury is typically proportionate to the rate of foliar accumulation. Nutritional imbalance is caused by high salinity inducing nutritional

deficiencies in the plant through replacement and competition. Soil structure decline is created by sodium in saline water causing the soil to decline in hydraulic conductivity and permeability.

Plants that have evolved to tolerate the effects of salt are known as halophytes (Flowers et al., 1977). The two main mechanisms of salt tolerance are those that reduce the rate of salt entering the plant, also known as salt exclusion, and the ability to minimize the concentration of salt accumulated in the cytoplasm, also known as ion compartmentalization (Cheeseman, 1988; Munns, 2002). Salt exclusion in plants prevents salt ions from entering the roots of the plant. Salt compartmentalization sequesters salt ions into cell vacuoles of the leaf. This compartmentalization within the vacuoles allows the leaf to have a high salt content while still functioning normally. This makes salt exclusion and/or compartmentalization imperative for plants that survive in saline environments.

Saline Landscape Irrigation

While it is understood that greywater contains high salinity, very little research has been done to study the effects of saline greywater on landscape plants (Cassaniti et al., 2009). Research conducted on food crops has been more focused on health concerns, such as biological contaminants, associated with the use of greywater rather than the ability to produce healthy, highly productive plants. Previous research did find that in tomato, lettuce, carrot, pepper, olive, bean, corn, sunflower, and millet, there was no reduction in growth when plants were irrigated with varying rates of treated and untreated greywater (Al-Hamaiedeh and Bino, 2010; Finley et al., 2009; Misra et al., 2010; Shanableh, 2012).

Greywater is not the only form of saline irrigation. Other saline irrigation sources include: municipally reclaimed water, saline groundwater, and agricultural drainage. Of these, municipally reclaimed water is the most common. Municipally reclaimed water is domestic water or municipal wastewater that has received secondary treatment (Boland, 2008; Cassaniti et al., 2012; Niu and Cabrera, 2010; Wu et al., 2001). The similarities in these forms of irrigation allow for crossover of greywater irrigation tolerance research to many irrigation sources. Plant species found to be tolerant of saline irrigation can potentially be recommended for landscapes affected by saline aerosols such as coastal regions and landscapes near roadways that receive deicing salts (Ferrante et al., 2011).

Salinity tolerance research conducted on landscape plant species has more commonly been conducted using artificial reclaimed wastewater (treated municipal effluent). This research has primarily been conducted in the arid climates of the American southwest and Australia where drought necessitates the reuse of water. Irrigation water in these areas is inherently saline and plants evaluated must be able to tolerate the increased salt from waste water. (Marcotte et al., 2004; Miyamoto et al., 2004; Niu et al., 2007, 2012; Niu and Rodriguez, 2006, 2010, 2012; Wu et al., 2000).

A study to determine the ability of common landscape plants of the southwestern United States used artificially created saline irrigation water (Miyamoto et al. 2004). Plants were irrigated with one of five levels of salinity; 800, 2000, 5000, 7500, or 10000 mg·L⁻¹, for 6 months. Results were analyzed to determine which level of salinity caused a 50% reduction in growth. Tested plants were then classified into five categories established by the US salinity laboratory classification: sensitive (0 – 3 dS m⁻¹), moderately sensitive (3 – 6 dS m⁻¹), moderately tolerant (6 – 8 dS m⁻¹), tolerant (8 – 10 dS m⁻¹) or highly tolerant (>10 dS m⁻¹).

Electrical conductivity (EC) values in salt tolerance classification were determined from soil saturation extract. This study found several species that are important to the southeastern United States with a US salinity laboratory classification level of tolerant or above. These species include: Bermuda grass [*Cynodon dactylon* (L.) Pers.], St. Augustine grass [*Stenotaphrum secundatum* (Walt.) Kuntze], Canary Island date palm (*Phoenix canariensis* Chabaud), and date palm (*Phoenix dactylifera* L.).

Research that incorporated reclaimed water irrigation into a commercial container nursery evaluated: vinca [*Catharanthus roseus* (L.) G. Don], salvia (*Salvia splendens* F. Sellow ex Roem. and Schult.), dwarf yaupon holly (*Ilex vomitoria* Ait. ‘Nana’), and Helliery holly (*Ilex crenata* Thunb. ‘Helliery’) for tolerance of reclaimed water irrigation (Yeager et al., 2010). Plants were irrigated with varying percentages of reclaimed water and deionized water. Shoot and root dry weights were measured at termination to determine growth. Researchers found that all plants were tolerant of reclaimed water irrigation, however it was stressed that growers monitor EC of container substrates to prevent salt accumulation.

In Texas, 10 herbaceous plants were drip irrigated with one of three saline solutions, 0.8 (tap water), 3.2, and 5.4 dS m⁻¹ (Niu et al., 2007). Salt solutions were created using a mix of sodium chloride, magnesium sulfate, and calcium chloride. This study identified several salt tolerant species that are frequently used in southeastern landscapes. These include: common Gaillardia (*Gaillardia aristata* Pursh), New Gold lantana (*Lantana* × *hybrid* L. ‘New Gold’), and Huntington Carpet Rosemary (*Rosmarinus officinalis* ‘Huntington Carpet’ L.). This research was similarly repeated with wildflowers native to the southwestern United States as well as annual bedding plants. Mexican Hat [*Ratibida columnifera* (Nutt.) Woot. & Standl.], Hooker’s Evening Primrose (*Oenothera elata* Kunth), mealycup sage (*Salvia farinacea* Benth.), Angelonia

(*Angelonia angustifolia* Benth.), ornamental pepper (*Capsicum annuum* L.), Helenium [*Helenium amarum* (Raf.) H. Rock], licorice plant (*Helichrysum petiolatum* Hilliard & B.L. Burt), Plumbago (*Plumbago auriculata* Lam.), snapdragon (*Antirrhinum majus* L.), Petunia (*Petunia* sp. Juss.), Verbena (*Verbena*×*hybrid*), zonal geranium (*Pelargonium*×*hortorum*), French marigold (*Tagetes patula* L.), Coleus [*Solenostemon scutellarioides* (L.) Codd], Fuchsia (*Fuchsia* sp. L.), and Rieger Begonia (*Begonia* × *hiemalis*) were identified to be moderately salt tolerant (Niu and Rodriguez, 2010, 2012; Villarino and Mattson, 2011).

An extensive greywater irrigation study conducted in Colorado concentrated on the long term effects of greywater irrigation on landscape plant growth and survival (Sharvelle et al., 2012). In a greenhouse experiment, four plant species {Meyer lemon (*Citrus*×*meyeri*), winter creeper *Euonymus* [*Euonymus fortunei* ‘Emerald Gaiety’ (Turcz.) Hand.-Maz.], tall fescue grass [*Festuca arundinacea* (Schreb.) Dumort., nom. cons.], and Bermuda grass (*Cynodon dactylon*)} were treated with synthetic greywater to reduce variability commonly observed in greywater. Synthetic greywater was created using the typical chemicals found in greywater, a combination of salts and surfactants and had an EC value of 1050 ($\mu\text{S}\cdot\text{cm}^{-1}$). Plant health was rated by collecting data on crown density, foliar color, turf quality, and above ground biomass. It was found that plants accumulated more biomass when treated with synthetic greywater, than when receiving plain tap water, and showed no signs of stress. This research also studied plant health in landscapes with greywater irrigation systems. Seven greywater irrigated landscapes were studied in the western United States. Of the species evaluated; common hackberry (*Celtis occidentalis* L.), fourwing saltbush [*Atriplex canescens* (Pursh) Nutt.], desert globemallow (*Sphaeralcea ambigua* A. Gray), honey mesquite (*Prosopis glandulosa* Torr.), hairyseed Bahia (*Bahia absinthifolia* Benth.), Juniper (*Juniperus* sp. L.), spindle tree (*Euonymus* sp. L.), rose of

Sharon (*Hibiscus syriacus* L.), daisy (*Chrysanthemum* L.), St. Augustine grass (*Stenotaphrum secundatum*), California valerian (*Valeriana californica* A. Heller), and Plum (*Prunus* sp. L.) were considered tolerant of greywater irrigation.

Methods of screening for plant salt tolerance focus on either greenhouse, outdoor container, or in field or landscape trials. Greenhouse trials provide for control over environmental factors, are less labor intensive, and less costly (Niu and Cabera, 2010). Outdoor container studies allow for outdoor environmental conditions while removing variation that can be found in field and landscape trials as well as allowing for easy removal of salinized substrates.

With current long term drought in the southeastern United States it is imperative to identify additional plant species that may be suitable for inclusion in greywater irrigated landscapes. As indicated above, plants previously studied are adapted for use in arid regions and are not suited for conditions in the hot and humid southeast. Further evaluation of species for tolerance of greywater salinity levels need to be conducted. Greywater has the capability of reducing the requirement of potable water by more than 50 percent (Christova-Boal et al., 1996). Studying plant species that can be irrigated with greywater gives the opportunity to put greywater systems into use and limit current waste of potable water.

Therefore the objectives of this research are to evaluate the tolerance of six landscape plants, commonly used in the southeastern United States, to saline irrigation water.

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Chapter II

Saline Irrigation Affects Growth and Leaf Tissue Chemical Composition of Three Native Landscape Plants

Index Words: Sustainability, greywater, water, sodium chloride, wastewater

Abstract: Drought and population growth strain the potable water supply in the southeastern United States. Greywater is a renewable irrigation alternative to potable water, however, its use as an irrigation source is limited by the potential for salt injury to plants. Research was conducted to determine the salt tolerance of three common landscape species, *Illicium parviflorum*, *Itea virginica*, ‘Henry’s Garnet’, and *Muhlenbergia capillaris*. Two experiments were performed, one with high NaCl concentrations and one with low NaCl concentrations. Plants received daily irrigation of tap water containing one of the following NaCl concentrations: 0 (tap water), 2000, 4000, 6000, 8000, or 10000 ppm ($\text{mg}\cdot\text{L}^{-1}$) (high NaCl conc. exp.) or 0 (tap water), 250, 500, or 1000 ppm ($\text{mg}\cdot\text{L}^{-1}$) (low NaCl conc. exp.) for 15 weeks. Plants were harvested after 5, 10 or 15 weeks. Root dry weight (RDW) and shoot dry weight (SDW) were determined at each harvest; survival was determined at experiment termination. Leaf tissue was analyzed for tissue macronutrient (N, P, K, Ca, and Mg), Na, and Cl concentrations in high NaCl concentration experiment. Substrate leachate pH and electrical conductivity (EC) were measured at termination in low NaCl concentration experiment. Experiments took place from June 2011 – Jan. 2013. In high NaCl concentration experiment RDW and SDW decreased with increasing NaCl in all species and harvest dates with the exception of *M. capillaris*. Mortality was observed in *I. parviflorum* and *I. virginica* at 8000 and 10000 ppm ($\text{mg}\cdot\text{L}^{-1}$). In general, leaf macronutrient, Na, and Cl increased with increasing NaCl concentration. In low NaCl

concentration experiment there was no effect of NaCl concentration on RDW or SDW at the majority of harvest dates in all species. All three species continued to grow between harvest dates in lower NaCl concentration experiment. Electrical conductivity increased linearly with increasing NaCl concentration in all species. *Illicium parviflorum* and *M. capillaris* were tolerant of saline irrigation that could be expected from greywater. *Itea virginica* showed symptoms of salt stress at the lowest NaCl concentration [250 ppm ($\text{mg}\cdot\text{L}^{-1}$)] and should not be irrigated with saline water.

Introduction

Alternative water sources provide an opportunity to reduce the demand for potable water. Greywater irrigation can provide homeowners and municipalities with an alternative use for this resource instead of it being wasted. One common chemical characteristic of greywater is high salt content, usually in the form of sodium chloride (NaCl). Reviews of several greywater studies indicate that sodium concentrations of greywater range from 7.4-480 ppm ($\text{mg}\cdot\text{L}^{-1}$), while chlorine concentrations range from 9-88 ppm ($\text{mg}\cdot\text{L}^{-1}$) (Christova-Boal et al., 1996; Eriksson et al., 2002). Salinity tolerance research conducted on landscape plant species has commonly been conducted using simulated reclaimed wastewater (treated municipal effluent) (Marcotte et al., 2004; Miyamoto et al., 2004; Niu et al., 2006, 2007; Niu and Rodriguez, 2012; Wu et al., 2000).

Limited information is available on salt tolerance of woody landscape species native to the southeastern United States (Jordan et al. 2001, Wu et al. 2001). Previous evaluation of salt tolerance of woody landscape plant species commonly used in the southeastern United States has utilized both native and non-native species. For example, *Ilex crenata* Thunb. ‘Helleri’ (Helleri holly), *Ilex crenata* Thunb. (Japanese holly), *Ilex vomitoria* Aiton (yaupon holly), *Juniperus*

chinensis L. (Chinese juniper), *Cercis canadensis* L. var. *mexicana* (Rose) M. Hopkins (Mexican red bud), and *Lagerstroemia* sp. L. (crapemyrtle) have all been screened and found to be tolerant of saline irrigation water (Cabrera, 2009; Niu et al. 2010; Valdez-Aguilar et al., 2011; Yeager et al., 2010). Evaluations of monocots have been mostly limited to palms and turf with few ornamental landscape grasses (Marcotte et al., 2004; Miyamoto et al., 2004; Niu et al., 2007; Niu and Rodriguez, 2006, 2010, 2012; Niu et al., 2012; Wu et al., 2000). Evaluation of additional landscape species, native to the southeastern United States, for tolerance of greywater salinity could increase plant selection options for a greywater irrigated landscape. Therefore, the objective of this research was to evaluate the tolerance of three landscape plant species, native to the southeastern United States, to saline irrigation water.

Materials and Methods

High Salt Concentrations

Run I

Liners [2in (5.1 cm)] of *Illicium parviflorum* Michx. ex Vent. (small anise tree)(propagated at Auburn University), *Itea virginica* L. ‘Henry’s Garnet’ (Virginia sweetspire) (Spring Meadow Nursery, Inc., Grand Haven, MI), and *Muhlenbergia capillaris* (Lam.) Trin. (pink muhly grass) (Magnolia Gardens Nursery, Magnolia, TX) were planted in 2.5 L (trade gallon) containers in a 5:3:1 pinebark:peat:perlite, by volume, substrate. Substrate was pre-plant amended with $9.1 \text{ lb}\cdot\text{yd}^{-3}$ ($4.52 \text{ kg}\cdot\text{m}^{-3}$) controlled-release fertilizer [Polyon with micros 17N-5P₂O₅-11K₂O (Harrell’s LLC, Lakeland, FL)], and dolomitic limestone [$4 \text{ lb}\cdot\text{yd}^{-3}$ ($1.8 \text{ kg}\cdot\text{m}^{-3}$)]. Plants were irrigated by hand daily with 10 oz (300 mL) of tap water containing one of the following concentrations (treatments) of sodium chloride (NaCl): 0 (tap water), 2000, 4000, 8000, or 1000 ppm ($\text{mg}\cdot\text{L}^{-1}$) resulting in approximately 10 – 15% leachate (SNA, 2007). While

these rates are much higher than those normally found in greywater, they were used so that results could be applicable to other more saline irrigation sources (Christova-Boal et al., 1996; Eriksson et al., 2002). Plants received tap water irrigation (no NaCl) on weekends. Plants were grown under natural photoperiods on raised benches in a polycarbonate greenhouse at Paterson Horticulture Teaching and Research Greenhouse Complex at Auburn University in Auburn, AL. Temperatures ranged from 72 - 65°f during the day and 80 - 67°f at night.

There were ten single container replications per treatment per species. Experimental design was a completely randomized design with each species in a separate experiment. Three plants of each species in each treatment were harvested 5 and 10 weeks after treatment initiation, and the remaining plants (n=4) were harvested 15 weeks after treatment initiation (experiment termination). Root dry weight (RDW), and shoot dry weight (SDW) were determined at each harvest, survival was determined at experiment termination. Recently matured leaf tissue samples were collected from SDW samples of three plants per treatment per species at experiment termination and analyzed for tissue macronutrient (N, P, K, Ca, and Mg), Na, and Cl concentrations using Inductively Coupled Plasma analysis (Brookside Laboratories Inc., New Bremen, OH). Treatments were initiated on 20 June 2011, and experiments were terminated on 30 Sept. 2011. Data were subjected to analysis of variance and regression analysis using PROC GLM in SAS (SAS Institute, Cary, NC).

Run II

On 12 Sept. 2011, Run I was repeated with the same procedures described above with only, *I. parviflorum* and *M. capillaris*, and with the omission of leaf tissue nutrient analysis. *Itea virginica* was omitted based on its lack of tolerance to NaCl concentrations found in run 1. Run II was terminated on 19 Dec. 2011.

Low Salt Concentrations

Run I

On 10 July 2012, an experiment similar to the high salt concentration experiment described above was initiated. Based on results from high salt concentration experiment, NaCl treatments were decreased to 0 (tap water), 250, 500, or 1000 ppm ($\text{mg}\cdot\text{L}^{-1}$) to evaluate seemingly less salt tolerant *I. virginica* and *I. parviflorum*. *Muhlenbergia capillaris* was also evaluated. Substrate pH and electrical conductivity (EC) were measured 25 Oct. 2012 (termination) from leachate collected using the pour-through nutrient extraction procedure (Wright, 1986). All other materials and methods were the same as described above with the omission of tissue analysis.

Run II

Low salt concentration experiment was repeated with the same procedures on 15 Oct. 2012; experiment termination was on 30 Jan. 2013.

Results

High Salt Concentrations

Illicium parviflorum

Root dry weight and SDW decreased linearly with increasing NaCl concentration at each harvest date (Figs. 1-2). There was no mortality at any NaCl concentration. Leaf macronutrient, Na, and Cl concentrations increased linearly with increasing NaCl concentration (Table 1). Leaf Ca concentration responded quadratically, initially increasing then decreasing, with increasing NaCl concentration. Leaf Mg concentration also responded quadratically, initially decreasing then increasing with increasing NaCl concentration. When repeated, there was a linear reduction in SDW with increasing NaCl concentration at week 5 (first harvest date) (Fig. 3). There was no

effect of treatment on RDW or SDW at other harvest dates. There was 10% mortality at 8000 ppm ($\text{mg}\cdot\text{L}^{-1}$) and 80% mortality at 10000 ppm ($\text{mg}\cdot\text{L}^{-1}$) at termination.

Itea virginica ‘Henry’s Garnet’

Root dry weight and SDW decreased linearly with increasing NaCl concentration at each harvest date (Figs. 1-2). Foliar damage was observed visually at lowest NaCl concentration of 2000 ppm ($\text{mg}\cdot\text{L}^{-1}$). There was 100% plant mortality at NaCl concentrations of 8000 and 10000 ppm ($\text{mg}\cdot\text{L}^{-1}$) at experiment termination. Plants irrigated with NaCl concentrations higher than 4000 ppm lacked sufficient leaf tissue for nutrient analysis. Leaf macronutrient, Na, and Cl concentrations increased linearly with increasing NaCl concentration (Table 1).

Muhlenbergia capillaris

Root dry weight decreased linearly with increasing NaCl concentration at weeks 10 and 15, however there was no effect of NaCl concentration at week 5. (Fig. 1). Shoot dry weight decreased linearly with increasing NaCl concentration at week 5, while there was no effect of NaCl concentration at weeks 10 and 15 (Fig. 2). Leaf N, P, Na, and Cl concentrations increased linearly with increasing NaCl concentration, while there was no effect on leaf K, Ca, and Mg concentrations (Table 1).

When repeated there was a linear reduction in RDW and SDW at final harvest (termination), however there was no effect of treatment on RDW and SDW at other harvest dates. (Fig. 3).

Low Salt Concentrations

There was no effect of NaCl concentration on RDW or SDW at any harvest date for all species during the first run (Figs. 4-5). When the experiment was repeated, there was a linear reduction in RDW of *I. virginica* with increasing NaCl concentration at week 10 (Fig. 6).

Illicium parviflorum SDW at week 10 responded quadratically, initially increasing then decreasing, with increasing NaCl concentration. *Itea virginica* SDW at week 5 responded quadratically, initially decreasing then increasing, with increasing NaCl concentration. *Muhlenbergia capillaris* substrate leachate pH increased linearly, with increasing NaCl concentration in both runs (Fig. 8). There was no effect of NaCl concentration on substrate pH with *I. parviflorum* or *I. virginica*. Electrical conductivity increased linearly with increasing NaCl concentration in all species and runs (Fig. 9).

Discussion

Increases in leaf macronutrient, Na, and Cl concentrations with increasing NaCl concentration observed in all species are likely attributed to a reduction in SDW with increasing NaCl concentration (Fig. 2, Table 1). *M. capillaris* had 100% survival even at NaCl irrigation rates of up to 20 times higher than generally found in greywater (Christova-Boal et al., 1996; Eriksson et al., 2002). *Illicium parviflorum* had 100% survival in all experiments with the exception of one run, where it had survival rates of 90% at 6000 ppm ($\text{mg}\cdot\text{L}^{-1}$), 80% at 8000 ppm ($\text{mg}\cdot\text{L}^{-1}$), and 20% at 10000 ppm ($\text{mg}\cdot\text{L}^{-1}$). Generally, plants that can tolerate saline irrigation levels of 2000-3000 ppm ($\text{mg}\cdot\text{L}^{-1}$) are considered to be salt tolerant (Watling, 2007). High survival rates of *I. parviflorum* and *M. capillaris* are likely due to their native habitats' proximity to saline environments (Dirr, 1998; Schroeder et al., 1976). *Muhlenbergia capillaris* leaf Na and Cl concentrations were up to 10 times higher in plants irrigated with the highest NaCl concentrations than in plants irrigated with tap water (Table 1). Likewise, *Illicium parviflorum* irrigated with 10000 ppm ($\text{mg}\cdot\text{L}^{-1}$) had leaf Na and Cl concentrations 200 and 50 times higher, respectively, than plants irrigated with tap water (Table 1). *Itea virginica* leaf concentrations of Na and Cl were 4 and 15 times higher, respectively, in plants irrigated with 4000 ppm ($\text{mg}\cdot\text{L}^{-1}$)

NaCl than plants irrigated with tap water (Table 1). The tolerance of these three species to saline irrigation may be related, at least in part, to their ability to accumulate Na and Cl in leaf tissue without toxicity as suggested by foliar damage of *I. virginica*, but not *I. parviflorum* or *M. capillaris*.

Differences in SDW and RDW between runs 1 and 2 of high NaCl concentration experiments are likely due to time of year of experimentation. While run 1 was grown in the summer and early fall (20 June 2011 - 30 Sept. 2011), run 2 was grown in late fall and early winter (On 12 Sept. 2011 - 19 Dec. 2011) when plant growth rates tend to be slower, even in a heated greenhouse.

Illicium parviflorum and *M. capillaris* continued to grow throughout all experiments despite prolonged (15 weeks) application of NaCl concentrations suggesting these species would be appropriate candidates for use in greywater irrigated landscapes. Additionally, despite growth reductions, both species still appeared “marketable” (healthy foliage and roots) by the end of each experiment indicating they could also be irrigated with saline water during production. Conversely, even at lower concentrations of NaCl, *I. virginica* exhibited growth reductions and tissue damage, indicating it may not be suitable for inclusion in a landscape receiving frequent greywater irrigation or for highly saline landscapes. Symptoms of salt stress observed in *I. virginica* included wilting, chlorosis, leaf necrosis, defoliation, and root damage.

Elevated EC levels observed in *I. parviflorum* and *I. virginica* irrigated with 0 ppm ($\text{mg}\cdot\text{L}^{-1}$) NaCl concentration can be attributed to lower growth rates and thus lower nutrient uptake resulting in increased dissolved salts in the substrate solution. Electrical conductivity was however within sufficiency ranges (Reed, 1996). Substrate EC above $6 \text{ ds}\cdot\text{m}^{-1}$ are considered highly saline, while $2 - 6 \text{ ds}\cdot\text{m}^{-1}$ are considered moderately saline (Watling, 2007). Electrical

conductivity in the lower NaCl concentration experiments were within the range of moderately saline, with the exception of 1000 ppm ($\text{mg}\cdot\text{L}^{-1}$) which were highly saline. Elevations in EC of substrate leachate with increasing NaCl concentrations were expected (Fig. 9).

Weekend irrigation of all plants with tap water (no NaCl) was used to mimic rain events or an irrigation cycle of tap water and appeared to aid in the prevention of NaCl accumulation in substrate. This technique could be used as a management tool in greywater irrigated landscapes. Periodically irrigating with tap water could reduce salt accumulation in soil or substrate. Likewise, less salt-tolerant plants could still be used in greywater irrigated landscapes if frequency of application is reduced.

There are concerns other than NaCl associated with greywater application. Surfactants, phosphorus, lint, and biological contaminants all represent potential environmental problems (Al-Hamaiedeh and Bino, 2010; Christova-Boal et al., 1996; Eriksson et al. 2002; Gross et al., 2005). Some of these concerns can be mitigated with subsurface irrigation and decreasing frequency of application. Research to address these concerns could be coupled with results of this work to facilitate greywater irrigation implementation in the landscape as well as future legislation. This research evaluated plant tolerance of a range of NaCl concentrations beyond those observed in greywater application. Responses to NaCl concentrations used in these experiments could also be applied to conditions that would be observed when irrigating with reclaimed wastewater or collected stormwater, or in landscapes in coastal regions and perhaps even arid environments. Additional research to identify native landscape species that are salt tolerant will have application for utilizing greywater for landscape irrigation as well as utilizing saline irrigation water in general. It is likely that potential plant species for use in greywater irrigated landscapes could be identified from coastal environments and species with drought

tolerance, for example, *Conradina canescens* (Torr. & A. Gray ex Benth.) A. Gray (false rosemary), *Morella cerifera* (L.) Small (southern wax myrtle), and *Uniola paniculata* L. (seaoats).

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Table 1. Effect of NaCl concentration in irrigation water on concentration of macronutrients, sodium, and chlorine in leaves of *Illicium parviflorum*, *Itea virginica*, and *Muhlenbergia capillaris* grown containers in a greenhouse in Auburn, AL.

| <i>I. parviflorum</i> | | | | | | | |
|----------------------------|---------------------------------------|-----------|-----------|-----------|-----------|--------------|------|
| NaCl (mg•L ⁻¹) | Leaf tissue element concentration (%) | | | | | | |
| | N | P | K | Ca | Mg | Na | Cl |
| 0 | 1.11 | 0.10 | 0.74 | 0.17 | 0.12 | 0.01 | 0.07 |
| 2000 | 1.23 | 0.12 | 0.82 | 0.15 | 0.11 | 0.11 | 0.32 |
| 4000 | 1.85 | 0.19 | 0.96 | 0.24 | 0.12 | 0.50 | 0.88 |
| 6000 | 2.19 | 0.20 | 0.87 | 0.18 | 0.09 | 0.60 | 0.96 |
| 8000 | 1.96 | 0.19 | 0.95 | 0.16 | 0.11 | 0.96 | 1.63 |
| 10000 | 2.27 | 0.19 | 1.10 | 0.18 | 0.12 | 2.26 | 4.07 |
| | L*** ^z | L*** | L** | Q* | Q* | L*** | L*** |
| Sufficiency Range | 0.99-2.08 | 0.12-0.18 | 0.67-0.95 | 0.20-0.28 | 0.11-0.15 | 0.0057-0.011 | N/A |
| <i>I. virginica</i> | | | | | | | |
| NaCl (mg•L ⁻¹) | N | P | K | Ca | Mg | Na | Cl |
| 0 | 1.13 | 0.09 | 0.51 | 0.53 | 0.22 | 0.04 | 0.21 |
| 2000 | 1.28 | 0.15 | 1.06 | 0.79 | 0.32 | 0.17 | 1.99 |
| 4000 | 1.66 | 0.23 | 1.07 | 0.94 | 0.35 | 0.68 | 3.26 |
| | L** | L*** | L*** | L*** | L*** | L*** | L*** |
| Sufficiency Range | 1.64-4.50 | 0.10-0.24 | 0.36-0.80 | 0.38-1.09 | 0.13-0.20 | 0.003-0.0139 | N/A |
| <i>M. capillaris</i> | | | | | | | |
| NaCl (mg•L ⁻¹) | N | P | K | Ca | Mg | Na | Cl |
| 0 | 0.64 | 0.16 | 0.77 | 0.18 | 0.12 | 0.24 | 0.54 |
| 2000 | 0.79 | 0.16 | 0.68 | 0.18 | 0.11 | 0.96 | 1.56 |
| 4000 | 0.65 | 0.15 | 0.54 | 0.11 | 0.08 | 1.17 | 1.74 |
| 6000 | 0.89 | 0.23 | 0.76 | 0.17 | 0.11 | 2.66 | 4.76 |
| 8000 | 0.91 | 0.22 | 0.82 | 0.18 | 0.11 | 3.39 | 6.07 |
| 10000 | 0.87 | 0.20 | 0.70 | 0.16 | 0.10 | 3.14 | 5.93 |
| | L** | L** | NS | NS | NS | L*** | L*** |

^zSignificance (SAS Institute, Cary, NC) of linear (L) or quadratic (Q) trend associated with effect of NaCl concentration in irrigation water on leaf tissue element concentration (n=3); trends were significant at $P \leq 0.05$ (*), 0.01 (**), 0.001(***), or not significant (NS).

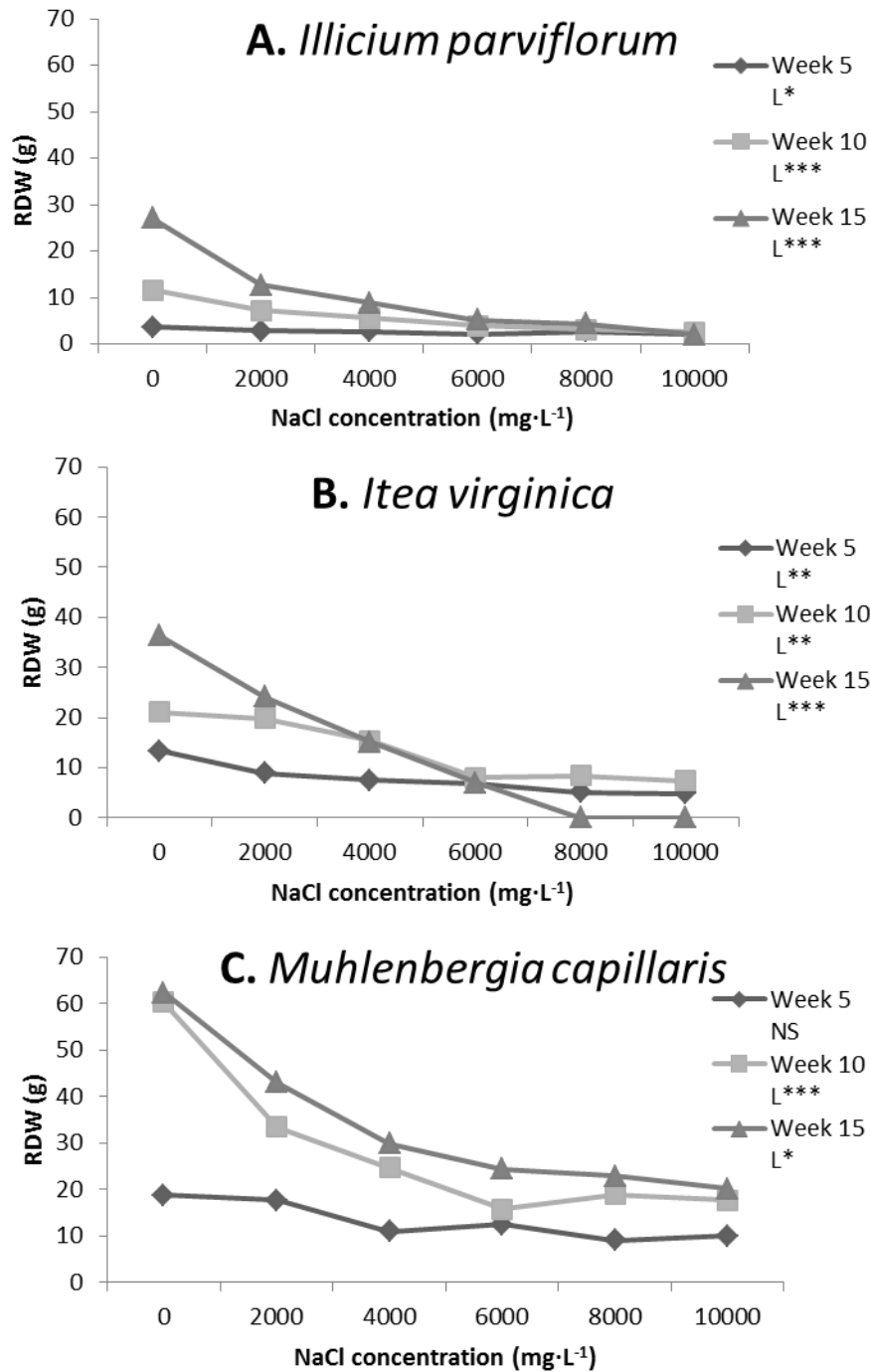


Figure 1. Effect of NaCl concentration in irrigation water on root dry weight (RDW) of A) *Illicium parviflorum*, B) *Itea virginica*, and C) *Muhlenbergia capillaris* grown in a greenhouse in Auburn, AL and harvested at 5, 10 and 15 weeks after treatment initiation from 20 June 2011 – 30 Sept. 2011. Data were subjected to regression analysis using PROC GLM in SAS (SAS Institute, Cary, NC).

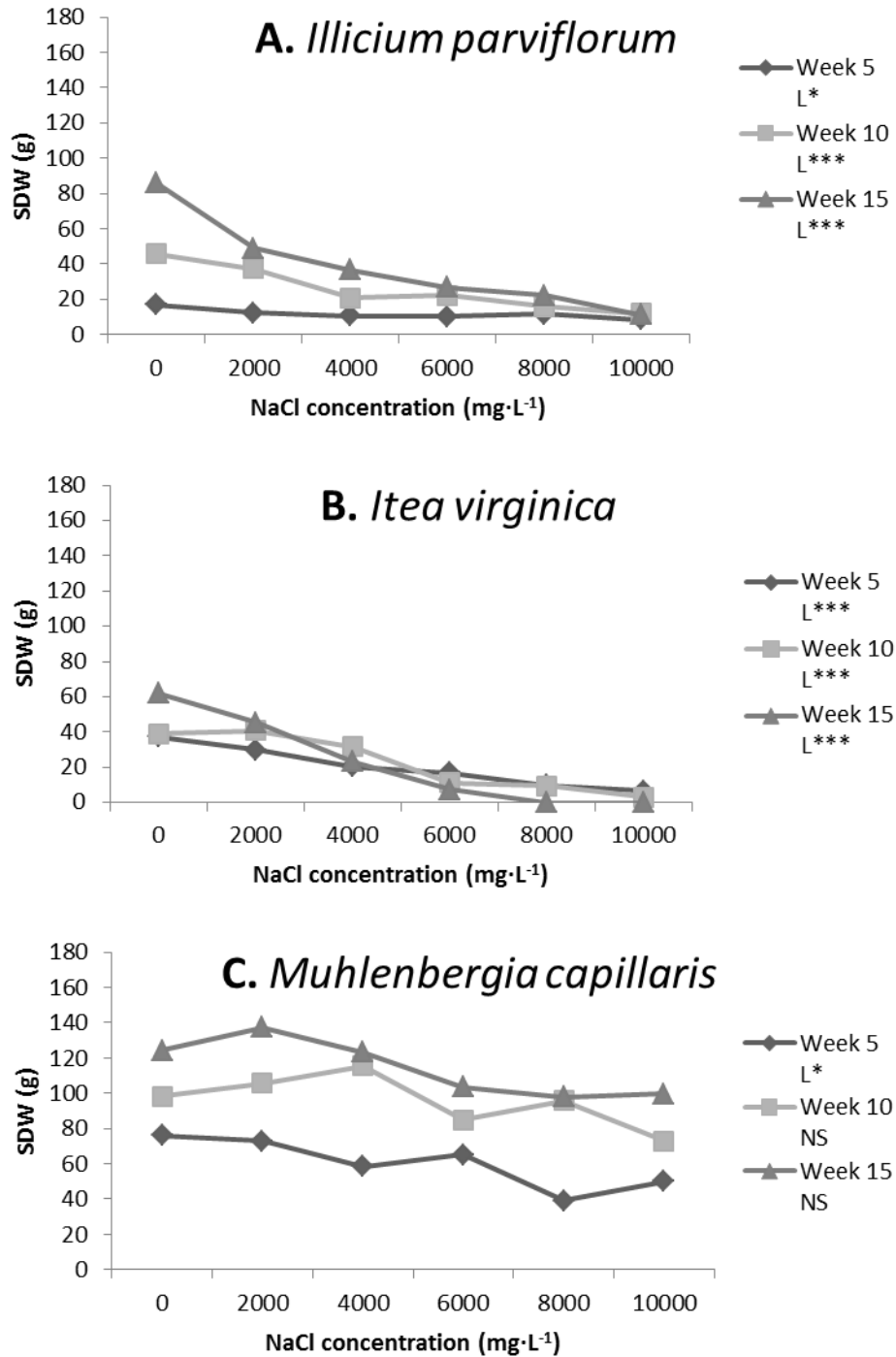


Figure 2. Effect of NaCl concentration in irrigation water on shoot dry weight (SDW) of A) *Illicium parviflorum*, B) *Itea virginica*, and C) *Muhlenbergia capillaris* grown in a greenhouse in Auburn, AL and harvested at 5, 10 and 15 weeks after treatment initiation from 20 June 2011 – 30 Sept. 2011. Data were subjected to regression analysis using PROC GLM in SAS (SAS Institute, Cary, NC).

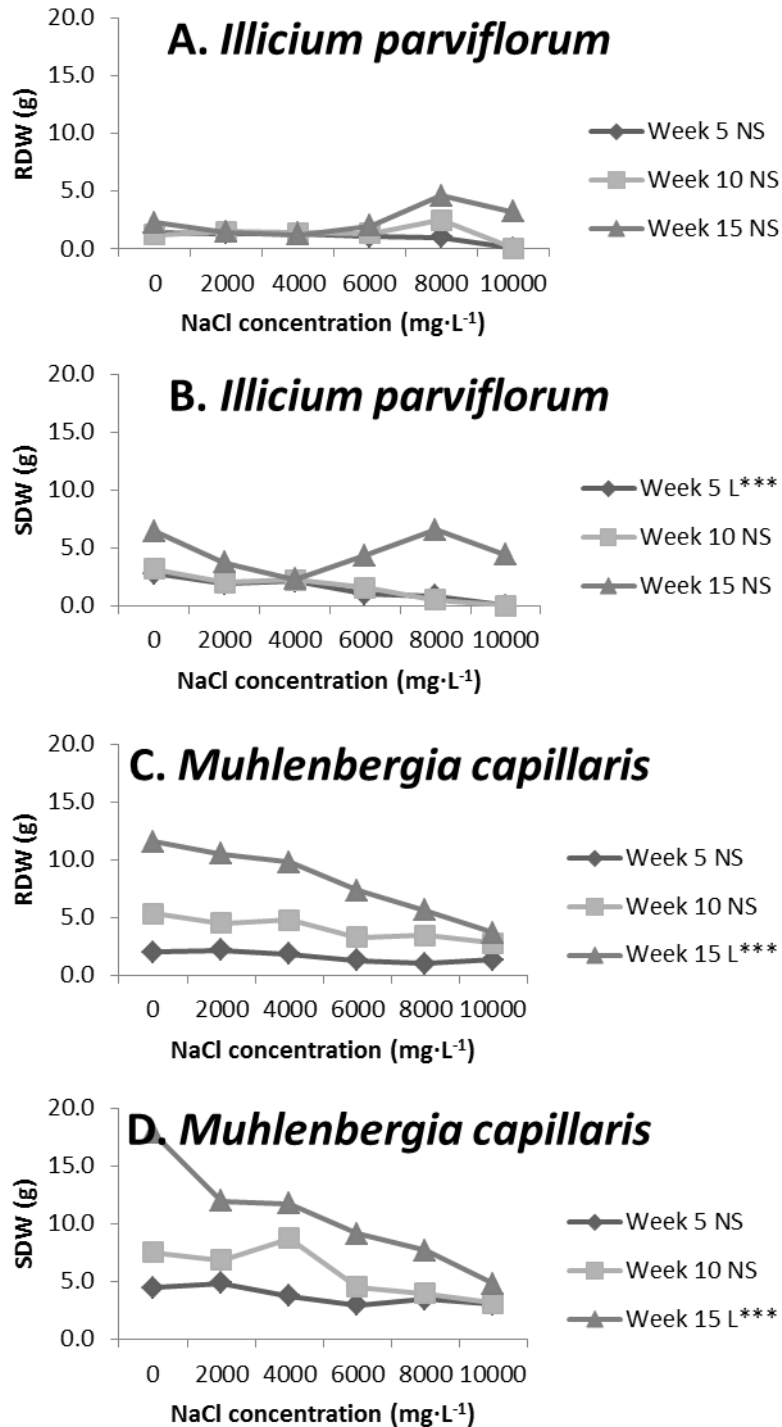


Figure 3. Effect of NaCl concentration in irrigation water on root dry weight (RDW) and shoot dry weight (SDW) of *Illicium parviflorum* and *Muhlenbergia capillaris* grown in a greenhouse in Auburn, AL and harvested at 5, 10, and 15 weeks after treatment initiation from 12 Sept. 2012 – 19 Dec. 2011. Data were subjected to regression analysis using PROC GLM in SAS (SAS Institute, Cary, NC).

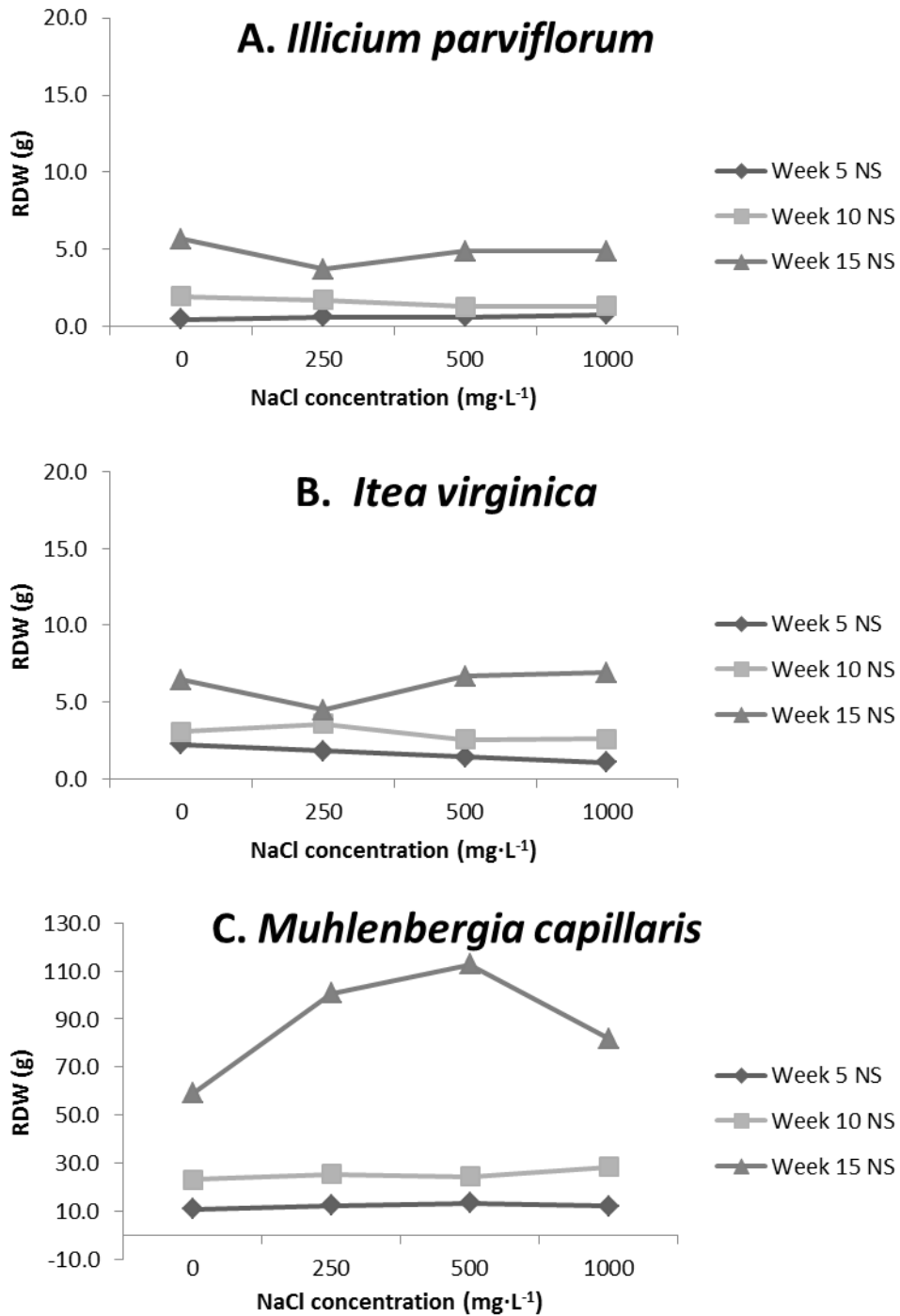


Figure 4. Effect of NaCl concentration in irrigation water on root dry weight (RDW) of A) *Illicium parviflorum*, B) *Itea virginica*, and C) *Muhlenbergia capillaris* grown in a greenhouse in Auburn, AL and harvested at 5, 10, and 15 weeks after treatment initiation from 10 July 2012 – 25 Oct. 2012. Data were subjected to regression analysis using PROC GLM in SAS (SAS Institute, Cary, NC).

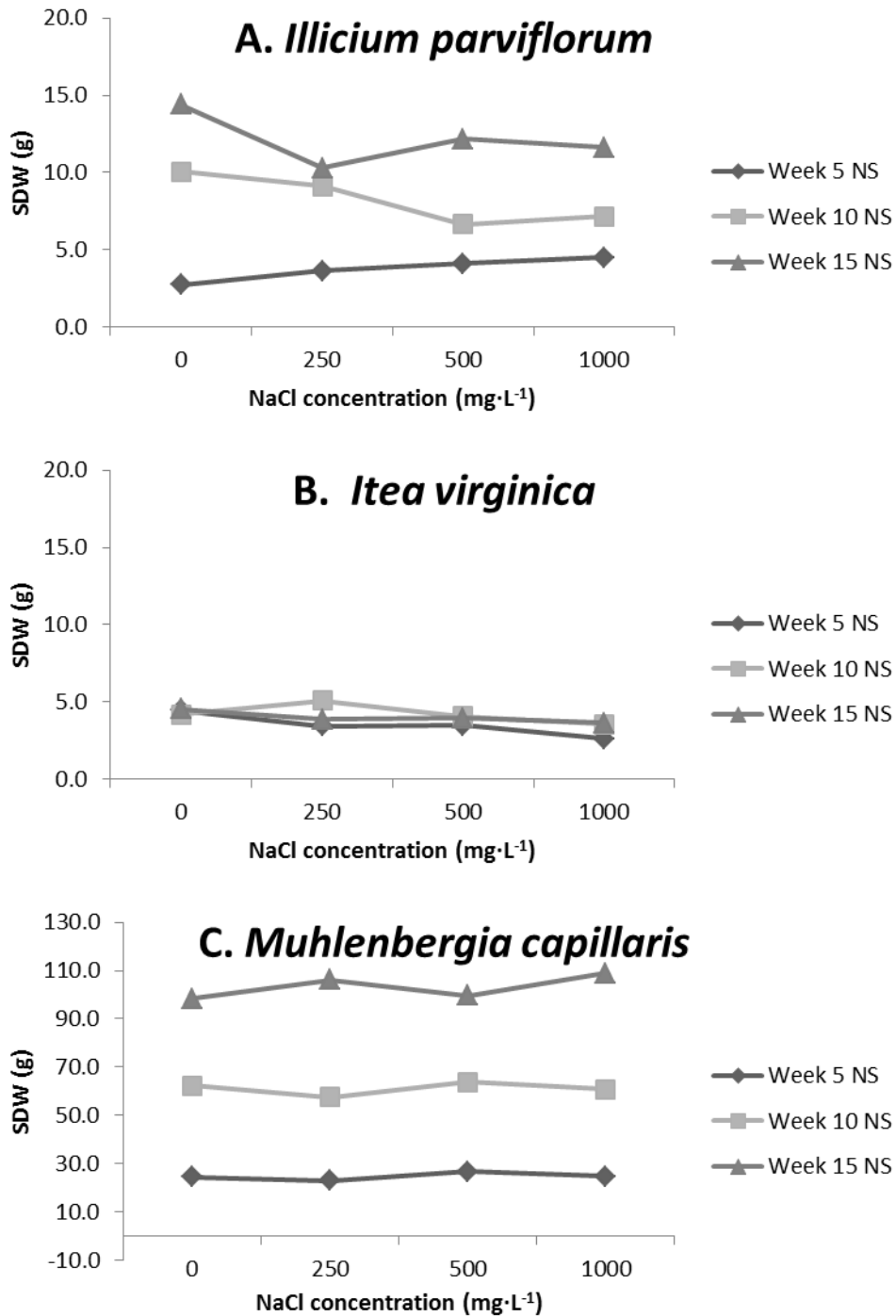


Figure 5. Effect of NaCl concentration in irrigation water on shoot dry weight (SDW) of A) *Illicium parviflorum*, B) *Itea virginica*, and C) *Muhlenbergia capillaris* grown in a greenhouse in Auburn, AL and harvested at 5, 10, and 15 weeks after treatment initiation from 10 July 2012 – 25 Oct. 2012. Data were subjected to regression analysis using PROC GLM in SAS (SAS Institute, Cary, NC).

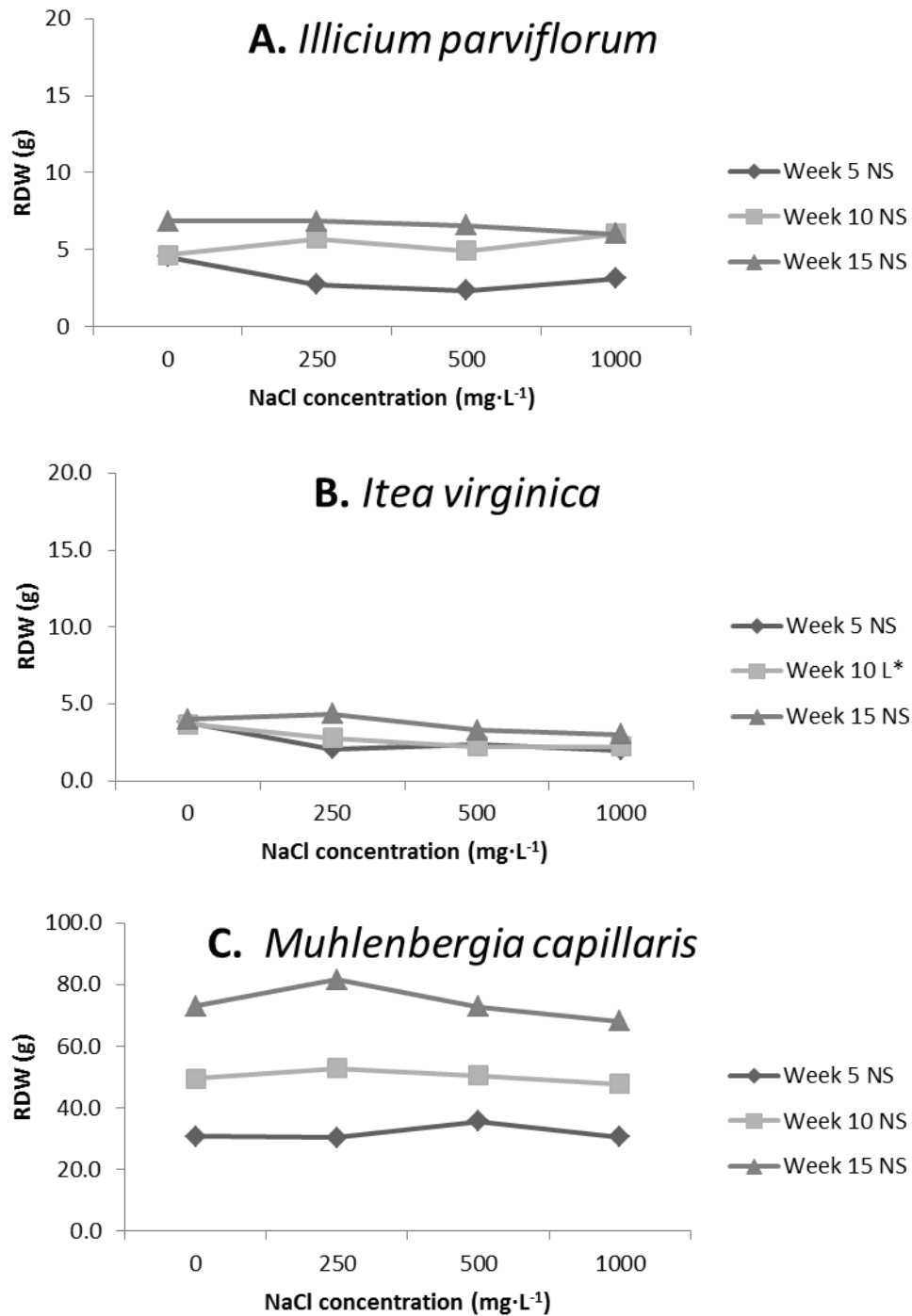


Figure 6. Effect of NaCl concentration in irrigation water on root dry weight (RDW) of A) *Illicium parviflorum*, B) *Itea virginica*, and C) *Muhlenbergia capillaris* grown in a greenhouse in Auburn, AL and harvested at 5, 10, and 15 weeks after treatment initiation from 15 Oct. 2012 - 30 Jan. 2013. Data were subjected to regression analysis using PROC GLM in SAS (SAS Institute, Cary, NC).

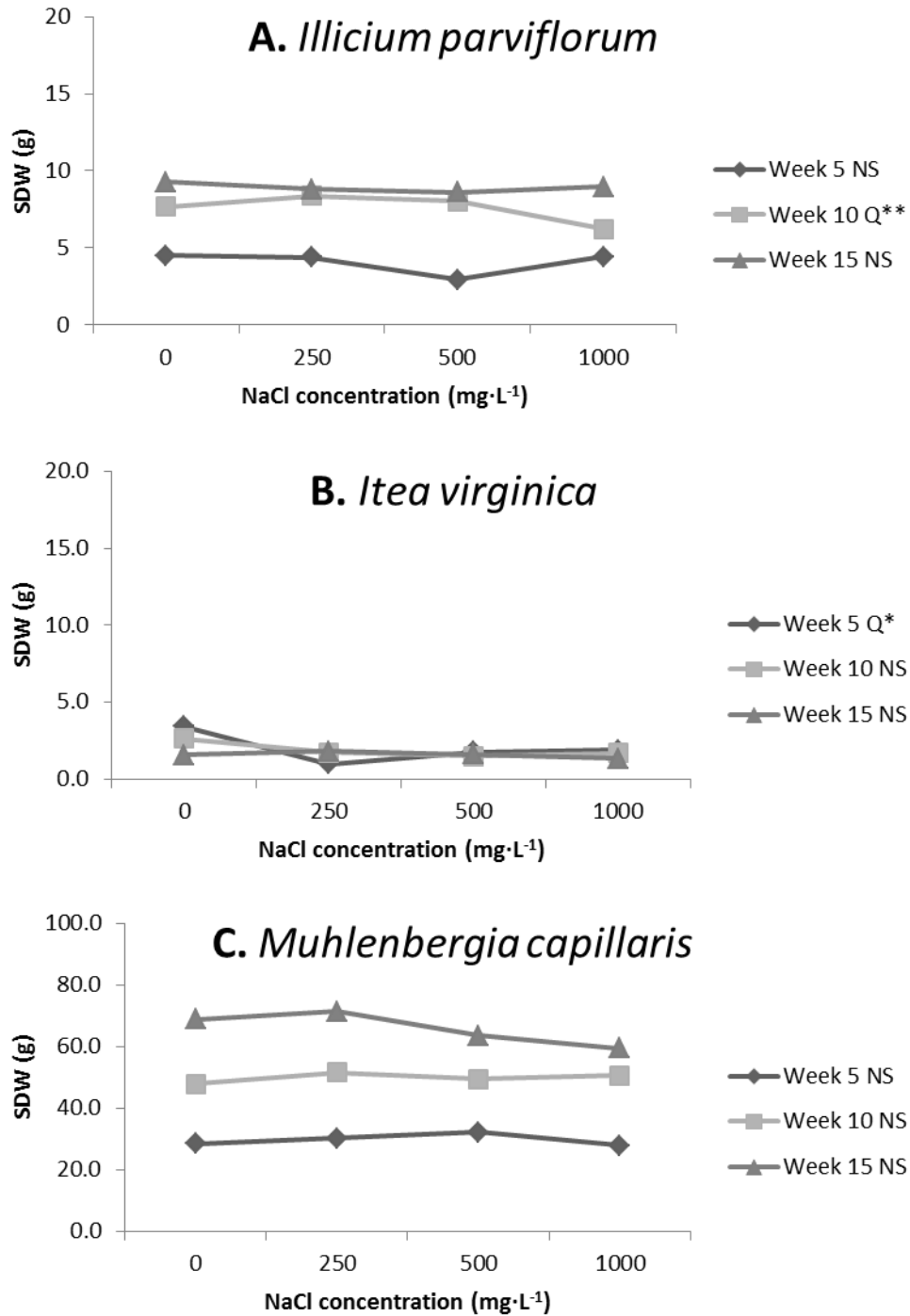


Figure 7. Effect of NaCl concentration in irrigation water on shoot dry weight (SDW) of A) *Illicium parviflorum*, B) *Itea virginica*, and C) *Muhlenbergia capillaris* grown in a greenhouse in Auburn, AL and harvested at 5, 10, and 15 weeks after treatment initiation from 15 Oct. 2012 - 30 Jan. 2013. Data were subjected to regression analysis using PROC GLM in SAS (SAS Institute, Cary, NC).

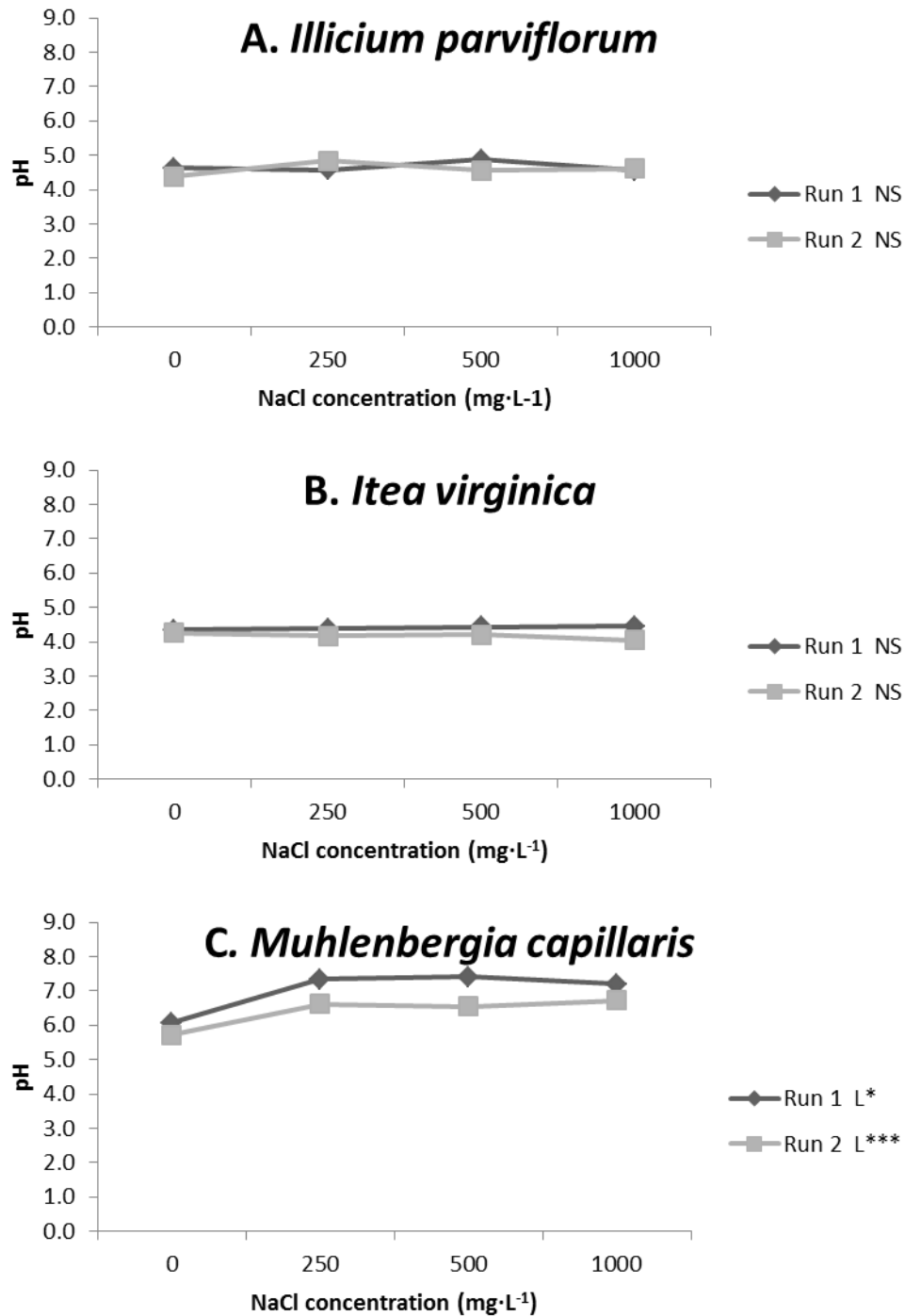


Figure 8. Effect of NaCl concentration in irrigation water on substrate pH of A) *Illicium parviflorum*, B) *Itea virginica*, and C) *Muhlenbergia capillaris* grown in a greenhouse in Auburn, AL from 10 July 2012 – 15 Oct. 2012 (Run 1) and 15 Oct. 2012 – Jan. 30 2013 (Run 2). Substrate pH measured 15 weeks after treatment initiation (termination) from leachate using the pour-through nutrient extraction procedure. Data were subjected to regression analysis using PROC GLM in SAS (SAS Institute, Cary, NC).

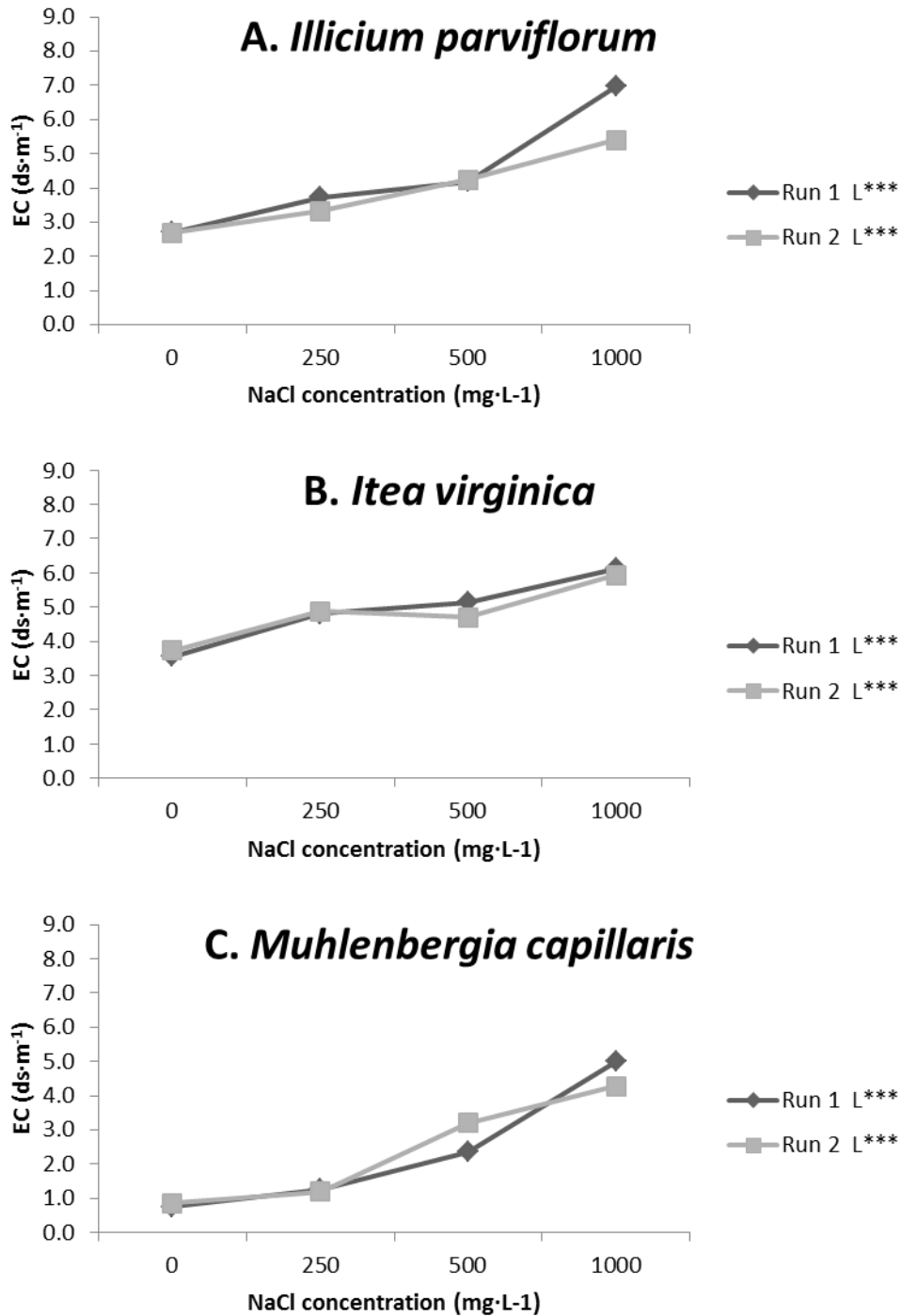


Figure 9. Effect of NaCl concentration in irrigation water on substrate electrical conductivity (EC) of A) *Illicium parviflorum*, B) *Itea virginica*, and C) *Muhlenbergia capillaris* grown in a greenhouse in Auburn, AL from 10 July 2012 – 15 Oct. 2012 (Run 1) and 15 Oct. 2012 – Jan. 30 2013 (Run 2). Substrate EC measured 15 weeks after treatment initiation (termination) from leachate using the pour-through nutrient extraction procedure. Data were subjected to regression analysis using PROC GLM in SAS (SAS Institute, Cary, NC).

Chapter III

Saline Irrigation Effects on Growth of Two Annual Bedding Plant Species for the Southeastern United States

Index Words: Sustainability, greywater, water, sodium chloride, wastewater

Abstract:

Annual bedding plant production has a large economic impact on economies of the southeastern United States. Generally, bedding plants require more irrigation than woody plants in the landscape. Drought and population growth strain the potable water supply. Greywater is a renewable irrigation alternative to potable water, however, its use as an irrigation source is limited by the potential for salt injury to plants. Research was conducted to determine salt tolerance of two annual bedding plants, *Begonia×semperflorens-cultorum* and *Portulaca oleracea*. Two experiments were performed, one with high NaCl concentrations and one with low NaCl concentrations. Plants received daily irrigation of tap water containing one of the following NaCl concentrations: 0 (tap water), 2000, 4000, 6000, 8000, or 10000 ppm ($\text{mg}\cdot\text{L}^{-1}$) (high NaCl conc. exp.) or 0 (tap water), 250, 500, or 1000 ppm ($\text{mg}\cdot\text{L}^{-1}$) (low NaCl conc. exp.) for 6 weeks. Root dry weight (RDW), shoot dry weight (SDW), and survival were determined at experiment termination. Substrate leachate pH and electrical conductivity (EC) were measured at termination in low NaCl concentration experiment. Experiments took place from July 2011 – March 2013. In high NaCl concentration experiment, RDW and SDW, of both species, decreased linearly with increasing NaCl concentration. *Begonia×semperflorens-cultorum* had mortality at 6000 (10% mortality) and 10000 ppm ($\text{mg}\cdot\text{L}^{-1}$) NaCl (50% mortality). In low NaCl concentration experiment there was no effect of NaCl concentration and no mortality in either species.

Substrate leachate EC increased linearly with increasing NaCl concentration with both species. Both species evaluated were tolerant of NaCl concentrations expected from greywater.

Introduction

In 2009, bedding plant production in Alabama had a \$79.1 million economic impact on the state of Alabama (AAES, 2009). Generally, these plants require more irrigation during production and in the landscape than woody plants due to poor drought tolerance. As a result, growers and landscapers could benefit from alternative irrigation sources to relieve the demand for potable water. Greywater is one such potential source. Greywater is wastewater from bathtubs, showers, hand basins, laundry machines, and kitchen sinks without input from toilets. Greywater is less polluted than combined wastewater due to the absence of feces, urine, and toilet paper (Eriksson et al., 2002; Valee et al., 2010). One concern with greywater reuse for irrigation is the potential for high salinity, usually in the form of sodium chloride (NaCl). Sodium concentrations of greywater can range from 7.4-480 ppm ($\text{mg}\cdot\text{L}^{-1}$), while chlorine concentrations can range from 9-88 ppm ($\text{mg}\cdot\text{L}^{-1}$) (Christova-Boal et al., 1996; Eriksson et al., 2002).

There has been extensive research on salt tolerance of bedding plants, herbaceous perennials, and wildflowers. The majority of species evaluated were commonly used in the southwestern United States and the Mediterranean. Salinity tolerance research with these plants was primarily conducted using simulated reclaimed wastewater (treated municipal effluent) (Miyamoto et al., 2004; Niu et al., 2007, 2012; Niu and Rodriguez, 2006, 2010, 2012; Wu et al., 2000; Yeager et al., 2010). Examples of species from those studies found tolerant of saline irrigation are: *Angelonia angustifolia* Benth. (summer snapdragon), *Catharanthus roseus* (L.) G. Don (vinca), *Delosperma cooperi* (Hook.f.) L.Bolus (ice plant), *Gazania rigens* (L.)

Gaertn. (treasure flower), and *Salvia splendens* Sellow ex Roem. & Schult. (annual salvia). Salt tolerant plant species have also been identified for landscapes in coastal regions (Black, 1997; Black, 2003). Including: *Solenostemon scutellarioides* (L.) Codd (Coleus), *Zinnia* sp. L. (Zinnia), *Pelargonium×hortorum* L.H. Bailey (pro sp.) (zonal geranium), *Senecio cineraria* DC. (dusty miller), and *Pentas* sp. Benth. (Pentas).

Begonia×semperflorens-cultorum Hort. (wax begonia) and *Portulaca oleracea* L. (purslane) are horticultural annual species used as bedding and container plants in the southeastern United States. While *B.×semperflorens-cultorum* has been recommended as a salt tolerant annual in coastal regions of Florida (Black, 1997), other research found that *B.×semperflorens-cultorum* was sensitive to saline irrigation (Miyamoto, 2004). *Portulaca* sp. have been recommended for inclusion in landscapes in coastal regions (Black, 2003; Grieve and Suarez, 1997; Hamidov et al., 2007). *Portulaca oleracea*, which has previously been considered a weed in the South, has more recently been utilized as an annual bedding plant (Everest et al., 1998; Everest and Williams, 2003).

Evaluation of additional annual bedding plants for tolerance of greywater salinity could increase plant selection options for a greywater irrigated landscape in the southeastern United States. Therefore, the objective of this research was to evaluate the tolerance of two annual bedding plant species to saline irrigation water.

Materials and Methods

High Salt Concentrations

Run I

Liners [2in (5.1 cm)] of *Begonia×semperflorens-cultorum* ‘Cocktail series’ and *Portulaca oleracea* ‘Toucan series’ (Ball Horticultural Company, West Chicago, IL) were

planted in 2.5 L (trade gallon) containers in a 5:3:1 pinebark:peat:perlite, by volume, substrate. Substrate was pre-plant amended with 9.1 lb·yd⁻³ (4.52 kg·m⁻³) controlled-release fertilizer [Polyon with micros 17N-5P₂O₅-11K₂O (Harrell's LLC, Lakeland, FL)], and dolomitic limestone [4 lb·yd⁻³ (1.8 kg·m⁻³)]. Plants were irrigated by hand daily with 10 oz (300 mL) of tap water containing one of the following concentrations (treatments) of sodium chloride (NaCl): 0 (tap water), 2000, 4000, 8000, or 1000 ppm (mg·L⁻¹) resulting in approximately 10 – 15% leachate (SNA, 2007). While these rates are higher than those normally found in greywater, they were used so that results could be applicable to other more saline irrigation sources (Christova-Boal et al., 1996; Eriksson et al., 2002). Plants received tap water irrigation (no NaCl) on weekends. Plants were grown under natural photoperiods on raised benches in a polycarbonate greenhouse at Paterson Horticulture Teaching and Research Greenhouse Complex at Auburn University in Auburn, AL. Temperatures ranged from 72 - 65°f during the day and 80 - 67°f at night.

There were ten single container replications per treatment per species. Experimental design was a completely randomized design with each species in a separate experiment. Plants were harvested 6 weeks after treatment initiation (experiment termination). Root dry weight (RDW) and shoot dry weight (SDW) were determined at experiment termination. Treatments were initiated on 11 July 2011, and experiment was terminated on 22 Aug. 2011. Data were subjected to analysis of variance and regression analysis using PROC GLM in SAS (SAS Institute, Cary, NC).

Run II

On 29 Sept. 2011, run I was repeated with the same procedures described above with only *P. oleracea*. *Begonia*×*semperflorens-cultorum* was omitted based on its lack of tolerance to these NaCl concentrations. Run II was terminated on 10 Oct. 2011.

Low Salt Concentrations

Run I

On 3 April 2012, (*B.*×*semperflorens-cultorum*) and 17 Sept. 2012 (*P. oleracea*) experiments similar to the high salt concentration experiment described above were initiated. Based on results from high salt concentration experiment, NaCl treatments were decreased to 0 (tap water), 250, 500, or 1000 ppm ($\text{mg}\cdot\text{L}^{-1}$) to evaluate seemingly less salt tolerant *Begonia*×*semperflorens-cultorum*. *Portulaca oleracea* was also evaluated. Substrate pH and electrical conductivity (EC) of *P. oleracea* were measured at termination (29 Oct. 2012) from leachate collected using the pour-through nutrient extraction procedure (Wright, 1986). *Begonia*×*semperflorens-cultorum* was terminated on 15 May 2012. All other materials and methods were the same as described above.

Run II

Low salt concentration experiment was repeated on 13 Feb. 2013; experiment termination was on 27 March 2013. Substrate pH and electrical conductivity (EC), from both species, were measured at termination.

Results

High Salt Concentrations

Begonia×*semperflorens-cultorum*

Root and shoot dry weight decreased linearly with increasing NaCl concentration (Fig. 1). Plants exhibited foliar damage at 6000 ppm (personal observation) and mortality at 6000 (10% mortality) and 10000 ppm ($\text{mg}\cdot\text{L}^{-1}$) NaCl (50% mortality).

Portulaca oleracea

Root and shoot dry weight decreased linearly with increasing NaCl concentration in both runs (Figs. 1-2). There were no visible signs of salt stress or mortality in either run.

Low Salt Concentrations

In both species, there was no effect of NaCl concentration on root or shoot dry weight in either run (Figs. 3-4). There were no symptoms of salt stress or mortality for either species in both runs. There was no effect of NaCl concentration on substrate leachate pH in both species (Fig. 5). Substrate leachate EC increased linearly with increasing NaCl concentration in both species (Fig. 6).

Discussion

Results for *P. oleracea* were consistent with previous recommendations for inclusion in saline environments (Black, 2003; Grieve and Suarez, 1997; Hamidov et al., 2007). *Portulaca oleracea* had 100% survival in all experiments (high and low NaCl), even at NaCl concentrations of up to 20 times higher than generally found in greywater. Despite growth reductions with increasing NaCl concentrations in high NaCl experiments, there were no signs of salt stress and plants continued to flower. *P. oleracea* still appeared to be “marketable” by the end of each experiment. Results from this experiment show that *P. oleracea* could be included in a greywater landscape and, potentially, other saline environments.

Begonia×*semperflorens-cultorum* was not tolerant of high NaCl concentrations, however, there was no effect on growth of *B.*×*semperflorens-cultorum* irrigated at lower NaCl

concentrations. There were also no visible signs of salt stress or reduced flowering when irrigated with low NaCl concentrations. Plants also appeared to be “marketable” in the low NaCl experiment suggesting that *B. ×semperflorens-cultorum* could be included in a greywater irrigated landscape as long as irrigation salinity and frequency is closely monitored.

Differences in growth between runs, in both species, can be attributed to runs occurring at different times of year. Despite being grown in a greenhouse, plants grown in the summer were larger than plants grown in the fall/spring due to warmer temperatures and increased solar radiation. However, results indicate that plant response to saline irrigation was not affected by time of year indicating that greywater irrigation could be used throughout the year. Increased irrigation demand during summer months and increased evaporation rates may increase salt accumulation in soil. Greywater application may need to be supplemented, with tap water, and closely monitored to avoid salt toxicity.

Elevated EC levels observed with *B. ×semperflorens-cultorum* irrigated with 0 ppm ($\text{mg}\cdot\text{L}^{-1}$) NaCl concentration can be attributed to lower growth rates and thus lower nutrient uptake resulting in increased dissolved salts in the substrate solution. Electrical conductivity was however within sufficiency ranges (Reed, 1996). General limits for salt concentrations in irrigation water range from 435 ppm ($\text{mg}\cdot\text{L}^{-1}$) for salt sensitive plants to 3485 ppm ($\text{mg}\cdot\text{L}^{-1}$) for salt tolerant crops (Watling, 2007). Substrate EC above $6 \text{ ds}\cdot\text{m}^{-1}$ is considered highly saline, while EC of 2 - $6 \text{ ds}\cdot\text{m}^{-1}$ is considered moderately saline (Watling, 2007). Electrical conductivity in the lower NaCl concentration experiments were within the range of moderately saline, with the exception of *B. ×semperflorens-cultorum* plants irrigated with 1000 ppm ($\text{mg}\cdot\text{L}^{-1}$) NaCl concentration, which were highly saline. Elevations in EC of substrate leachate with increasing NaCl concentrations, in both species, were expected. NaCl concentrations of 1000 ppm ($\text{mg}\cdot\text{L}^{-1}$)

had an EC of $1.6 \text{ ds}\cdot\text{m}^{-1}$ and caused an increase in substrate leachate EC of $1.1 - 1.72 \text{ ds}\cdot\text{m}^{-1}$ (Fig. 6) showing that there was little salt accumulation within the substrate.

The method of irrigation application can impact the effects of saline irrigation. The use of drip irrigation or subsurface irrigation allows for reduced contact with plant above-ground tissue, limiting the effects of saline irrigation on foliage. In this research, tap water irrigation (no NaCl), applied weekly, was used to mimic a rain event or tap water irrigation cycle and appeared to prevent the accumulation of NaCl in the substrate. Periodically irrigating with non-saline water could be used as a management tool in greywater-irrigated landscapes.

NaCl concentrations used in this research (even in the low NaCl concentration experiment) were higher than those observed in greywater. Results from these experiments could be applicable for other saline irrigation sources, landscapes in coastal regions, and perhaps landscapes in arid environments because plant response to salt stress is often similar to that of drought stress. Additional research to identify annual landscape species that are salt tolerant will have application for utilizing greywater for landscape irrigation as well as utilizing saline irrigation water in general. Previous research has shown that it may be beneficial to use plants that are native to saline environments or species that are drought tolerant (LeCompte, 2013). Examples of plants native to the Southeastern Gulf Coast that may be tolerant of greywater irrigation are: *Lupinus westianus* Small (Gulf Coast lupine), *Helianthus debilis* Nutt. (cucumberleaf sunflower), *Sesuvium portulacastrum* (L.) L. (shoreline seapurslane), *Borrchia frutescens* (L.) DC., (bushy seaside tansy), and *Heterotheca subaxillaris* (Lam.) Britton & Rusby (camphorweed). Evaluation of *B. ×semperflorens-cultorum* and *P. oleracea* found both species tolerant of NaCl concentrations expected from greywater. Results from this

and future research can be used to promote greywater-irrigated landscapes, and decrease the demand for potable water.

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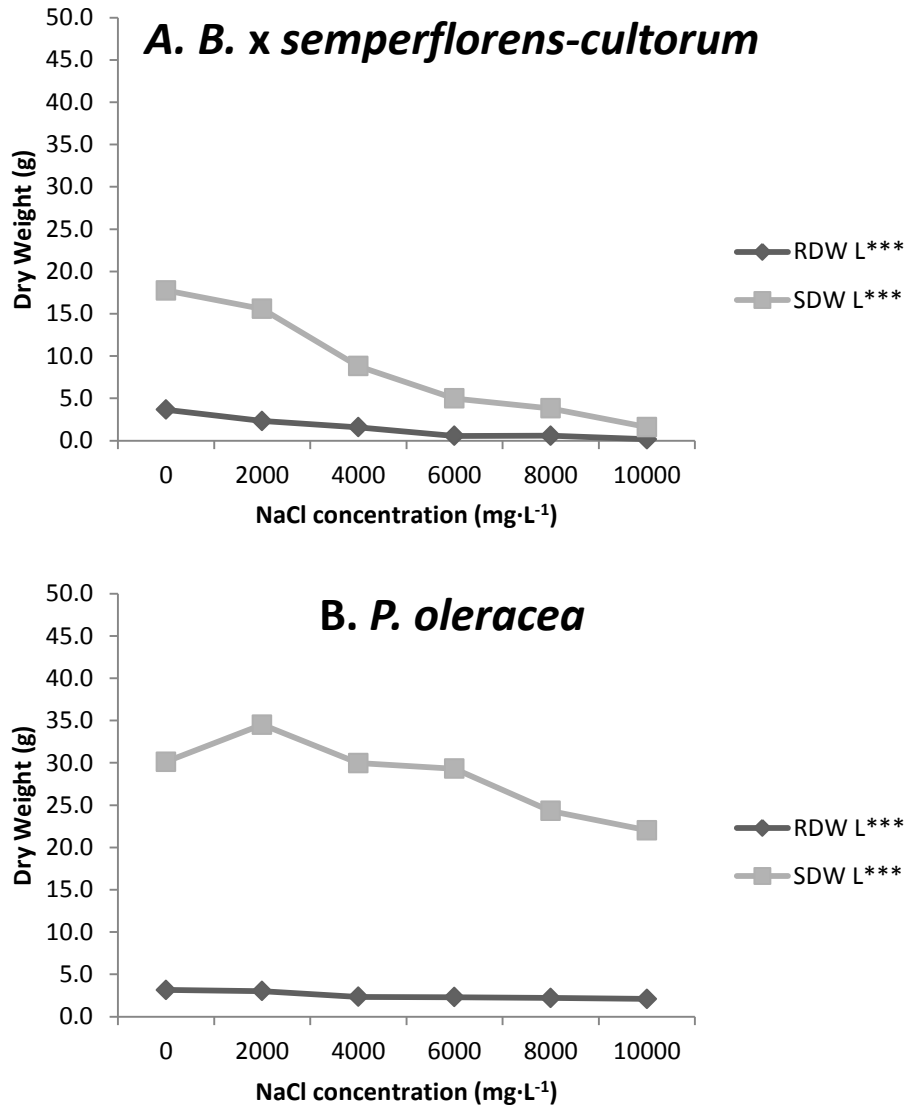


Figure 1. Effect of NaCl concentration in irrigation water on root dry weight (RDW) and shoot dry weight (SDW) of A) *Begonia x semperflorens-cultorum* and B) *Portulaca oleracea* grown in a greenhouse in Auburn, AL and harvested at 6 weeks after treatment initiation from 11 July 2011 - 22 Aug. 2011. Data were subjected to regression analysis using PROC GLM in SAS (SAS Institute, Cary, NC).

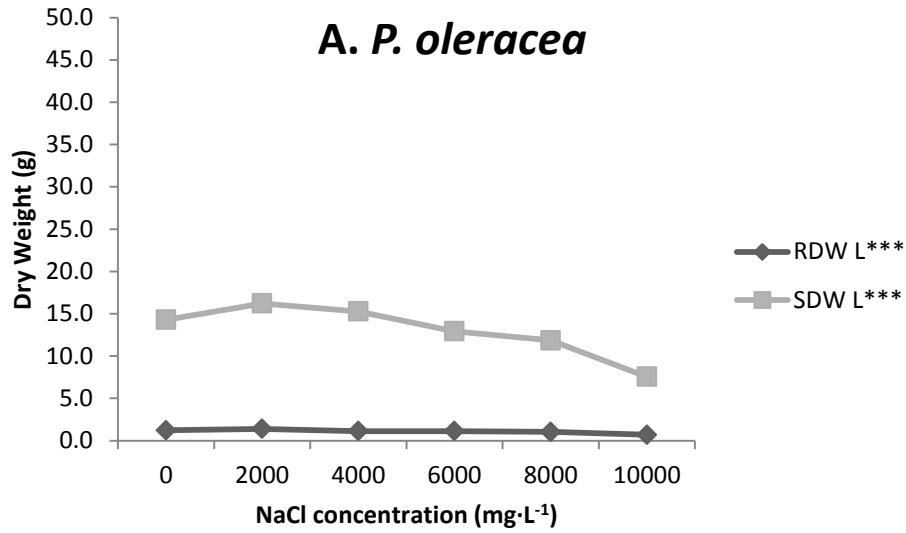


Figure 2. Effect of NaCl concentration in irrigation water on root dry weight (RDW) and shoot dry weight (SDW) of *A. Portulaca oleracea* grown in a greenhouse in Auburn, AL and harvested at 6 weeks after treatment initiation from 29 Sept. 2011 – 10 Oct. 2011. Data were subjected to regression analysis using PROC GLM in SAS (SAS Institute, Cary, NC).

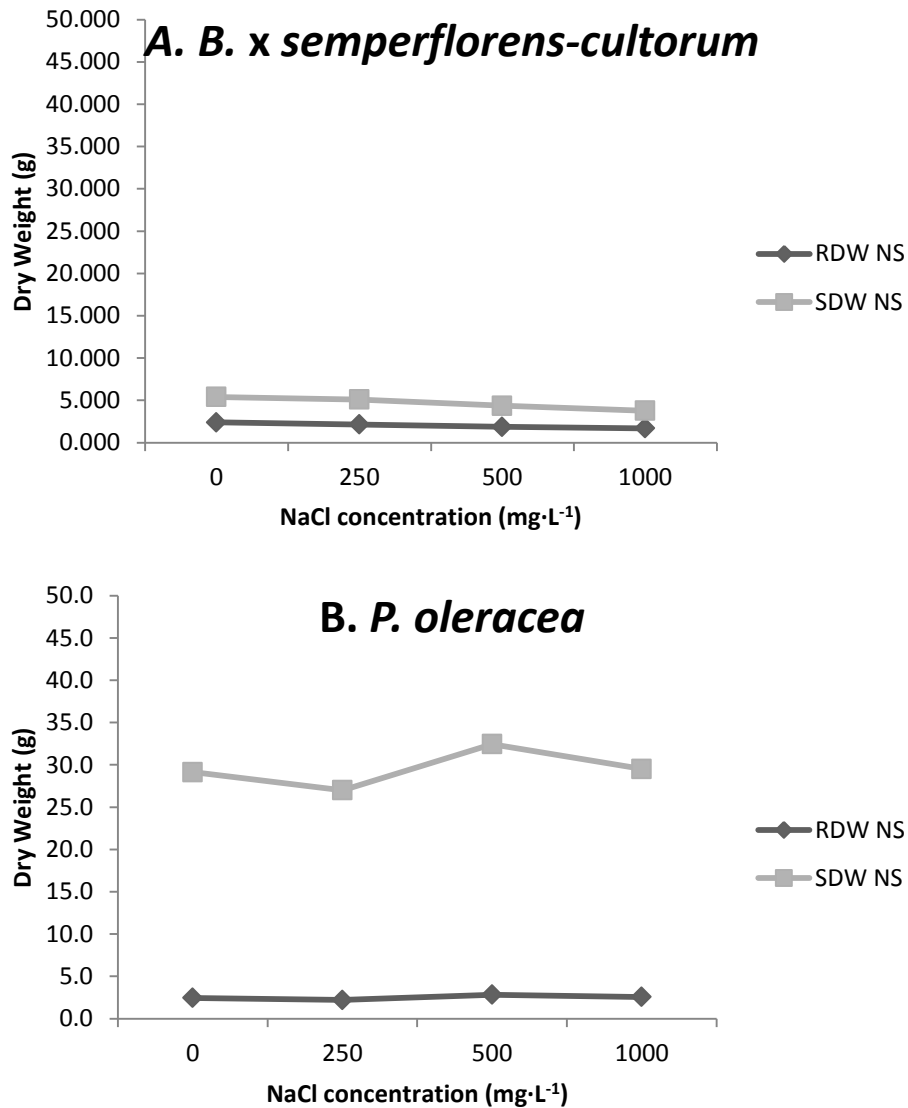


Figure 3. Effect of NaCl concentration in irrigation water on root dry weight (RDW) and shoot dry weight (SDW) of A) *Begonia x semperflorens-cultorum* and B) *Portulaca oleracea* grown in a greenhouse in Auburn, AL and harvested at 6 weeks after treatment initiation from 3 April 2012 - 15 May 2012 (*B. x semperflorens-cultorum*) and 17 Sept. 2012 - 29 Oct. 2012 (*P. oleracea*). Data were subjected to regression analysis using PROC GLM in SAS (SAS Institute, Cary, NC).

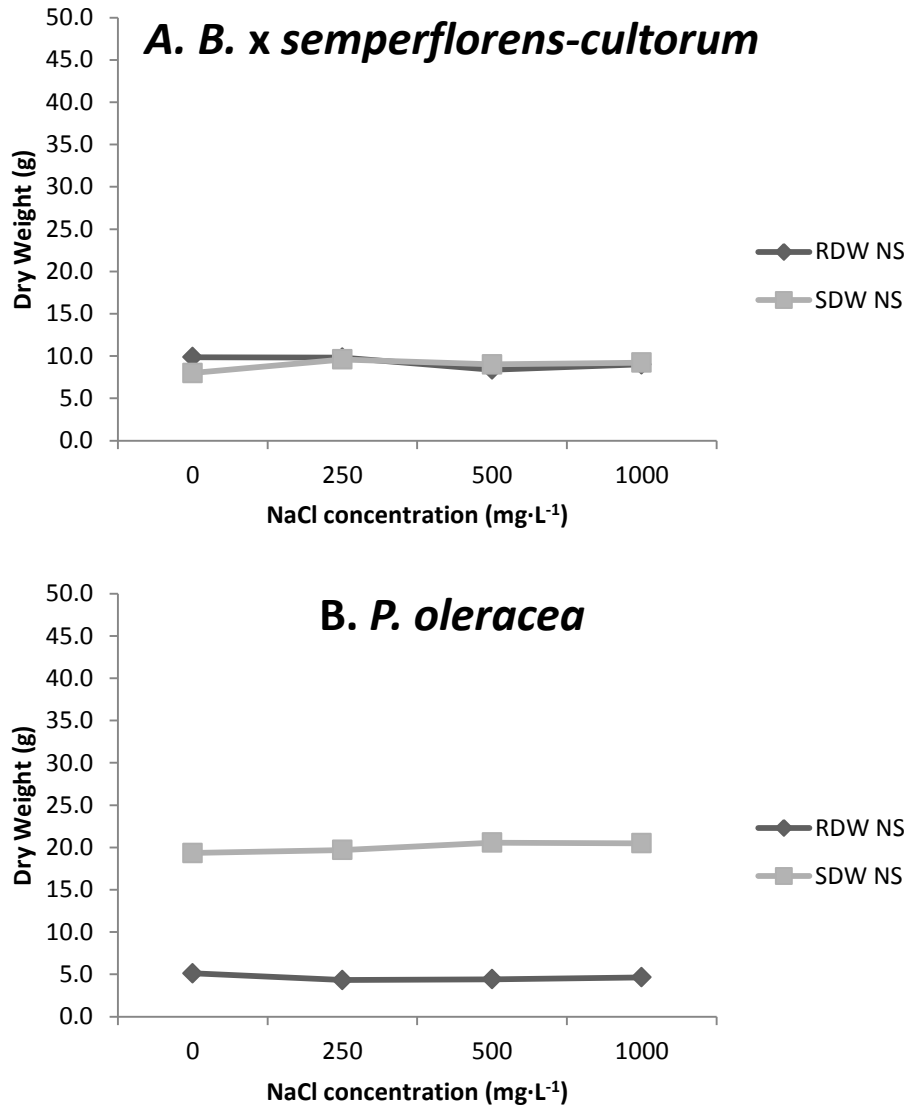


Figure 4. Effect of NaCl concentration in irrigation water on root dry weight (RDW) and shoot dry weight (SDW) of A) *Begonia x semperflorens-cultorum* and B) *Portulaca oleracea* grown in a greenhouse in Auburn, AL and harvested at 6 weeks after treatment initiation from 13 Feb. 2013 - 27 March 2013. Data were subjected to regression analysis using PROC GLM in SAS (SAS Institute, Cary, NC).

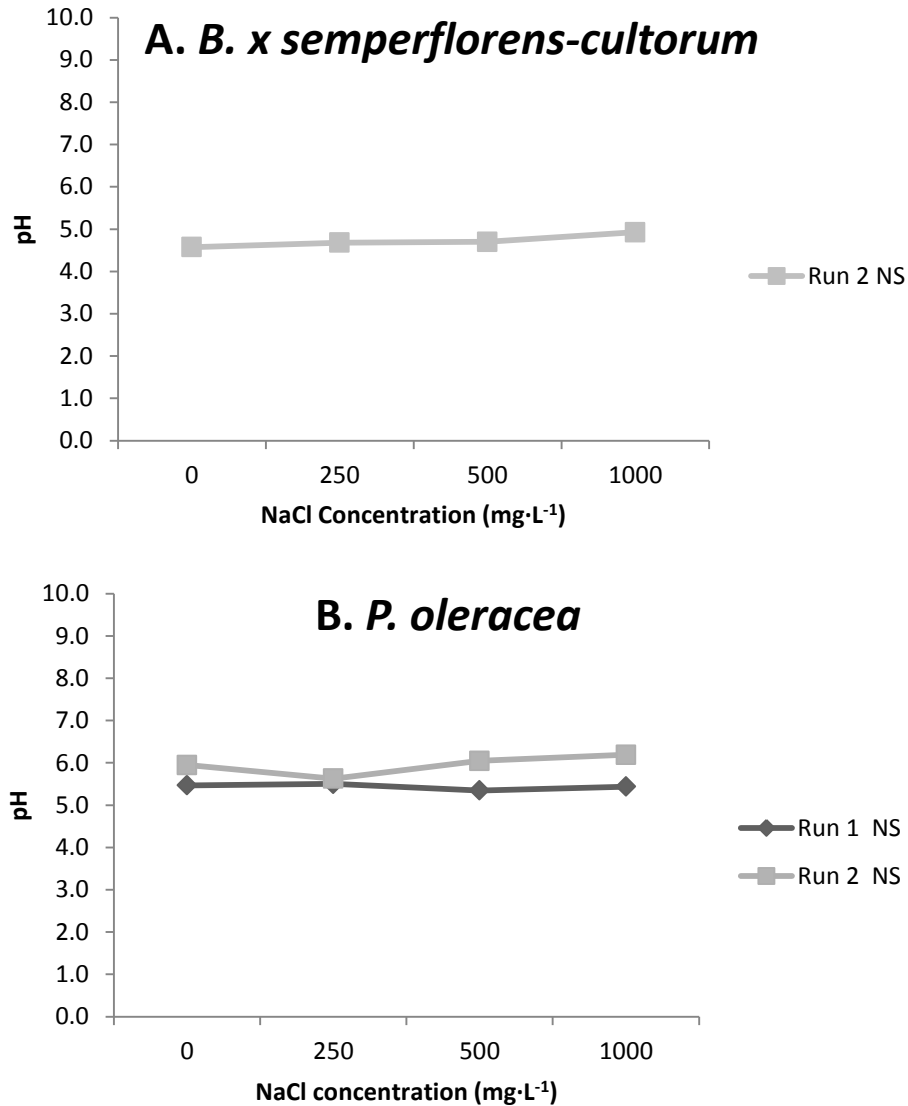


Figure 5. Effect of NaCl concentration in irrigation water on substrate pH of A) *Begonia x semperflorens-cultorum* and B) *Portulaca oleracea* grown in a greenhouse in Auburn, AL from 17 Sept. 2012 - 29 Oct. 2012 (Run 1 *P. oleracea*) and Feb. 2013 - 27 March 2013 (Run 2 *B. x semperflorens-cultorum* and *P. oleracea*). Substrate pH measured 6 weeks after treatment initiation (termination) from leachate using the pour-through nutrient extraction procedure (Wright, 1986). Data were subjected to regression analysis using PROC GLM in SAS (SAS Institute, Cary, NC).

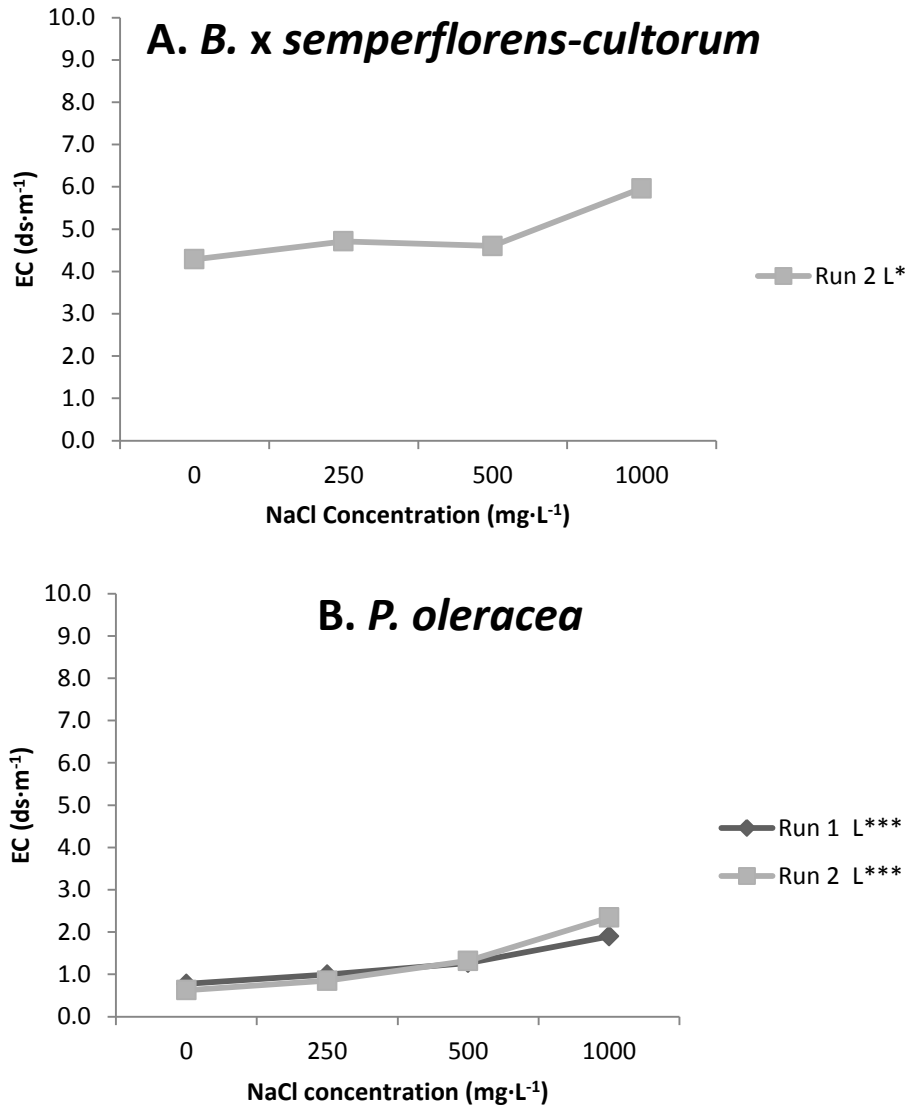


Figure 6. Effect of NaCl concentration in irrigation water on substrate electrical conductivity (EC) of A) *Begonia x semperflorens-cultorum* and B) *Portulaca oleracea* grown in a greenhouse in Auburn, AL from 17 Sept. 2012 - 29 Oct. 2012 (Run 1 *P. oleracea*) and Feb. 2013 - 27 March 2013 (Run 2 *B. x semperflorens-cultorum* and *P. oleracea*). Substrate EC measured 6 weeks after treatment initiation (termination) from leachate using the pour-through nutrient extraction procedure (Wright, 1986). Data were subjected to regression analysis using PROC GLM in SAS (SAS Institute, Cary, NC).

Chapter IV

Final Discussion

This research evaluated landscape species used in the southeastern United States for tolerance to saline irrigation. Currently, alternative irrigation sources are not widely used the South due to low cost of potable water (EPA, 2004). With increasing demand on an already stressed potable water supply homeowners and property managers will need to use alternative irrigation sources to maintain current water requirements of landscapes. It is imperative that plant species are continued to be evaluated to prepare for the potential shift in irrigation sources.

Results from chapter 2 identified two species tolerant of NaCl concentrations observed in greywater irrigation. *Illicium parviflorum* and *Muhlenbergia capillaris* were tolerant of NaCl concentrations higher than generally observed in greywater. *Itea virginica* was not tolerant of even the lowest NaCl concentrations.

M. capillaris had 100% survival even at NaCl irrigation rates of up to 20 times higher than generally found in greywater (Christova-Boal et al., 1996; Eriksson et al., 2002). *Illicium parviflorum* had 100% survival in all experiments with the exception of one run, where it had survival rates of 90% at 6000 ppm ($\text{mg}\cdot\text{L}^{-1}$), 80% at 8000 ppm ($\text{mg}\cdot\text{L}^{-1}$), and 20% at 10000 ppm ($\text{mg}\cdot\text{L}^{-1}$) NaCl. Generally, plants that can tolerate saline irrigation levels of 2000-3000 ppm ($\text{mg}\cdot\text{L}^{-1}$) NaCl are considered to be salt tolerant (Watling, 2007). High survival rates of *I. parviflorum* and *M. capillaris* were likely due to their native habitats' proximity to saline environments (Dirr, 1998; Schroeder et al., 1976). *Muhlenbergia capillaris* leaf Na and Cl concentrations were up to ten times higher in plants irrigated with the highest NaCl concentrations than in plants irrigated with tap water. Likewise, *Illicium parviflorum* irrigated with 10000 ppm ($\text{mg}\cdot\text{L}^{-1}$) had leaf Na and Cl concentrations 200 and 50 times higher,

respectively, than plants irrigated with tap water. *Itea virginica* leaf concentrations of Na and Cl were 4 and 15 times higher, respectively, in plants irrigated with 4000 ppm ($\text{mg}\cdot\text{L}^{-1}$) NaCl than plants irrigated with tap water. Even at lower concentrations of NaCl, *I. virginica* exhibited growth reductions and tissue damage, indicating it may not be suitable for inclusion in a landscape receiving frequent greywater irrigation or for highly saline landscapes. Signs of salt stress observed in *I. virginica* included wilting, chlorosis, leaf necrosis, defoliation, and root damage.

Chapter 3 evaluated two annual bedding plants to NaCl concentrations that would be observed in greywater. *Begonia*×*semperflorens-cultorum* and *Portulaca oleracea* were both tolerant of NaCl concentrations higher than would be expected from greywater. *Portulaca oleracea* had 100% survival in all experiments (high and low NaCl), even at NaCl concentrations of up to 20 times higher than generally found in greywater. Despite growth reductions with increasing NaCl concentrations in high NaCl experiments, there were no signs of salt stress and plants continued to flower. *P. oleracea* still appeared to be “marketable” by the end of each experiment. Results from this experiment show that *P. oleracea* could be included in a greywater landscape and, potentially, other saline environments.

Begonia×*semperflorens-cultorum* was not tolerant of high NaCl concentrations, however, there was no effect on growth of *B.*×*semperflorens-cultorum* irrigated at lower NaCl concentrations. There were also no visible signs of salt stress or reduced flowering when irrigated with low NaCl concentrations. Plants also appeared to be “marketable” in the low NaCl experiment suggesting that *B.*×*semperflorens-cultorum* could be included in a greywater irrigated landscape as long as irrigation salinity and frequency is closely monitored.

The method of irrigation application can impact the effects of saline irrigation. The use of drip irrigation or subsurface irrigation allows for reduced contact with plant above-ground tissue, limiting the effects of saline irrigation on foliage. In this research, tap water irrigation (no NaCl), applied weekly, was used to mimic a rain event or tap water irrigation cycle and appeared to prevent the accumulation of NaCl in the substrate. This technique could be used as a management tool in greywater irrigated landscapes. Periodically irrigating with tap water could reduce sodium accumulation in soil or substrate. Likewise, less salt-tolerant plants could still be used in greywater irrigated landscape if frequency of application is reduced.

There are concerns other than NaCl associated with greywater application. Surfactants, phosphorus, lint, and biological contaminants all represent potential environmental problems (Al-Hamaiedeh and Bino, 2010; Christova-Boal et al., 1996; Eriksson et al. 2002; Gross et al., 2005). Some of these concerns can be mitigated with subsurface irrigation and decreasing frequency of application. Research to address these concerns could be coupled with results of this work to facilitate greywater irrigation implementation in the landscape as well as future legislation.

In hindsight, additional data collection would have strengthened results. It would have been beneficial to collect pH and electrical conductivity of substrate leachate at every termination, instead of only lower NaCl concentration experiments. Growth indices, marketability ratings and the addition of a field study would have benefited results and given a better evaluation of performance in the landscape.

Ultimately, greywater irrigation application is dependent on the value for the consumer. Current prices of potable water in the United States are low when compared to other goods and services such as cable television, telephone service, and electricity (EPA, 2004). Alternative

water sources will potentially be considered for incorporated into the landscape when there is a financial benefit. By researching additional plants for tolerance of saline irrigation, alternative irrigation sources could promote future growth in the green industry.

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