Article

Relative Salt Tolerance of Four Herbaceous Perennial Ornamentals

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Received: 26 March 2019; Accepted: 8 May 2019; Published: 11 May 2019

Abstract: Salt tolerant ornamental plants can be irrigated with alternative water sources that are typically saline as a sustainable practice for urban landscaping, especially in arid and semi-arid regions. However, the salt tolerance of many ornamentals is not known. An eight-week greenhouse experiment was conducted to assess the relative salt tolerance of four perennial ornamentals, ‘Angelina’ (*Sedum rupestre*), ‘Autumn Joy’ (*S. telephium*), ‘Blue Spruce’ (*S. reflexum*), and ‘Blue Daze’ (*Evolvulus glomeratus*). The plants were grown in pots with potting mix substrate and irrigated with control or saline solutions. The electrical conductivities (EC) of the saline solutions were 5.0 and 10.0 mS/cm. Data collected included relative shoot, root, and total dry weight (DW), visual score, shoot tissue concentrations of Na^+ , Cl^- , K^+ , and Ca^{2+} , and the K^+/Na^+ ratio. There were significant differences in treatment and varieties for all response variables, and some interactions were also significant, indicating different responses to salinity for the four varieties. Shoot, root, and total DW decreased with increasing salinity for all varieties. Visual score was highest in Autumn Joy and Blue Spruce when treated with EC5 and EC10 and lowest in Angelina and Blue Daze, the latter of which showed symptoms of moderate foliar damage including leaf necrosis, or “burn”, due to salt stress. The concentrations of Na^+ and Cl^- in the shoot tissue increased with increasing salinity while K^+ and Ca^{2+} and the K^+/Na^+ ratio tended to decrease. Of the four varieties of herbaceous perennial ornamentals evaluated in this study, Autumn Joy and Blue Spruce were considered the most relatively salt tolerant while Angelina and Blue Daze were least tolerant.

Keywords: ornamentals; herbaceous; perennials; salt tolerance; salinity

1. Introduction

Water is increasingly becoming the most important resource in the world today as population and urbanization increase. Not only is water essential for all life, it impacts nearly every aspect of society, including food production, human health, and even ecological health [1,2]. Because fresh water sources are limited, the use of alternative water sources, such as recycled wastewater, is important for sustainable water management [3,4]. Moreover, recycled wastewater is considered a viable and sustainable option for urban landscaping, especially in arid and semi-arid regions [5,6]. However, recycled wastewater contains elevated salt levels and harmful ions, such as sodium and chloride, which are detrimental to plant growth and aesthetic quality of many ornamental plants [7–9]. Therefore, there is a need to identify salt tolerant ornamentals that can be irrigated with alternative water sources, such as saline recycled wastewater. Selecting for and improving salt tolerance has long been pursued for agricultural crops [10], but less attention has been devoted to evaluating salt tolerance in ornamental plants [11].

Salts, primarily sodium and chloride, can impose two types of stress on plants: osmotic and ionic [12]. Osmotic stress occurs when salts accumulate in the soil near the root zone causing a reduction in water potential which impairs the plant’s ability to uptake water. Plants rapidly respond

to osmotic stress in a cascade of physiological alterations, including reduced stomatal conductance, transpiration, photosynthesis, and ultimately reduced growth [13]. Ionic stress occurs when sodium and chloride accumulate in the shoots to toxic levels where they can disrupt cytosolic metabolism leading to tissue damage [14]. This specific ion damage manifests in the leaves as chlorosis, followed by necrosis and eventually defoliation [15]. This foliar damage, or “burn”, is especially a concern for ornamental plants because of its detrimental impact on aesthetic quality [16].

The Crassulaceae family, also known as stonecrops, includes the genus *Sedum*, which have fleshy, succulent leaves and typically yellow, pink, or red flowers that bloom in the summer [17]. Many species of *Sedum* are low-growing and have been used as ground cover in rock gardens and green roofs. Monterusso et al. [18] researched nine species of *Sedum* and found them suitable for green roofs in Michigan because of their ability to tolerate cold and drought. *Sedum* spp. have been shown to use Crassulacean acid metabolism (CAM) which allows the plant to open stomata at night instead of during the day to reduce water loss through transpiration [19]. Convolvulaceae, also known as the morning glory family, includes the genus *Evolvulus* which are herbaceous perennials. A popular ornamental, *E. glomeratus* (‘Blue Daze’) produces vibrant blue flowers and is also considered a good candidate for green roofs [20,21]. However, little is known about the salt tolerance of both *Sedum* spp. and *E. glomeratus*. Here in this study, we evaluated the salt tolerance of four herbaceous perennial ornamentals, including three varieties of *Sedum* and one variety of *Evolvulus*.

2. Materials and Methods

2.1. Plant Materials and Culture

Four varieties of perennial ornamentals were selected for this study: ‘Angelina’ (*S. rupestre*), ‘Autumn Joy’ (*S. telephium*), ‘Blue Spruce’ (*S. reflexum*), and ‘Blue Daze’ (*E. glomeratus*). Rooted cuttings were donated by Southwest Perennials (Dallas, TX, USA). The cuttings were transplanted to 6-in pots filled with Metro Mix 360 potting substrate (Sungro, Agawam, MA, USA). Fifteen days after transplanting, Marathon 1% granular (OHP Inc., Mainland, PA, USA), a systemic insecticide, was applied as a top dressing to the substrate at a rate of 2 grams per pot as a preventative measure against any potential pests.

2.2. Treatments and Irrigation

A total of three treatments were used, including two saline solutions and a control. The control was a nutrient solution prepared with reverse osmosis (RO) water and Peter’s Excel 15-5-15 (JR Peters Inc., Allentown, PA, USA) at a rate of 200 ppm N. The electrical conductivity (EC) of the control solution was 1.5 mS/cm and the pH was 6.5. The two saline solutions, referred to as EC5 and EC10, were prepared in the same manner as the control with the addition of NaCl to increase the EC to 5.0 and 10.0 mS/cm, respectively. These saline solutions represent moderate and high levels of salinity that exceed the tolerance threshold of most plants [22]. The treatments were initiated 29 days after transplanting for a duration of eight weeks and applied over-head on an as-needed basis. Each plant received 1 L of treatment solution per irrigation which provided an approximate 30% leaching fraction to minimize the accumulation of salts in the substrate while also minimizing waste of the treatment solutions.

2.3. Greenhouse Environmental Conditions

The experiment was conducted in a temperature-controlled greenhouse located at Texas A&M AgriLife Research Center in El Paso, Texas. Throughout the experiment, average day/night air temperature was 28 °C/19 °C and the average relative humidity ranged from 17 to 46%. The average daily photosynthetic photon flux density (PPFD) was 450 $\mu\text{mol}/\text{m}^2/\text{s}$ and the maximum was 1021 $\mu\text{mol}/\text{m}^2/\text{s}$.

2.4. Visual Quality and Leachate

The visual quality of the foliage was assessed for damage caused by the saline treatments, including leaf burn and defoliation, and plants were scored on a scale of 1–5, where 1 = severe leaf burn and defoliation; 2 = moderately-severe leaf burn and defoliation; 3 = moderate leaf burn, 4 = minimal leaf burn; and 5 = no leaf burn. Increments of 0.5 were used to improve the assessment for plants that were difficult to score. Here, a final rating of 1–2 is considered sensitive to salinity, 3 is considered moderately sensitive to salinity, and 4–5 is considered tolerant to salinity. Visual score was recorded weekly throughout the experiment. Leachate was also collected weekly according to the pour-through method [23] from six random plants per treatment and the leachate EC was recorded.

2.5. Growth Measurements

At the termination of the study, which was eight weeks after treatment initiation, shoots were separated from roots at the substrate level and the roots were gently shaken to remove the substrate, rinsed in RO water, and blotted dry. All tissue samples were brought to complete dryness in a forced-air oven at 60 °C and then dry weight was recorded. Due to the varying growth habits of the four varieties, comparisons were made using the relative values of shoot, root, and total dry weight (DW). Relative values were calculated as follows:

$$(y_{i,\text{salt}}/\bar{y}_{\text{control}}) \times 100$$

2.6. Tissue Mineral Analysis

Tissue samples were ground in a Wiley mill (Thomas Scientific, Swedesboro, NH, USA) to pass a 40-mesh screen and the dry powder samples were used for analysis. The samples were digested with nitric acid and then analyzed for Na⁺, K⁺, and Ca²⁺ content using inductively coupled plasma (ICP) optical emission spectrometry [24,25]. Chloride content was determined using a 2% acetic acid extraction [26] and a M926 Chloride Analyzer (Cole Parmer Instrument Company, Vernon Hills, IL, USA).

2.7. Experimental Design and Statistical Analysis

A total of three treatments, four varieties, and six replications ($n = 72$) were randomized and all data were analyzed as a two-way analysis of variance (ANOVA) at $\alpha = 0.05$ using JMP 14.2 (SAS Inc., Cary, NC, USA). Means were separated using Tukey's Honest Significant Difference (HSD) test at $\alpha = 0.05$.

3. Results

3.1. Leachate

The mean leachate EC throughout the experiment of control, EC5, and EC10 treatments was 2.7, 7.3, and 12.8 mS/cm, respectively (Figure 1). The leachate EC was relatively constant in the control but varied in the EC5 and EC10 treatments. For the EC5 treatment, the leachate EC increased up to 8.0 mS/cm by the final week of the experiment. For the EC10 treatment, the leachate EC fluctuated from 15.4 mS/cm at week four to 12.9 mS/cm at week eight.

3.2. Relative Tissue DW

There were significant effects of treatment and varieties, but not their interactions, for relative shoot, root, and total DW (Table 1). The relative tissue DW of all four varieties decreased with increasing salinity compared to the control (Table 2). Shoot growth appeared to be more sensitive than root growth only in Angelina and Blue Daze when treated with EC5. Relative shoot DW was highest in Blue Daze and lowest in Angelina and Blue Spruce. The relative shoot DW of Autumn Joy was not significantly different from the other varieties, indicating high variability of shoot growth in response to the EC5 treatment. The relative root DW was highest in Blue Daze and lowest in Angelina, Autumn

Joy, and Blue Spruce. The relative total DW was most reflective of shoot growth in that Blue Daze had the highest, Angelina and Blue Spruce had the lowest, and Autumn Joy was not significantly different from the other varieties. For plants treated with EC10, relative shoot, root, and total DW was highest in Blue Daze and lowest for Angelina, Autumn Joy, and Blue Spruce.

Figure 1. Mean electrical conductivity (EC) of leachate throughout the experiment. Six random containers per treatment were measured on a weekly basis. Bars represent standard deviation.

Table 1. Analysis of Variance (ANOVA) summary of all response variables measured from four varieties of perennial ornamentals irrigated with control or saline solutions for eight weeks.

Source	Relative Shoot DW	Relative Root DW	Relative Total DW	Visual Score	Na ⁺	Cl ⁻	Ca ²⁺	K ⁺	K ⁺ /Na ⁺ Ratio
Model	<0.0001 *	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001
Treatment	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001
Variety	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001
Treatment x Variety	0.2455	0.8780	0.2191	<0.0001	<0.0001	<0.0001	00.0057	<0.0001	<0.0001

* Significance levels of ANOVA.

Table 2. Relative means of shoot, root, and total dry weight (DW) of four varieties of perennial ornamentals irrigated with control or saline solutions for eight weeks.

Varieties	EC5		EC10	
Relative shoot DW (%)				
Angelina	37.29	B a ^z	19.85	B b
Autumn Joy	53.28	AB a	15.70	B b
Blue Daze	72.47	A a	42.42	A b
Blue Spruce	43.93	B a	20.30	B b
Relative root DW (%)				
Angelina	57.94	B a	19.19	B b
Autumn Joy	53.78	B a	14.69	B b
Blue Daze	91.46	A a	57.93	A b
Blue Spruce	47.89	B a	14.06	B b
Relative total DW (%)				
Angelina	38.44	B a	19.81	B b
Autumn Joy	53.42	AB a	15.40	B b
Blue Daze	74.92	A a	44.42	A b
Blue Spruce	44.12	B a	19.88	B b

^z Means followed by different letters indicate a significant difference according to Tukey's HSD test ($p < 0.05$); uppercase letters among varieties (within columns) and lowercase letters among treatments (within rows).

3.3. Visual Score

There were significant effects of treatment and varieties, including their interaction, for visual score (Table 1). The interaction indicated different responses in visual quality of the four varieties to the salt treatments (Figure 2). In general, the salt treatments reduced the visual score in all varieties except Autumn Joy (Figure 3). For the control plants, there were no significant variety differences in visual score. For plants treated with EC5, Autumn Joy and Blue Spruce had the highest visual score (5.00 and 4.83, respectively) and Angelina and Blue Daze had the lowest (4.33 and 4.17, respectively). For plants treated with EC10, Autumn Joy had the highest visual score (4.75), followed by Blue Spruce (4.08), while Angelina, and Blue Daze had the lowest (3.58 and 3.25, respectively). Declines in visual score were observed as early as week four when treated with EC5 and week three when treated with EC10.

Figure 2. Representative photos of the four varieties of perennial ornamentals (Angelina (A), Autumn Joy (B), Blue Daze (C), and Blue Spruce (D)) irrigated with control (CNT) or saline solutions for eight weeks. Photos were taken at termination of the experiment.

Figure 3. Mean visual score of foliar salt damage of four perennial ornamentals irrigated with control (A), EC5 (B), and EC10 (C) treatments for eight weeks. The visual score was based on a scale of 1–5, where 1 = severe leaf burn and defoliation; 2 = moderately-severe leaf burn and defoliation; 3 = moderate leaf burn; 4 = minimal leaf burn; 5 = no leaf burn. Letters indicate significant differences among varieties for the final week according to Tukey's HSD test ($p < 0.05$). Bars represent standard deviation.

3.4. Ion Concentrations

There were significant effects for treatment and varieties, including their interactions, for Na^+ , Cl^- , Ca^{2+} , and K^+ concentrations in the shoot tissue and also the K^+/Na^+ ratio (Table 1). The interactions indicated different responses in ion concentrations of the four varieties to the salt treatments. Overall, the concentrations of Na^+ and Cl^- in the shoot tissue increased with increasing salinity while Ca^{2+} and K^+ tended to decrease (Table 3). The K^+/Na^+ ratio decreased in the salt treatments compared to the control. Autumn Joy had the lowest concentrations of Na^+ in the shoot tissue and the highest K^+/Na^+ ratio. Blue Daze had the lowest concentrations of Cl^- , K^+ , and Ca^{2+} and the lowest K^+/Na^+

ratio. Numerically, Angelina had the highest concentrations of Na^+ and Cl^- in the shoot tissue when treated with EC10. Overall, the concentrations of Na^+ in the shoot tissue were lower than those of Cl^- , for all treatments.

Table 3. Mean concentrations of Na^+ , Cl^- , Ca^{2+} , and K^+ , and the K^+/Na^+ ratio in the shoot tissue of four varieties of perennial ornamentals irrigated with control or saline solutions for eight weeks.

Varieties	Control		EC5		EC10	
Na ⁺ (mg/g)						
Angelina	0.19	B c *	11.25	AB b	35.78	A a
Autumn Joy	0.19	B c	7.96	C b	16.35	C a
Blue Daze	0.46	A c	13.53	A b	26.38	B a
Blue Spruce	0.16	B c	9.23	BC b	32.15	AB a
Cl ⁻ (mg/g)						
Angelina	2.70	A c	34.49	A b	56.57	A a
Autumn Joy	1.56	B c	24.60	B b	46.49	A a
Blue Daze	0.86	C c	10.58	C b	26.85	B a
Blue Spruce	2.71	A c	24.79	B b	52.01	A a
Ca ²⁺ (mg/g)						
Angelina	49.90	A a	41.29	A b	42.91	A b
Autumn Joy	45.94	A a	43.87	A ab	38.05	B b
Blue Daze	14.01	C a	10.78	C b	8.82	D b
Blue Spruce	33.29	B a	32.18	B a	32.19	C a
K ⁺ (mg/g)						
Angelina	46.39	B a	41.34	B b	35.49	B c
Autumn Joy	56.65	A a	53.38	A ab	48.26	A b
Blue Daze	33.20	D a	19.07	C b	13.53	C c
Blue Spruce	38.97	C a	36.79	B a	32.34	B b
K ⁺ /Na ⁺ ratio						
Angelina	279.23	A a	3.73	B b	1.02	B b
Autumn Joy	304.20	A a	6.75	A b	2.99	A b
Blue Daze	73.91	B a	1.42	C b	0.52	C b
Blue Spruce	262.03	A a	4.19	B b	1.01	B b

* Means followed by different letters indicate a significant difference according to Tukey's HSD test ($p < 0.05$); uppercase letters among varieties (within ions and columns) and lowercase letters among treatments (within rows).

4. Discussion

4.1. Leachate

The leachate EC of the salt treatments tended to increase throughout the duration of the experiment indicating that salinity was increasing in the potting substrate and was representative of the dynamic nature of salinity. Salts can accumulate due to constant influx (e.g., irrigation) and insufficient outflux (i.e., leaching). Under natural field conditions, concentration of salts in the soil can also vary by means of evaporation, irrigation water, rising water tables, and rainfall [12,27].

4.2. Plant Growth

All four varieties showed reduced growth in response to salinity but no mortality was recorded throughout this experiment. Suppression of growth is the initial and most obvious response to the stress imposed on plants by salinity and can even lead to plant death [28]. For landscaping, reduced growth that does not lead to mortality of the plant can potentially be beneficial by limiting excessive growth and reducing upkeep. Shoot and root growth appeared to be similarly affected by salinity except in Angelina and Blue Daze when treated with EC5. The EC10 treatment showed similar relative

reductions in both shoot and root growth for all varieties, even though shoot tissue is generally considered to be more sensitive to salinity than root tissue [12]. Overall, the reductions in relative total DW between Blue Daze (*E. glomeratus*) and the three varieties of *Sedum* spp. indicated some variation in salt tolerance between the genera.

4.3. Visual Quality

For agricultural crops, yield is considered the most important trait for salt tolerance. However, for ornamental plants, aesthetic quality is the most important trait [16]. Therefore evaluating ornamentals for salt tolerance based on visual quality is practical [7]. In this study, symptoms of foliar damage were observed in the form of leaf necrosis and ranged from minimal to moderate. This was likely due to ion toxicity from the high concentrations of Na^+ and Cl^- in the shoots. However, the foliar damage was not severe enough to cause defoliation in any of the plants throughout this experiment. No foliar damage was observed in Autumn Joy and only minimal foliar damage was observed in Blue Spruce, Angelina, and Blue Daze when treated with EC5. When treated with EC10, minimal foliar damage was observed in Autumn Joy and Blue Spruce, while moderate foliar damage was observed in Angelina and Blue Daze. Based on these findings, Autumn Joy and Blue Spruce were considered relatively salt tolerant and Angelina and Blue Daze were less tolerant.

4.4. Ion Concentrations

The lower Na^+ concentrations that were observed in Autumn Joy indicate mechanisms for exclusion of Na^+ from the shoot tissue and possible retention in the root tissue, although this could not be confirmed in this study. An important feature of salt tolerant halophytes is the ability to withstand high concentrations of Na^+ in the shoot tissue where it is effectively compartmentalized in the vacuoles of cells [12,14]. However, Autumn Joy also had the highest concentrations of K^+ in the shoots and subsequently the highest K^+/Na^+ ratio compared to the other three varieties. The maintenance of high K^+ in the shoot tissue is important for osmotic adjustment and is a component of salt tolerance [12,29]. Conversely, Autumn Joy had high concentrations of Cl^- in the shoot tissue. Even though Cl^- is an essential macronutrient in plants, it can be toxic in the shoot tissue at concentrations ranging as low as 4–7 mg/g [30]. For Ca^{2+} , the decrease in concentrations in the shoot tissue that were observed in the salt-treated plants agrees with reports that sodium can displace calcium in the cell wall and plasma membrane of plants [29,31]. Blue Spruce had high concentrations of both Na^+ and Cl^- along with minimal foliar damage when treated with EC10 and indicates that this species was able to effectively sequester and tolerate these high concentrations in the shoot tissue. In contrast, the moderate foliar damage that was observed in Angelina and Blue Daze was likely due to cytosolic damage from these ions.

5. Conclusions

Salt tolerance in plants is a surprisingly complex trait and has garnered the attention of much research over many decades. In this study, we evaluated the relative salt tolerance of four varieties of herbaceous perennial ornamentals, three *Sedum* spp. and one *Evolvulus* species. Variation in relative salt tolerance was observed in the response variables measured among genera and species. Overall, Autumn Joy (*S. telephium*) and Blue Spruce (*S. reflexum*) showed salt tolerant traits, including the ability to tolerate high concentrations of Na^+ and/or Cl^- in the shoot tissue while maintaining good aesthetic quality with minimal damage observed in the shoot tissue.

Author Contributions: Conceptualization, G.N.; Data curation, T.H.; Formal analysis, T.H.; Funding acquisition, G.N.; Investigation, T.H.; Methodology, T.H.; Project administration, G.N.; Writing—original draft, T.H.; Writing—review & editing, G.N.

Funding: This research was partially funded by the Agriculture Research Service (ARS) Floriculture and Nursery Research Initiative (FNRI) and the USDA National Institute of Food and Agriculture Hatch Project TEX090450.

Acknowledgments: The authors thank Southwest Perennials, Inc. for donating plant materials for this study.

Conflicts of Interest: The authors declare no conflict of interest.

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