

ABSTRACT

NORDEN, DANIEL SCOTT. Development of Protocols for Containerized Culture of Seabeach Amaranth (*Amaranthus pumilus*) and Nitrogen Nutrition of Southern Seaoats (*Uniola paniculata*) Grown in the Float System. (Under the direction of Drs. Frank A. Blazich and Stuart L. Warren.)

Fresh seeds of seabeach amaranth (*Amaranthus pumilus* Raf.) were treated with K-GA₃ at 0, 100, 500, or 1000 mg•L⁻¹ for 24 hours and germinated at 25 °C or an 8/16-hour thermoperiod of 30/20 °C with daily photoperiods at each temperature of 0 (total darkness) or 16 hours. Germination was recorded every 3 days for 30 days. Nontreated seeds did not germinate. When germinated at 25 °C the response of seeds to K-GA₃ was linear for both photoperiods with significantly greater total germination in the dark for seeds treated with K-GA₃ at 100, 500, or 1000 mg•L⁻¹. At 25 °C, 84% germination was observed for seeds treated with K-GA₃ at 1000 mg•L⁻¹ and maintained in darkness, whereas for seeds exposed to a 16-hour photoperiod, maximum germination was 72%. The response to K-GA₃ at 30/20 °C was quadratic with maximum germination at predicted rates of K-GA₃ at 882 and 875 mg•L⁻¹ (93% and 91%, respectively) for photoperiods of 0 and 16 hours, respectively. Treatment of seeds with K-GA₃ removed physiological dormancy. For a second study, seeds of seabeach amaranth were moist-prechilled for 90 days at 4 °C or treated with K-GA₃ at 1000 mg•L⁻¹ for 24 hours. Both groups of seeds were sown in containers of two differing volumes, 139 or 635 cm³, with a substrate of 1 peat : 1 pine bark (v/v) amended with pulverized dolomitic limestone at 2.24 or 4.48 kg•m⁻³. The containers were maintained in a greenhouse and after seedling emergence, seedlings were fertilized with a 20N-4.4P-8.2K acidic, water soluble fertilizer or a 15N-2.2P-12.3K basic, water soluble fertilizer. Each fertilizer was applied thrice weekly at N application rates (NARs) of 75 to 300 mg•L⁻¹. Eight weeks after sowing, data were recorded. Regardless of fertilizer, top dry weight and leaf area of seabeach amaranth increased linearly with increasing NAR, and

maximum top dry weight and leaf area occurred with N at $300 \text{ mg}\cdot\text{L}^{-1}$, whereas root dry weight was unaffected by NAR. Both fertilizers increased electrical conductivity (EC) linearly with increasing NAR, and EC values of 1.15 to $1.18 \text{ dS}\cdot\text{m}^{-1}$ should be adequate for maximum growth. Substrate pH decreased linearly with increasing NAR 21, 43, and 57 days after initiation. Top and root dry weights, and leaf area were greater for seedlings from seeds treated with K-GA_3 . Seabeach amaranth grown in the large containers had top and root dry weights and leaf area 61%, 33%, and 57% larger, respectively, than plants grown in the smaller container volume. Top N concentration increased linearly with increasing NAR for acidic and basic fertilizers with N concentrations of 58.4 and $50.4 \text{ mg}\cdot\text{g}^{-1}$, respectively, at maximum top dry weight. Seabeach amaranth can be produced successfully utilizing an acid or basic fertilizer having a 4.5N-1P-1.9K or 6.8N-1P-5.6K ratio, respectively. As a third experiment, seeds of southern sea oats were sown in styrofoam float trays filled with a vermiculite-based hydroponic substrate. Trays were floated in plastic tubs (one tray per tub) containing a complete nutrient solution with N at 10 to $240 \text{ mg}\cdot\text{L}^{-1}$ from a 2N-3.5P-1K ratio (8N-14.1P-4.1K) liquid slow-release fertilizer. After 10 weeks data were recorded. Total plant, top, leaf, stem, and root dry weights increased quadratically with increasing N application rate (NAR) with maximum dry weights calculated to occur with N at 140 to $150 \text{ mg}\cdot\text{L}^{-1}$, respectively. Leaf area, root length, and root area were maximized with N at 157, 140, and $140 \text{ mg}\cdot\text{L}^{-1}$, respectively. Calculated leaf N concentration at maximum top dry weight was $31 \text{ mg}\cdot\text{g}^{-1}$. Southern sea oats can be grown successfully using the float system with optimum N rates of 140 to $150 \text{ mg}\cdot\text{L}^{-1}$ provided by a fertilizer having a 2N-3.5P-1K ratio.

Development of Protocols for Containerized Culture of Seabeach Amaranth (*Amaranthus pumilus*) and Nitrogen Nutrition of Southern Seaots (*Uniola paniculata*)
Grown in the Float System

by
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A thesis submitted to the Graduate Faculty of
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DEDICATION

I would like to dedicate my thesis to my entire family for their love and support.

To my parents, I can never begin to repay what you have done for me. Ever.

To my older brother, David, thank you for introducing me to horticulture and letting me follow
in your footsteps.

To my younger brother, Peter, thank you for being my source of inspiration; you have proven
that anything is possible.

BIOGRAPHY

Daniel Scott Norden was born November 18, 1980, to Roger and Carol Norden in Raleigh, North Carolina. Following in his older brother's footsteps, he quickly became interested with youth horticulture after winning his first Grand National Award for gardening from the National Junior Horticulture Association (NJHA) at the age of six. This led to Daniel becoming involved with horticulture through NJHA and 4-H organizations at the local, state, and national levels. He often competed in horticultural related contests sponsored by these organizations in topics such as photography, gardening, and environmental science and won over 30 National and Grand National Awards. Daniel attended primary and secondary schools in Raleigh and graduated from Enloe High School in 1999. He also began playing cello in the third grade and continued to play throughout high school and college.

In August 1999, he enrolled at NC State University, Raleigh, to pursue a BS degree in Horticultural Science. While enrolled at State, Daniel served for several years as a national officer in the NJHA including President from 2000 to 2001. He also played cello in the Raleigh Symphony Orchestra for 3 ½ years. Daniel was awarded a BS degree cum laude in Horticultural Science, May 2003.

Daniel was admitted in January 2004 to the Graduate School at North Carolina State University to pursue a MS degree in Horticultural Science. Following completion of the MS program he plans to pursue a career in the plant sciences.

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Fig. 1

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Fig. 2

Cumulative (30-day) seed germination of nonstratified seeds of seabeach amaranth as influenced by temperature, photoperiod, and K-GA₃ treatments. Following treatment with 0, 100, 500, or 1000 mg/liter (ppm) K-GA₃ for 24 hr, seeds were germinated at 25C or at an 8/16-hr thermoperiod of 30/20C with daily photoperiods at each temperature of 0 (total darkness) or 16-hr. Symbols represent mean percent germination of four petri dishes each containing 50 seeds and vertical bars = ± 1 SE. An asterisk denotes a significant difference at $P \leq 0.05$ between means of dark (0-hr photoperiod) and light (16-hr photoperiod) germination within each K-GA₃ rate at 25C (77F) or 30/20C (86/68F). 22

Chapter 3

Fig. 1

Influence of nitrogen application rate (NAR) on total plant, top, leaf, stem, and root dry weights of southern sea oats grown in the float system. Data points are means of six observations and vertical bars = ± 1 SE. 87

GENERAL INTRODUCTION

Seabeach amaranth (*Amaranthus pumilus* Raf.) is a summer annual native to the beaches and barrier islands of the Atlantic Coast and once ranged from Massachusetts to South Carolina (Weakley et al., 1996). By 1990 the species had disappeared from six of the nine states of its original range with the only remaining populations occurring in New York, North Carolina, and South Carolina (Weakley et al., 1996). Elimination from two-thirds of its historic range, and vulnerability of the plant to various threats, both natural and human, caused seabeach amaranth to be listed as “threatened” by the U.S. Fish and Wildlife Service (1993). As a result of the threatened status of the plant, a recovery plan was developed by Weakley et al. (1996).

One concern regarding loss of the species is that seabeach amaranth plays an important role in the initial stages of the development of sand dunes by trapping and binding sand on the beach (U.S. Fish and Wildlife Service N.Y. Field Office, 2004; Weakley et al., 1996). Ecologists also view the plant as an indicator species which allows one to access the vitality and vigor of a beach ecosystem. Thus, various state and federal agencies are interested in restoring the species to areas where it once grew. In addition, beach restoration and sand renourishment projects have created a demand for seedling transplants of seabeach amaranth that are currently unavailable.

To establish seabeach amaranth in locations where it was once endemic and to meet the demand for transplants will require protocols for propagation and culture. One logical approach would involve containerized production of seedlings that can then be planted in suitable beach environments. Countless studies have been published regarding production of various bedding plants and much of this research may be applicable to seabeach amaranth (Dole and Wilkins,

2005). Some research has been reported regarding sexual propagation of seabeach amaranth, specifically seed germination (Baskin and Baskin, 1998; Blazich et al., 2005), but more research is needed. If production protocols are developed, these would provide opportunities for growers to produce and sell plants to federal, state, and private agencies for recovery efforts.

Freshly harvested seeds of seabeach amaranth are physiologically dormant (have embryo dormancy) and require a period of stratification (moist-prechilling) to break (release) dormancy (Baskin and Baskin, 1998; Blazich et al., 2005). Stratification of 84 to 120 days is necessary to remove physiological dormancy completely followed by germination at high temperatures (e.g., 8/16-hour thermoperiod of 30/20 °C) with light (e.g., a daily 16 hour photoperiod) to achieve maximum germination (Baskin and Baskin, 1998; Blazich et al., 2005). The benefit of stratification raises the issue of whether a growth regulator such as gibberellic acid (GA) can ease germination by eliminating the need to stratify. Physiological seed dormancy of many species can be removed by treatment with various gibberellins, most notably GA₃ or GA₄₊₇ (Bewley and Black, 1994; Mayer and Poljakoff-Mayber, 1989).

Other factors that require investigation for development of protocols for production of seedling transplants of seabeach amaranth involve culture. Little if any quantitative data have been published regarding culture of the species. Skaradek and Murray (2005) reported successful greenhouse culture of seedlings of seabeach amaranth in “5 cm²” containers with “a mixture of half peat and half sand thoroughly moistened to saturation.” The seedlings were later transplanted to the field and grown in a loamy soil overlaid with 5 to 8 cm of sand. The plants grew in the field to maturity producing seeds. Unfortunately, the report of Skaradek and Murray (2005) is extremely sketchy and does not include such information as the volume of the containers in which the seedlings were grown, fertilization of the seedlings, and substrate pH.

Another dune-forming species, southern seaoats (*Uniola paniculata* L.) is a perennial dune grass that in most of its natural range (southern Virginia to the Yucatan Peninsula) is the dominant, coastal sand-binding plant species (Ricciuti, 1984). The species is generally subtropical and its native range is determined by climate as it is intolerant of extremely hot summers or cold winters (Ricciuti, 1984). In southern Virginia and northern North Carolina, southern seaoats is at the northern limit of its range and the plants usually die back to ground level and resprout from rhizomes in the spring. Seed germination occurs in late spring and little growth takes place until adequate sand surrounds the culms (stems), usually by the end of the second year (Woodhouse and Hanes, 1966). Seaoats has the ability to stabilize dunes upon establishment by utilizing culms and extensive root systems to trap sand (Ricciuti, 1984; Rogers and Nash, 2003). Thus, it has been planted extensively to build and stabilize coastal sand dunes (Latham, 2001; Woodhouse and Hanes, 1966).

In recent years, transplants of southern seaoats have been in demand to restore beaches and stabilize sand dunes along the southern Atlantic and Gulf Coasts of the United States damaged by tropical storms and erosion. Demand has in turn created a need for information regarding propagation and culture. Considerable research has been reported on propagation, specifically seed germination (Burgess et al., 2002, 2005; Seneca, 1969, 1972; Westra and Loomis, 1966). However, with few exceptions research on culture has been limited (Bachman and Whitwell, 1995; Hester and Mendelssohn, 1990; Seneca, 1972), particularly with respect to mineral nutrition of container-grown plants. Cultural information, coupled with previous work on seed germination, would result in protocols for production of transplants that may prove profitable to growers such as former and current tobacco farmers who are seeking alternative crops that can be grown in the float system.

Tobacco and vegetable transplants can be grown successfully using the float system (Jones et al., 1993; Liptay et al., 1992; Rideout, 2004; Rideout et al., 1994; Soundy et al., 2001). This system involves constructing shallow wooden frames on the floor of a greenhouse. The frames are lined with polyethylene sheeting and filled with nutrient solution. Seeds are sown in peat- or vermiculite-based soilless substrates in styrofoam trays (flats), floating on the nutrient solution. Irrigation is by capillary movement of nutrient solution into the substrate. Potential advantages of the float system over conventional overhead irrigation include lower production costs, more efficient use of water and mineral nutrients, reduced disease pressure (dry plant foliage), and elimination of nutrient leaching to groundwater below the greenhouse (Rideout, 2004). Frantz and Welbaum (1998) also noted if other crops could be produced successfully using the float system, float systems could potentially produce high-value horticultural crops to supplement farm incomes. Soundy et al. (2001) reported that although production of vegetable transplants using the float system has several advantages including improved health of seedlings, production of lettuce (*Lactuca sativa* L.) transplants using the float system resulted in poor root systems.

Some research has been conducted on growing alternative crops using the float system (Frantz and Welbaum, 1998; Rideout, 2004; Soundy et al., 2001; Welbaum et al., 2001). Nash [as reported by Latham (2001)], an Agricultural Extension Agent in New Hanover County, N. C., has produced transplants of seaots successfully utilizing the float system. Initially, he attempted to grow southern seaots using conventional container production but he encountered many problems particularly with foliar fungal diseases due to irrigating over the tops of plants (David L. Nash, personal communication). Using the float system reduced dramatically foliar infestations. However, despite successful culture of southern seaots using the float system, little

quantitative information has been published on this means of culture to produce transplants of seaoats.

In summary, research is needed for development of protocols for production of containerized transplants of seabeach amaranth in addition to research on mineral nutrition of southern seaoats when grown in a float system. The following research was therefore conducted to (A) study the influence of temperature, light, and K-GA₃ treatment on seed germination of seabeach amaranth, (B) develop protocols for containerized production of seedling transplants of seabeach amaranth by investigating such factors as the influence of K-GA₃ treatment of seeds on subsequent seedling growth, container volume, substrate pH, and N source and rate, and (C) study the influence of N nutrition on vegetative growth of southern seaoats when grown in the float system.

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Chapter 1

Seed Germination of Seabeach Amaranth (*Amaranthus pumilus*) in Response to Temperature,
Light, and Gibberellin A₃ Treatments

(In the format appropriate for submission to the
Journal of Environmental Horticulture)

Seed Germination of Seabeach Amaranth (*Amaranthus pumilus*) in Response to Temperature,
Light, and Gibberellin A₃ Treatments¹

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Abstract

Seeds of seabeach amaranth (*Amaranthus pumilus* Raf.) stored at 4C (39F) for approximately 1 year (harvested September 2003) and freshly harvested seeds (October 2004) were soaked in November 2004 in solutions of the potassium (K) salt (K-salt) of gibberellin A₃ (K-GA₃) at 0, 100, 500, or 1000 mg/liter (ppm) for 24 hr in darkness. After treatment, seeds were germinated at 25C (77F) or at an 8/16-hr thermoperiod of 30/20C (86/68F) with daily photoperiods at each temperature of 0 (total darkness) or 16 hr. Germination was recorded every 3 days for 30 days. Stored and fresh seeds responded similarly. However, the 2003 seeds had greater viability (percent germination) and vigor (germinated faster) and these data are presented. Regardless of germination temperature and photoperiod, nontreated seeds [0 mg/liter (ppm) K-GA₃] did not germinate. When germinated at 25C (77F) the response of seeds to K-GA₃ treatment was linear for both photoperiods with significantly greater total (30-day) germination occurring in the dark for seeds treated with 100, 500, or 1000 mg/liter (ppm) K-GA₃. At 25C (77F), the greatest total germination (84%) was observed for seeds treated with 1000 mg/liter (ppm) K-GA₃ and maintained in darkness, whereas for seeds exposed to a 16-hr photoperiod, maximum germination was 72%. At 30/20C (86/68F) the response to K-GA₃ was quadratic with maximum germination at predicted rates of 882 and 875 mg/liter (ppm) K-GA₃ (93% and 91%, respectively) for photoperiods of 0 and 16 hr, respectively. Treatment of nonstratified seeds of seabeach amaranth with K-GA₃ removed physiological (embryo) dormancy and eliminates the need for stratification (moist-prechilling). Treatment also reduced sensitivity of the seeds to light, and appeared to broaden the range of temperatures for germination.

Index words: sexual propagation, beach restoration, seed dormancy, seed stratification, Amaranthaceae, recovery plans, threatened species, dune species.

Significance to the Nursery Industry

To remove physiological (embryo) seed dormancy of seabeach amaranth (*Amaranthus pumilus*), a species federally listed as “threatened,” the seeds must be stratified (moist-prechilled) for 84 to 120 days. Results herein indicate treatment of seeds for 24 hr with a solution of 1000 mg/liter (ppm) K-GA₃ will remove physiological dormancy without the need of lengthy stratification. Treatment with K-GA₃ also eliminates the need for light to maximize germination and may broaden the range of temperatures at which germination occurs.

Introduction

Seabeach amaranth is a summer annual native to the beaches and barrier islands of the Atlantic Coast. The plant once ranged from Massachusetts to South Carolina (9). By 1990 it no longer occurred in six of the nine states of its original range with the remaining populations occurring in New York, North Carolina, and South Carolina (9). Elimination of two-thirds of its historic range, and vulnerability of the plant to various threats, both natural and human, caused seabeach amaranth to be listed as “threatened” by the U.S. Fish and Wildlife Service in 1993 (7). As a result of the threatened status of the plant, a recovery plan was developed by Weakley et al. (9).

One concern regarding loss of the species is that seabeach amaranth plays an important role in the initial stages of the development of sand dunes by trapping and binding sand on the beach (8,9). Ecologists also view the plant as an indicator of the vitality and vigor of a beach ecosystem. Thus, various state and federal agencies are interested in restoring the species to

areas where it once grew. In addition, beach restoration and sand renourishment projects have created a demand for seedling transplants of seabeach amaranth that are currently unavailable.

To establish seabeach amaranth in locations where it was once endemic and to meet the demand for transplants will require protocols for propagation and culture. One approach may involve production of seedlings that can then be planted in suitable beach environments. Some research has been reported regarding sexual propagation, specifically seed germination (2,4), but more research is needed. If production protocols are developed, these would provide opportunities for growers to produce and sell plants to federal, state, and private agencies for recovery efforts.

Researchers have determined that freshly harvested seeds of seabeach amaranth are physiologically dormant (have embryo dormancy) and require a period of stratification to break (release) dormancy (2,4). Stratification of 84 to 120 days is necessary to remove physiological dormancy completely followed by germination at high temperatures [e.g., 8/16-hr thermoperiod of 30/20 (86/68F)] with light (e.g., a daily 16 hr photoperiod) to achieve maximum germination (2,4). The benefit of stratification raises the issue of whether a growth regulator such as gibberellic acid (GA) can ease germination by eliminating the need to stratify. Physiological seed dormancy of many species can be removed by treatment with various gibberellins, most notably GA₃ or GA₄₊₇ (3,5). Therefore, the following research was conducted to study the influence of temperature, light, and GA treatment on seed germination of seabeach amaranth.

Materials and Methods

Mature utricles (one-seeded, small, indehiscent, bladderly fruit) of seabeach amaranth were collected from plants growing on Oak Island, North Carolina on September 15, 2003. The plants were growing on the beach berm in the area between the high tide line of the Atlantic Ocean and the toe of the frontal dune. As the utricles were collected they were placed in a paper

bag, and left to dry at 21C (70F). On October 6, 2003, the utricles, still in the paper bag, were transported to Raleigh and the fruit were placed in a plastic dish pan for additional drying at 21C (70F). From October 16 to October 20, 2003, the seeds were extracted by hand by rubbing them between the palms. Extracted seeds were placed in a covered glass petri dish and stored in the dark at 21C (70F). After extraction, seeds were cleaned manually by removal of chaff and other debris, and left in the dark at 21C (70F). On November 18, 2003, the seeds were placed in a glass bottle, the bottle was sealed, and the bottle placed in a refrigerator maintained at 4C (40F).

On October 22, 2004, mature fruit of seabeach amaranth were collected from plants growing on Emerald Isle, North Carolina. These plants were also growing in a beach environment like that of the plants from which fruit were collected in September 2003. The utricles were handled like those harvested in September 2003 and they were transported the same day collected to Raleigh for drying, seed extraction, and cleaning as described for the 2003 seeds. Following cleaning, these seeds were stored in the dark at 21C (70F).

On November 3, 2004, the 2003 and 2004 seeds were removed from storage and graded under a dissecting scope to remove abnormal or damaged seeds and any debris not removed by previous cleaning. From the graded seeds, four lots consisting of 800 seeds per lot of the year 2003 seeds, and four lots consisting of 800 seeds each of the year 2004 seeds were removed from the graded seeds. Each lot of 800 seeds was placed in a 125-ml Erlenmeyer flask containing 50 ml of solution of the potassium (K) salt (K-salt) of GA_3 (K- GA_3) at 0, 100, 500, or 1000 mg/liter (ppm). The solutions were prepared by dissolving K- GA_3 in distilled water (pH of distilled water = 6.3) and the control [0 mg/liter (ppm)] was distilled water. Flasks were wrapped with aluminum foil to exclude light, and placed on a rotary shaker (100 revolutions per minute) for 24 hr. Next, seeds were removed from the flasks and placed in covered, 9-cm diameter glass petri dishes (50 seeds per dish) with each dish containing two germination blotters uniformly

moistened with tap water. The blotters had been soaked previously in tap water for 48 hr. Dishes were placed in black sateen cloth bags and left overnight at 21C (70F). The next day, dishes were randomized within two growth chambers [C-chambers (6)] at the Southeastern Plant Environment Laboratory (NC State University Phytotron, Raleigh). The chambers were maintained at 25C (77F) or an 8/16-hr thermoperiod of 30/20C (86/68F). Chamber temperatures varied within 0.5C (0.9F) of the set point.

Within each temperature regime, seeds were subjected to daily photoperiods of 0 (total darkness) or 16 hr. Regardless of temperature, the 16-hr photoperiod treatment was imposed the same time each day. The photoperiod treatment for the alternating temperature of 30/20C (86/68F) began with the transition to the high temperature portion of the cycle.

Growth chambers were equipped with cool-white fluorescent lamps that provided a photosynthetic photon flux (400-700 nm) of approximately $32 \mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$ (2.4 klx) as measured outside the dishes at dish level with a cosine-corrected LI-COR LI-185 quantum/radiometer/photometer (LI-COR, Lincoln, NE). Regardless of photoperiod, temperature within the petri dishes, as measured with a thermocouple, never exceeded ambient temperature by more than $\pm 0.5\text{C}$ (0.9F) of the set point. The constant darkness treatment was imposed by keeping the petri dishes in black sateen cloth bags throughout the experiment, and all germination counts and moistening of the blotters for this treatment were performed utilizing a fluorescent lamp equipped with a #122 Roscolux green diffusion filter (Rosco Laboratories, Inc., Stamford, CT). Germination blotters were kept moist throughout the experiment. Seeds showing signs of decay were removed from the dishes when recording germination.

For each temperature and year of seed collection (2003 and 2004), all photoperiod and K-GA₃ treatments were replicated four times with a replication consisting of a petri dish containing 50 seeds. Germination counts were recorded every 3 days for 30 days and germinated seeds

were removed from the dishes. A seed was considered germinated when radicle emergence was ≥ 1 mm (0.04 in). Percent germination was calculated as a mean of four replications per treatment. Within each temperature regime, the experiment was a $2 \times 2 \times 4 \times 10$ factorial in a completely random design. The main plots were 2 collection dates/locations, 2 photoperiods, 4 concentrations of K-GA₃, and 10 germination counts (every 3 days for 30 days). All data were subjected to analysis of variance (ANOVA) procedures. Data were analyzed following arsin transformation and results were similar to nontransformed data. Hence, results presented are based on nontransformed data.

Results and Discussion

Even though seeds differed in location and year of collection, and duration of storage, statistical analysis revealed both groups responded similarly although the 2003 seeds had greater viability (percent germination) and vigor (germinated faster). Therefore, subsequent statistical analysis focused on data of the year 2003 seeds and are discussed and presented.

ANOVA showed that within each temperature regime, light, K-GA₃, time (days), and their interactions were highly significant ($P < 0.001$). Thus, within each temperature, K-GA₃ concentration data were fitted to simple linear and quadratic equations within each time \times photoperiod combination. The maximum of the polynomial curve was calculated as a first order derivative of the independent variable where the dependent variable equaled zero. Likewise, within each temperature, cumulative (30-day) germination as influenced by photoperiod within each K-GA₃ concentration was separated by the F test.

Nontreated seeds [0 mg/liter (ppm) K-GA₃] did not germinate regardless of temperature and photoperiod (Figs. 1 and 2). At 25C (77F) the response of seeds to K-GA₃ was linear with significantly greater germination occurring in darkness for seeds treated with 100, 500, or 1000

mg/liter (ppm) K-GA₃ compared to the 16-hr photoperiod (Fig. 2). The greatest total germination at 25C (77F) was 84% for seeds treated with 1000 mg/liter (ppm) K-GA₃ and kept in darkness, whereas for seeds exposed to a 16-hr photoperiod, 72% germination was realized at 1000 ppm K-GA₃. Previous research by Blazich et al. (4) demonstrated seed germination of seabeach amaranth is negligible at 25C (77F) whether or not the seeds are first stratified prior to being placed at 25C and whether or not stratified seeds are subjected to a 16-hr photoperiod at 25C (77F). When nonstratified seeds of seabeach amaranth were treated in the present investigation with K-GA₃, they not only germinated at 25C (77F) but greater germination occurred in darkness as opposed to seeds exposed to light.

At 30/20C (86/68F) the response to K-GA₃ was quadratic with maximum total germination at predicted rates of 882 and 875 mg/liter (ppm) K-GA₃ (93% and 91%, respectively) for seeds exposed to photoperiods of 0 or 16 hr, respectively (Fig. 2). Photoperiod had no influence on germination of seeds treated with 500 or 1000 mg/liter (ppm) K-GA₃ and germinated at 30/20C (86/68F), but seeds treated with 100 mg/liter (ppm) K-GA₃ and germinated in light had significantly greater germination than seeds in darkness. These results, like those at 25C (77F) are also intriguing because Blazich et al. (4) reported that following seed stratification of seabeach amaranth for 120 days and germination in the dark at 30/20C (86/68F), germination of 49% was observed by day 12 increasing to 50% by day 30, whereas for seeds exposed to a daily 16-hr photoperiod, 82% germination was realized by day 12 and increased to 85% by day 30.

As mentioned previously, the present research included seeds collected in 2003 and stored dry for approximately a year at 4C (40F) and seeds collected in 2004 which were stored briefly under dry conditions at 21C (70F) and can be regarded as fresh seeds. Statistical analysis revealed both groups responded in a similar manner although the 2003 seeds had greater viability

and vigor. Since the 2003 seeds did not germinate unless treated with K-GA₃, indicates physiological seed dormancy of seabeach amaranth persists over time.

Treatment of seeds with 100 mg/liter (ppm) K-GA₃ was only slightly effective in removing physiological dormancy of seabeach amaranth in comparison to treatment of seeds with 500 or 1000 mg/liter (ppm) K-GA₃ (Figs. 1 and 2). Reduced effectiveness of K-GA₃ may have simply been a dose response. Another possible explanation may have been related to the pH of the 100 mg/liter (ppm) K-GA₃ solution. Following preparation of the K-GA₃ solutions with distilled water (pH = 6.3), the pH of the 100, 500, and 1000 mg/liter (ppm) K-GA₃ solutions was 5.8, 3.9, and 3.7, respectively. Arnold et al. (1) reported removal of embryo dormancy of seeds of dwarf snapdragon [*Chaenorrhinum minus* (L.) Lange] by treatment with various gibberellins was influenced by many factors including solution pH, concentration of the solutions, duration of treatment, and the kind/type of giberellin used to treat the seeds. They also observed pH optimum varied between seeds stored for 10 months at room temperature and freshly harvested seeds.

Since a seed was considered germinated when radical emergence was ≥ 1 mm (0.04 in), the authors did not observe subsequent development of the germinated seeds to determine if the K-GA₃ treatments, particularly at the higher concentrations of 500 and 1000 ppm, had any deleterious affects on subsequent seedling growth. However, observations from a later nonreplicated study indicated seedlings resulting from seeds treated with 1000 ppm K-GA₃ grew normally.

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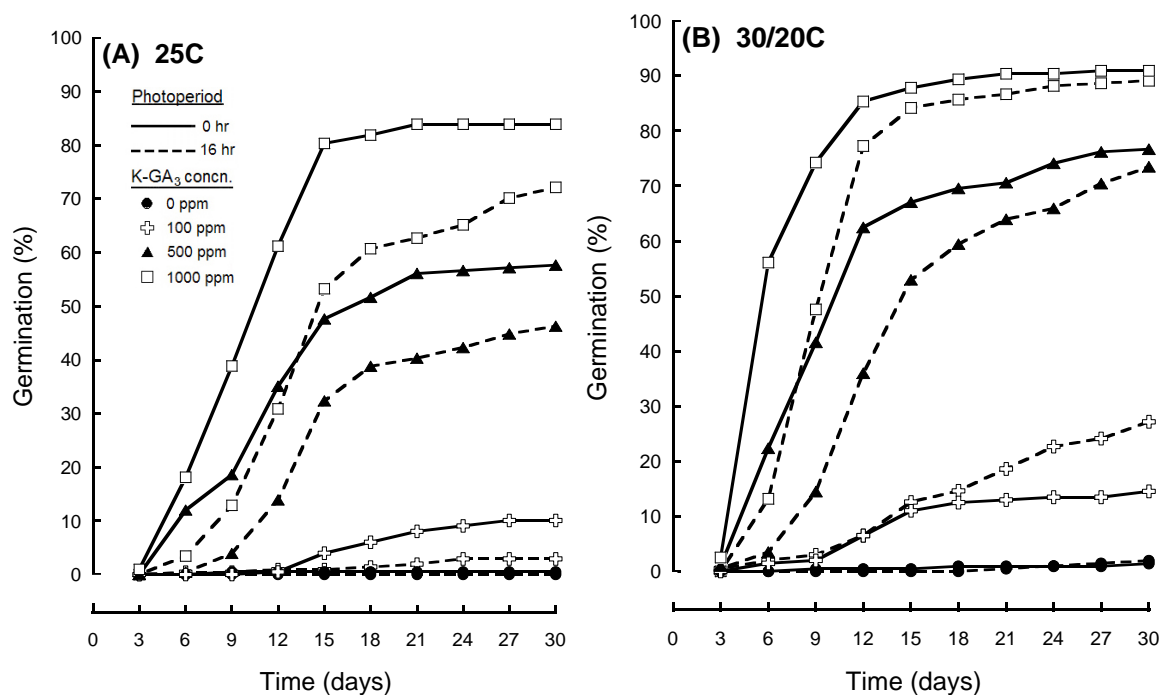


Fig. 1. Influence of temperature, photoperiod, and K-GA₃ treatments on time course of seed germination of nonstratified seeds of seabeach amaranth. Following treatment with 0, 100, 500, or 1000 mg/liter (ppm) K-GA₃ for 24 hr, seeds were germinated at (A) 25C (77F) or at an (B) 8/16-hr thermoperiod of 30/20C (86/68F) with daily photoperiods at each temperature of 0 (total darkness) or 16-hr. Symbols in (A) and (B) represent mean percent germination of four petri dishes each containing 50 seeds.

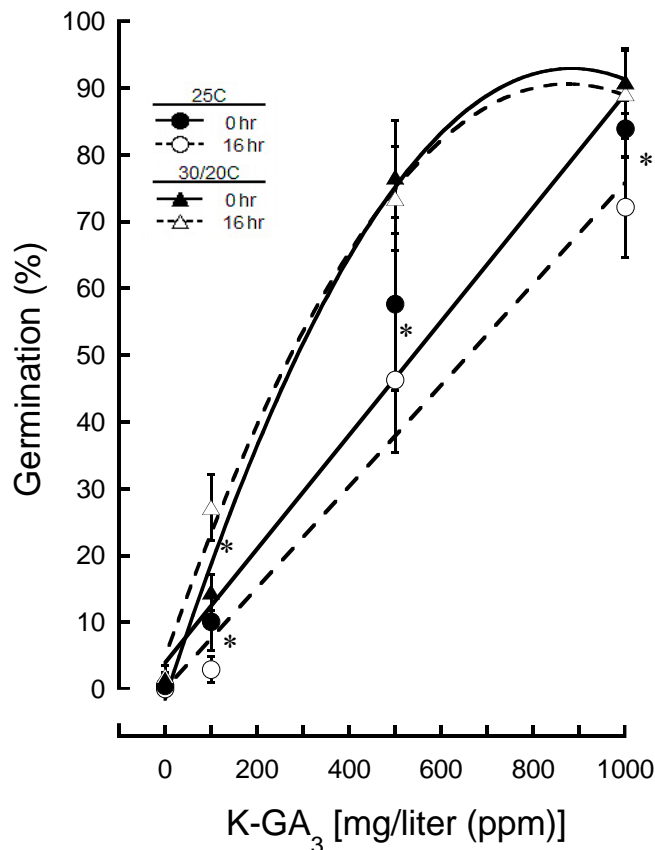


Fig. 2. Cumulative (30-day) seed germination of nonstratified seeds of seabeach amaranth as influenced by temperature, photoperiod, and K-GA₃ treatments. Following treatment with 0, 100, 500, or 1000 mg/liter (ppm) K-GA₃ for 24 hr, seeds were germinated at 25C or at an 8/16-hr thermoperiod of 30/20C with daily photoperiods at each temperature of 0 (total darkness) or 16-hr. Symbols represent mean percent germination of four petri dishes each containing 50 seeds and vertical bars = ± 1 SE. An asterisk denotes a significant difference at $P \leq 0.05$ between means of dark (0-hr photoperiod) and light (16-hr photoperiod) germination within each K-GA₃ rate at 25C (77F) or 30/20C (86/68F). Regression equations are as follows: 25C (77F), 0-hr photoperiod, $y = 3.92 + 0.085x$, $R^2 = 0.98$; 25C (77F), 16-hr photoperiod, $y = -0.001 + 0.076x$, $R^2 = 0.99$; 30/20C (86/68F), 0-hr photoperiod, $y = -1.5 + 0.214x - 0.0001x^2$, $R^2 = 0.99$; 30/20C (86/68F), 16-hr photoperiod, $y = 4.8 + 0.196x - 0.0001x^2$, $R^2 = 0.99$.

Chapter 2

Gibberellin A₃ Treatment of Seeds, Container Volume, Substrate pH, and Nitrogen Source and Rate, Influence Growth of Containerized Seabeach Amaranth (*Amaranthus pumilus*)

(In the format appropriate for submission to HortScience)

Subject Category: Crop Production-Nursery and Landscape Plants

Gibberellin A₃ Treatment of Seeds, Container Volume, Substrate pH, and Nitrogen Source and Rate, Influence Growth of Containerized Seabeach Amaranth (*Amaranthus pumilus*)

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Subject Category: Crop Production-Nursery and Landscape Plants

Gibberellin A₃ Treatment of Seeds, Container Volume, Substrate pH, and Nitrogen Source and Rate, Influence Growth of Containerized Seabeach Amaranth (*Amaranthus pumilus*)

Additional index words: beach restoration, Amaranthaceae, recovery plans, threatened species, dune species, mineral nutrition, sexual propagation

Abstract. Seeds of seabeach amaranth (*Amaranthus pumilus* Raf.), a species federally listed as ‘threatened,’ were stratified (moist-prechilled) for 90 days at 4°C or treated with a solution of K-GA₃ at 1000 mg•L⁻¹ for 24 hours. After treatment, both groups of seeds were sown in containers of two differing volumes, 139 or 635 cm³ with a substrate of 1 peat : 1 pine bark (v/v) amended with one of two rates of pulverized dolomitic limestone (2.24 or 4.48 kg•m⁻³). The containers were maintained in a greenhouse and after seedling emergence, seedlings were fertilized with a 20N-4.4P-8.2K acidic, water soluble fertilizer or a 15N-2.2P-12.3K basic, water soluble fertilizer. Each fertilizer was applied thrice weekly at N application rates (NARs) of 75, 150, 225, or 300 mg•L⁻¹. Eight weeks after initiation (sowing of seeds), the study was terminated and data recorded. Regardless of fertilizer, acidic or basic, top dry weight and leaf area of seabeach amaranth increased linearly with increasing NAR, and maximum top dry weight and leaf area occurred with N at 300 mg•L⁻¹, whereas root dry weight was unaffected by NAR. Both fertilizers increased electrical conductivity (EC) linearly with increasing NAR, and EC values of 1.15 to 1.18 dS•m⁻¹ should be adequate for maximum growth. Substrate pH decreased linearly with increasing NAR 21, 43, and 57 days after initiation. Top and root dry weights, and leaf area were greater for seedlings resulting from seeds treated with K-GA₃ compared to seedlings produced from stratified seeds. Seabeach amaranth grown in the large containers had top and root dry weights and leaf area 61%, 33%, and 57% larger, respectively, compared to plants

grown in the smaller container volume. Top N concentration increased linearly with increasing NAR for acidic and basic fertilizers with N concentrations of 58.4 and 50.4 mg•g⁻¹, respectively, at maximum top dry weight. Similarly, top nutrient content of N increased linearly with NAR, however, top N content was unaffected by either rate of limestone or type of fertilizer. Seabeach amaranth can be produced successfully in containerized production with maximum top growth occurring with N at 300 mg•L⁻¹ provided by an acid or basic fertilizer having a 4.5N-1P-1.9K or 6.8N-1P-5.6K ratio, respectively.

Seabeach amaranth is a summer annual native to the beaches and barrier islands of the Atlantic Coast and once ranged from Massachusetts to South Carolina (Weakley et al., 1996). However, by 1990 it no longer occurred in six of the nine states of its original range with the remaining populations occurring in New York, North Carolina, and South Carolina (Weakley et al., 1996). Disappearance of the species from a large portion of its historic range and vulnerability of the plant to various threats, both natural and human, resulted in seabeach amaranth being listed as ‘threatened’ by the U.S. Fish and Wildlife Service (1993). The threatened status of the plant resulted in development of a recovery plan by Weakley et al. (1996).

Loss of seabeach amaranth from many areas where it was once endemic has raised concerns as it plays an important role in the initial stages of the development of sand dunes by trapping and binding sand on the beach (U.S. Fish and Wildlife Service New York Office, 2004; Weakley et al., 1996). Weakley et al. (1996) described the plant as “a classic example of a fugitive species”—“an inferior competitor which is always excluded locally under interspecific competition, but which persists in newly disturbed habitats by virtue of its high dispersal ability; a

species of temporary habitats [Lincoln et al. (1982)].” The plant is also regarded by ecologists as an indicator species which allows one to assess the vitality and vigor of a beach ecosystem. Thus, various state and federal agencies are interested in restoring seabeach amaranth to areas where it once grew. Beach restoration and sand renourishment projects have also created a demand for seedling transplants of the species that are unavailable. Therefore, to reestablish the plant in locations where it was once endemic and to meet the demand for transplants will require protocols for propagation and culture. One logical approach would involve production of seedlings that can be planted in suitable beach environments. Countless studies have been published regarding production of various bedding plants and much of this research may be applicable to seabeach amaranth (Dole and Wilkins, 2005). If production protocols are developed for the species they may provide opportunities for growers to produce and sell plants to federal, state, and private agencies for recovery efforts.

Seeds of seabeach amaranth are relatively easy to germinate provided one is aware of previous research (Baskin and Baskin, 1998; Blazich et al. 2005). Freshly harvested seeds of the species are physiologically dormant (have embryo dormancy) and require a period of stratification (moist-prechilling) to break (release) dormancy (Baskin and Baskin, 1998; Blazich et al., 2005). Stratification of 84 to 120 d is necessary to remove physiological dormancy completely followed by germination at high temperatures (e.g., 8/16 h thermoperiod of 30/20 °C) with light (e.g., a daily 16-h photoperiod) to achieve maximum germination. A recent report by Norden et al. (2007) noted the need for lengthy stratification to remove embryo dormancy can be eliminated by treatment of the seeds with the potassium (K) salt (K-salt) of gibberellin A₃ (K-GA₃). Treatment of the seeds for 24 h with a solution of K-GA₃ at 1000 mg•L⁻¹ will remove physiological dormancy without the need for stratification. Treatment of the seeds with K-GA₃

also reduces sensitivity of the seeds to light and appears to broaden the range of temperatures at which germination will occur. Although Norden et al. (2007) reported K-GA₃ treatment will remove physiological seed dormancy of seabeach amaranth, they did not observe subsequent growth of seedlings to determine if this treatment has any deleterious effects on seedling growth.

Although protocols for seed germination have been published, little if any quantitative information has been published regarding culture. Skaradek and Murray (2005) reported successful greenhouse culture of seedlings of seabeach amaranth in “5 cm²” containers with “a mixture of half peat and half sand thoroughly moistened to saturation.” The seedlings were later transplanted to the field and grown in a loamy soil overlaid with 5 to 8 cm of sand. The plants grew in the field to maturity producing seeds. Unfortunately, the report of Skaradek and Murray (2005) is extremely sketchy and does not include such information as the volume of the containers in which the seedlings were grown, fertilization of the seedlings, and substrate pH. Therefore, the following research was conducted to develop protocols for containerized production of seedling transplants of seabeach amaranth. To develop such protocols various factors were investigated including the influence of K-GA₃ treatment of seeds on subsequent seedling growth, container volume, substrate pH, and nitrogen (N) source and rate.

Materials and Methods

The study was a 2 x 2 x 2 x 4 x 2 factorial with six replications in a split-plot design. The main plots were two container volumes, two rates of limestone substrate amendment, and two fertilizers with differing sources of N with four rates of each fertilizer. The sub-plot was two treatments for removing seed embryo dormancy, stratification at 4 °C for 90 d or treatment with K-GA₃ at 1000 mg•L⁻¹ for 24 h prior to sowing.

Containers included 635 cm³ Regal 45G plastic pots (Kord Products, Inc., Brampton, Ontario, Canada) or a 2 x 2 cell square (individual cell volume = 139 cm³) from modified Traymaster Rosepot flats, (Mackenzie Nursery Supply, Inc., Perry, Ohio). The substrate was 1 peat : 1 pine bark (v/v) amended with one of two rates of pulverized dolomitic limestone (2.24 or 4.48 kg•m⁻³) (also referred to as low or high rates). The containers were filled with the appropriate substrate and tapped twice on a bench to settle the substrate. The filled containers were then moistened with tap water.

Seeds of an Oak Island, North Carolina, population of seabeach amaranth collected in Sept. 2003 and placed in dry storage at 4 °C in Nov. 2003 were removed from storage on 2 Feb. 2005 and graded. The seeds were graded under a dissecting scope to remove abnormal or damaged seeds and any debris not removed by previous cleaning prior to storage. From the graded seeds, two lots consisting of 650 seeds per lot were removed from the graded seeds. One lot of seeds was mixed with 200 mL of moist sand [10 dry sand : 1 tap water (v/v)] and the seed/sand mixture was placed in a nonvented 3.8-L polyethylene food storage bag. The sand had been sieved dry through a 16-mesh (1.59 mm) screen and the fine separate retained for this study. The polyethylene bag was sealed with a twist tie and the bag was placed in the dark at 4 °C where it remained for 90 d to allow for seed stratification. The other lot of 650 seeds was returned to dry storage at 4 °C for 89 d. On 1 May 2005 this lot of seeds was removed from dry storage at 4 °C and the seeds were placed in a 125-mL Erlenmyer flask containing 50 mL of a solution of K-GA₃ at 1000 mg•L⁻¹. The solution had been prepared by dissolving K-GA₃ in distilled water (pH of distilled water = 6.3). The flask was wrapped with aluminum foil to exclude light and was placed on a rotary shaker (100 revolutions per minute) for 24 h at 21 °C.

Following treatment of both seed lots, seeds were sown (the experiment was initiated) on 2 May 2005 in containers of two differing volumes.

Prior to seeding, the 635 cm³ containers were partitioned with a wooden stake placed horizontally in the center of the pot with one partition being for K-GA₃ treated seeds and the other for stratified seeds. Three seeds of seabeach amaranth were then sown (covered to the minimum diameter of the seeds) in both container volumes, in each respective partition of the 635 cm³ container and two of four designated cells in the 2 x 2 cell squares. The two designated cells in a 2 x 2 square were diagonally opposite and in one cell stratified seeds were sown and in the other cell seeds treated with K-GA₃ were sown.

Following sowing, containers were moved to the Department of Horticultural Science Greenhouses and maintained under natural photoperiod and irradiance with days/nights of 28 ± 2/19 ± 2 °C. A pressure compensated Chapin E0W60 emitter (Chapin Watermatics, Inc., Watertown, N.Y.) was placed on each side of the partition in the large 635 cm³ containers. In the 2 x 2 cell square containers, an emitter was placed in each of the cells in which seeds were sown. Two fertilizers were selected based on the sources of N: Peter's 20N-4.4P-8.2K (20N-10P₂O₅-20K₂O) Professional Water Soluble Peat-lite Special (Scotts-Sierra Hort. Products Co., Marysville, Ohio) with N derived from ammonium nitrate and potassium nitrate (an acidic fertilizer) and Peter's 15N-2.2P-12.3K (15N-5P₂O₅-15K₂O) Excel Cal-Mag Water Soluble Fertilizer (Scotts-Sierra Hort. Products Co.) with N derived from ammonium nitrate, calcium nitrate, potassium nitrate, urea phosphate, and magnesium nitrate (a basic fertilizer). Micronutrients were present in both fertilizers and no source of S was added other than as an anion. Each fertilizer was applied at N application rates (NARs) of 75, 150, 225, or 300 mg•L⁻¹ via a D16 Dosatron (Dosatron, Clearwater, Fla.) using a 1:100 ratio (1 fertilizer : 100 tap water)

to maintain a leaching fraction (volume leached \div volume applied) > 0.2 . To simply discussion of the effects of rate of fertilization, only the N rate will be listed but the reader should be cognizant a 4.5N-1P-1.9K or 6.8N-1P-5.6K ratio, for the acid or basic fertilizer, respectively was maintained at all rates of N.

Containers were fertigated three-times weekly starting 16 May 2005 except for when substrate was too moist. No other irrigation was required. Prior to fertigation, containers were misted daily with tap water to prevent seed washout. Four weeks after sowing, seedlings were thinned to one seedling per cell (2 x 2 cell square) or per partition in the 635 cm³ plastic pots. Substrate solution was collected 21, 30, 36, 43, 50, and 57 d after initiation of the experiment via the pour-through nutrient extraction technique (Wright, 1986). Solution pH and electrical conductivity (EC) of the extracted solutions were measured using a pH and electrical conductivity (EC) meter (Accumet Benchtop pH and Electrical Conductivity Meter, Fisher Scientific Co. LLC., Pittsburgh, Penn.).

During the study the substrate was preventatively treated with a Banrot 40WP (etridiazole and thiophanate-methyl) drench for damping off at a rate of 0.95 g•L⁻¹ twice at three week intervals. In previous studies, fungus gnats (*Orfelia* Costa spp.) were observed on seedlings of seabeach amaranth where larvae would enter young stems and feed on the vascular tissue. Therefore, all containers were drenched with Gnatrol (*Bacillus thuringiensis* subsp. *israelensis*) (Valent BioSciences Corp., Libertyville, Ill.) weekly at a rate of 10.4 mL•L⁻¹. Plants were also treated as needed with Merit 2F {Imidacloprid, 1-[(6-Chloro-3-pyridinyl)methyl]-N-nitro-2-imidazolidinimin} (Bayer Environmental Sci., Res. Triangle Park, N.C.) at a rate of 0.33 mL•L⁻¹ for adult fungus gnats.

On 29 June 2005, 8 weeks after initiation (8 weeks after sowing of seeds), the study was terminated and various data recorded including survivability. Survivability was used as a quantitative index to determine the percentage of seedlings still surviving at the conclusion of the study. Plants were separated into roots, stems, and leaves and leaf area was measured for replications 1 to 4 using a LI-COR LI-3100 Leaf Area Meter (LI-COR Biosciences, Lincoln, Nebr.). Leaf, stem, and root dry weights were also recorded following drying at 60 °C until weights were stable. Top dry weight (leaf + stem dry weight) and root:top ratio [RTR (root dry weight ÷ top dry weight)] were calculated from these data.

After recording dry weights, stems and leaves were combined (tops) and ground using a Foss Tecator Cyclotec™ 1093 sample mill (Analytical Instruments, LLC, Golden Valley, Minn.) to pass a ≤ 0.5 mm (0.02 in) sieve. Tops were analyzed for N, P, K, Ca, Mg, S, Na, Mn, Zn, Cu, Fe, and B by the North Carolina Department of Agriculture and Consumer Services, Raleigh. Nitrogen concentrations were determined by oxygen combustion with an elemental analyzer (NA 1500, CE Elantech Instruments, Milan, Italy). All other mineral nutrient concentrations were determined by EPA method 200.7 with an ICP spectrophotometer (Optima 3300 DV ICP Emission Spectrometer; Perkin Elmer Corp., Wellesley, Mass.) following open-vessel nitric acid (HNO₃) digestion in a microwave digestion system (CEM Corp., Matthews, N.C.). Mineral nutrient contents were calculated based on percentage concentration of a nutrient divided by 100 and multiplied by top dry weight.

All data were subjected to analysis of variance procedures (ANOVA) and regression analysis, where appropriate (SAS Inst. Inc., Cary, N.C.). All three-way and four-way interactions were nonsignificant and any significant two-way interactions are presented in tables. Treatments means were separated by the F test, $P= 0.05$, where appropriate. When significant (P

≤ 0.05), simple linear and polynomial curves were fitted to the N rate data. The maximum of the polynomial curve was calculated as the zero point in a first-order derivative of the independent variable.

Results and Discussion

pH. As anticipated, substrate pH was affected by NAR and rate of limestone, and surprisingly container volume, whereas there were no significant interactions (Tables 1 and 2). Substrate pH decreased linearly with increasing NAR 21, 43, and 57 d after initiation (Table 1). The high rate of limestone produced significantly higher substrate pH at all sample times (Table 2). In addition, pH in the larger containers was significantly greater than the smaller volume containers at the high rate of limestone 30, 36, 50, and 57 d after initiation (data not presented). There was no difference in substrate pH between the container volumes at the low rate of limestone. Most substrate pH values observed herein were within substrate pH recommendations for soilless media (5.4 to 6.0) (Dole and Wilkins, 2005).

Electrical conductivity. EC was affected by NAR, fertilizer, and the NAR x fertilizer interaction (Table 3). EC was affected similarly at all sample times, therefore, EC values were averaged over all sample times. Both fertilizers increased EC linearly with increasing NAR. From N at 150 to 300 mg•L⁻¹ EC was similar for both fertilizers, however, with N at 75 mg•L⁻¹ the basic fertilizer had higher EC. Maximum top growth occurred with N at 300 mg•L⁻¹ (Table 4), therefore EC values of 1.15 to 1.18 dS•m⁻¹ should be adequate for maximum growth. Recommended EC values for cock's comb (*Celosia L. sp.*) range from 1.0 to 2.6 dS•m⁻¹ (Whipker et al., 2003). Likewise, optimal EC values for 'Gloria Scarlet' silver cock's comb (*Celosia argentea L. 'Gloria Scarlet'*) were 1.1 to 1.5 dS•m⁻¹ (Kang and van Iersel, 2002).

Growth. Stem and leaf dry weights responded in a similar fashion to all treatments; therefore only top dry weight data are presented. Top dry weight of seabeach amaranth was affected by all treatments, however, there were no significant interactions so only the main effects are presented. Top dry weight of seabeach amaranth increased linearly with increasing NAR, and maximum top dry weight occurred with N at $300 \text{ mg}\cdot\text{L}^{-1}$ (Table 4). At $300 \text{ mg}\cdot\text{L}^{-1}$, top dry weight increased 106% compared to top dry weight with N at $75 \text{ mg}\cdot\text{L}^{-1}$. While N at $300 \text{ mg}\cdot\text{L}^{-1}$ is high, plant production N recommendations for annuals can be as high as $255 \text{ mg}\cdot\text{L}^{-1}$ when applied with every irrigation (Nelson, 2003). Similar to top dry weight, leaf area increased linearly with increasing NAR resulting in maximum leaf area at $300 \text{ mg}\cdot\text{L}^{-1}$ which was 131% greater than leaf area at $75 \text{ mg}\cdot\text{L}^{-1}$. In contrast, root dry weight was unaffected by NAR.

RTR decreased linearly with increasing NAR for both fertilizers, acidic and basic (Table 4). This was not surprising as increasing NARs typically reduce RTR (Friend et al., 1994). As plants transition from N deficient to adequate N, most plants typically allocate a larger fraction of carbohydrates to top growth (Friend et al., 1994). When grown with the acidic fertilizer, RTR decreased 59% from 75 to $300 \text{ mg}\cdot\text{L}^{-1}$, whereas RTR of seabeach amaranth grown with the basic fertilizer decreased 29% from 75 to $300 \text{ mg}\cdot\text{L}^{-1}$.

Top and root dry weight, leaf area, and survivability were affected significantly by fertilizer (excluding leaf area), container volume (excluding survivability), seed treatment, and rate of limestone, and all interactions were nonsignificant (Table 5). The basic fertilizer, larger container volume (635 cm^3), K-GA₃ treated seeds, and the $4.5 \text{ kg}\cdot\text{m}^3$ limestone amendment resulted in increased top and root dry weight compared to the acidic fertilizer, smaller container volume (139 cm^3), stratified seeds, and $2.24 \text{ kg}\cdot\text{m}^3$ limestone amendment, respectively. Top and root dry weights of seabeach amaranth were 17% and 19% larger when grown with the basic

fertilizer compared to the acidic. However, the basic fertilizer and acidic fertilizer-grown plants had similar leaf areas. Contrary to these results, survivability was higher (5%) with the acidic fertilizer than the basic (Table 5).

Seabeach amaranth grown in the large containers had top and root dry weights and leaf area 61%, 33%, and 57% larger, respectively, compared to plants grown in the smaller container volume (Table 5). Likewise, plants of ‘Sweet Charlie’ strawberry (*Fragaria x ananassa* Duch. ‘Sweet Charlie’) were larger when grown in 300 cm³ containers compared to 150 cm³ containers (Bish et al., 2002).

Top and root dry weights, and leaf area of plants grown from K-GA₃ treated seeds were 24%, 25%, and 24% larger, respectively, than plants grown from stratified seeds. Plants from K-GA₃ treated seeds also had 7% higher survival than plants from stratified seeds. Why the K-GA₃ treated seeds produced larger plants is not completely understood, however, one hypothesis is the K-GA₃ treatment affected the metabolic rate of the embryos. A second and more likely explanation is the more rapid germination of the K-GA₃ treated seeds resulted in a time advantage over seedlings resulting from stratified seeds. Research by Norden et al. (2007), indicated the higher the K-GA₃ treatment, the faster seeds of seabeach amaranth will germinate.

Plants of seabeach amaranth grown in the high rate of limestone amendment had top and root dry weights both 33% higher than plants grown in the low rate of limestone amendment. This is not surprising as the pH of dune soil from North Carolina beaches ranges from 7.4 to 7.9 (Oosting and Billings, 1942). Leaf area was also 20% higher with the high rate of limestone, whereas survivability was 7% higher at the higher rate.

Mineral nutrient concentrations and contents. Top N concentration increased linearly with increasing NAR for acidic and basic fertilizers with N concentrations of 58.4 and 50.4

$\text{mg}\cdot\text{g}^{-1}$, respectively, at maximum top dry weight (Tables 4 and 6). Mills and Jones (1996) reported a range of foliar N concentrations of 37.1 to 41.3 $\text{mg}\cdot\text{g}^{-1}$ for silver cock's comb (*Celosia argentea* L.), a member of the Amaranthaceae. However, it is not possible to determine if these N concentrations represented values at maximum growth. Only top P concentrations of seabeach amaranth grown with the acidic fertilizer increased linearly with increasing NAR, whereas foliar P concentrations of amaranth grown with the basic fertilizer and foliar K concentrations of amaranth grown with either fertilizer were unaffected by NAR. This is contrary to recent reports with herbaceous and woody perennial plants where foliar P and K concentrations increased quadratically or linearly with increasing NAR (Conden et al, 2003; Harvey et al, 2004).

Top Ca concentration of seabeach amaranth grown with the acidic fertilizer decreased linearly with increasing NAR, while top Ca concentration of plants grown with the basic fertilizer increased linearly with increasing NAR (Table 6). Cabrera and Devereaux (1998) reported a decrease in foliar Ca concentration where $\text{NH}_4\text{-N}$ was a significant fraction of the N supply (Cabrera and Devereaux, 1998) as NH_4 competes with Ca for uptake. The major component of the basic fertilizer was $\text{NO}_3\text{-N}$, and it would not have competed with Ca uptake. In addition, the basic fertilizer also contained Ca. Even though the basic fertilizer also contained Mg, top Mg concentrations were unaffected by NAR or fertilizer (Table 6).

Sulfur is a major component of enzymes and proteins associated with growth, therefore S concentrations might be expected to increase with NAR (Marschner, 1995; Mengel and Kirkby, 1987). However, top S concentrations of seabeach amaranth decreased linearly with increasing NAR for both fertilizers (Table 6). These results are similar to those of Cabrera and Devereaux (1998) who reported increasing NAR decreased foliar S concentrations.

Both fertilizers resulted in a linear decrease in top Na concentration of seabeach amaranth (Table 6). The Na levels reported herein are considerably higher than levels reported for silver cock's comb (Mills and Jones, 1996). The higher Na levels are most likely attributed to seabeach amaranth being a halophyte (Skaradek and Murray, 2005), but no research appears to have been reported regarding the salt-exclusion mechanism in seabeach amaranth. There are several reported salt-exclusion mechanisms occurring in grasses such as members of subfamily Chloridoideae which possess salt-excreting 'microhairs' (Chapman, 1996), as well as vacuolar Na^+/H^+ antiporters that sequester Na into vacuoles to prevent toxic salt accumulation in the cytoplasm (Saqib et al., 2005). Most halophytes tightly regulate Na uptake at salinity levels below or equivalent to seawater, thus, a decreasing top Na concentration could be due to dilution caused by increasing growth (Gorham, 2007). Foliar Mn, Zn, and B concentrations of seabeach amaranth were affected by NAR only when grown with the acidic fertilizer (Table 6). Both top Mn and Zn concentrations decreased linearly with increasing NAR, while foliar B concentrations increased linearly with increasing NAR. Foliar Fe (mean = $122.8 \text{ mg}\cdot\text{g}^{-1} \pm 5.5$) and Cu (mean = $9.7 \text{ mg}\cdot\text{g}^{-1} \pm 0.9$) concentrations were unaffected by NAR or fertilizer.

Rate of limestone significantly affected top mineral nutrient concentrations (Table 7). At the low lime rate, top N, P, and Mn concentration of amaranth were significantly higher than plants grown at the high lime treatment. In contrast, top Ca, Mg, and Na concentration of amaranth grown with the high rate of limestone were significantly higher than amaranth grown with the low limestone rate. In contrast, top K (Table 7), and top S, Fe, Cu, Zn, and B concentrations were unaffected by rate of limestone (data not presented).

Mineral nutrient contents of tops were affected by all main effects, but all interactions were not significant. Top nutrient content of N, P, K, Ca, Mg, S, Na, Fe, Mn, Zn, and B

increased linearly with increasing NAR (data not presented), whereas Cu was the only nutrient unaffected by NAR (data not presented). As previously mentioned, top concentrations of Ca, Mn, and Zn for only the acidic fertilizer, as well as S for both fertilizers decreased with increasing NAR. This is contrary to an increase of all top nutrient contents. Thus, the decreasing top concentrations were probably due to dilution as the plants increased in dry weight.

Top nutrient content of Ca, Mg, and Mn were affected by rate of limestone and Ca, Mg, and Mn were affected by fertilizer (Table 8). Top Ca and Mg content were significantly higher at the high lime rate which can be attributed to a doubling of the input of dolomitic limestone at the high lime treatment. However, top Mn content of amaranth when grown with the high rate of limestone decreased compared to plants grown with the low lime rate probably due to decreasing Mn availability with increasing pH. The basic fertilizer yielded significantly higher top Ca and Mg content than the acidic fertilizer which can be attributed to the nutrient sources of the basic fertilizer. However, top N, P, K, S, Zn, Cu, Fe, and B contents were unaffected by either rate of limestone or fertilizer (data not presented).

Summary. Seabeach amaranth can be produced successfully in containerized production with maximum top growth occurring with N at $300 \text{ mg} \cdot \text{L}^{-1}$ provided by an acid or basic fertilizer with a 4.5N-1P-1.9K or 6.8N-1P-5.6K ratio, respectively. Limiting fertilizer inputs to the lowest nutrient concentrations consistent with adequate growth is an important consideration for growers. It should be implemented, whenever possible, because it is a cost-saving technique that can significantly reduce the levels of nutrient runoff from nurseries (Tyler, 1995). Growth data indicated larger plants can be grown with a basic fertilizer, a larger container, and a high rate of

limestone (Table 5). Further, results herein indicate seed treatment with K-GA₃ does not appear to cause any undesirable morphological effects and results in larger plants.

Finally, a question warranting future research is what size plants are desirable for coastal restoration plantings? Large plants can be produced in a short amount of time. However, are large plants more advantageous for transplanting into the beach environment? It may be plants produced in smaller containers and at a lower N rate might be more desirable for beach plantings since they would be less susceptible to physical injury. This situation mirrors results reported by Liptay et al. (1992) where ‘TH-318’ tomato [*Solanum lycopersicum* L. var. *lycopersicum* (syns. *Lycopersicon lycopersicum* Karst., *Lycopersicon esculentum* Miller) ‘TH-318’] transplants produced using the float system at a high N rate (350 mg•L⁻¹) exhibited lower survivability in comparison to transplants produced at lower rates of N (100 to 200 mg•L⁻¹).

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Table 1. Effect of nitrogen application rate (NAR) on substrate pH of containerized seabeach amaranth.^z

NAR (mg•L ⁻¹)	Days after initiation (DAI)					
	21	30	36	43	50	57
75	5.84 ± 0.57	6.07 ± 0.54	5.93 ± 0.47	6.08 ± 0.45	5.92 ± 0.42	6.04 ± 0.42
150	5.79 ± 0.56	6.00 ± 0.56	5.96 ± 0.48	6.03 ± 0.46	5.78 ± 0.43	5.96 ± 0.45
225	5.62 ± 0.55	5.84 ± 0.60	5.91 ± 0.47	5.88 ± 0.60	5.79 ± 0.41	5.79 ± 0.58
300	5.52 ± 0.55	5.81 ± 0.56	5.78 ± 0.46	5.77 ± 0.49	5.71 ± 0.38	5.81 ± 0.40
Significance						
Linear ^y	*	NS	NS	*	NS	*
Quadratic	NS	NS	NS	NS	NS	NS

^zData are means of 192 observations ± 1 SE.

^yRegression equations are: 21 DAI, $y = 6.0 - 0.002x$, $R^2 = 0.98$; 43 DAI, $y = 6.2 - 0.001x$, $R^2 = 0.98$; 57 DAI, $y = 6.2 - 0.00x$, $R^2 = 0.92$.

NS, * Nonsignificant or significant at $P \leq 0.05$, respectively.

Table 2. Effect of rate of limestone substrate amendment and container volume on substrate pH of containerized seabeach amaranth.^z

	Days after initiation					
	21		30		36	
	<u>Container volume (cm³)</u>		<u>Container volume (cm³)</u>		<u>Container volume (cm³)</u>	
Rate of limestone (kg•m ⁻³)	139	630	139	630	139	630
2.24	5.23 ± 0.06	5.25 ± 0.07	5.46 ± 0.04	5.39 ± 0.07	5.54 ± 0.05	5.49 ± 0.06
4.48	6.08 ± 0.07	6.22 ± 0.07	6.35 ± 0.07	6.51 ± 0.06	6.19 ± 0.06	6.36 ± 0.05
Significance	*	*	*	*	*	*

Table 2 (continued).

Days after initiation					
43		50		57	
<u>Container volume (cm³)</u>		<u>Container volume (cm³)</u>		<u>Container volume (cm³)</u>	
139	630	139	630	139	630
5.55 ± 0.05	5.47 ± 0.07	5.48 ± 0.06	5.55 ± 0.06	5.58 ± 0.06	5.52 ± 0.07
6.29 ± 0.07	6.44 ± 0.03	5.98 ± 0.06	6.19 ± 0.06	6.15 ± 0.07	6.35 ± 0.06
*	*	*	*	*	*

^zData are means of 96 observations ± 1 SE.

* Significant at $P \leq 0.05$.

Table 3. Effect of nitrogen application rate (NAR) on substrate EC of containerized seabeach amaranth grown with an acidic or basic fertilizer.^z

NAR (mg•L ⁻¹)	Acidic fert. ^y	Basic fert. ^x
	-----dS•m ⁻¹ -----	
75	0.397 ± 0.025	0.624 ± 0.027
150	0.608 ± 0.042	0.677 ± 0.038
225	0.912 ± 0.023	0.966 ± 0.020
300	1.150 ± 0.050	1.180 ± 0.055
Significance		
Linear ^w	***	***
Quadratic	NS	NS

^zData are means of 48 observations ± 1 SE.

^yAcid fert. = 20N-4.4P-8.2K with N derived from ammonium nitrate and potassium nitrate.

^xBasic fert. = 15N-2.2P-12.3K with N derived from ammonium nitrate, calcium nitrate, potassium nitrate, urea phosphate, and magnesium nitrate.

^wRegression equations are: acidic fert., $y = 0.13 + 0.003x$, $R^2 = 0.99$; basic fert., $y = 0.37 + 0.003x$, $R^2 = 0.97$.

NS, *** Nonsignificant or significant at $P \leq 0.001$, respectively.

Table 4. Effect of nitrogen application rate (NAR) on top and root dry weight, root:top ratio (RTR), and leaf area of containerized seabeach amaranth.^z

NAR (mg·L ⁻¹)	Top dry weight (g)	Root dry weight (g)	RTR ^y		Leaf area (cm ²)
			Acidic fert. ^x	Basic fert. ^w	
75	0.93 ± 0.60	0.16 ± 0.09	0.22 ± 0.09	0.14 ± 0.05	92.2 ± 50.5
150	1.32 ± 0.66	0.19 ± 0.11	0.15 ± 0.08	0.15 ± 0.05	130.3 ± 58.9
225	1.57 ± 1.12	0.17 ± 0.13	0.12 ± 0.12	0.09 ± 0.05	162.5 ± 111.9
300	1.92 ± 1.13	0.19 ± 0.11	0.09 ± 0.04	0.10 ± 0.03	213.2 ± 107.5
Significance ^v					
Linear	***	NS	***	***	***
Quadratic	***	NS	NS	NS	***

Table 4 (continued).

^zData for top dry weight, root dry weight, and leaf area are means of 96 observations \pm 1 SE. Data for RTR are means of 48 observations \pm 1 SE.

^yRTR = root dry weight \div top dry weight.

^xAcid fert. = 20N-4.4P-8.2K with N derived from ammonium nitrate and potassium nitrate.

^wBasic fert. = 15N-2.2P-12.3K with N derived from ammonium nitrate, calcium nitrate, potassium nitrate, urea phosphate, and magnesium nitrate.

^vRegression equations are: top dry weight, $y = 0.63 + 0.004x$, $R^2 = 0.99$; RTR (acidic fert.), $y = 0.25 - 0.0006x$, $R^2 = 0.97$; RTR (basic fert.), $y = 0.17 - 0.0002x$, $R^2 = 0.79$; leaf area, $y = 50.7 + 0.527x$, $R^2 = 0.99$.

NS, *, **, *** Nonsignificant or significant at $P \leq 0.05$, 0.01, or 0.001, respectively.

Table 5. Effect of fertilizer, container volume, seed treatment, and rate of substrate limestone amendment on top and root dry weight, leaf area, and survivability of containerized seabeach amaranth.^z

	Top dry weight (g)	Root dry weight (g)	Leaf area (cm ²)	Survivability (%)
<u>Fertilizer^y</u>				
Acidic	1.32 ± 0.95	0.16 ± 0.11	147.6 ± 102.0	96 ± 20
Basic	1.55 ± 0.99	0.19 ± 0.12	151.4 ± 92.2	91 ± 28
Significance	*	*	NS	*
<u>Volume (cm³)</u>				
139	1.10 ± 0.61	0.15 ± 0.09	116.6 ± 61.7	92 ± 28
630	1.77 ± 1.15	0.20 ± 0.13	182.5 ± 113.7	95 ± 21
Significance	*	*	*	NS
<u>Seed treatment</u>				
K-GA ₃ ^x	1.59 ± 1.02	0.20 ± 0.11	165.3 ± 104.8	97 ± 16
Stratified ^w	1.28 ± 0.91	0.16 ± 0.11	133.7 ± 86.2	90 ± 31
Significance	*	*	*	*

Table 5 (continued).

<u>Rate of limestone ($\text{kg}\cdot\text{m}^{-3}$)</u>				
2.24	1.23 ± 0.99	0.15 ± 0.11	136.0 ± 105.1	90 ± 31
4.48	1.63 ± 0.92	0.20 ± 0.11	163.1 ± 86.6	97 ± 16
Significance	*	*	*	*

^zData are means of 192 observations \pm 1 SE.

^yAcid fert. = 20N-4.4P-8.2K with N derived from ammonium nitrate and potassium nitrate; basic fert. = 15N-2.2P-12.3K with N derived from ammonium nitrate, calcium nitrate, potassium nitrate, urea phosphate, and magnesium nitrate.

^xK-GA₃ = seeds treated with K-GA₃ at $1000 \text{ mg}\cdot\text{L}^{-1}$ for 24 h prior to sowing.

^wStratified = seeds stratified (moist-prechilled) for 90 d prior to sowing.

NS, * Nonsignificant or significant at $P \leq 0.05$, respectively.

Table 6. Effect of nitrogen application rate (NAR) and type of fertilizer on top mineral nutrient concentrations of containerized seabeach amaranth.^z

NAR (mg•L ⁻¹)	N		P		K	
	Acidic fert. ^y	Basic fert. ^x	Acidic fert.	Basic fert.	Acidic fert.	Basic fert.
	----- mg•g ⁻¹ -----					
75	36.1 ± 3.3	44.0 ± 4.7	6.9 ± 0.6	7.0 ± 1.0	32.2 ± 2.7	30.5 ± 2.7
150	48.1 ± 3.9	41.2 ± 6.7	9.3 ± 0.7	6.1 ± 1.9	30.4 ± 2.1	31.0 ± 2.9
225	51.8 ± 6.4	50.9 ± 5.0	10.7 ± 1.7	8.0 ± 1.3	32.8 ± 5.2	33.2 ± 2.1
300	58.4 ± 4.6	50.4 ± 5.2	11.7 ± 0.6	7.3 ± 0.5	33.3 ± 2.8	31.0 ± 1.9
Significance						
Linear ^w	***	**	***	NS	NS	NS
Quadratic	NS	NS	NS	NS	NS	NS

Table 6 (continued).

^zData are means of 48 observations \pm 1 SE.

^yAcid fert. = 20N-4.4P-8.2K with N derived from ammonium nitrate and potassium nitrate.

^xBasic fert. = 15N-2.2P-12.3K with N derived from ammonium nitrate, calcium nitrate, potassium nitrate, urea phosphate, and magnesium nitrate.

^wRegression equations are: N (acidic), $y = 30.9 + 0.094x$, $R^2 = 0.97$; N (basic), $y = 39.4 + 0.039x$, $R^2 = 0.78$; P (acidic), $y = 5.7 + 0.021x$, $R^2 = 0.98$; Ca (acidic), $y = 15.1 - 0.020x$, $R^2 = 0.94$; Ca (basic), $y = 11.7 + 0.007x$, $R^2 = 0.90$; S (acidic), $y = 6.0 - 0.008x$, $R^2 = 0.96$; S (basic), $y = 4.8 - 0.005x$, $R^2 = 0.99$; Na (acidic), $y = 8.3 - 0.008x$, $R^2 = 0.90$; Na (basic), $y = 8.3 - 0.008x$, $R^2 = 0.97$; Mn (acidic), $y = 297.4 - 0.405x$, $R^2 = 0.98$; Zn (acidic), $y = 153.4 - 0.199x$, $R^2 = 0.89$; B (acidic), $y = 31.4 + 0.032x$, $R^2 = 0.90$.

NS, *, **, *** Nonsignificant or significant at $P \leq 0.05$, 0.01, or 0.001, respectively.

Table 7. Effect of rate of limestone on top nutrient concentration of containerized seabeach amaranth.^z

Rate of limestone (kg•m ⁻³)	N	P	K	Ca	Mg	Mn	Na
	-----mg•g ⁻¹ -----						
2.24	49.8 ± 8.2	8.7 ± 2.2	31.5 ± 3.6	11.5 ± 2.0	8.8 ± 1.0	0.3 ± 0.05	6.5 ± 1.0
4.48	45.4 ± 7.6	8.0 ± 2.1	32.1 ± 2.4	12.8 ± 2.1	9.6 ± 0.9	0.1 ± 0.03	6.9 ± 1.0
Significance	*	*	NS	*	*	*	*

^zData are means of 192 observations ± 1 SE.

NS, * Nonsignificant or significant at $P \leq 0.05$, respectively.

Table 8. Effects of rate of limestone and type of fertilizer on top nutrient content of containerized seabeach amaranth.^z

	Ca	Mg	Mn
Rate of limestone (kg•m ⁻³)	-----mg-----		-----µg-----
2.24	16.4 ± 8.4	12.9 ± 7.0	393 ± 200
4.48	21.3 ± 8.4	16.0 ± 5.8	239 ± 92
Significance	*	*	*
Fertilizer			
Acidic ^y	15.2 ± 7.4	12.7 ± 6.9	305 ± 188
Basic	22.5 ± 8.4	16.2 ± 5.8	327 ± 159
Significance	*	*	NS

^zData are means of 192 observations ± 1 SE.

^yAcidic fert. = 20N-4.4P-8.2K with N derived from ammonium nitrate and potassium nitrate; basic fert. = 15N-2.2P-12.3K with N derived from ammonium nitrate, calcium nitrate, potassium nitrate, urea phosphate, and magnesium nitrate.

NS, * Nonsignificant or significant at $P \leq 0.05$, respectively.

Chapter 3

Nitrogen Nutrition of Southern Seaoats (*Uniola paniculata*) Grown in the Float System

(In the format appropriate for submission to the
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Nitrogen Nutrition of Southern Seaoads (*Uniola paniculata*) Grown in the Float System¹

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Abstract

Seeds of southern seaoats (*Uniola paniculata* L.) were removed from storage in July 2004, surface disinfested with 2.6% sodium hypochlorite (NaOCl) for 15 min, and sown in styrofoam tobacco (*Nicotiana tabacum* L.) float trays (flats) filled with a vermiculite-based hydroponic substrate. Trays were floated in plastic tubs (one tray per tub) containing a complete nutrient solution with nitrogen (N) at 10, 60, 120, 180, or 240 mg•L⁻¹ (ppm) from a 2N-3.5P-1K ratio (8N-32P₂O₅-5K₂O) liquid slow-release fertilizer. After 10 weeks the study was terminated and data recorded. Total plant, top, leaf, stem, and root dry weights increased quadratically with increasing nitrogen application rate (NAR) with maximum dry weights calculated to occur with N at 140 to 150 mg•L⁻¹, respectively. Other growth indexes of leaf area, root length, root area, plant height, crown growth index, tiller number, and leaf number also increased quadratically with increasing NAR similar to dry weight data. Leaf area, root length, and root area were maximized with N at 157, 140, and 140 mg•L⁻¹, respectively. Root to top ratio and specific leaf area were both unaffected by NAR. Leaf mineral nutrient concentrations of N and phosphorous responded quadratically with increasing NAR whereas, foliar mineral nutrient concentrations of potassium, calcium, sulfur, sodium (Na), manganese, zinc, and copper responded linearly to increasing NARs. With the exception of Na and iron, foliar nutrient content for all analyzed nutrients increased quadratically with increasing NAR. Calculated leaf N concentration at maximum top dry weight was 31 mg•g⁻¹. Southern seaoats can be grown successfully using the float system with optimum N rates of 140 to 150 mg•L⁻¹ provided by a fertilizer having a 2N-3.5P-1K ratio.

Index words: beach restoration, dune species, Poaceae, mineral nutrition, fertility.

Significance to the Nursery Industry

Transplants of southern seaoats (*Uniola paniculata*), a major coastal dune stabilizing species of the southern Atlantic and Gulf Coasts of the United States, are currently in great demand. However, research on culture has been limited particularly with respect to mineral nutrition. Research herein demonstrated southern seaoats can be produced successfully using the float system, and nitrogen (N) in the nutrient solution at 140 to 150 mg•L⁻¹ (ppm) will maximize vegetative growth. Use of the float system for production of transplants could provide opportunities for former and current tobacco (*Nicotiana tabacum*) farmers to produce alternative crops using the float system. Production of the species may also provide an additional means to supplement farm incomes.

Introduction

Southern seaoats is a perennial dune grass that in most of its natural range (southern Virginia to the Yucatan Peninsula) is the dominant, coastal sand-binding plant species (30). The species is generally subtropical and its native range is determined by climate as it is intolerant of extremely hot summers or cold winters (30). In southern Virginia and northern North Carolina, southern seaoats is at the northern limit of its range and the plants usually die back to ground level and resprout from rhizomes in the spring. Seed germination occurs in late spring and little growth takes place until adequate sand surrounds the culms (stems), usually by the end of the second year (41). Seaoats has the ability to stabilize dunes upon establishment by utilizing culms and extensive root systems to trap sand (30, 33). Thus, it has been planted extensively to build and stabilize coastal sand dunes (23, 41).

In recent years, transplants of southern seaoats have been in demand to restore beaches and stabilize sand dunes along the southern Atlantic and Gulf Coasts of the United States that have been damaged by tropical storms and erosion. Demand has in turn created a need for

information regarding propagation and culture. Considerable research has been reported on propagation, specifically seed germination (3, 4, 34, 35, 40). However, with few exceptions research on culture has been limited (1, 18, 35), particularly with respect to mineral nutrition of container-grown plants. Cultural information, coupled with previous work on seed germination, would result in protocols for production of transplants that may prove profitable to growers such as former and current tobacco farmers who are seeking alternative crops that can be grown in the float system.

Tobacco and vegetable transplants can be grown successfully using the float system (21, 24, 31, 32, 36). This system involves constructing shallow wooden frames on the floor of a greenhouse. The frames are lined with polyethylene sheeting and filled with nutrient solution. Seeds are sown in peat- or vermiculite-based soilless substrates in styrofoam trays (flats), floating on the nutrient solution. Irrigation is by capillary movement of nutrient solution into the substrate. Potential advantages of the float system over conventional overhead irrigation include lower production costs, more efficient use of water and mineral nutrients, reduced disease pressure (dry plant foliage), and elimination of nutrient leaching to groundwater below the greenhouse (31). Frantz and Welbaum (11) also noted if other crops could be produced successfully using the float system, float systems could potentially produce high-value horticultural crops to supplement farm incomes. Soundy et al. (36) reported that although production of vegetable transplants using the float system has several advantages including improved health of seedlings, production of lettuce (*Lactuca sativa* L.) transplants using the float system resulted in poor root systems.

Some research has been conducted on growing alternative crops using the float system (11, 31, 36, 39). Nash [as reported by Latham (23)], an Agricultural Extension Agent in New Hanover County, NC, has produced transplants of sea oats successfully utilizing the float system.

Initially, he attempted to grow southern seaoats using conventional container production but he encountered many problems particularly with foliar fungal diseases due to irrigating over the tops of plants (David L. Nash, personal communication). Using the float system reduced dramatically foliar infestations. However, despite successful culture of southern seaoats using the float system, little information has been published on this means of culture to produce transplants of seaoats. Therefore, the following research was conducted to study the influence of N nutrition on vegetative growth of southern seaoats in a float system.

Materials and Methods

On July 12, 2004, seeds of southern seaoats, collected in Fall 2003 from Oak Island, NC, were removed from storage at 4C (39F) and surface disinfested with a solution of 2.6% sodium hypochlorite (NaOCl) for 15 min. Following treatment, the seeds were rinsed with tap water and sown in modified Standard Carolina Greenhouse 288-cell styrofoam float trays (Carolina Greenhouse Co., Kinston, NC) with each cell having a volume of 14 cm³ (0.85 in³). The trays were modified by cutting them with a serrated knife which reduced a tray from 24 x 12 cells [67 x 34 x 6.5 cm (26 x 13 x 3 in)] to 15 x 12 cells [42 x 34 x 6.5 cm (17 x 13 x 3 in)]. Each modified tray was filled with Carolina's Choice Tobacco Mix (Carolina Soil Co., Kinston, NC), a vermiculite-based hydroponic substrate and floated in gray plastic tubs [50 x 36 x 12 cm (20 x 14 x 5 in)] (Consolidated Plastics Company, Inc., Twinsburg, OH), each tub containing 10 L (2.6 gal) of tap water. Prior to floating the trays, the tubs were rinsed three times with distilled water to which was added Liqui-Nox, a phosphate-free analytical washing agent (Alconox, Inc., White Plains, NY). Cells of trays that were dry after 3 days of floating in the tap water were altered by removing any obstructions preventing water movement or removing air pockets in the growing substrate. The trays were removed from the tubs and three seeds were sown per cell in a 6 x 6

cell square in the center of a modified tray [center was determined from the top (shorter dimension side) of tray counting five cells right and four cells down] and refloated in a gray plastic tub containing 10 L (2.6 gal) of a complete nutrient solution with varying rates of N. The tubs were maintained under natural photoperiod and irradiance in a greenhouse (Dept. of Hort. Sci., NC State Univ., Raleigh) maintained at days/nights of $27 \pm 2\text{C} / 21\text{C} \pm 1\text{C}$ ($81/70 \pm 4\text{F}$). Temperatures of the nutrient solutions averaged 26C (79F).

Treatments included five rates of N at 10, 60, 120, 180, or $240 \text{ mg}\cdot\text{L}^{-1}$ from a 2N-3.5P-1K ratio ($8\text{N}-32\text{P}_2\text{O}_5-5\text{K}_2\text{O}$) liquid slow-release fertilizer (Growth Products, Ltd., White Plains, NY) which also contained calcium (Ca), magnesium (Mg), and micronutrients. To simplify discussion of the effects of rate of fertilization, only the N rate will be listed but the reader should be cognizant a 2N-3.5P-1K ratio was maintained at all rates of N. Nutrient sources of N, P, and K in the fertilizer were urea, methylene urea, potassium carbonate, diammonium phosphate, and phosphoric acid. The nutrient solution in each tub was replaced weekly, and solutions were prepared with tap water. Tap water averages for $\text{NO}_3\text{-N}$, $\text{NH}_4\text{-N}$, phosphorous (P), potassium (K), Ca, Mg, and alkalinity were 0.10, 0.96, 0.5, 7.0, 10.0, 4.0, and $20.0 \text{ mg}\cdot\text{L}^{-1}$, respectively, with a pH of 7.4.

Seedling emergence first occurred on July 16, 2004 and was essentially complete by July 23 when seedlings were thinned to one per cell. On July 28, the outer rows of cells in each float tray were reseeded because of limited emergence. These seeds, also from the same seed lot sown initially on July 12, were surface disinfested with a solution of NaOCl as described previously.

The experiment was a randomized complete block design with six replications and five treatments (N rates). A tub was considered a single experimental unit. Tubs were oriented on a greenhouse bench parallel to the cooling pads to direct uniform airflow across all treatments. Electrical conductivity (EC) and pH of the nutrient solutions were recorded using a HI 9811

Hanna Meter (Hanna Instruments, Inc., Woonsocket, RI). This was done every 3 days before (sample time 1) and after (sample time 2) weekly replacement of the nutrient solutions. EC at sample time 1 averaged 0.401, 0.811, 1.305, 1.803, and 2.300 $\text{dS}\cdot\text{m}^{-1}$ for NARs of 10, 60, 120, 160, and 240 $\text{mg}\cdot\text{L}^{-1}$, respectively. Likewise, pH at sample time 1 for NARs of 10, 60, 120, 160, and 240 $\text{mg}\cdot\text{L}^{-1}$ averaged 6.9, 6.7, 6.8, 6.9, and 6.9, respectively. EC averages for sample time 2 (before refilling) were 0.409, 0.859, 1.484, 2.076, and 2.692 $\text{dS}\cdot\text{m}^{-1}$, for N at 10, 60, 120, 160, and 240 $\text{mg}\cdot\text{L}^{-1}$, respectively, whereas averages for pH were 5.6, 5.6, 6.0, 6.3, and 6.5 for NARs of 10, 60, 120, 160, and 240 $\text{mg}\cdot\text{L}^{-1}$, respectively.

Algal growth and insect and potential disease problems were concerns of the investigators and were minimized during the study. One particular concern was the potential for algal growth in the nutrient solutions. Control was achieved by several means. First, the Standard Carolina 288-cell styrofoam float trays were modified (reduced in size) to occupy the maximum space (area) in a plastic tub to minimize light penetration to the nutrient solutions. The nutrient solutions were also replaced weekly to provide additional algal control and prior to the tubs being refilled, they were thoroughly rinsed to remove any visible signs of algal growth. Cutrine Plus (Applied Biochemists, Germantown, WI), a chelated copper algaecide, was also added to each nutrient solution at 0.48 ml/10 L (0.0076 fl oz/gal) [from a stock solution of 25 ml Cutrine Plus/250 ml tap water (0.85 fl oz Cutrine Plus/8.45 fl oz tap water)] starting Aug. 5, 2004 and was increased to 0.65 ml/10 L (0.0083 fl oz/gal) of nutrient solution on Aug. 15. Fungus gnats (*Orfelia* Costa spp.) were observed on the seedlings and growing medium which required treatment on two occasions. On Aug. 6, the trays were removed from the plastic tubs and sprayed with Merit 2F [Imidacloprid, 1-[(6-Chloro-3-pyridinyl)methyl]-N-nitro-2-imidazolidinimin] (Bayer Environ. Sci., Res. Triangle Park, NC) at a rate of 0.33 $\text{mg}\cdot\text{L}^{-1}$. To further control fungus gnats, the float trays were treated on Sept. 10 with Talstar Flowable

Bifenthrin (FMC Corp., Philadelphia, PA), at a rate of $0.65 \text{ mg}\cdot\text{L}^{-1}$. In early September root samples were tested for the presence of *Pythium* Pringsh. spp. root rot and results were negative.

On Sept. 9, 2004, the study was terminated and data recorded. Seedlings in the outer rows of each modified float tray were discarded to remove edge effects leaving a 4 x 4 square of 16 cells containing 16 plants. Three plants were chosen randomly from the 4 x 4 square and crown growth index (widest diameter + diameter perpendicular to widest diameter \div 2), plant height, number of tillers, leaf number, leaf length, leaf area, root length, and root area were recorded. Leaf length and leaf area, and root length and root area were recorded using a Monochrome Agvision System 286 Image Analyzer (Decagon Devices, Inc., Pullman, WA). Leaf, stem, and root dry weights were also recorded following drying at 60C (140F) to a constant weight (48 hr). Top dry weight (leaf + stem dry weight), root:top ratio [RTR (root dry weight \div top dry weight)], total plant dry weight (leaf + stem + root dry weight), specific leaf area [SLA (leaf area \div leaf dry weight)], and root diameter [(root length \div root area) \div 3.14] were calculated.

Leaves of plants were ground separately using a Foss Tecator Cyclotec™ 1093 sample mill (Analytical Instruments, LLC, Golden Valley, Minn.) to pass a $\leq 0.5 \text{ mm}$ (0.02 in) sieve. Mineral nutrient analysis [N, P, K, Ca, Mg, sulfur (S), sodium (Na), manganese (Mn), zinc (Zn), copper (Cu), iron (Fe), and boron (B)] of leaves of five replications was conducted by the North Carolina Department of Agriculture and Consumer Services, Raleigh. Nitrogen concentrations were determined by oxygen combustion with an elemental analyzer (NA 1500, CE Elantech Instruments, Milan, Italy). All other mineral nutrient concentrations were determined by EPA method 200.7 with an ICP spectrophotometer (Optima 3300 DV ICP Emission Spectrometer; Perkin Elmer Corp., Wellesley, Mass.), following open-vessel nitric acid (HNO_3) digestion in a microwave digestion system (CEM Corp., Matthews, NC). Mineral nutrient contents of the

leaves were based on percentage concentration of a nutrient divided by 100 and multiplied by the leaf dry weight.

Data were subjected to regression analysis in SAS version 8.01 (SAS Inst. Inc., Cary, NC). When significant ($P \leq 0.05$), simple linear and polynomial curves were fitted to data. The maximum of the polynomial curve was calculated as a first order derivative of the independent variable where the dependent variable equaled zero.

Results and Discussion

Growth. Total plant, top, leaf, stem, and root dry weights of southern sea oats responded quadratically to increasing NAR with predicted maximum total plant, top, leaf, stem, and root dry weights at 145, 145, 141, 151, and 143 $\text{mg}\cdot\text{L}^{-1}$, respectively (Fig. 1). At maximum dry weight, total plant, top, leaf, stem, and root dry weights increased 196%, 237%, 208%, 316%, and 121%, compared to N at 10 $\text{mg}\cdot\text{L}^{-1}$. Hester and Mendelsohn (18) reported total dry weight of sea oats grown in containers with dune sand increased 230% when fertilized with macro-nutrients (10N-10P₂O₅-10K₂O, 732 $\text{kg}\cdot\text{ha}^{-1}$) compared to no fertilizer. In contrast, Bachman and Whitwell (1) reported top dry weight of sea oats was less when grown in a peat:perlite substrate with N at 1.8 $\text{kg}\cdot\text{m}^{-3}$ (3 lb/yd^3) in comparison to N at 0.9 $\text{kg}\cdot\text{m}^{-3}$ (1.5 lb/yd^3). The differences may have resulted from the substrates used and the frequency of irrigation as the float system provides constant irrigation and nutrients versus an organic substrate which is irrigated less frequently and has lower moisture and nutrient retention.

Research with the container-grown herbaceous perennials, blackfoot daisy (*Melampodium leucanthum* Torr. and Gray) (20), ‘Margarete’ fall flowering anemone (*Anemone x hybrida* Paxton ‘Margarete’) (10), autumn sage (*Salvia greggii* Gray) (19), and ‘Scarlet Sage’ salvia (*Salvia splendens* F. Sellow ex Roem. & Schult. ‘Scarlet Sage’) (22) found maximum growth was achieved with N at 166, 150, 150, and 210 $\text{mg}\cdot\text{L}^{-1}$, respectively, similar to results

herein for southern sea oats. In contrast, N was required at 400 or 399 $\text{mg}\cdot\text{L}^{-1}$ applied weekly to maximize growth of container-grown ‘Stella de Oro’ daylily (*Hemerocallis* L. x ‘Stella de Oro’) or ‘Parigo Pink’ Inca lily (*Alstroemeria* L. ‘Parigo Pink’), respectively (29, 37). Optimum NARs are dependent upon not only the N rate but also frequency of application. The optimum NAR usually decreases with increasing frequency of application.

The similar response of all plant parts to NAR (Fig. 1) was unexpected as roots and tops frequently respond differently to increasing NAR. Griffin et al. (15) and Cabrera and Devereaux (5) reported for containerized ‘Green Giant’ arborvitae (*Thuja* X ‘Green Giant’) and ‘Tonto’ crape myrtle (*Lagerstroemia indica* L. x *fauriei* Koehne ‘Tonto’) that root dry weight decreased quadratically or linearly, respectively, with increasing NARs, whereas top dry weight increased quadratically. In addition, it is unusual to observe root dry weight peaking at a similar NAR as top dry weight since root growth is often maximized at a lower NAR (15, 28). Response of root growth to NAR appears to be very species specific. However, similar to data herein, Dubois et al. (10) working with ‘Margarete’ fall flowering anemone and Conden et al. (8) working with Japanese ternstroemia (*Ternstroemia gymnanthera* Thunb.) reported root dry weight increased quadratically with increasing NAR with calculated maximum root dry weight occurring with N at 119 and 86 $\text{mg}\cdot\text{L}^{-1}$, respectively, which was similar to NAR that maximized top dry weights for each species. Based on our results, sea oats appear to require high rates of N to maximize growth.

Similar to plant dry weights, leaf area, root length, and root area responded quadratically to NAR (Table 1). Maximum leaf area, root length, and root area were predicted with N at 157, 140, and 140 $\text{mg}\cdot\text{L}^{-1}$ (ppm), respectively. At the maximum value, leaf area, root length, and root area increased 241%, 153%, and 146% compared to 10 $\text{mg}\cdot\text{L}^{-1}$. Root diameter (mean = 2.67 mm \pm 0.12 SE) was unaffected by NAR (data not presented) indicating NARs did not alter root

morphology. Plant height, crown growth index, tiller number, and leaf number responded quadratically to NAR with calculated maximum height (mean = 167.7 cm), crown growth index (mean = 3.9 mm), number of tillers (mean = 1.2), and number of leaves (mean = 7.2) occurring at 162, 148, 140, and 139 mg•L⁻¹ (ppm), respectively (data not presented).

RTR was unaffected by NAR (data not presented) indicating carbon allocation between the top and roots was unaffected by NAR. In contrast, most plants typically allocate a larger fraction of carbohydrates to top growth with increasing NAR (11). RTR of lettuce grown in the float system was less when grown at a NAR of 100 mg•L⁻¹ (ppm) compared to 60 mg•L⁻¹ (36).

Likewise, SLA was unaffected by NAR indicating leaf thickness was unaffected by NAR (Table 1). SLA is a morphological index of leaf expansion with a high ratio corresponding to a thinner leaf (13). However, contrary to our results, SLA of lettuce transplants grown in the float system increased linearly with increasing rates of K (36).

Mineral nutrient concentrations and contents. Leaf mineral nutrient concentrations of N and P responded quadratically with increasing NAR, whereas foliar mineral nutrient concentrations of K, Ca, S, Na, Mn, Zn, and Cu responded linearly to increasing NARs (Table 2). Nitrogen concentrations in tops of ‘Parigo Pink’ Inca lily and ‘Scarlet Sage’ salvia responded similarly to increasing NAR (22, 37). Nitrate is absorbed continually by plants as long as it is present in the substrate solution with excess nitrate being stored when supply exceeds demand for growth (9). Maximum top dry weight occurred at a calculated NAR of 145 mg•L⁻¹, with a corresponding foliar N concentration of 31 mg•g⁻¹ (Table 3). Thus, foliar N concentration \geq 31 mg•g⁻¹ appears adequate for maximum growth. This foliar N concentration is higher than leaf N concentration at 21.4 mg•g⁻¹ and 18.5 mg•g⁻¹ reported for sea oats by Hester and Mendelsohn (18) and Bachman and Whitwell (1), respectively (Table 3). However, it is lower than N at 45

and $47 \text{ mg}\cdot\text{g}^{-1}$ required for maximum growth of 'Parigo Pink' Inca lily and 'Margarete' fall flowering anemone, respectively (10, 37).

At maximum top dry weight, foliar P and K concentrations were $6.3 \text{ mg}\cdot\text{g}^{-1}$ and $20.9 \text{ mg}\cdot\text{g}^{-1}$, respectively (Table 3). Concentrations of P and K increased with increasing NAR. Thus, increasing foliar P and K concentrations might be expected with increasing NAR. Foliar K concentrations of lettuce transplants grown in the float system (36), also increased with increasing rate of K application. In recent studies, foliar P and K concentrations increased quadratically or linearly with increasing NARs for both an herbaceous and a woody perennial plant (8, 16). The lowest foliar P concentration reported herein [$3.7 \text{ mg}\cdot\text{g}^{-1}$ (Table 2)] was greater than the foliar P concentrations (1.3 and $1.6 \text{ mg}\cdot\text{g}^{-1}$) reported by Hester and Mendelssohn (18) and Bachman and Whitwell (1) indicating even the lowest P rate ($17.5 \text{ mg}\cdot\text{L}^{-1}$) was not limiting growth in this study. In addition, P at 2.5 and $5 \text{ mg}\cdot\text{L}^{-1}$ (lowest rate applied) was adequate for maximum growth of *Rhododendron* 'Victor' and 'Helleri' holly (*Ilex crenata* Thunb. 'Helleri'), respectively (17, 42).

Leaf Ca concentrations decreased with increasing NAR (Table 2). Reductions of major cations (Ca and Mg) in leaf tissue concentration with increasing NARs have been reported in studies in which the NH_4^+ form is a significant fraction of the N supply (5). Reduced levels of Ca can be attributed to antagonistic effects between cations in the substrate solution competing for uptake by the roots or dilution due to increased growth with increasing NARs. Foliar Ca content increased quadratically with increasing NAR with a predicted maximum at $141 \text{ mg}\cdot\text{L}^{-1}$ (Table 4), indicating decreasing foliar Ca concentration with increasing NAR was due to dilution. Foliar nutrient content for the other mineral nutrients increased quadratically with increasing NARs except for Fe (data not presented) and Na, which were unaffected by NARs (Table 4).

Increasing leaf Mg concentrations with increasing NARs might be expected as it is a vital

component of chlorophyll and a cofactor for many regulatory enzymes (27) all of which should increase from chlorotic, N stressed to healthy, N sufficient plants due to increasing NARs.

However, in the present investigation top Mg concentration was unaffected by NAR (Table 2).

Since S is a constituent of many proteins and enzymes associated with growth, foliar S concentration might be expected to increase with increasing NAR (25, 26) which is what occurred herein (Table 2). However, Cabera and Devereaux (5) reported foliar S concentrations decreased with increasing NAR, which impacted the N:S ratio. They attributed a decrease in growth to the increasing N:S ratio. The N:S ratio was unaffected by NARs in the present investigation (data not presented).

Leaf Na concentration decreased linearly with NAR (Table 2). This decrease in Na concentration could be due to the ability of a halophyte to tightly regulate Na uptake at salinity levels below or equivalent to seawater (14). Southern seaoats belongs to the subfamily Chloridoideae (7). Most members of this subfamily are regarded as salt tolerant (halophytes) and have the ability to absorb salt from the soil and then exude the salts through microhairs on the leaf surface that function as salt glands after translocation through the grass (6).

Concentrations of Mn, Zn, and Cu increased linearly with increasing NAR (Table 2). Foliar mineral nutrient concentrations of Fe (mean = $101.44 \mu\text{g}\cdot\text{g}^{-1} \pm 33.92$) and B (mean = $8.69 \mu\text{g}\cdot\text{g}^{-1} \pm 0.50$) were unaffected by NAR. From this we conclude foliar N concentration $\geq 31 \text{ mg}\cdot\text{g}^{-1}$ was adequate for growth, whereas for all other nutrients (P, K, Ca, Mg, S, B, Cu, Fe, Mn, Na, and Zn) reported means (Table 3) should be considered indicative of good plant vigor, although optimal levels were not determined directly.

Both foliar N concentration and plant dry weight responded quadratically to NARs (Table 2, Fig. 1). Thus, the effects of NARs on dry weight can be explained by direct effects of leaf N concentration. In addition, top dry weight was highly correlated to top N concentration ($P =$

0.004, $r = 0.61$). In particular, the decrease in dry weight at higher than optimal NARs appeared to be related to the EC in the nutrient solution.

Summary. Southern seaoats can be produced successfully using the float system with optimum N rates of 140 to 150 mg•L⁻¹ provided by a 2N-3.5P-1K ratio liquid slow-release fertilizer. Although the dune environment is relatively nutrient sterile, Broome et al. (2) noted dune grasses, such as seaoats, respond positively to fertilization, even though their extensive fibrous root system allows them to exploit the low nutrient conditions in their native habitat. Limiting fertilizer inputs to the lowest nutrient concentrations consistent with adequate growth is an important consideration for growers. It should be implemented, whenever possible, because it is a cost-saving technique that can significantly reduce the levels of nutrient runoff from nurseries (38).

Results herein provide needed information regarding N nutrition of southern seaoats when grown in the float system, however, survivability and vigor of plants after transplanting warrants investigation. Liptay et al. (24) reported ‘TH-318’ tomato [*Solanum lycopersicum* L. var. *lycopersicum* (syns. *Lycopersicon lycopersicum* Karst., *Lycopersicon esculentum* Miller) ‘TH-318’] transplants produced using the float system at a high N rate (350 mg•L⁻¹) exhibited lower survivability in comparison to transplants produced at lower rates (100 to 200 mg•L⁻¹). Similarly, Welbaum et al. (39) observed ‘Krispy King’ sweet corn (*Zea mays* L. var. *rugosa* Bonaf. ‘Krispy King’) transplants grown in the float system flowered earlier and produced fruit earlier than conventional culture, however, fruit and flower quality were lower than direct-seeded methods. Nash [as reported by Latham (23)], has achieved much success with dune establishment of seaoats produced using the float system. Whether differences exist in establishment among seaoats produced using the float system versus other means of culture have yet to be determined. Nevertheless, culture of the species using the float system may allow

tobacco farmers to utilize float beds at times of the year when the beds are not in use. Also, sea oats might serve as a possible alternative crop to tobacco or an additional crop to supplement farm incomes.

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Table 1. Effect of nitrogen application rate (NAR) on leaf area, specific leaf area (SLA), root area, and root length of southern seaoads grown in the float system.^z

NAR (mg•L ⁻¹)	Leaf area (cm ²)	SLA ^y (cm ² •g ⁻¹)	Root	
			Length (cm)	Area (cm ²)
10	40 ± 7 ^y	313 ± 25	77 ± 15	10 ± 2
60	91 ± 13	313 ± 30	164 ± 25	20 ± 2
120	137 ± 31	316 ± 55	179 ± 46	24 ± 7
180	123 ± 20	321 ± 48	192 ± 17	25 ± 3
240	108 ± 7	385 ± 21	125 ± 27	16 ± 5
Significance ^x				
Linear	*	NS	NS	NS
Quadratic	**	NS	**	**

^zData are means of six observations ± 1 SE.

^ySLA = leaf area (cm²) ÷ leaf dry weight (g).

^xNS,*, ** Nonsignificant or significant at $P \leq 0.05$ or 0.01 , respectively. Regression equations are: leaf area, $y = 26.6 + 1.40x - 0.0045x^2$, $R^2 = 0.99$; root length, $y = 62.9 + 1.90x - 0.0068x^2$, $R^2 = 0.98$; root area, $y = 7.9 + 0.249x - 0.0009x^2$, $R^2 = 0.99$.

Table 2. Effect of nitrogen application rate (NAR) on foliar mineral nutrient concentrations of southern seaoats grown in the float system.^z

NAR	N	P	K	Ca	Mg	S
(mg•L ⁻¹)	-----mg•g ⁻¹ -----					
10	20.8 ± 0.7	3.7 ± 0.2	17.3 ± 0.8	2.2 ± 0.10	1.5 ± 0.08	1.5 ± 0.10
60	27.3 ± 0.4	5.4 ± 0.1	20.8 ± 0.6	1.6 ± 0.09	1.4 ± 0.05	2.0 ± 0.03
120	30.2 ± 0.9	6.0 ± 0.1	20.7 ± 1.5	1.4 ± 0.07	1.4 ± 0.05	2.1 ± 0.07
180	31.3 ± 0.8	6.3 ± 0.2	21.9 ± 0.3	1.3 ± 0.04	1.3 ± 0.03	2.2 ± 0.03
240	30.6 ± 0.8	5.8 ± 0.1	21.7 ± 0.7	1.3 ± 0.03	1.4 ± 0.05	2.6 ± 0.40
Significance ^y						
Linear	***	***	**	***	NS	***
Quadratic	***	***	NS	**	NS	NS

Table 2 (continued).

Na	Mn	Zn	Cu	Fe	B
-----mg·g ⁻¹ -----	-----μg·g ⁻¹ -----				
1.8 ± 0.10	95.0 ± 3.4	35.2 ± 2.0	8.3 ± 0.4	54.4 ± 4.4	7.4 ± 0.4
1.6 ± 0.06	104.2 ± 3.9	48.8 ± 5.5	9.7 ± 0.1	65.0 ± 3.6	8.7 ± 1.7
1.0 ± 0.07	124.8 ± 8.8	62.7 ± 2.6	11.5 ± 1.0	81.6 ± 18.3	8.9 ± 1.3
0.6 ± 0.01	125.6 ± 3.6	76.6 ± 4.2	12.5 ± 1.0	242.6 ± 166.2	7.8 ± 0.5
0.6 ± 0.02	141.5 ± 9.1	78.9 ± 2.7	11.5 ± 0.5	63.6 ± 2.0	10.7 ± 0.8
***	***	***	**	NS	NS
*	NS	NS	*	NS	NS

Table 2 (continued).

^zData are means of six observations \pm 1 SE.

^yNS, *, **, *** Nonsignificant or significant at $P \leq 0.05$, 0.01, or 0.001, respectively. Regression equations are: N, $y = 20.0 + 0.131x - 0.0004x^2$, $R^2 = 0.99$; P, $y = 3.5 + 0.035x - 0.0001x^2$, $R^2 = 0.99$; K, $y = 18.4 + 0.017x$, $R^2 = 0.83$; Ca, $y = 2.0 - 0.004x$, $R^2 = 0.89$; S, $y = 1.6 + 0.004x$, $R^2 = 0.95$; Na, $y = 1.9 - 0.006x$, $R^2 = 0.96$; Mn, $y = 92.1 + 0.255x - 0.0002x^2$, $R^2 = 0.98$; Zn, $y = 36.3 + 0.198x$, $R^2 = 0.98$; Cu, $y = 8.8 + 0.016x$, $R^2 = 0.86$.

Table 3. Reported foliar mineral nutrient concentrations of field- and container-grown southern seaots and foliar mineral nutrient concentrations at optimal N rate for top growth of southern seaots grown in the float system.

Mineral nutrient	Reported foliar concn. (mg•g ⁻¹)		Predicted foliar concentration (mg•g ⁻¹) at maximum top growth (N at 145 mg•L ⁻¹)	Predicted maximum foliar concn. (mg•g ⁻¹)
	Field-grown ^z	Container-grown ^y		
N	17.7 to 21.4	14.5 to 18.5	31.2	31.7
P	0.7 to 1.3	1.0 to 1.6	6.3	6.4
K	15.5 to 16.0	12.5 to 15.1	20.9	22.5
Ca	1.2 to 1.3	2.8 to 5.0	1.5	2.0
Mg	1.4 to 1.9	1.1 to 1.6	NA	NA
S	NA	NA	2.2	2.6
Na	2.2 to 4.1	NA ^x	1.0	1.8
Mn	0.02 to 0.05	0.04 to 0.05	0.12	0.16
Zn	0.01 to 0.02	0.01 to 0.02	0.07	0.08
Cu	0.02 to 0.03	0.003 to 0.004	0.01	0.01
Fe	NA	NA	NA	NA
B	NA	NA	NA	NA

^zMeans for southern seaots reported by Hester and Mendelssohn (18).

^yMeans for southern seaots reported by Bachman and Whitwell (1).

^xNA = Not available.

Table 4. Effect of nitrogen application rate (NAR) on foliar mineral nutrient contents of southern seaots grown in the float system.^z

NAR (mg•L ⁻¹)	N	P	K	Ca	Mg	S	Na
	-----mg-----						
10	66 ± 5	12 ± 1	55 ± 4	7.0 ± 0.7	4.8 ± 0.4	4.6 ± 0.5	5.8 ± 0.6
60	178 ± 22	36 ± 4	134 ± 15	10.4 ± 0.9	8.8 ± 1.1	13.1 ± 1.6	10.5 ± 1.3
120	286 ± 54	58 ± 11	191 ± 30	13.5 ± 2.5	13.0 ± 2.4	19.8 ± 3.7	10.3 ± 2.7
180	222 ± 40	46 ± 10	158 ± 33	9.0 ± 1.7	9.6 ± 2.1	16.1 ± 3.3	4.4 ± 0.9
240	181 ± 33	34 ± 6	127 ± 21	7.4 ± 1.2	8.3 ± 1.2	15.1 ± 2.6	3.6 ± 0.6
Significance ^y							
Linear	NS	NS	NS	NS	NS	*	NS
Quadratic	**	**	**	**	**	**	NS

Table 4 (continued).

Mn	Zn	Cu	Fe	B
-----mg-----				
0.3 ± 0.02	0.1 ± 0.01	0.03 ± 0.002	0.17 ± 0.02	0.02 ± 0.001
0.7 ± 0.07	0.3 ± 0.05	0.06 ± 0.009	0.42 ± 0.05	0.05 ± 0.010
1.2 ± 0.20	0.6 ± 0.10	0.10 ± 0.020	0.70 ± 0.07	0.08 ± 0.010
0.9 ± 0.20	0.5 ± 0.10	0.09 ± 0.020	1.26 ± 0.67	0.06 ± 0.010
0.8 ± 0.09	0.5 ± 0.06	0.07 ± 0.008	0.37 ± 0.05	0.06 ± 0.020
*	*	NS	NS	NS
**	**	**	NS	*

Table 4 (continued).

^zData are means of six observations \pm 1 SE.

^yNS, *, **, *** Nonsignificant or significant at $P \leq 0.05$, 0.01, or 0.001, respectively. Regression equations are: N, $y = 37.2 + 3.11x - 0.11x^2$, $R^2 = 0.96$; P, $y = 5.2 + 0.67x - 0.0023x^2$, $R^2 = 0.97$; K, $y = 38.4 + 2.00x - 0.0069x^2$, $R^2 = 0.98$; Ca, $y = 6.3 + 0.094x - 0.0004x^2$, $R^2 = 0.89$; Mg, $y = 3.8 + 0.11x - 0.0004x^2$, $R^2 = 0.93$; S, $y = 3.1 + 0.20x - 0.0007x^2$, $R^2 = 0.96$; Mn, $y = 0.18 + 0.011x - 0.00004x^2$, $R^2 = 0.95$; Zn, $y = 0.03 + 0.007x - 0.00002x^2$, $R^2 = 0.98$; Cu, $y = 0.01 + 0.001x - 0.000004x^2$, $R^2 = 0.97$; B, $y = 0.02 + 0.0007x - 0.000002x^2$, $R^2 = 0.89$.

NS, *, **, *** Nonsignificant or significant at $P \leq 0.05$, 0.01, or 0.001, respectively.

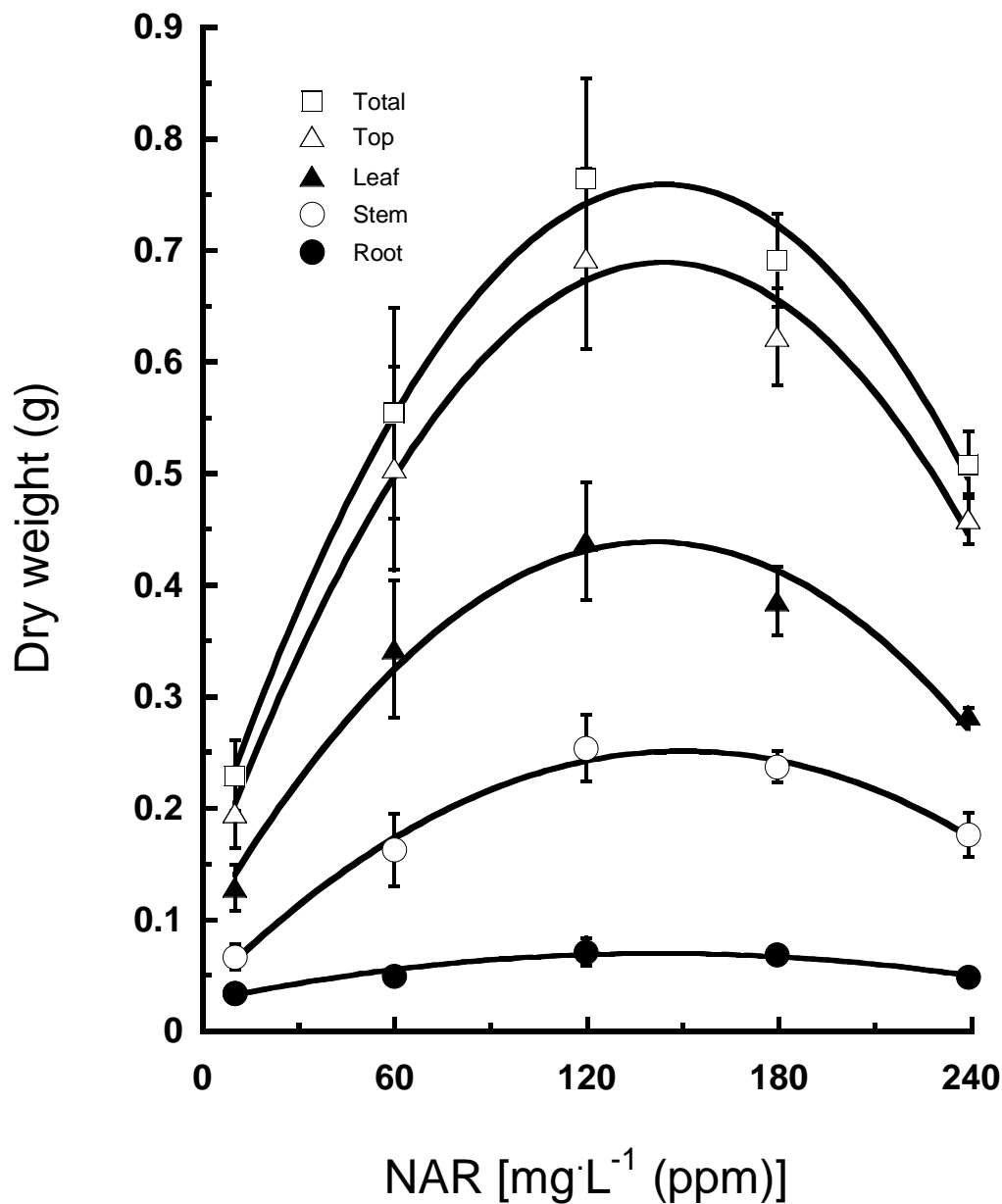


Fig. 1. Influence of nitrogen application rate (NAR) on total plant, top, leaf, stem, and root dry weights of southern sea oats grown in the float system. Data points are means of six observations and vertical bars = ± 1 SE. Regression equations are: total plant dry weight $y = 0.15 + 0.008x - 0.00003x^2$, $R^2 = 0.99$; top dry weight $y = 0.13 + 0.008x - 0.00003x^2$, $R^2 = 0.99$; leaf dry weight $y = 0.09 + 0.005x - 0.00002x^2$, $R^2 = 0.99$; stem dry weight $y = 0.04 + 0.003x - 0.0000095x^2$, $R^2 = 0.99$; root dry weight $y = 0.03 + 0.0006x - 0.000002x^2$, $R^2 = 0.98$.