

**CWPPRA - ENHANCEMENT OF BARRIER ISLAND AND SALT
MARSH VEGETATION DEMONSTRATION (TE-53) PROJECT**

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EXECUTIVE SUMMARY

Barrier islands are unique environments comprising a variety of habitat types, each exhibiting unique environmental stressors. The geomorphology of Louisiana barrier islands developed as a result of deltaic processes associated with the Mississippi River, and the ephemerality of these islands remains associated with the delta cycle. There is a focus on the restoration and maintenance of these important ecosystems, which are critical components of Louisiana's comprehensive coastal restoration plans. Such restoration efforts are tremendously expensive because they require long-distance transport of vast volumes of sand and other sediment. Therefore, a crucial component of barrier island restoration is the rapid establishment and expansion of vegetation to stabilize these newly placed sediments. The research chapters in the following report evaluate the efficacy of soil amendments, planting techniques, and propagule/seed dispersal methods in improving the success and cost efficiency of plant restoration efforts in key barrier island habitats.

Humic acid, an operationally defined component of organic matter, is known in the agricultural literature to improve soil quality in marginal soils when applied as a soil amendment, but previously it has not been thoroughly investigated for use in coastal plant restoration efforts. The greenhouse studies in this report were conducted to determine the beneficial and deleterious application ranges of this soil amendment.

- Substantial variation in the response of individual plant species to humic acid amendment was detected, however, applications of 2,700 ml m⁻² and higher detrimentally affected all plant species.
- Low to moderate humic acid amendment dosages (100 to 300 ml m⁻²) resulted in some increased growth response in the species assessed, with the exception of *Distichlis spicata* (saltgrass) and *Baccharis halimifolia* (groundsel bush).
- An additional greenhouse study examining the potential synergy of fertilizer regime and humic acid amendment detected a significant benefit of fertilizer regime, but no clear synergy between these treatments.

A field investigation of planting density, fertilizer regime, and humic acid amendment in dune and swale environments yielded several clear and important findings relevant to barrier island restoration, in spite of multiple environmental anomalies. The winter preceding the field study's implementation was atypically cold and delayed the readiness of the nursery stock for planting in early spring as is desired for such projects. The Deepwater Horizon oil spill clean up efforts also delayed some of the plantings until the fall. Additionally, two years of unusually below normal precipitation subsequent to the planting effort may have obfuscated some treatment effects. Nonetheless, several important findings were discernible.

- Increasing the planting density of *Uniola paniculata* (sea oats) from 1.52 m centers (low density) to 0.76 m centers (high density) resulted in an obvious and sustained benefit of increased vegetative coverage.

- The low density planting treatment for *Panicum amarum* (bitter panicum) quickly became equivalent to the high density planting treatment, suggesting that there would be no long-term benefit to increasing the planting density of this rapidly expanding species.
- Broadcast fertilizer increased vegetative coverage of all species. This effect was somewhat masked in the first year, as the planting contractor had also applied fertilizer to all plantings.
- Little benefit of humic acid amendment was discernible in the field study, likely due to a combination of factors including the minimal precipitation during the study and lack of soil components to retain the applied humic acid.
- The presence of *Cynodon dactylon* (bermuda grass), which had been seeded as a portion of the restoration effort, appeared to limit the establishment and expansion of target species, particularly *Spartina patens* (marshhay cordgrass).
- *Cynodon dactylon* might restrict sand movement, thereby limiting the development of a well-defined primary dune by interfering with aeolian transport.
- Finally, the field components of this research project underscored the importance of incorporating flexibility in barrier island planting schedules to coincide with the suitability of the created physical environment to optimize initial survival and subsequent growth enhancing amendments that may increase plant vigor and expansion.

Key insights regarding the restoration ecology of *B. halimifolia* were:

- Optimal seed germination (64%) occurs at the soil surface, whereas seed burial of greater than 0.5 cm results in less than 3% germination.
- Shade is found to significantly reduce *B. halimifolia* germination response.
- Efforts to establish *B. halimifolia* from seed in swale environments necessitates locating these restoration efforts in protected areas where the potential for the burial of seeds by sand or impact of shade is minimal.
- Importantly, *B. halimifolia* seeds have no required dormancy period and can germinate immediately after leaving the mother plant without any pretreatment, limiting the seed preparation requirements prior to initiating a restoration effort.
- Hydromulch significantly increases seed germination in sediments containing no organic matter, but simulated drought conditions negatively affect germination response regardless of the treatment assessed.

- Successful field establishment of *B. halimifolia* employing seed and hydromulch (hydroseeding) will require sufficient soil moisture post hydroseeding, and hence a subsequent rainfall event or watering for germination to occur.

Several findings concerned the targeted restoration of back-barrier salt-marsh habitat plant species. Greenhouse studies demonstrated benefits of treatments for *Avicennia germinans* and *Spartina alterniflora*.

- Humic acid amendment (500 ml m^{-2}) resulted in increased *S. alterniflora* (smooth cordgrass) biomass and cumulative height without resulting in differences in leaf tissue nitrogen.
- A very thin (nonsmothering) layer of hydromulch has potential to enhance survival and establishment of *A. germinans* (black mangrove) propagules.
- Hydromulch application in the upper intertidal range assists in propagule establishment in selected locations.
- Natural *A. germinans* establishment was influenced by marsh platform elevation, and tidal creeks that have developed will likely provide a conduit for future black mangrove dispersal into the project area.

INTRODUCTION

The development of methods and techniques to enhance the restoration of coastal environments is an area of continuing need for the natural resource managers responsible for maintaining these valuable habitats. This need is particularly pressing in Louisiana, where a high rate of coastal wetland loss is well documented (Dahl 2000; Bjerstedt 2011). Barrier island and headland habitats are also being lost and are in need of improved restoration techniques and approaches (Khalil et al. 2010). Barrier islands are well known to be geomorphically important components of comprehensive coastal ecosystem restoration as well as provide a multitude of ecosystem services (Swilling et al. 1997; Stone and McBride 1998), particularly reductions in storm energies for the mainland areas they protect (Stone and McBride 1998; Petrolia and Kim 2009; Wamsley et al. 2009). Barrier islands include a number of unique habitats (Hester et al. 2005), each possessing specific environmental stressors and providing specific ecosystem services. A key component of the effective restoration of many coastal habitats is the successful establishment and subsequent expansion of appropriate vegetation, which acts to protect restored areas by stabilizing sediment and mitigating erosive forces (Brown and Hafenrichter 1948; Broome et al. 1982; Shepard et al. 2011). Therefore, the investigation of potential mechanisms and techniques to enhance the effectiveness of restoration plantings in coastal environments is an area of crucial need in the field of coastal restoration.

Dune habitats form immediately landward of the beach environment on barrier islands as a result of aeolian transport of sands (Nordstrom 2008). Vegetation and nonliving structures such as sand fencing are integral to the formation of dunes, for the presence of such structures reduces wind velocity, causing sand particles to accumulate in these areas (Dahl and Woodward 1977). Because the substrate of dune habitats is almost entirely sand, a number of environmental stressors act to limit both the plant species that can survive in these habitats as well as the growth of these species (Dahl and Woodward 1977; Hester et al. 2005). The low soil organic matter typical of dune habitats, especially those that have been recently rehabilitated with newly placed materials, results in low availability of both water and nutrients (Mendelssohn and Hester 1988; Hester et al. 2005; Lane et al. 2008). Additionally, the proximity of dune habitats to the ocean can result in salinity impacts due to salt spray (Oostings 1945; Oostings and Billings 1942; Boyce 1954). However, salt spray has been shown to be beneficial by providing critical micronutrients (Boyce 1954; Van der Valk 1974). Vegetation growing in dune habitats can also be subject to frequent abrasion, burial, and excavation due to the shifting nature of the sandy substrate (Oostings 1945; Oostings and Billings 1942). In addition to these chronic stressors, acute episodes such as storm-related overwash events represent a major obstacle to successful establishment and maintenance of vegetation (Rosati and Stone 2009). Ritchie and Penland (1988) found that the dune systems at the Caminada-Moreau Headland followed a 10- year cycle over which time they increased sand storage within the dune habitat, with a large storm rapidly removing this storage at the end of the cycle.

Swale habitats occur behind (i.e., interior to) rear dune habitats, and they typically have higher soil organic matter content. They can, though, be subject to many of the same stressors as dune habitats, but at a lower intensity, as they are protected by the higher elevation foredune (Hester et al. 2005; Lane et al. 2008). The lower elevation of swale habitats can result in longer-term elevated soil salinities after overwash events, as a greater volume of precipitation is

required to reduce surface soil salinities than in sandy dune soils. The lower intensity of stressors in swale habitat typically results in a different assemblage of plant species than that of the more stressful dune environment (Hester et al. 2005; Lane et al. 2008). *Spartina patens* (marshhay cordgrass) becomes a dominant plant species in the rear dune and swale habitats of the Gulf of Mexico, replacing foredune species such as *U. paniculata* (sea oats) and *P. amarum* (bitter panicum) (Ritchie and Penland 1988; Hester et al. 2005). Similarly, *D. spicata* (saltgrass) and *Paspalum vaginatum* (seashore paspalum) rarely occur in foredune environments but can often occur more extensively in swale habitats. Woody species, such as *B. halimifolia* (groundsel bush), also become more prevalent in the swale environment (Hester et al. 2005; Lane et al. 2008). The presence of woody vegetation is important, for it provides an additional dimension of habitat beyond the herbaceous vegetation more typical of foredune areas on Louisiana barrier islands. Although the benefits of woody species in swale habitats have not been scientifically assessed in Louisiana, similar dune scrub in other areas is regarded as important faunal habitat (Russell et al. 2009).

Back-barrier salt marshes are highly valuable habitats occurring in the lower energy environments on the bay side of barrier islands (Hester et al. 2005; Khalil et al. 2010). These ecosystems provide many of the same ecosystem services as salt marshes occurring on the mainland of Louisiana, including habitat for fish and invertebrates (Anderson et al. 2012), carbon sequestration, and transformation of nutrients (Hester et al. 2005). An important aspect of back-barrier marshes is that they provide the platform for migration of sediments from other components of the island during overwash events, thus, extending the longevity of the restored barrier island. Louisiana back-barrier marshes experience substantial erosion during the post-frontal phases of major storms (Khalil et al. 2010), and thus they require rehabilitation to an extent similar to that of the Gulf of Mexico-facing barrier island habitats. Natural back-barrier marshes tend to have a coarser substrate than their mainland analogues, but many of the same plant species dominate them. In particular, *S. alterniflora* (smooth cordgrass) and *A. germinans* (black mangrove) are key constituents of back-barrier salt marshes (Hester et al. 2005). Created back-barrier marshes are frequently characterized as having relatively low soil organic matter and nutrient content (Fearnley 2008). Additionally, back-barrier marshes may develop hypersaline surface soils immediately subsequent to sediment placement, because prior to compacting, the elevation of these sediments is frequently above the average tidal range, leading to salt pan formation due to evaporation. Once the created back-barrier salt marsh sediments have compacted and dewatered sufficiently to fall within target marsh elevations, soil anaerobiosis and salinity levels typical of a coastal salt marsh as a consequence of tidal action are primary drivers of vegetation establishment and expansion.

Although the environmental stressors limiting plant establishment and expansion in the dune, swale, and back-barrier marshes of barrier islands vary, as do the native plant species adapted to succeed in these environments, several general strategies to enhance the efficacy and cost-effectiveness of plant restoration efforts are discernible. For instance, the use of widely available soil amendments that ameliorate unfavorable soil conditions such as low soil fertility and organic matter content is worth consideration. Additionally, exploring alternative means of establishing vegetation either to increase the speed with which high vegetative cover can be achieved or to reduce the cost of establishing a plant species in these habitats should be investigated. Finally, studies both of native species to fill niches that are not addressed by

current planting approach and of species combinations, such as woody vegetation in swale habitats, should be undertaken. The research described in the following chapters represents multiple components of a CWPPRA Demonstration Project (TE-53) designed to address existing knowledge gaps detailed above in a combination of greenhouse studies and field trials in conjunction with a large-scale restoration effort.

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GREENHOUSE STUDIES

CHAPTER 1. GREENHOUSE INVESTIGATIONS OF HUMIC ACID AMENDMENT AND POTENTIAL SYNERGY WITH FERTILIZER IN COASTAL DUNE, SWALE, AND SALT MARSH VEGETATION

Introduction

A key aspect of barrier island restoration is the successful establishment of vegetation, which serves to stabilize the sands and sediments of the island (Dahl and Woodward 1977; Hester et al. 2005). However, the dune, swale, and back-barrier habitats that occur on barrier islands are harsh environments in which vegetative establishment is often challenging (Hester et al. 2005). The development and evaluation of novel methods that enable the successful, efficacious establishment of vegetation in these environments is therefore highly desirable. The humic acid soil amendment technique has been frequently reported for its capacity to improve the growth of agricultural plant species in marginal soils (Zhang et al. 2002; Farouk et al. 2012), but only limited information exists regarding its potential to accelerate coastal plant growth. To further investigate the potential benefits of humic acid amendment, two large, multispecies greenhouse studies were conducted. The first study assessed the beneficial and deleterious dosage range for humic acid. The second study investigated the possible synergy between optimal humic acid amendment levels and fertilizer application.

Humic acid is operationally defined as that portion of organic matter that is insoluble at a pH less than 2 and soluble at higher pH levels (Jackson 1993; Zhang et al. 2002). The two other primary portions of organic matter defined in this operational manner are fulvic acid, which is soluble both at low and high pH levels, and humin, which is insoluble at both at low and high pH levels (Jackson 1993). Humic acid can be extracted from a variety of organic materials, such as composts, agricultural waste products, and leonardite, which is also known as brown coal (Malcolm and MacCarthy 1986; De Santiago et al. 2010). As humic acid is an operationally defined group of polymers rather than a specific chemical compound with a defined chemical formula, variation in the exact chemical properties occurs among formulations (Jackson 1993; De Santiago et al. 2010). The source materials from which individual humic acid formulations are extracted, as well as the actual extraction process, exert a great deal of influence on the final chemical characteristics of the humic acid product (Malcolm and MacCarthy 1986; De Santiago et al. 2010). Nonetheless, commercially produced humic acid preparations generally possess certain consistent properties that result in the improvement of soil qualities (Jackson 1993). Because of these consistent properties, the use of commercially produced humic acid as a soil amendment has been extensively researched in marginal agricultural soils (e.g., Sanchez et al. 2005; Farouk et al. 2012) and horticultural plant management (Ferrini and Nicese 2002; Van Dyke et al. 2009).

A range of humic acid amendment levels has been found beneficial for horticultural and agricultural plant species (Sharif et al. 2002; Verlinden et al. 2009; Udrenas et al. 2011); however, there are relatively few analogous studies on coastal plant species (Willis and Hester 2008; Willis and Hester 2010). In studies assessing the benefits of humic acid on turf grasses and crop species, low to moderate levels of humic acid amendment are typically employed. For example, in a study focusing on turf grasses, Udrenas et al. (2011) found that a 1.5 L hectare⁻¹ application rate of a humic acid preparation (Humistar: 13.32% humic acids and 3.33% fulvic

acids) improved density and disease resistance in *Poa pratensis* and *Agrostis stolonifera*. Similarly, Sharif et al. (2002) found that the amendment of soils with 50 to 100 mg kg⁻¹ humic acid (Humifirst: 12% humic and 3% fulvic acids) resulted in a significant increase in dry weight of maize shoots and roots. Also, Verlinden et al. (2009) performed a number of experiments examining the benefits of applying humic acid at a rate of 50 L hectare⁻¹ (Humifirst: 8.25 kg humic substances hectare⁻¹) on both turf grass and vegetable species, including Italian ryegrass, maize, potato, and spinach, and evaluated these results using a meta-analysis approach. Humic acid amendment generally enhanced biomass yield for all species, with the size of the significant effects decreasing in the order of potato, grass species, and Italian rye (Verlinden et al. 2009). Verlinden et al. (2009) also found that the uptake of nitrogen and phosphorous increased for all species evaluated. Interestingly, Valdrighi et al. (1996) found that humic acid amendment increased biomass production of chicory, and attributed this to effects on the microbial community that in turn improved plant nutrition. Jindo et al. (2012) found that humic acid amendment promoted root growth through emulating plant growth regulators.

Low to moderate application rates of humic acid amendment have also been used in the few previous studies that focused on coastal plant species. Willis and Hester (2008) determined the efficacy of low level humic acid amendment (20, 40, and 80 ml m⁻²) and planting unit (aboveground partition, belowground partition, or whole plant) on *P. amarum* for coastal restoration efforts. Humic acid amendment was found to significantly increase aboveground biomass, but not belowground biomass or aboveground tissue nitrogen concentration (Willis and Hester 2008). Willis and Hester (2010) assessed the effects of low level humic acid amendment (5, 20, and 80 ml m⁻²) interactively with fertilizer on *U. paniculata*, *P. amarum*, *S. alterniflora*, and *A. germinans*. Growth of *P. amarum* and *S. alterniflora* was enhanced with all levels of humic acid amendment in these studies; however, no benefit of any level of humic acid amendment was found for *U. paniculata* or *A. germinans* (Willis and Hester 2010). Humic substances have been applied to dredged marine sediments in conjunction with the planting of *P. vaginatum* with the intent of enhancing phytoremediation processes through the chelation properties exhibited by these humic substances (Bianchi et al. 2010). Although the research focus in this case was not the establishment and growth of *P. vaginatum*, it is important to note that the application of humic substances to this species growing in a dredged marine sediment was not found to be deleterious (Bianchi et al. 2010).

The benefit of fertilizer application in increasing the growth of coastal plant species in dune and swale environments has long been recognized (Brown and Hafenrichter 1948; Broome et al. 1982). Humic acid is thought to be synergistic with fertilizer application by enhancing the effectiveness of nutrient uptake through a number of mechanisms (Yadav 1989; Rauthan and Schnitzer 1981; Piccolo et al. 1992; Sivakumar and Ponnusami 2011). Application of humic acid has been demonstrated to increase root membrane permeability, which results in enhanced nutrient uptake (Rauthan and Schnitzer 1981). Additionally, humic acid is known to increase the availability of nitrogen (Yadav 1989) and phosphorus (Rauthan and Schnitzer 1981) by the formation of soluble compounds, increasing the uptake of these nutrients. In research assessing humic acid amendment and fertilizer application interactively, Willis and Hester (2010) documented that fertilizer application invoked a more immediate growth response but that plants receiving humic acid amendment were statistically equivalent in terms of their growth response by the conclusion of the study. This suggests that humic acid may have acted in a fashion

analogous to a slow-release fertilizer or through some other, temporally similar effect that increased plant growth but required a longer period of time for this effect to be discerned (Willis and Hester 2010).

The first greenhouse experiment described, hereafter referred to as the range-finding study, was designed to provide greater insight into no-effect, beneficial, and toxic amendment levels of humic acid over a broad range of humic acid amendment concentrations in several key coastal plant species representing the dune, swale, and salt marsh environments. The results of this study were used to identify a narrower range of concentrations for the second greenhouse experiment, in which a select range of humic acid concentrations were applied in a factorial design with and without a beneficial fertilizer regime (Broome et al. 1982) to further investigate the benefits of humic acid amendment and possible synergies. These results were used as the basis of the humic acid amendment and fertilization portion of the field study described in a later chapter.

Materials and Methods

Range-Finding Study Experimental Approach

Bare root liners (small diameter seedlings) of *U. paniculata*, *P. amarum*, *D. spicata*, *P. vaginatum*, *S. patens*, and *S. alterniflora* were ordered from local suppliers. At the time of this study, *B. halimifolia* seedlings were not stocked by nurseries; therefore, all *B. halimifolia* seedlings were grown from locally collected seeds. Because of the excessively cold prior winter, no *A. germinans* seedlings were available from Louisiana suppliers, and were therefore acquired from a nursery in Florida. One seedling or bare root liner was transplanted into a standard gallon nursery pot lined with silt cloth to prevent substrate leakage and filled with fine river sand (grain size 0.12 – 0.25mm) from the Atchafalaya Basin. Experimental units were randomly assigned to one of six humic acid treatments, 0 ml m⁻², 100 ml m⁻², 300 ml m⁻², 900 ml m⁻², 2,700 ml m⁻² and 8,100 ml m⁻². Humic acid was acquired from a commercial supplier (3 Tier Technologies, Longwood, Florida) and carefully applied to the soil surface, avoiding aerial plant tissues. Each humic acid treatment was replicated five times for a total of 30 pots per species. The experiment was initiated on June 29, 2009 and was harvested after two months. The experiment was conducted under ambient conditions at the University of Louisiana Center for Ecology and Environmental Technology (CEET) greenhouse facility. Experimental vessels were watered twice weekly based on 1,600 ml yr⁻¹, which is representative of the average precipitation of the Caminada-Moreau Headland, Louisiana (Grand Isle Weather Station: GISL1 - 8761724).

Refinement Study Experimental Approach

As with the range-finding study, bare root liners of *U. paniculata*, *P. amarum*, *D. spicata*, *P. vaginatum*, *S. patens*, and *S. alterniflora* were ordered from local suppliers. *Baccharis halimifolia* seedlings were grown from locally collected seeds, whereas *A. germinans* seedlings were provided by a Florida supplier. One seedling or bare root liner was transplanted into a standard gallon nursery pot lined with silt cloth to limit leakage and filled with fine river sand (grain size 0.12 – 0.25mm) from the Atchafalaya Basin. Experimental units were randomly assigned to one of four humic acid treatments: 0 ml m⁻², 125 ml m⁻², 250 ml m⁻², and 500 ml m⁻²,

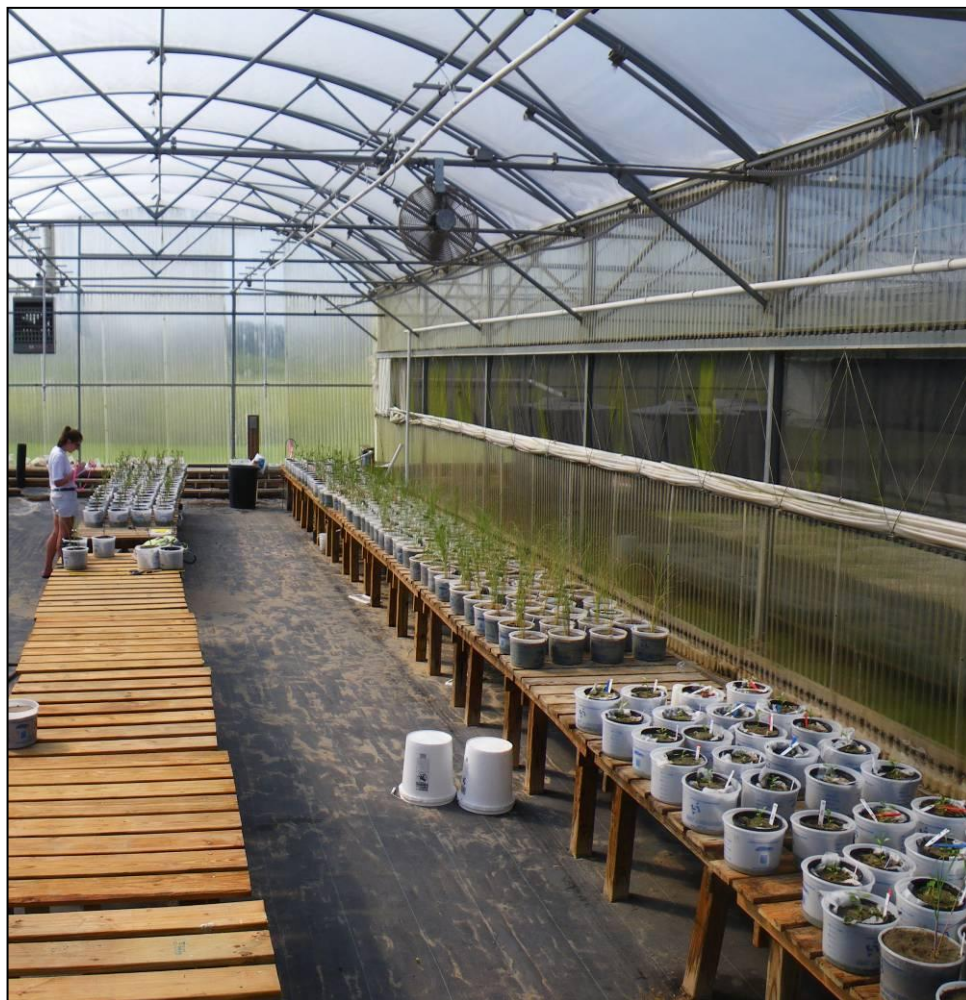


Image 1.1 Image of humic acid refinement study, which included a total of 256 experimental units.

and one of two fertilizer treatments: low fertilizer level (10% Hoaglands Solution; Hoagland 1950) or high fertilizer level (100% Hoaglands Solution) in a factorial fashion. Humic acid was acquired from a commercial supplier (3 Tier Technologies, Longwood, FL) and carefully applied to the soil surface, avoiding aerial plant tissues. Hoaglands Solution was prepared in the Coastal Plant Ecology Laboratory using reagent grade chemicals. Each treatment combination was replicated four times for a total of 32 pots per species. The experiment was initiated on September 21, 2009, at the University of Louisiana CEET greenhouse facility and was harvested ten months later. Experimental units were watered twice weekly based on $1,600 \text{ ml yr}^{-1}$, which is considered average annual rainfall for coastal Louisiana (Grand Isle Weather Station: GISL1 - 8761724).

Vegetative Characterization

Cumulative stem height was determined monthly for all species. For the range-finding study, substantial tissue injury was noted shortly after the initiation of the humic acid amendment treatment. Therefore, the amount of dead tissue of the total tissue was visually estimated weekly as an additional metric. At the conclusion of the study, plant material was harvested, separated into live aboveground, dead aboveground, and belowground biomass components. For the refinement study, leaf tissue was also collected for elemental analysis. For leaf elemental analysis, a subsample of leaf tissue was collected, dried to a constant weight, ground to pass through number 20 mesh using a Wiley Mill, and separated into two aliquots. The first aliquot was submitted to the LSU Soil Testing and Plant Analysis Laboratory for the determination of total carbon and nitrogen content following standard methods. The second aliquot was digested using trace-metal-grade hot nitric acid following the methods of Jones et al. (1991) and was then submitted to the LSU Soil Testing and Plant Analysis Laboratory for determination of elemental content using ICP-OES (EPA method 200.7).

Soil Physico-Chemical Characterization

Soil samples were collected at the conclusion of each of the studies for the determination of soil moisture, conductivity, pH, nutrient status, and organic matter. The soil samples were weighed and then dried at 65°C until a constant weight was achieved and soil percent moisture was calculated. Dried soil samples were homogenized and an approximately 2-g subsample was combusted at 500°C for 5 hours to determine percent organic matter (Parent and Caron 1993). Additional subsamples of the homogenized soil samples were subjected to two 1:2 (w:v) extraction procedures employing deionized water and 2M KCl, respectively. One aliquot of the deionized water extract was used for the determination of pH and conductivity (Rhoades 1990). The second aliquot of deionized water extract was submitted to the LSU Soil Testing and Plant Analysis Laboratory for the determination of total phosphorus, potassium, and other relevant cations using ICP-OES (EPA method 200.7). The KCl extract was submitted to the SLU Microbial Testing Laboratory for the determination of ammonium and nitrate-nitrite using colorimetric methods (EPA method 350.1 and 353, respectively).

Results: Range-Finding Study

Final Cumulative Stem Height

Highly significant effects of species, humic acid amendment, and the interaction thereof were detected in the range-finding study for final cumulative stem height (Figures 1.1 – 1.4; $F =$

17.2, $p < 0.01$, $F = 20.4$, $p < 0.01$, and $F = 5.2$, $p < 0.01$, respectively). Overall, humic acid amendment of 100 ml m^{-2} , 300 ml m^{-2} , and 900 ml m^{-2} demonstrated significantly greater final cumulative stem height than the 0 ml m^{-2} treatment (Figures 1.1 – 1.4; Contrast $F = 4.3$, $p < 0.05$). However, the humic acid amendment of 2,700 and $8,100 \text{ ml m}^{-2}$ showed a highly significant reduction overall in final cumulative stem height compared to the 0 ml m^{-2} treatment (Figures 1.1 – 1.4; Contrast $F = 23.4$, $p < 0.01$). *Uniola paniculata* final cumulative stem height was significantly greater in the 100 ml m^{-2} humic acid amendment treatments compared to the 0 ml m^{-2} treatment (Figure 1.1 Top Panel; Contrast $F = 44.7$, $p < 0.01$). *Spartina patens* final cumulative stem height was significantly greater in the 100 ml m^{-2} , 300 ml m^{-2} , and 900 ml m^{-2} humic acid amendment treatments compared to the 0 ml m^{-2} treatment (Figure 1.3 Top Panel; Contrast $F = 44.7$, $p < 0.01$).

Visually Observed Total Tissue Damage

Significant effects of time, as well as the interactions of time and humic acid amendment, time and plant species, and time, humic acid amendment, and plant species, were detected on visually estimated dead tissue (Figures 1.5 – 1.8; $F = 61.7$, $p < 0.01$, $F = 5.1$, $p < 0.01$, $F = 11.3$, $p < 0.01$, $F = 2.4$, $p < 0.01$, respectively). All species were found to experience a high level of visually evident tissue damage in the $8,100 \text{ ml m}^{-2}$ humic acid amendment treatment. At the conclusion of the study, *U. paniculata* and *P. amarum* displayed elevated levels of tissue damage only at $8,100 \text{ ml m}^{-2}$ humic acid amendment (Figure 1.5; $F = 19.1$, $p < 0.01$ and $F = 22.3$, $p < 0.01$, respectively), whereas *D. spicata* and *S. patens* demonstrated substantial damage at both $2,700 \text{ ml m}^{-2}$ and $8,100 \text{ ml m}^{-2}$ humic acid amendment concentrations (Figure 1.6 Top Panel; $F = 18.6$, $p < 0.01$ and Figure 1.7 Top Panel; $F = 82.8$, $p < 0.01$, respectively).

Biomass Partitioning

Highly significant main effects of species and humic acid amendment, as well as a significant interaction of species and humic acid amendment, on live aboveground biomass were detected (Figures 1.9 – 1.12; $F = 8.4$, $p < 0.01$, $F = 15.4$, $p < 0.01$, $F = 2.3$, $p < 0.05$, respectively). Overall humic acid amendment of 100 ml m^{-2} , 300 ml m^{-2} , and 900 ml m^{-2} significantly increased live aboveground biomass compared with the 0 ml m^{-2} treatment (Figures 1.9–1.12; Contrast $F = 5.6$, $p < 0.05$). As was seen with final cumulative stem height, however, humic acid amendment of 2,700 and $8,100 \text{ ml m}^{-2}$ resulted in a highly significant reduction of live aboveground biomass compared to the 0 ml m^{-2} treatment (Figures 1.9–1.12; Contrast $F = 63.2$, $p < 0.01$). Similarly, highly significant effects of species, humic acid amendment, and the interaction of species and humic acid amendment on belowground biomass were detected (Figures 1.9–1.12; $F = 43.9$, $p < 0.01$, $F = 3.8$, $p < 0.01$, $F = 2.0$, $p < 0.01$, respectively). Humic acid amendment of $2,700 \text{ ml m}^{-2}$ and $8,100 \text{ ml m}^{-2}$ resulted in an overall significant reduction of belowground biomass compared to the 0 ml m^{-2} treatment (Figures 1.9–1.12; Contrast $F = 4.2$, $p < 0.05$). A significant effect of humic acid addition was detected for *P. amarum* live biomass, in which live aboveground biomass was significantly increased in the 100 ml m^{-2} , 300 ml m^{-2} , and 900 ml m^{-2} humic acid treatments compared to the 0 ml m^{-2} treatment (Figure 1.9 bottom panel; Contrast $F = 6.9$, $p < 0.01$). Humic acid amendment levels of 100 ml m^{-2} , 300 ml m^{-2} , and 900 ml m^{-2} also significantly increased *P. amarum* belowground biomass (Figure 1.9 bottom panel; Contrast $F = 7.3$, $p < 0.05$). Interestingly, *S. patens* live aboveground biomass and belowground

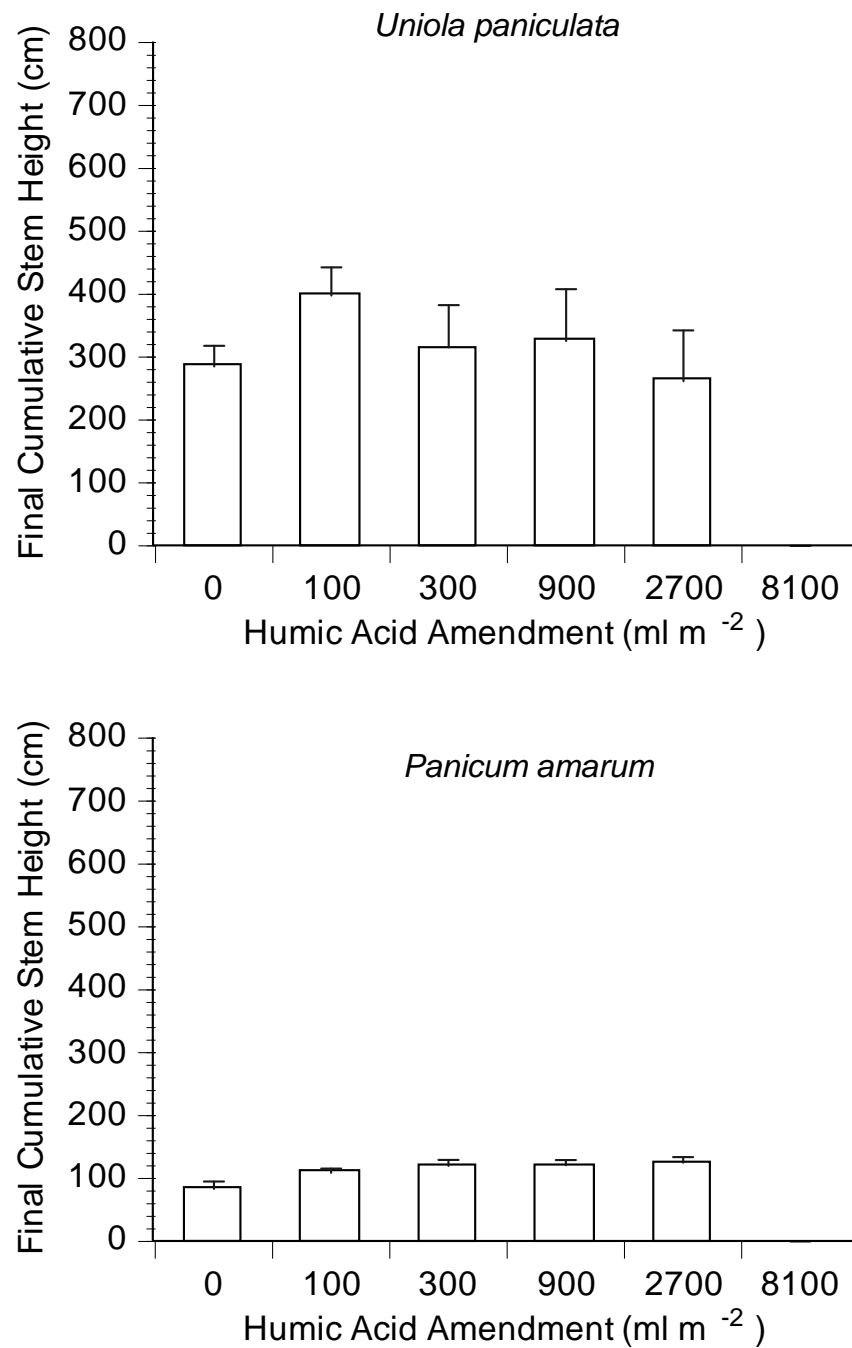


Figure 1.1 The effect of humic acid amendment on final cumulative stem height for *U. paniculata* (top panel; mean \pm SE, $n = 5$, LSD = 133.80) and *P. amarum* (bottom panel; mean \pm SE, $n = 5$, LSD = 133.80).

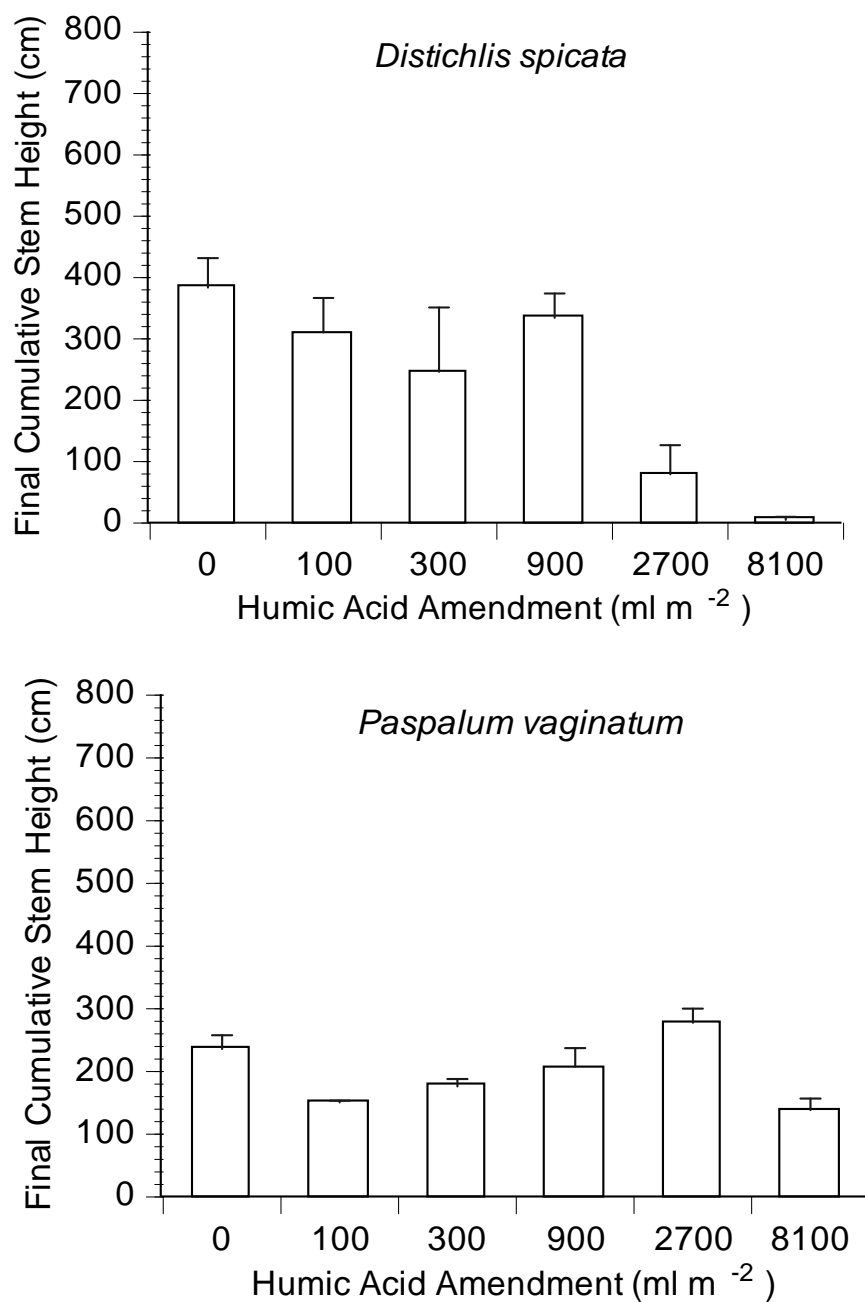


Figure 1.2 The effect of humic acid amendment on final cumulative stem height for *D. spicata* (top panel; mean \pm S.E. , n = 5, LSD = 133.80) and *P. vaginatum* (bottom panel; mean \pm S.E. , n = 5, LSD = 133.80).

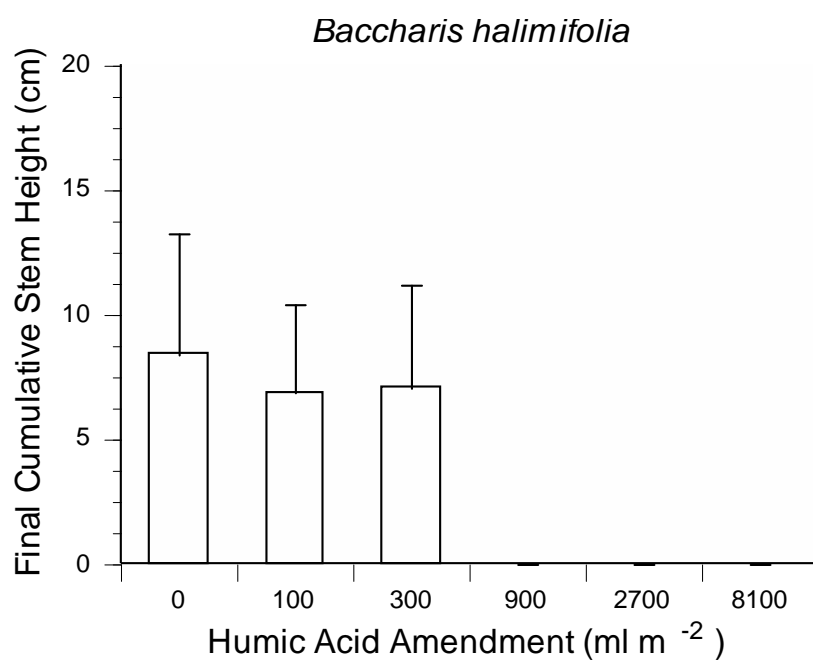
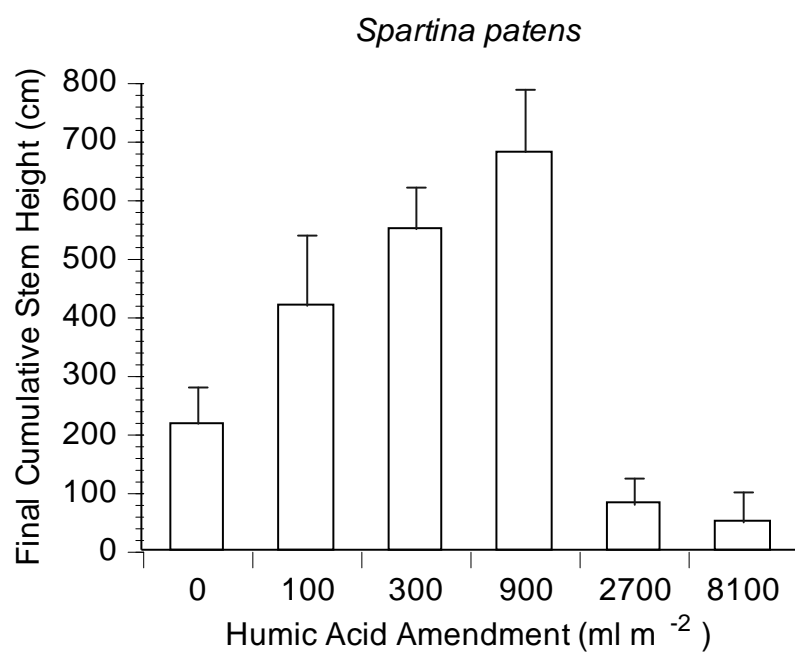


Figure. 1.3 The effect of humic acid amendment on final cumulative stem height for *S. patens* (top panel; mean \pm SE, $n = 5$, LSD = 133.80) and *B. halimifolia* (bottom panel; mean \pm SE, $n = 5$, LSD = 133.80).

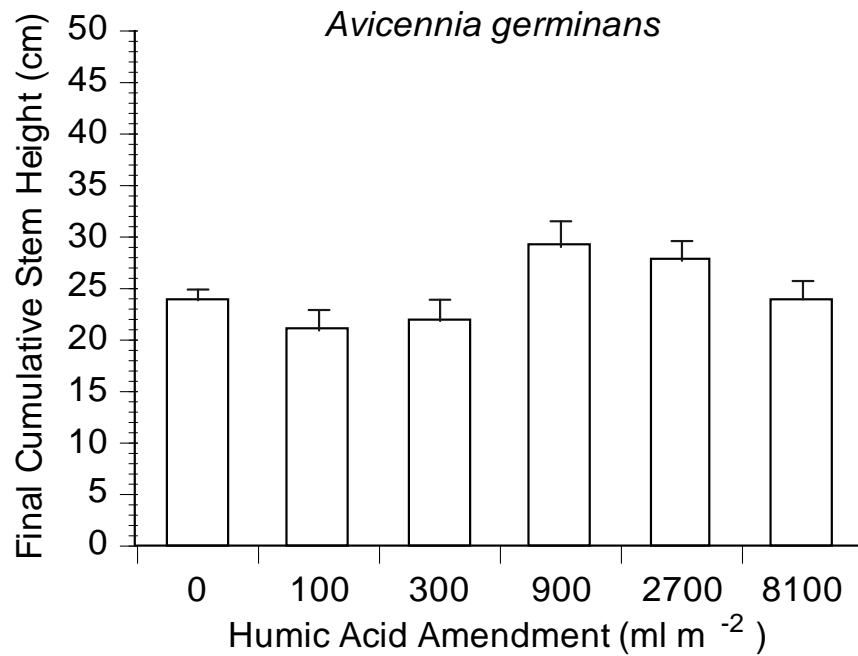
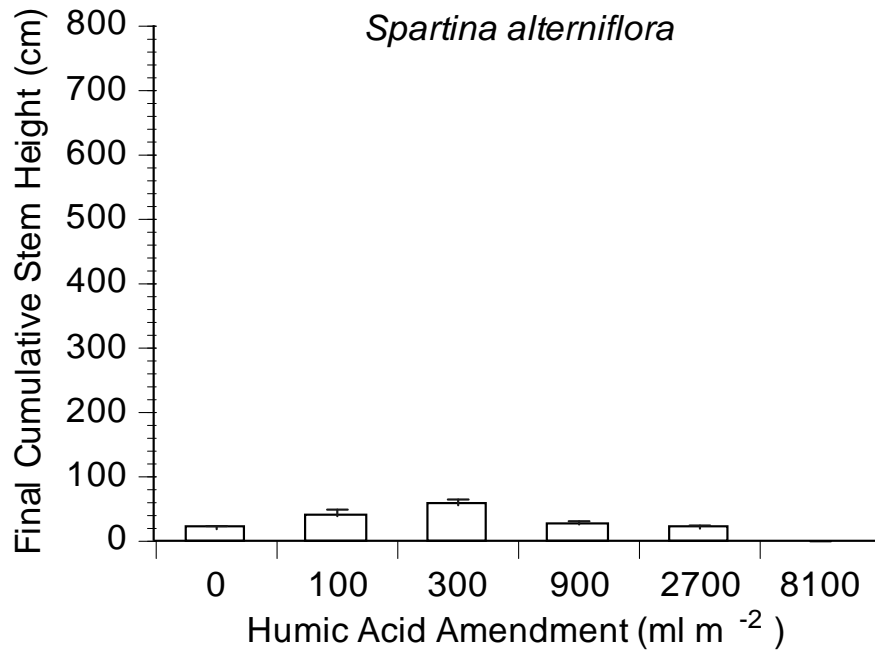


Figure 1.4 The effect of humic acid amendment on final cumulative stem height for *S. alterniflora* (top panel; mean \pm SE, $n = 5$, LSD = 133.80) and *A. germinans* (bottom panel; mean \pm SE, $n = 5$, LSD = 133.80).

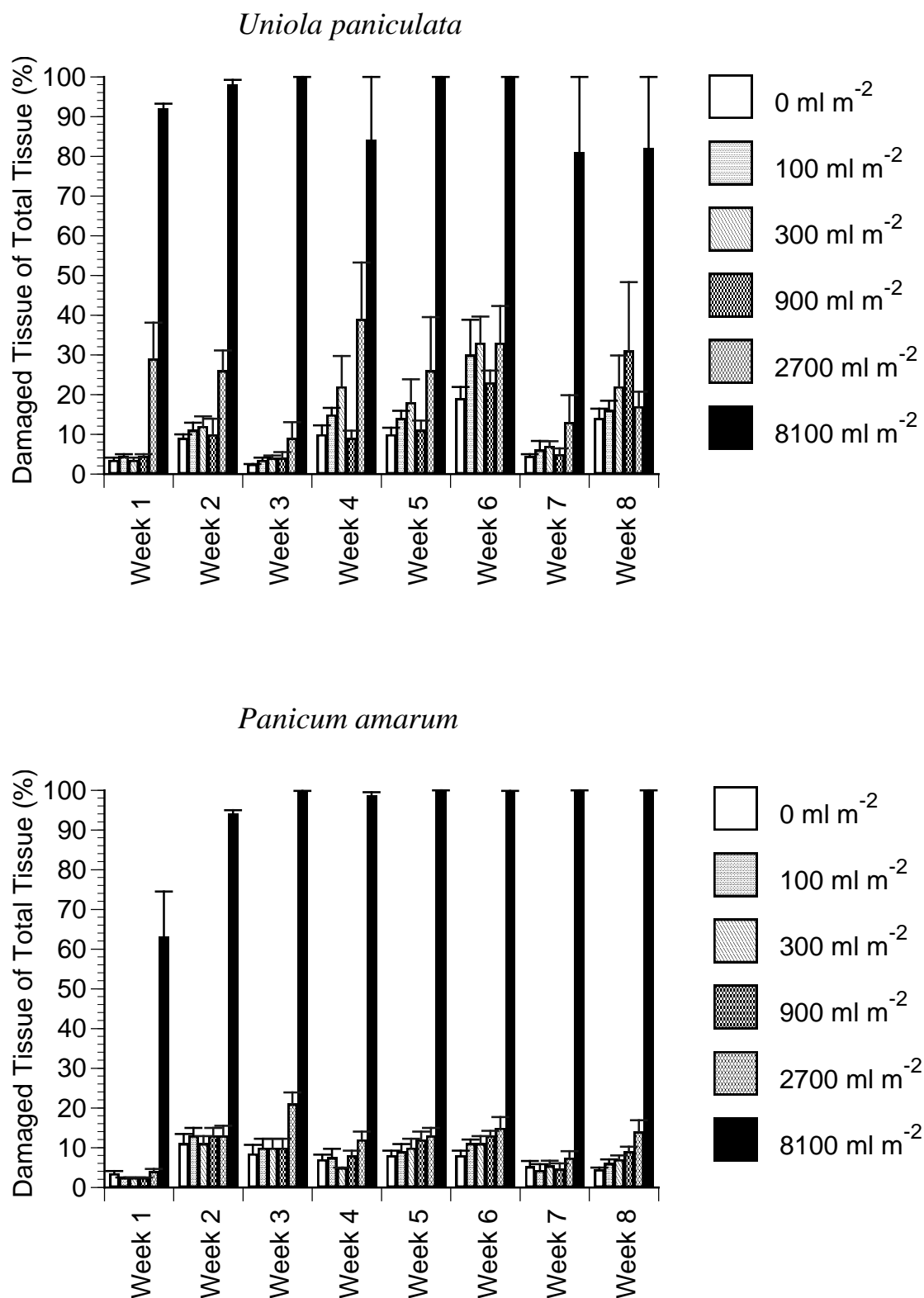
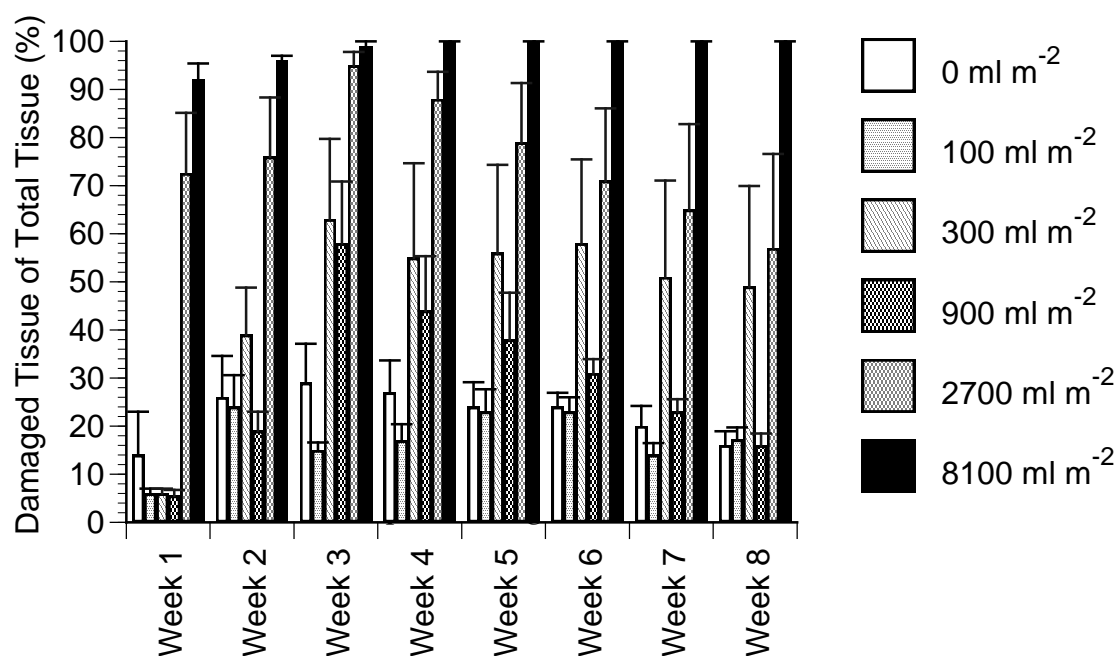


Figure 1.5 The effect of time and humic acid amendment on visually observed damaged tissue as a percentage of total tissue for *U. paniculata* (top panel; mean \pm SE, $n = 5$) and *P. amarum* (bottom panel; mean \pm SE, $n = 5$).

Distichlis spicata



Paspalum vaginatum

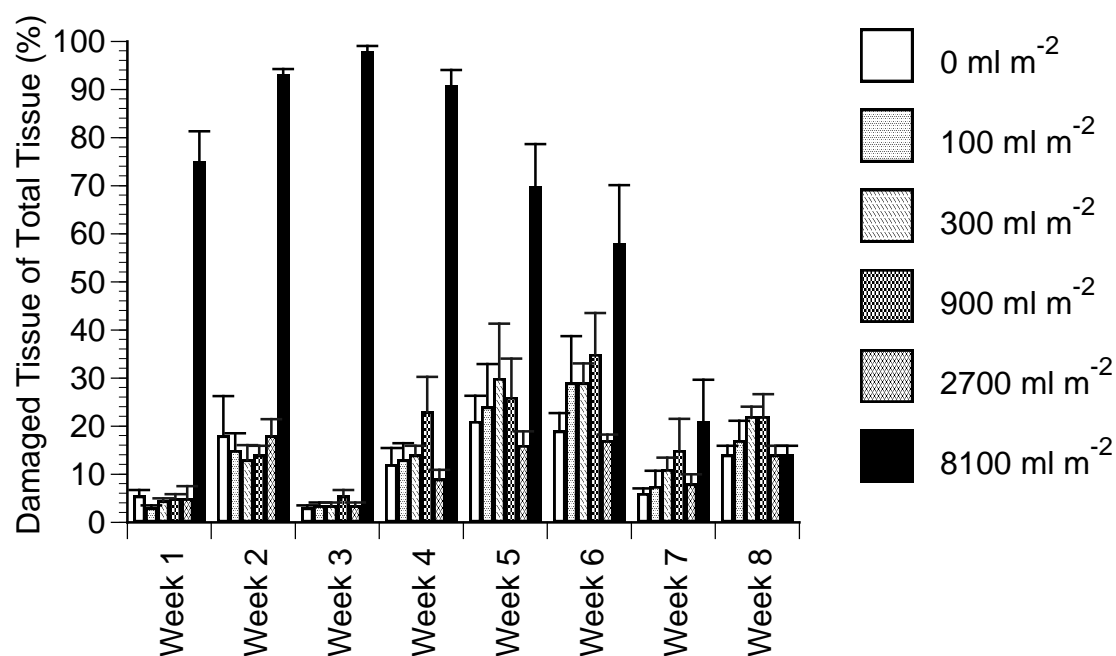
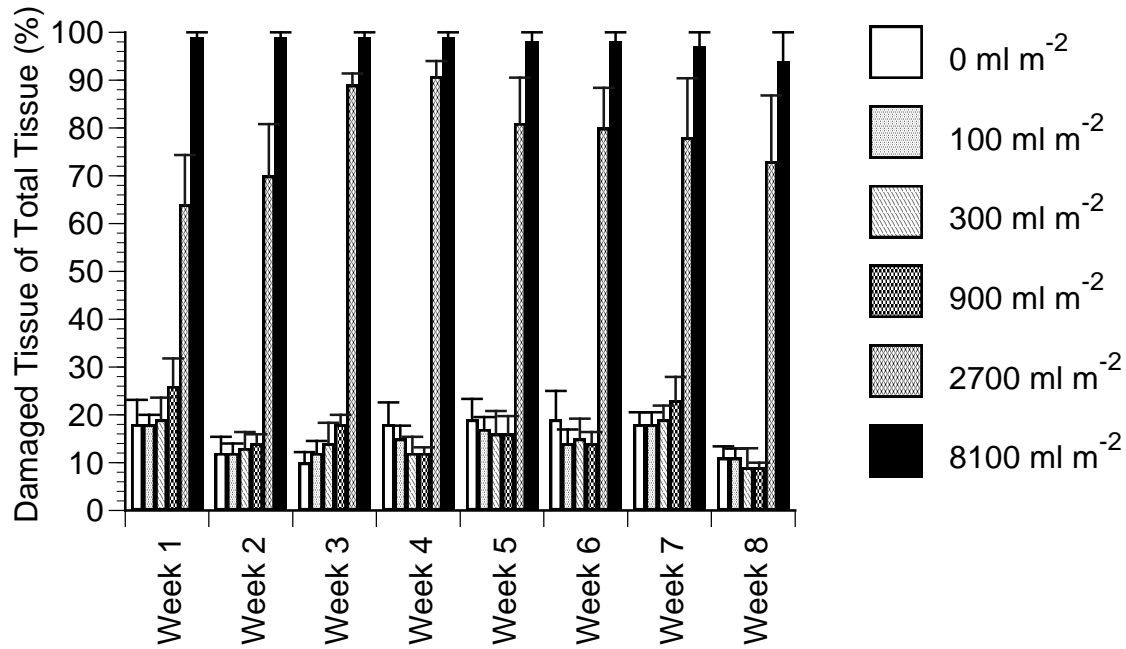


Figure 1.6. The effect of time and humic acid amendment on visually observed damaged tissue as a percentage of total tissue for *D. spicata* (top panel; mean \pm SE, n = 5) and *P. vaginatum* (bottom panel; mean \pm SE, n = 5).

Spartina patens



Baccharis halimifolia

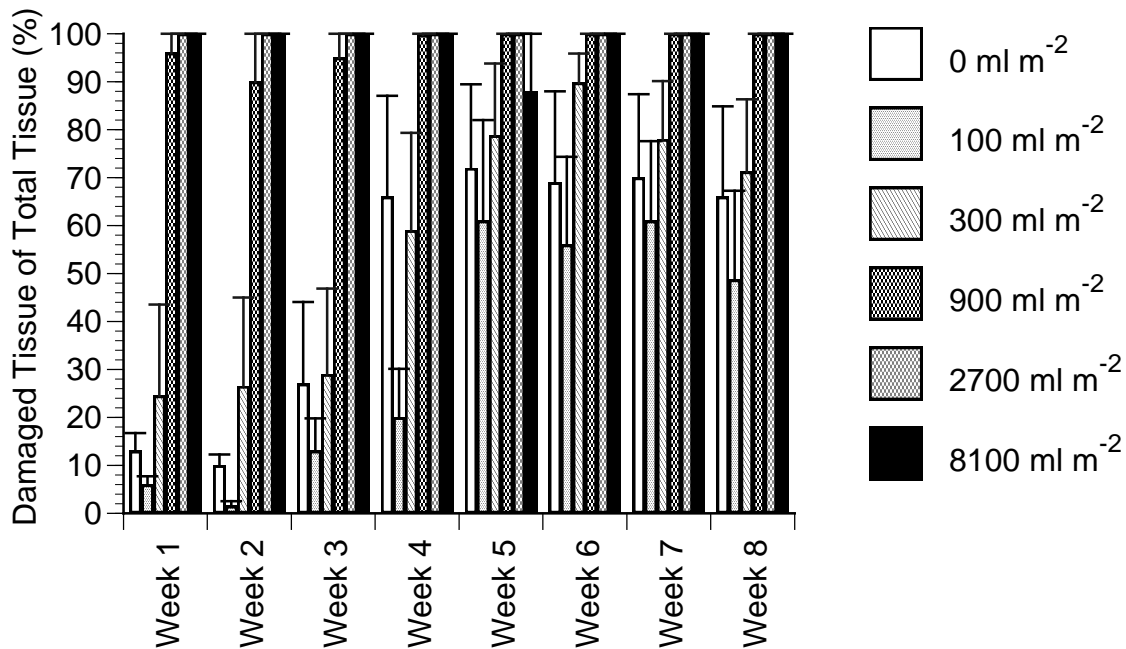
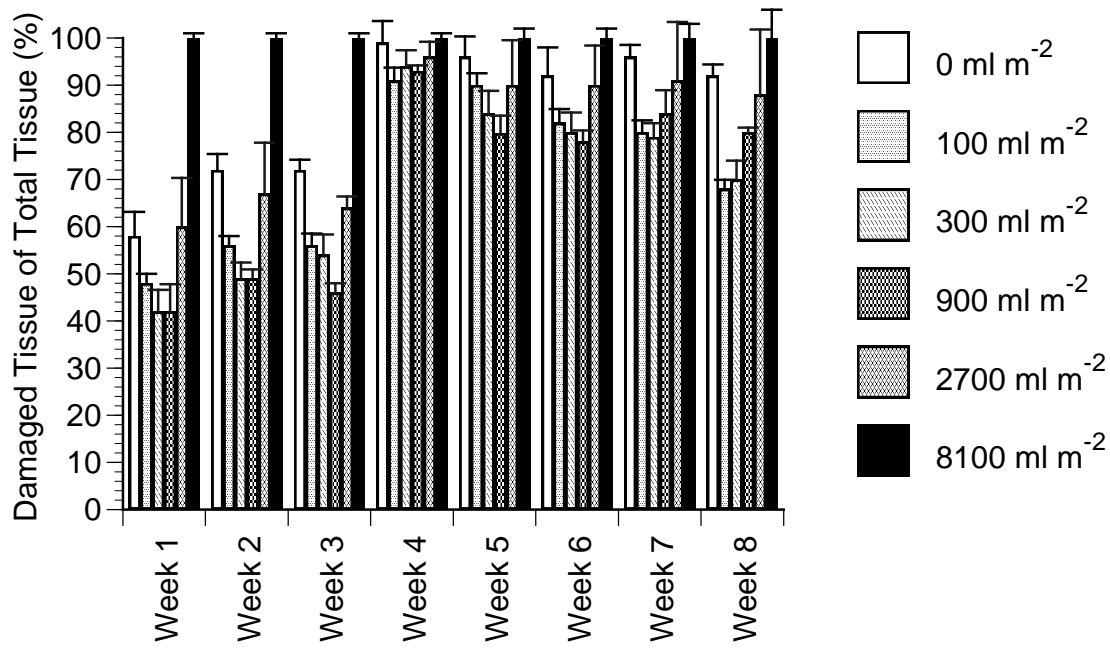


Figure 1.7 The effect of time and humic acid amendment on visually observed damaged tissue as a percentage of total tissue for *S. patens* (top panel; mean \pm SE, $n = 5$) and *B. halimifolia* (bottom panel; mean \pm SE, $n = 5$).

Spartina alterniflora



Avicennia germinans

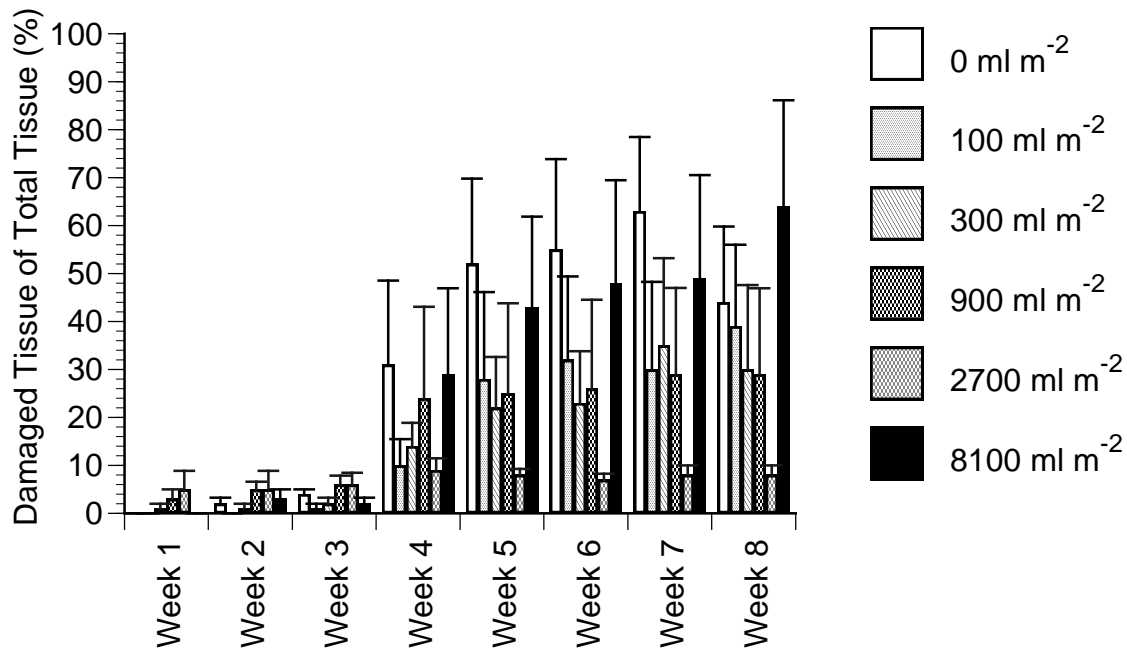


Figure 1.8 The effect of time and humic acid amendment on visually observed damaged tissue as a percentage of total tissue for *S. alterniflora* (top panel; mean \pm SE, n = 5) and *A. germinans* (bottom panel; mean \pm SE, n = 5).

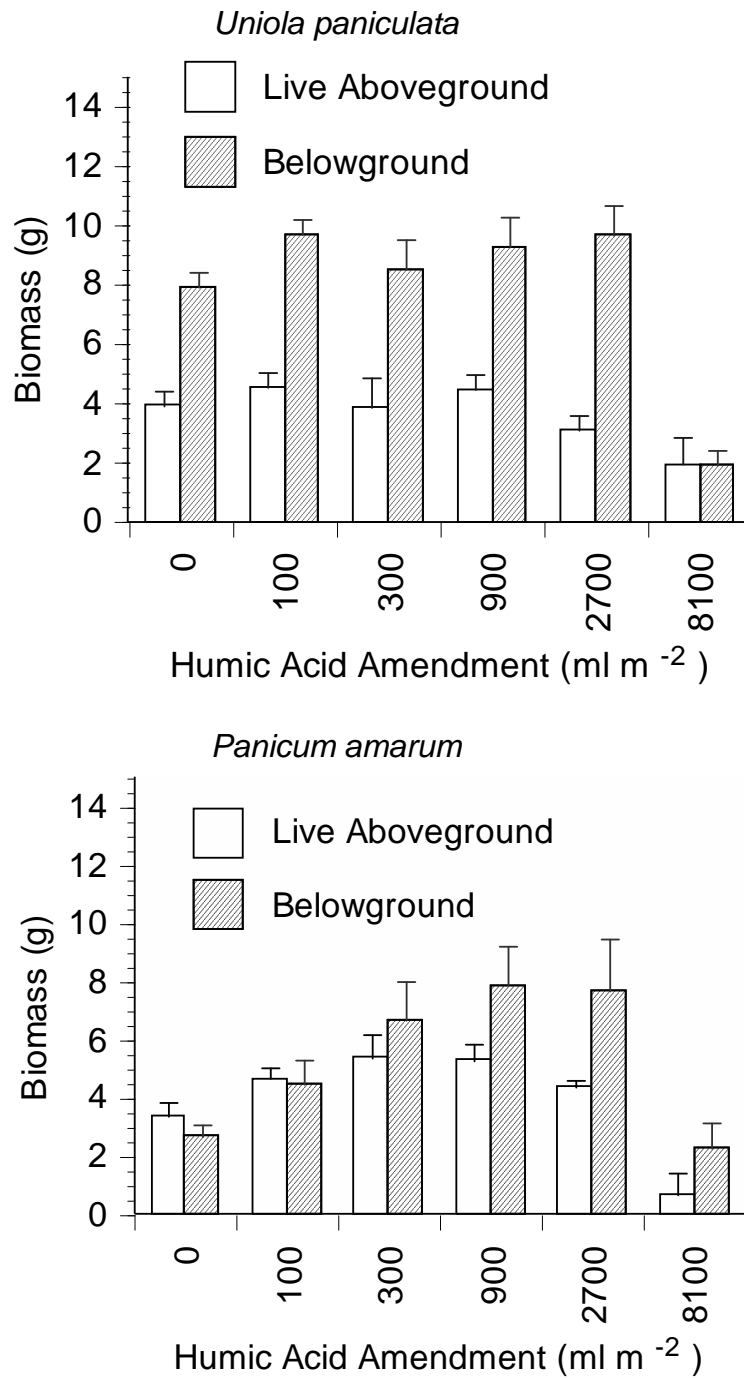


Figure 1.9 The effect of humic acid amendment on aboveground live and belowground biomass for *U. paniculata* (top panel; mean \pm SE, $n = 5$) and *P. amarum* (bottom panel; mean \pm SE, $n = 5$). Live aboveground LSD = 1.6358, Belowground LSD = 8.003

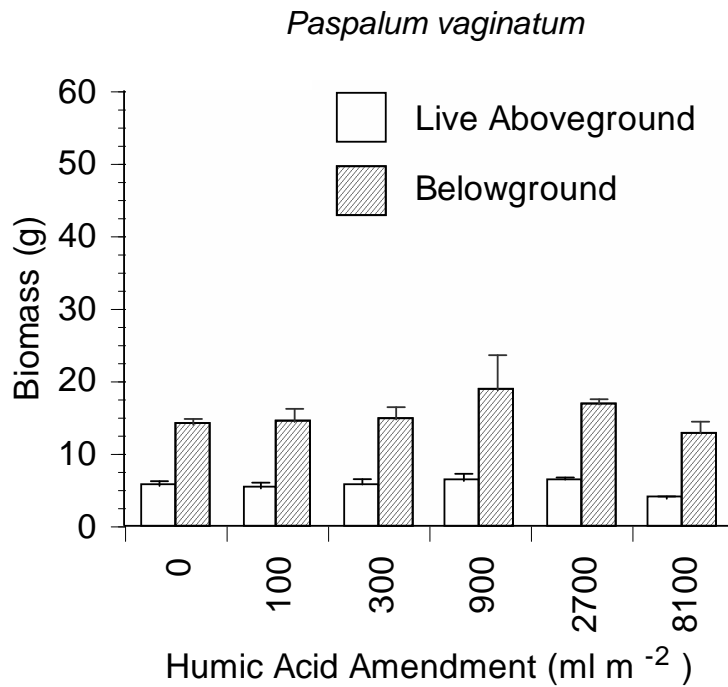
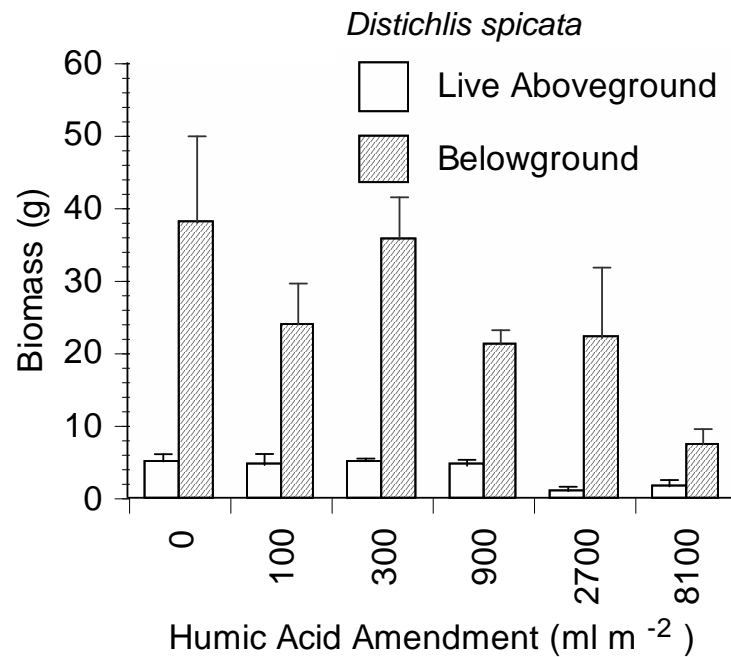


Figure. 1.10 The effect of humic acid amendment on aboveground live and belowground biomass for *D. spicata* (top panel; mean \pm SE, $n = 5$) and *P. vaginatum* (bottom panel; mean \pm SE, $n = 5$). Live aboveground LSD = 1.6358, Belowground LSD = 8.003

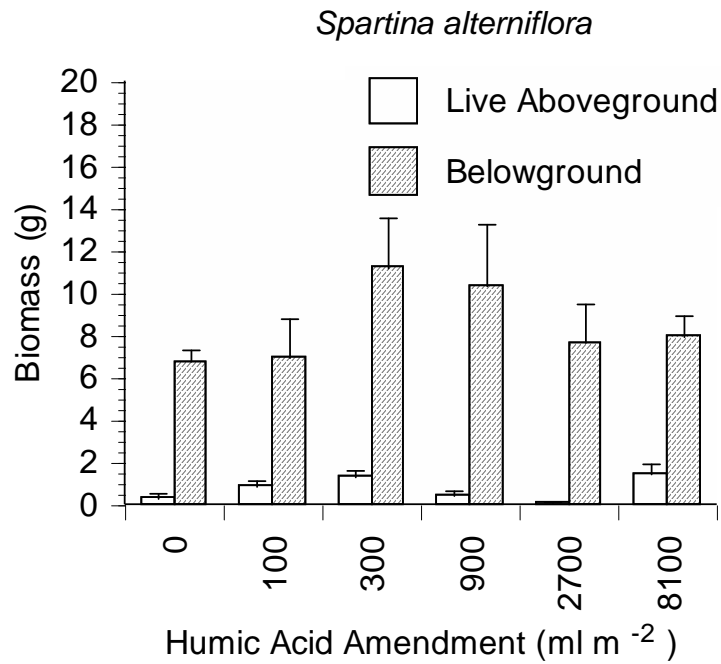
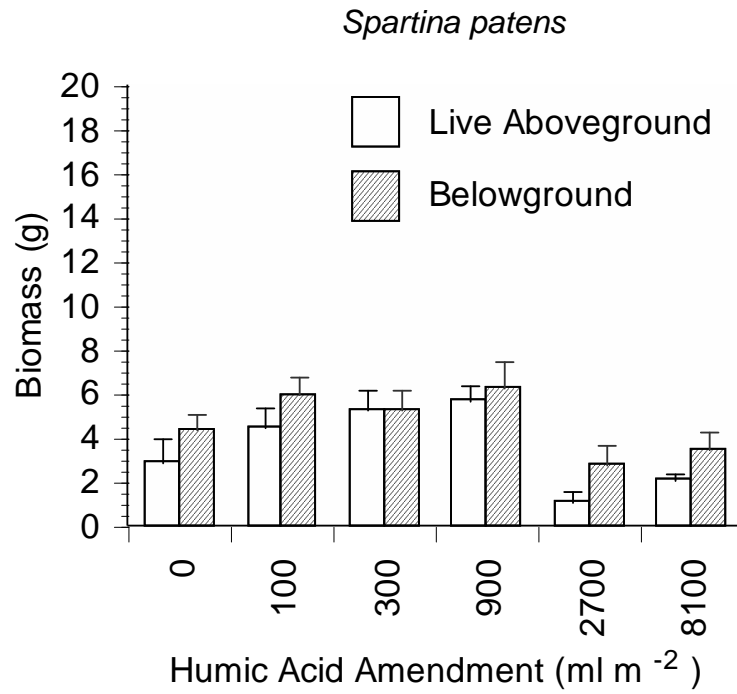


Figure 1.11 The effect of humic acid amendment on aboveground live and belowground biomass for *S. patens* (top panel; mean \pm SE, $n = 5$) and *S. alterniflora* (bottom panel; mean \pm SE, $n = 5$). Live aboveground LSD = 1.6358, Belowground LSD = 8.003

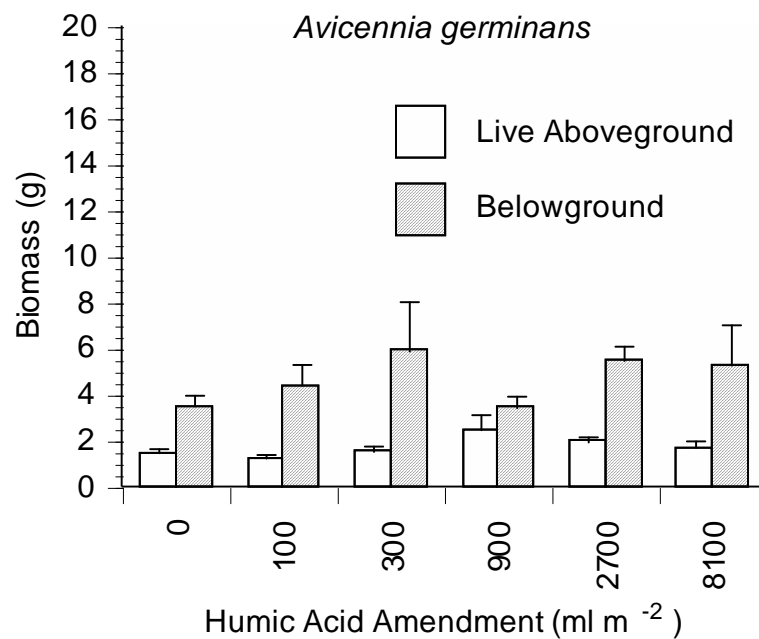
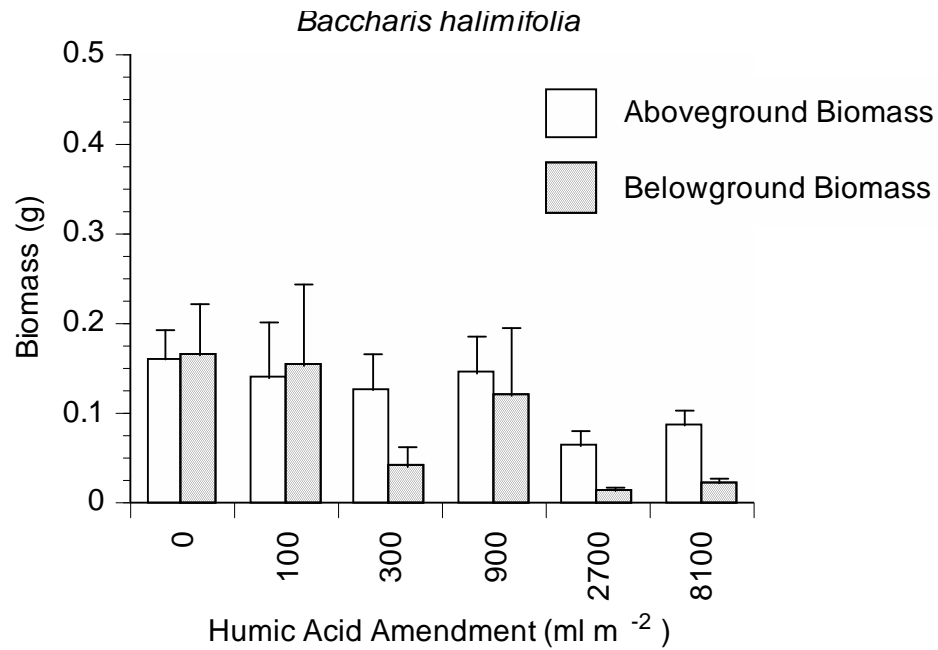


Figure 1.12 The effect of humic acid amendment on aboveground live and belowground biomass for *B. halimifolia* (top panel; mean \pm SE, $n = 5$) and *A. germinans* (bottom panel; mean \pm SE, $n = 5$). Live aboveground LSD = 1.6358, Belowground LSD = 8.003

biomass were also significantly greater in the 100 ml m⁻², 300 ml m⁻², and 900 ml m⁻² humic acid addition treatments than the 0 ml m⁻² treatment (Figure 1.11 top panel; Contrast F = 11.9, p < 0.01, Figure 1.11 bottom panel; Contrast F = 11.9, p < 0.01, respectively).

Soil Physico-Chemical Characterization

Uniola paniculata and *Paspalum vaginatum* were significantly (Tables A1.1–A1.6; Contrast F = 99.0, p < 0.01) more acidic than all other dune and swale species, and *A. germinans* was significantly (Tables A1.7 and A1.8; F = 5.7, p < 0.05) more acidic than *S. alterniflora*. The 8100 ml m⁻² humic acid amendment resulted in a significantly more basic soil pH than all other humic acid amendment levels for both the dune and swale species (Tables A1.1–A1.6; Contrast F = 172.9, p < 0.01) and salt marsh species (Tables A1.7 and A1.8; Contrast F = 11.9, p < 0.01). Interestingly, *S. patens* did not demonstrate elevated pH in the 8100 ml m⁻² treatment as was found for all other dune and swale species, leading to an interaction of species and humic acid amendment (Tables A1.1–A1.6; F = 5.3, p < 0.01). Importantly, all pH values fall within a normal pH range for dune (5.86 – 8.62) and salt marsh (7.18 – 8.33) habitats. Soil conductivity was significantly higher (Tables A1.1–A1.6; F = 25.8, p < 0.01) for *D. spicata* than for all other dune and swale species. A highly significant and clear trend of increasing soil conductivity with increasing humic acid amendment was noted (Tables A1.1–A1.8; F = 7.3, p < 0.01), although this trend was stronger in *U. paniculata* and *P. vaginatum* than in other dune and swale species, resulting in a highly significant interaction (F = 2.4, p < 0.01). As with pH, soil conductivity was found to fall within a normal range for dune and swale species (40 – 1079 $\mu\text{S cm}^{-1}$).

Baccharis halimifolia had significantly greater soil moisture than all other species, likely due to its slower growth (Tables A1.1–A1.6; Contrast F = 154.4, p < 0.01). Soil moisture was significantly greater in the 8100 ml m⁻² humic acid amendment treatment (Tables A1.1–A1.6; Contrast F = 86.5, p < 0.01), possibly as a consequence of the low plant growth in this treatment. *Uniola paniculata* and *B. halimifolia* in 8100 ml m⁻² humic acid amendment treatment had significantly greater soil moisture than other dune and swale species, resulting in a significant interaction (Tables A1.1–A1.6; F = 4.0, p < 0.01). No significant effect of species or humic acid amendment, or interaction thereof, was found for soil organic matter for the dune and swale species. A trend toward decreased soil nitrate-nitrite with humic acid amendment was found (Tables A1.1–A1.6; F = 3.2, p < 0.01) for dune and swale plants. Interestingly, no effect on humic acid amendment was detected for soil ammonium, but *S. patens* had significantly greater soil ammonium than other dune and swale species (Tables A1.1–A1.6; F = 59.1, p < 0.01). No significant effect of species, humic acid amendment, or interaction thereof, was found for soil phosphorus for the dune and swale species.

Avicennia germinans displayed slightly greater soil moisture than *S. alterniflora* (Tables A1.3 and A1.4; F = 46.2, p < 0.01), and as with the dune and swale species, the 8100 ml m⁻² humic acid amendment treatment had significantly greater soil moisture (Tables A1.7 and A1.8; F = 11.4, p < 0.01). No significant effect of species or humic acid amendment, or interaction thereof, was found for soil organic matter for the salt marsh species. No significant effect of species or humic acid amendment, or interaction thereof, was detected regarding conductivity for the salt marsh species. No significant effects of species, humic acid amendment, or interaction thereof, on soil nitrate-nitrite, ammonium, or phosphorus was found for the salt marsh species.

Results: Refinement Study

Final Cumulative Stem Height

Highly significant effects of species, fertilizer, and the interaction thereof were detected in the refinement study (Figures 1.13–1.16; $F = 284.8$, $p < 0.01$, $F = 225.9$, $p < 0.01$, and $F = 24.6$, $p < 0.01$, respectively). Fertilizer application benefited all species, but *S. patens* and *D. spicata* performed exceptionally well under this condition (Figure 1.14 top panel and Figure 1.15 top panel). Interestingly, an interaction of humic acid amendment with fertilizer level was detected in which final stem height was greater in the high fertilizer level for the 0, 125, and 250 ml m⁻² humic acid amendment levels, but was similar for the low and high fertilizer in 500 ml m⁻².

Biomass Partitioning

A highly significant effect of species on live aboveground biomass was detected in which *P. amarum* and *S. patens* had much greater live aboveground biomass than all other species (Figures 1.17–1.24; Contrast $F = 441.9$, $p < 0.01$). Also, a highly significant effect of fertilizer regime on live aboveground biomass was detected (Figures 1.17–1.24; $F = 274.4$, $p < 0.01$), with almost twice the live aboveground biomass produced by the high fertilizer treatment as the low fertilizer treatment. *Baccharis halimifolia* did not demonstrate enhanced live aboveground biomass with the high fertilization treatment, whereas all other species did, leading to a significant fertilizer regime by species interaction (Figures 1.17–1.24; $F = 23.9$, $p < 0.01$). Interestingly, a highly significant effect of species on belowground biomass (Figures 1.17–1.24; Contrast $F = 244.0$, $p < 0.01$) was driven by the substantial belowground biomass production of *D. spicata* and *S. patens*. As with live aboveground biomass, the high fertilizer regime had a highly significant effect, again almost doubling biomass production compared to the low fertilizer regime (Figures 1.17–1.24; $F = 67.1$, $p < 0.01$). A significant interaction of species and fertilizer regime was again noted (Figures 1.17–1.24; $F = 3.5$, $p < 0.01$), driven by biomass production not differing between high and low fertilized treatments for *B. halimifolia*.

Leaf Tissue Chemistry

A significant effect of species (Tables A1.9–A1.12; $F = 55.0$, $p < 0.01$) and a significant interaction of species and fertilizer were detected for leaf nitrogen content (Tables A1.9–A1.12; $F = 7.5$, $p < 0.01$), both resulting from the high leaf nitrogen content of *D. spicata* under high fertilizer conditions. No other significant main effects or interactions were detected in regard to leaf nitrogen, leaf phosphorus, or potassium.

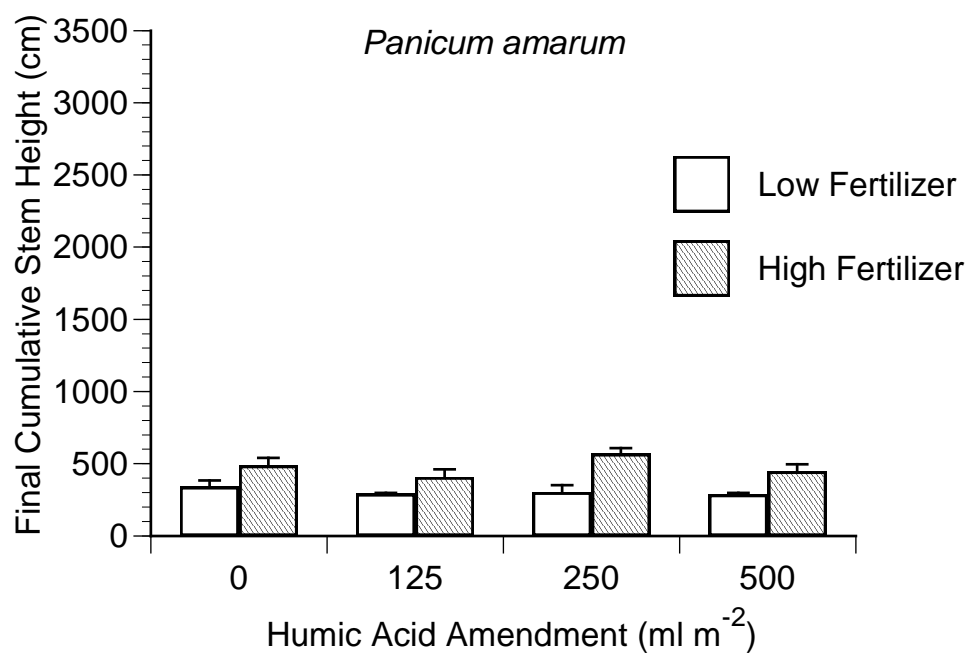
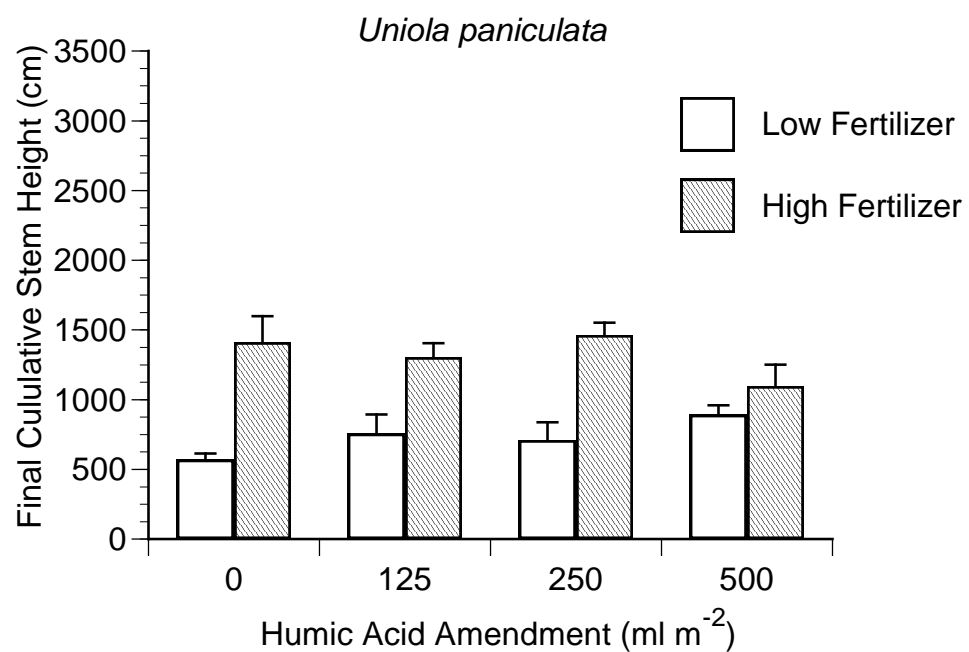


Figure 1.13 The effect of humic acid amendment and nutrient status on final stem height of *U. paniculata* (top panel; mean \pm SE, $n=4$, LSD = 333.2) and *P. amarum* (bottom panel; mean \pm SE, $n=4$, LSD = 333.2).

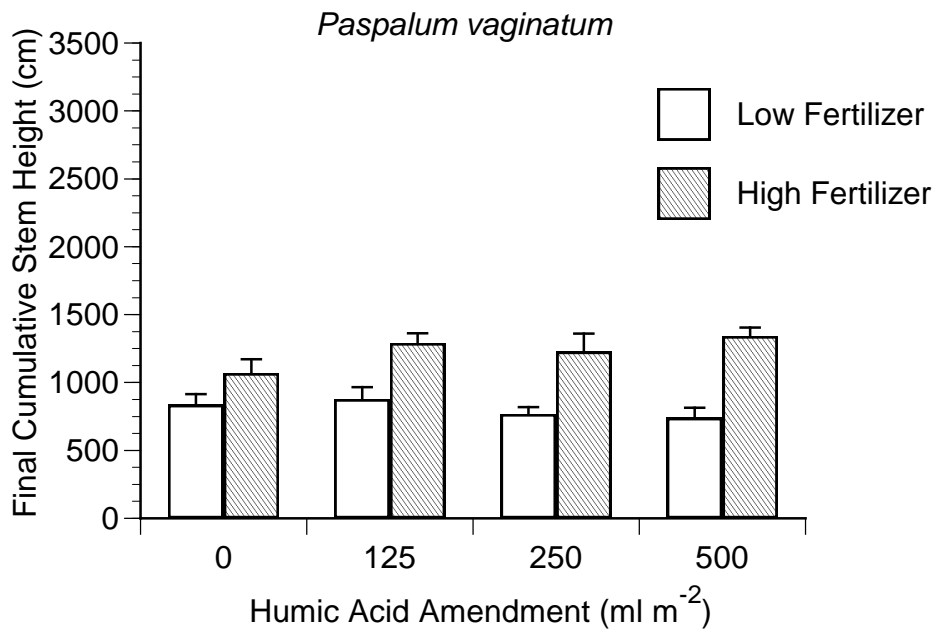
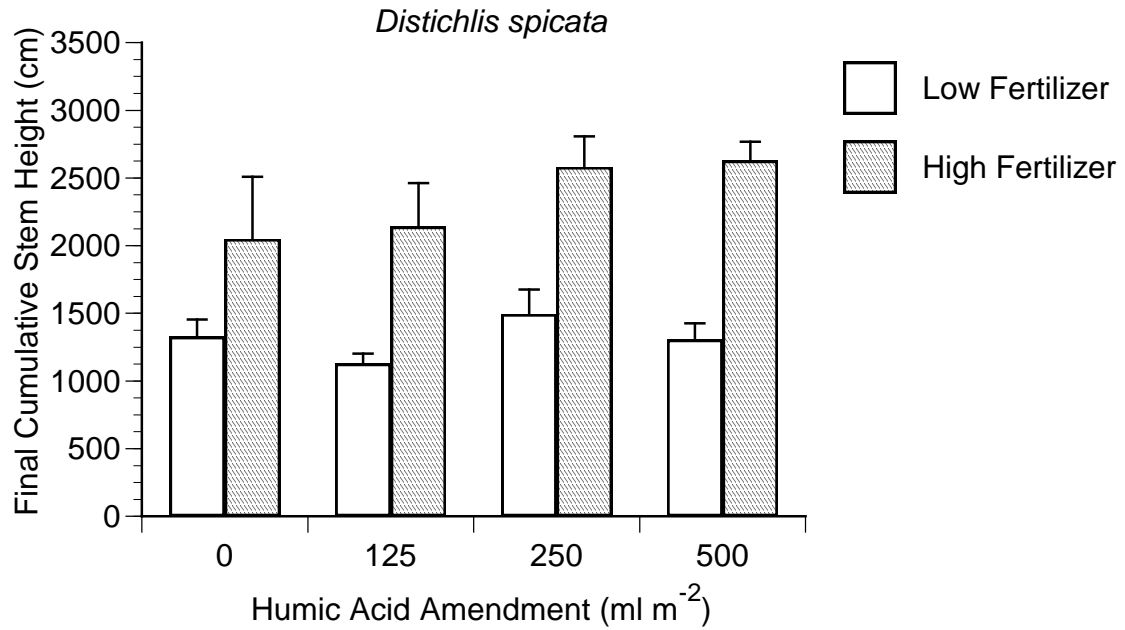


Figure 1.14 The effect of humic acid amendment and nutrient status on final stem height of *D. spicata* (top panel; mean \pm SE, $n=4$, LSD = 333.2) and *P. vaginatum* (bottom panel; mean \pm SE, $n=4$, LSD = 333.2).

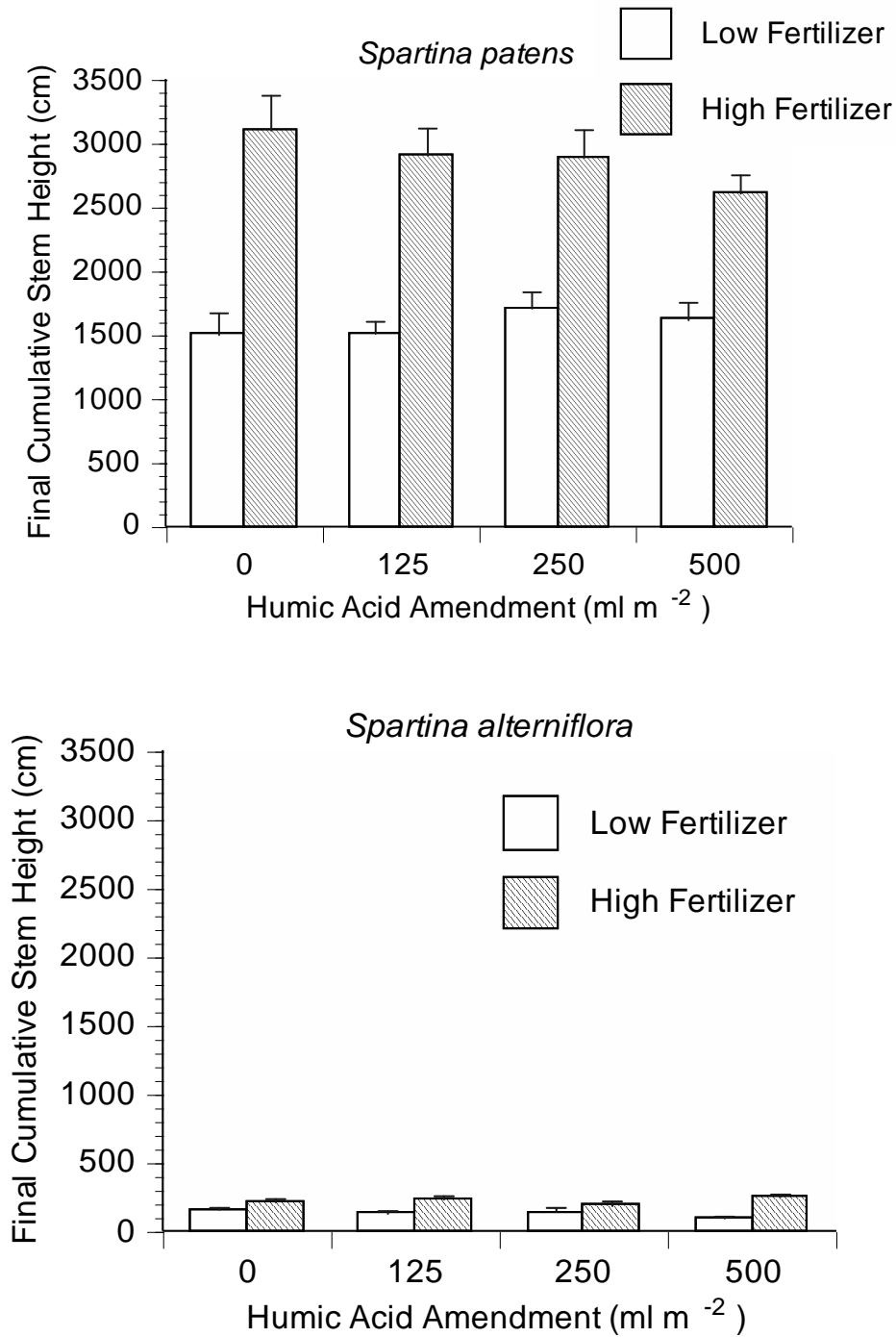


Figure 1.15 The effect of humic acid amendment and nutrient status on final stem height of *S. patens* (top panel; mean \pm SE, $n=4$, LSD = 333.2) and *S. alterniflora* (bottom panel; mean \pm SE, $n=4$, LSD = 333.2).

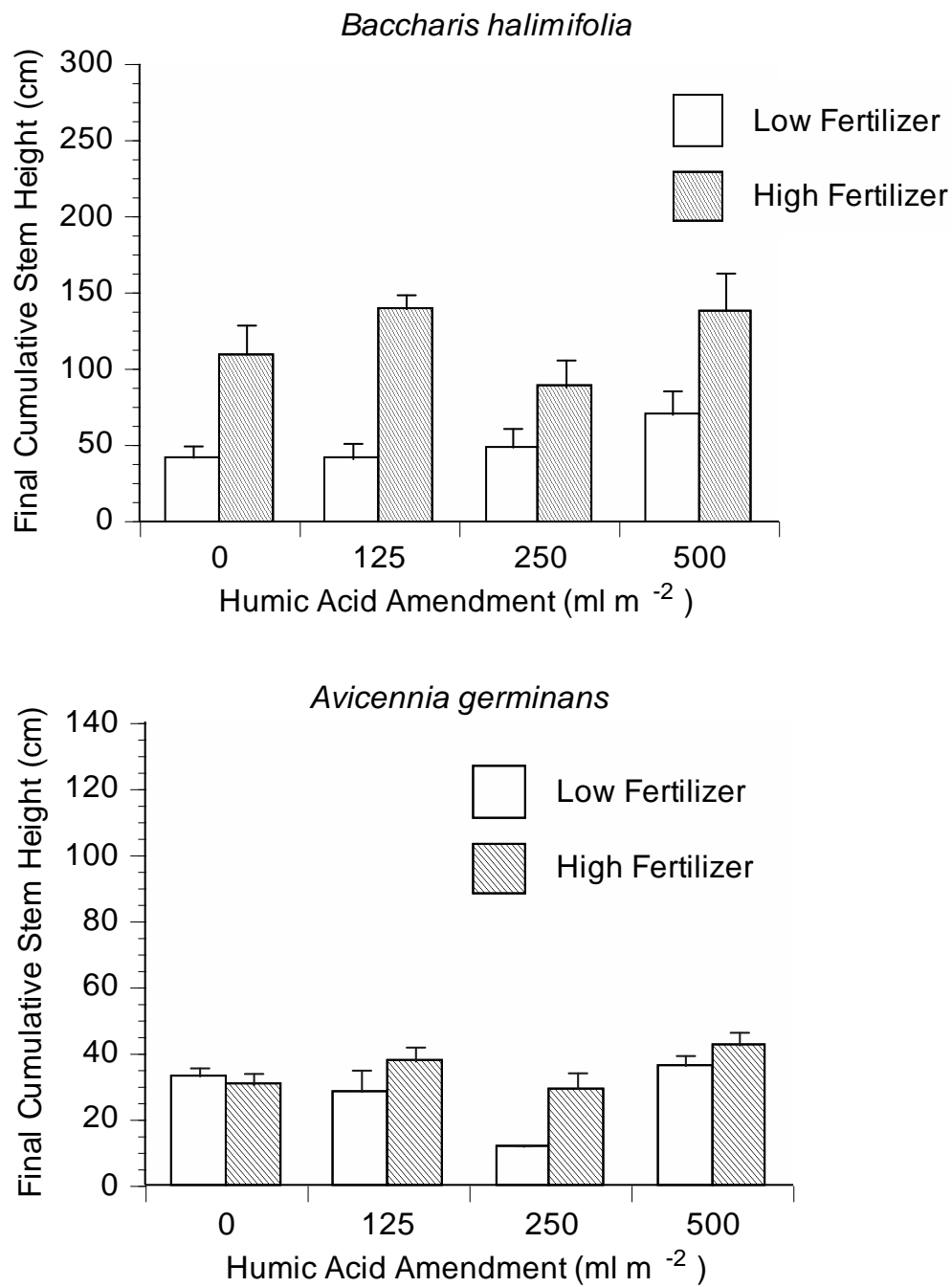


Figure 1.16 The effect of humic acid amendment and nutrient status on final cumulative height of *B. halimifolia* (top panel; mean \pm SE, $n=4$, LSD = 333.2) and *A. germinans* (bottom panel; mean \pm SE, $n=4$, LSD = 333.2).

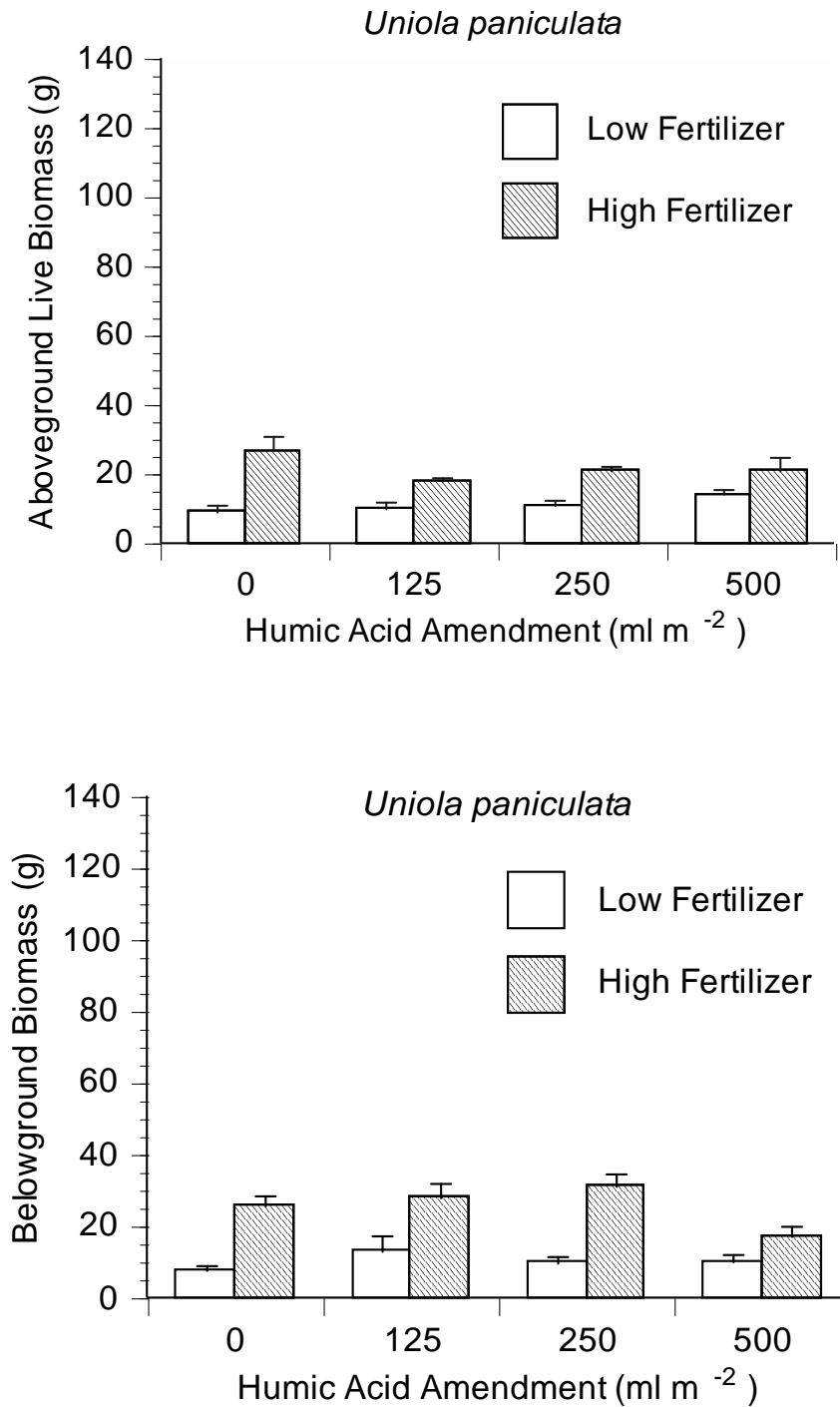


Figure 1.17 The effect of humic acid amendment and nutrient status on *U. paniculata* aboveground biomass (top panel; mean \pm SE, $n=4$, LSD = 4.614) and belowground biomass (bottom panel; mean \pm SE, $n=4$, LSD = 18.169).

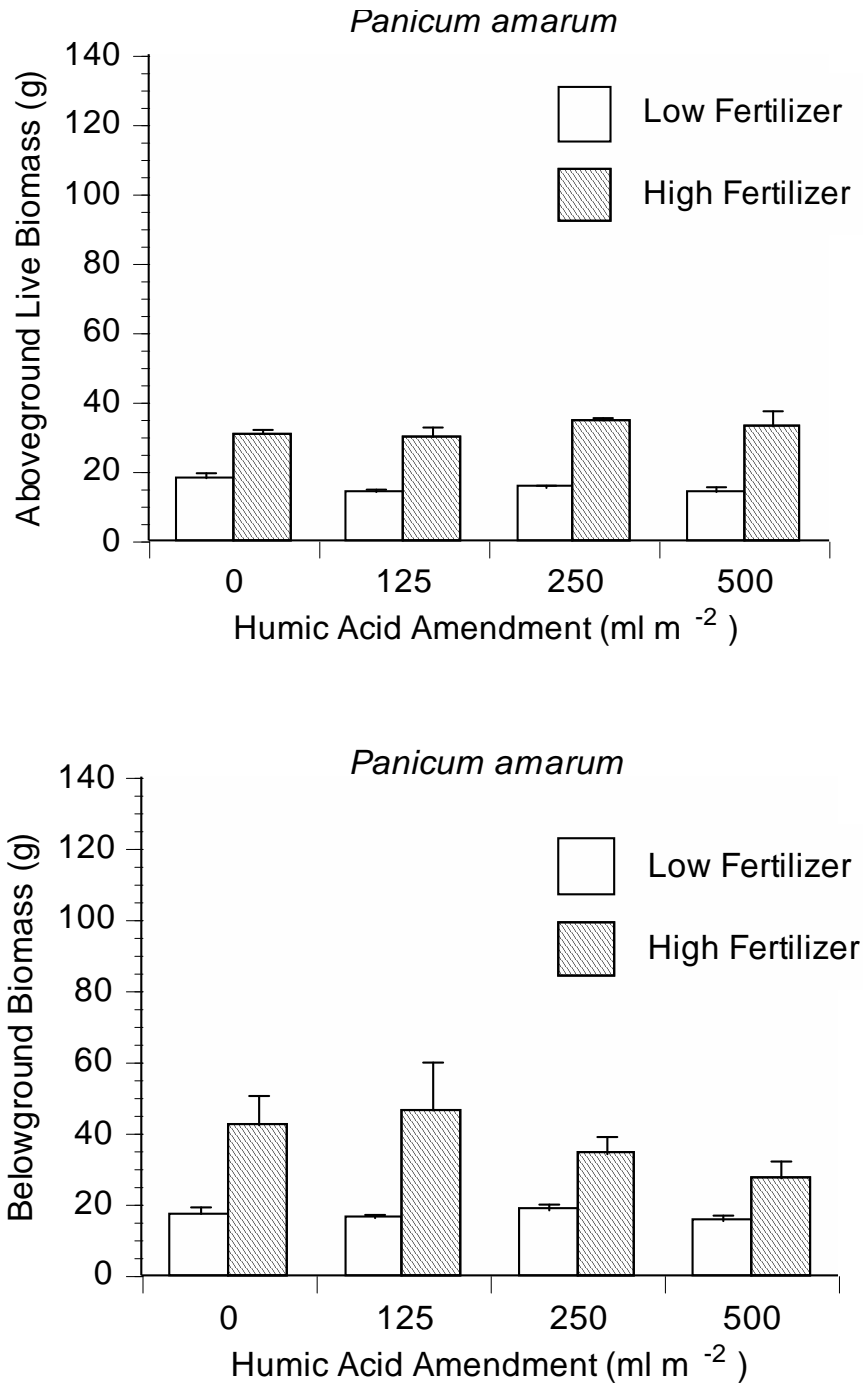


Figure. 1.18 The effect of humic acid amendment and nutrient status on *P. amarum* aboveground biomass (top panel; mean \pm SE, $n=4$, LSD = 4.614) and belowground biomass (bottom panel; mean \pm SE, $n=4$, LSD = 18.169).

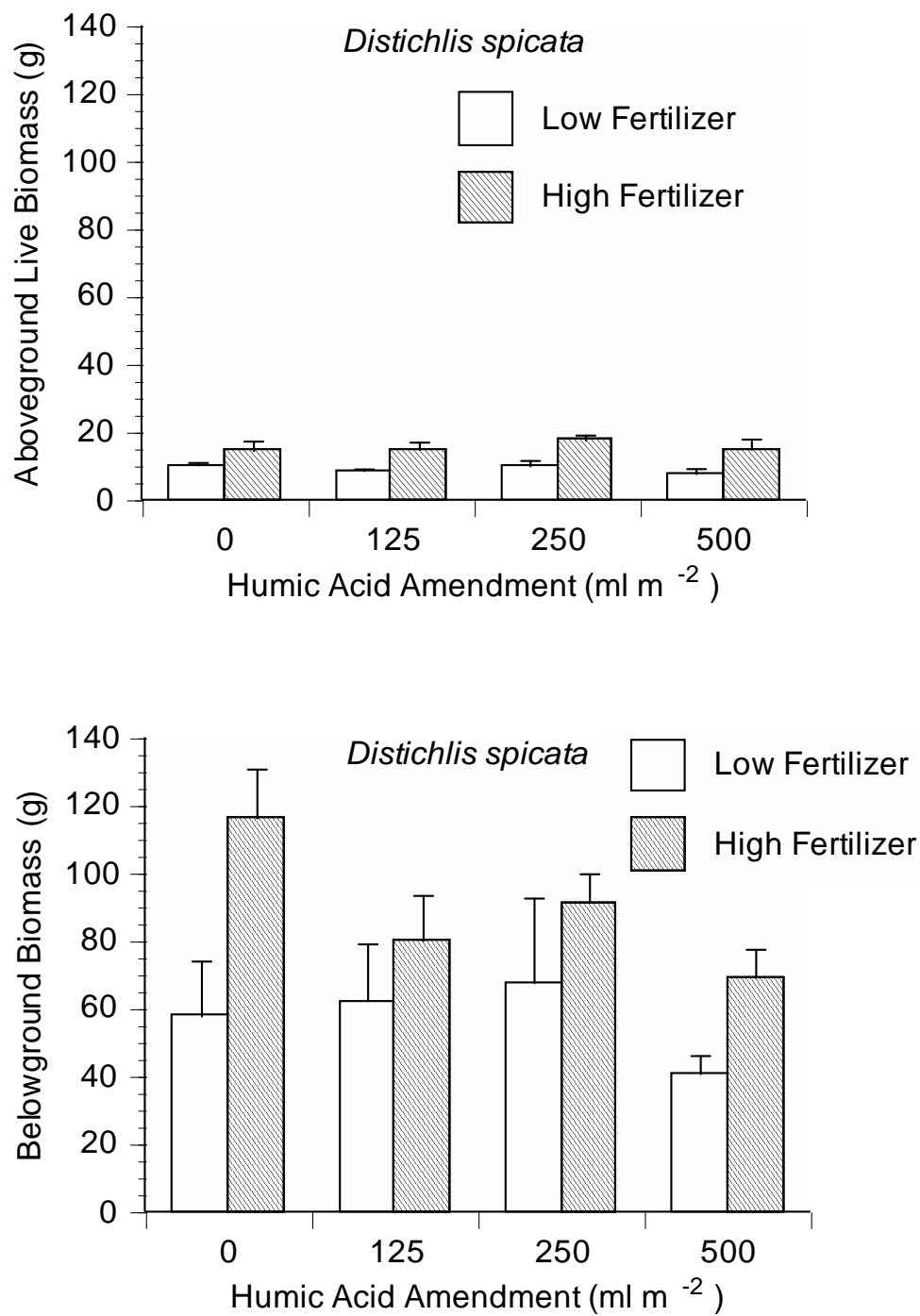


Figure 1.19 The effect of humic acid amendment and nutrient status on *D. spicata* aboveground biomass (top panel; mean \pm SE, $n=4$, LSD = 4.614) and belowground biomass (bottom panel; mean \pm SE, $n=4$, LSD = 18.169).

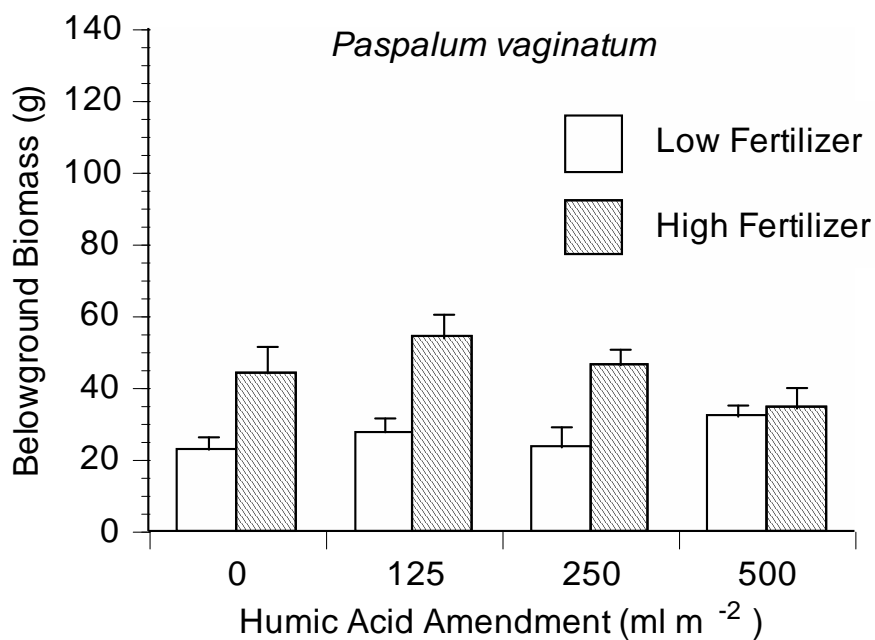
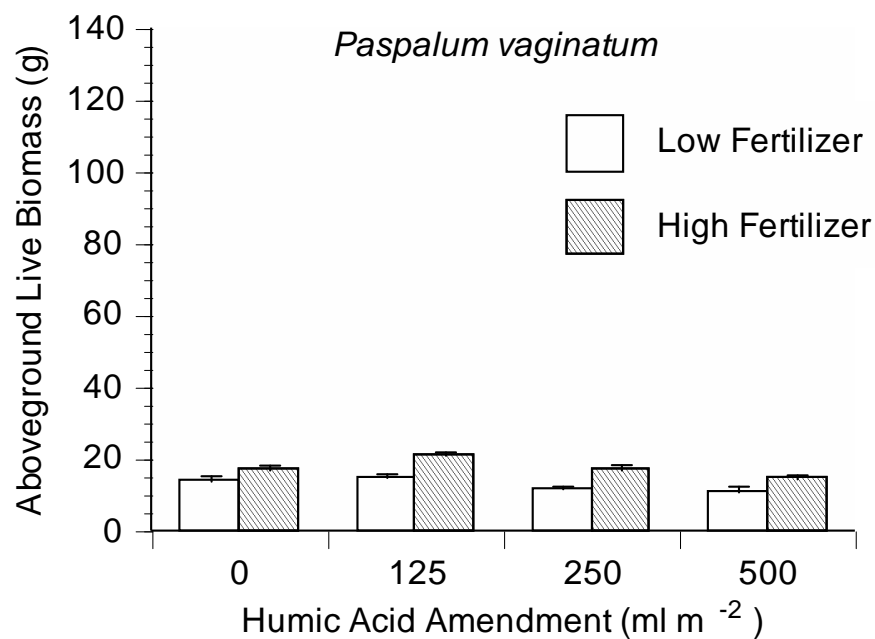


Figure 1.20 The effect of humic acid amendment and nutrient status on *P. vaginatum* aboveground biomass (top panel; mean \pm SE, $n=4$, LSD = 4.614) and belowground biomass (bottom panel; mean \pm SE, $n=4$, LSD = 18.169).

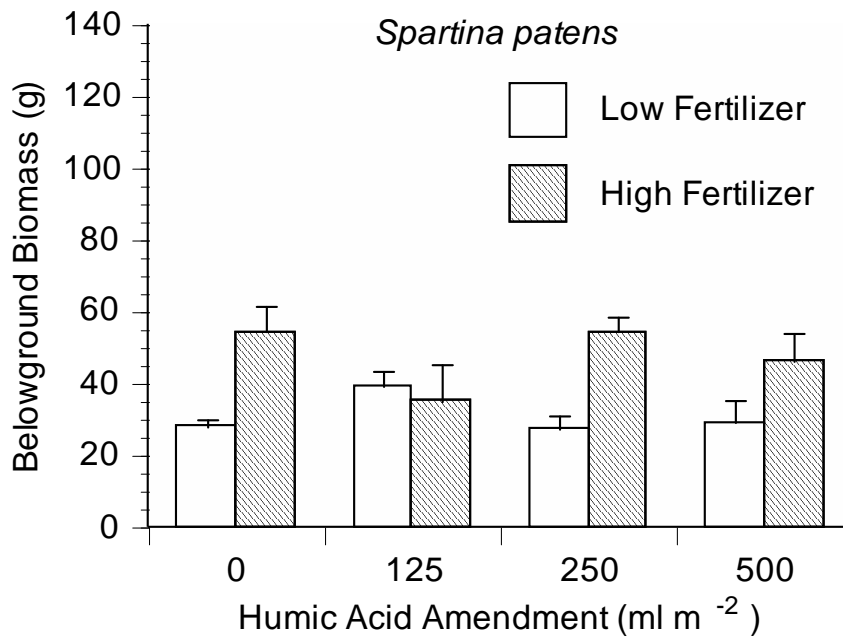
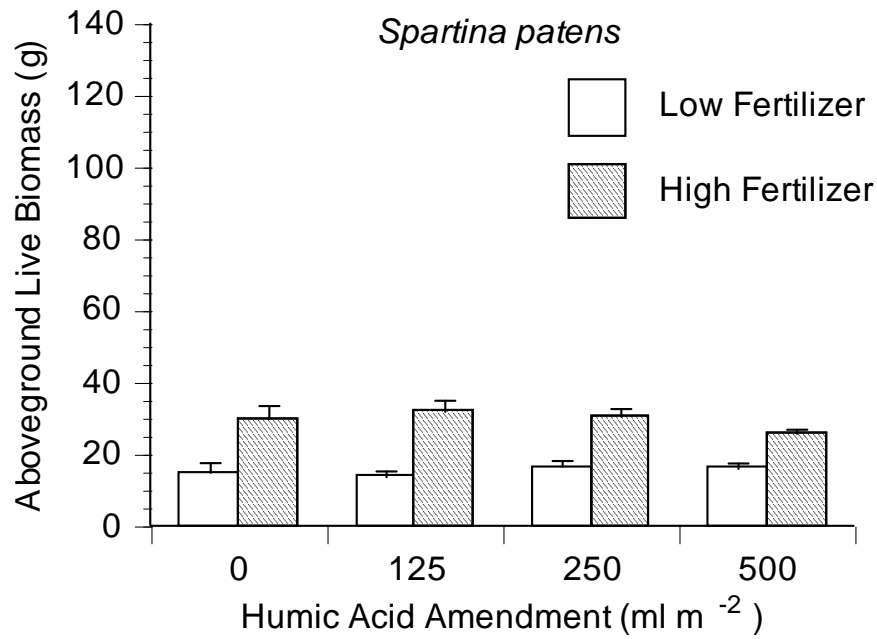


Figure 1.21 The effect of humic acid amendment and nutrient status on *S. patens* aboveground biomass (top panel; mean \pm SE, $n=4$, LSD = 4.614) and belowground biomass (bottom panel; mean \pm SE, $n=4$, LSD = 18.169).

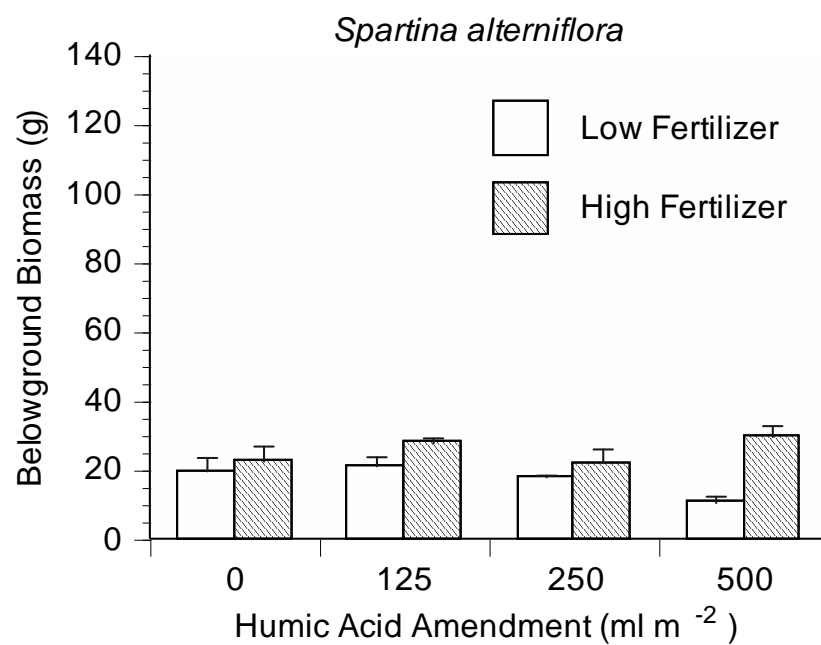
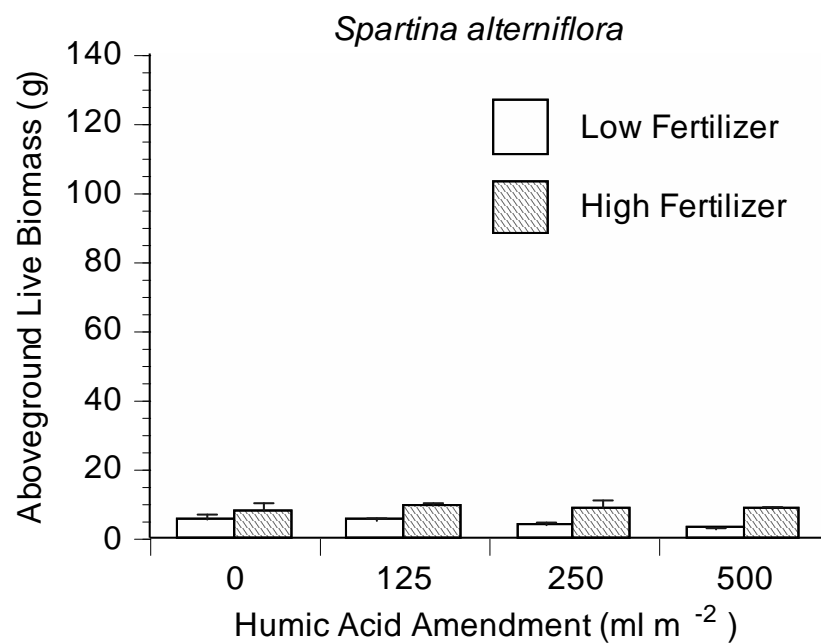


Figure 1.22. The effect of humic acid amendment and nutrient status on *S. alterniflora* aboveground biomass (top panel; mean \pm SE, $n=4$, LSD = 4.614) and belowground biomass (bottom panel; mean \pm SE, LSD = 18.169).

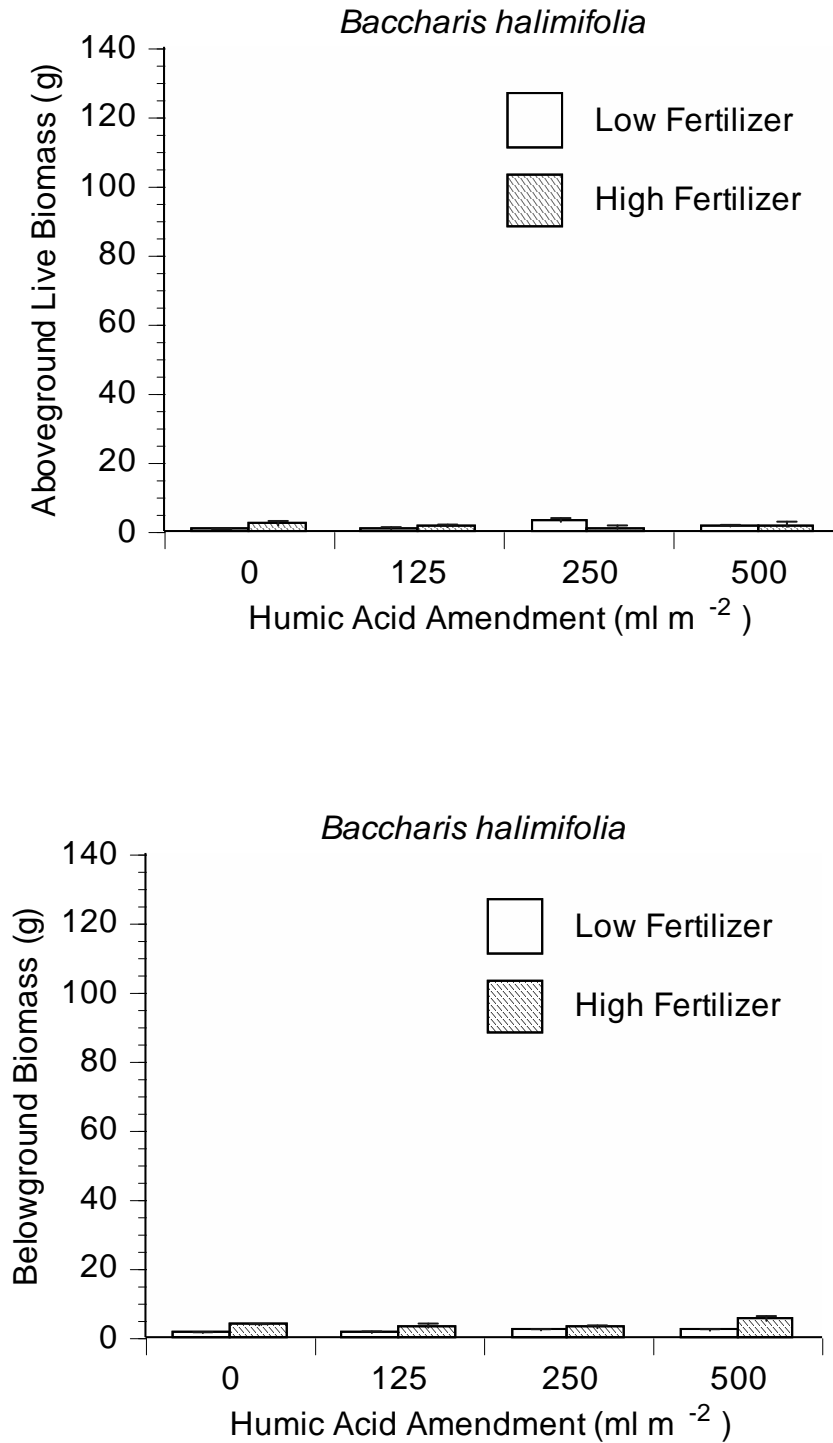


Figure 1.23 The effect of humic acid amendment and nutrient status on *B. halimifolia* aboveground biomass (top panel; mean \pm SE, $n=4$, LSD = 4.614) and belowground biomass (bottom panel; mean \pm SE, $n=4$, LSD = 18.169).

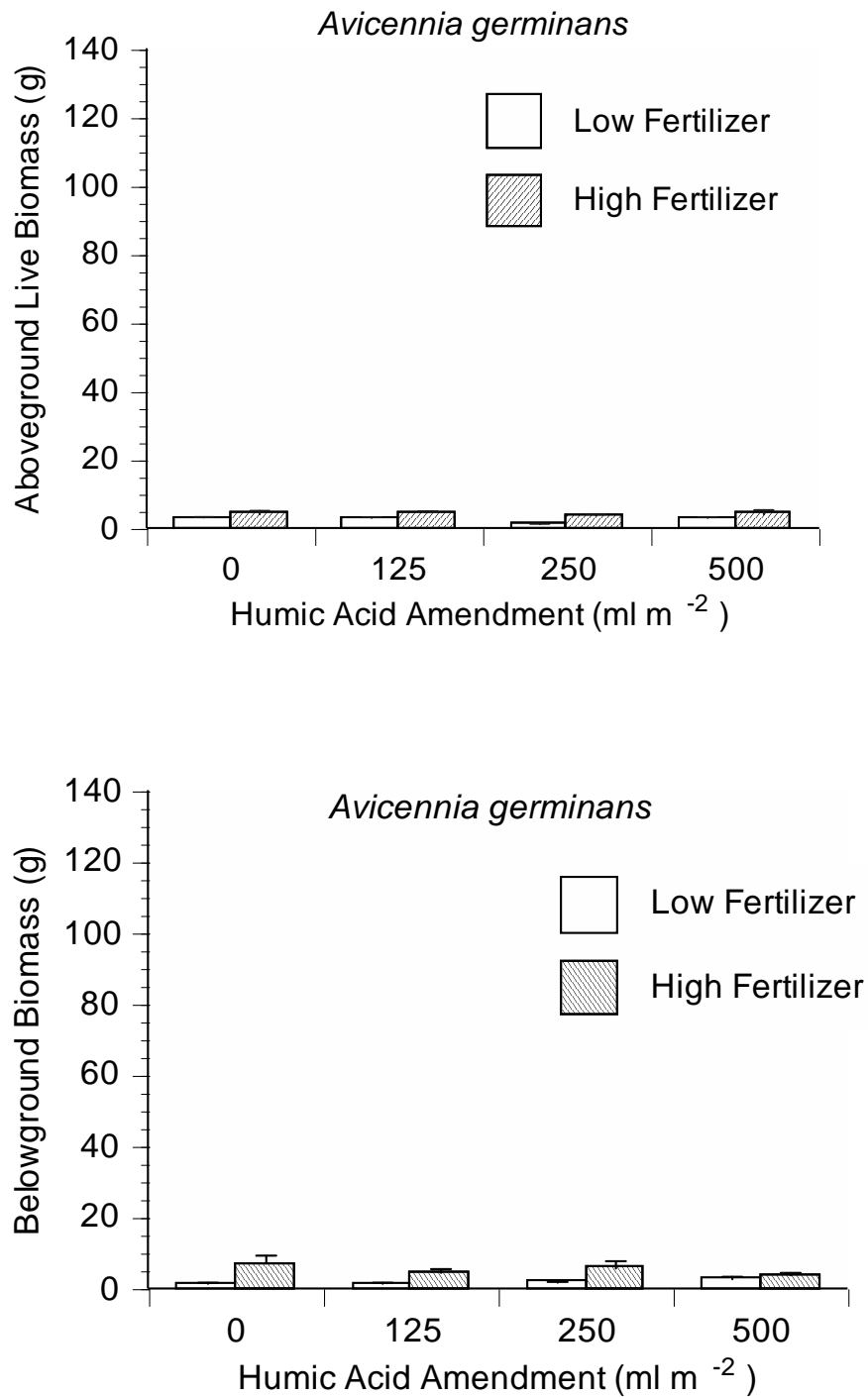


Figure 1.24. The effect of humic acid amendment and nutrient status on *A. germinans* aboveground biomass (top panel; mean \pm SE, $n=4$, LSD = 4.614) and belowground biomass (bottom panel; mean \pm SE, $n=4$, LSD = 18.169).

Soil Physico-Chemical Characterization

Soil organic matter was significantly higher in the 0 ml m⁻² humic acid amendment level than in the combination of humic acid amendments for dune and swale species (Tables A1.13–A1.18; Contrast F = 10.4, p < 0.01). No significant effects of species, fertilizer level, or interactions were found for soil organic matter. *Baccharis halimifolia* demonstrated significantly greater soil moisture than other dune and swale species (Tables A1.13–A1.18; F = 84.4, p < 0.01). Also, the low fertilizer level demonstrated lower soil moisture than the high fertilizer level (Tables A1.13–A1.18; F = 46.7, p < 0.01). No significant effect of humic acid amendment or any interactions on soil moisture were detected. A significant effect of species was detected in which *P. amarum* and *B. halimifolia* were more acidic than the other dune and swale species. No significant effects of fertilizer level, humic acid amendment, or interactions on soil pH, were detected. *Spartina patens* had significantly greater soil ammonium than all other dune and swale species (Tables A1.13–A1.18; Contrast F = 41.9, p < 0.01). No significant effects of fertilizer level, humic acid amendment, or interactions on soil ammonium were detected. *Distichlis spicata* had greater soil nitrate-nitrite than all other dune and swale species (Tables A1.13–A1.18; Contrast F = 15.2, p < 0.01). No significant effects of fertilizer level, humic acid amendment, or interactions on soil nitrate-nitrite were detected. Interestingly, *S. patens* and *D. spicata* had greater soil phosphorus than all other dune and swale species (Tables A1.13–A1.18; Contrast F = 15.4, p < 0.01). Also the 0 ml m⁻² humic acid amendment level had greater soil phosphorus than all humic acid amendment levels (Tables A1.13–A1.18; Contrast F = 11.5, p < 0.01). The high fertilizer level had greater soil phosphorus than the low fertilizer level (Tables A1.13–A1.18; F = 5.8, p < 0.05). No significant interactions were detected with regard to soil phosphorus for dune and swale species.

No significant effects of species, fertilizer level, humic acid amendment, or interactions thereof were detected regarding soil pH, conductivity, or organic matter for the salt marsh species. *Spartina alterniflora* had significantly greater soil ammonium than *A. germinans* (Tables A1.19–A1.20; F = 10.1, p < 0.01). No significant effects of species, fertilizer level, or humic acid amendment, or interactions thereof, were detected regarding soil nitrate-nitrite or phosphorus for the salt marsh species.

Discussion

The effects of humic acid amendment dosage and plant species discerned from the range-finding and refinement studies provide useful information both for the subsequent field studies reported herein and for further investigations of humic acid enhancement of plant growth processes. Although a range of plant species responses were apparent, moderately low dosages of humic acid were determined to be the most beneficial levels for the majority of the species studied, while extremely high dosage proved to be detrimental for all species. Humic acid amendment did not result in the dramatic increase in dune and swale plant growth observed in response to fertilizer addition, nor were clear synergies between humic acid amendment and fertilizer noted. Importantly, however, no reduction in plant growth was evident with any of the moderate humic acid amendment levels employed in the refinement study, and trends toward increased plant growth for certain species with humic acid amendment were discerned.

In the range-finding study, the foredune species *P. amarum* and *U. paniculata* demonstrated increased growth with moderate to high levels of humic acid amendment. For instance, *P. amarum* aboveground biomass was increased with humic acid amendment levels up to 900 ml m⁻², and belowground biomass was increased with humic acid amendment levels up to 2,700 ml m⁻². Humic acid amendment levels between 100 ml m⁻² and 2,700 ml m⁻² had a similar effect, significantly increasing *U. paniculata* belowground biomass; however, aboveground biomass did not show this same trend. These humic acid amendment levels are much higher than what are typically employed in marginal agricultural soil enhancement efforts (~ 4 ml m⁻²). As the foredune environment where *P. amarum* occurs is highly nutrient and organic matter deficient (Ehrenfeld 1990; Hester and Mendelsohn 1990), the benefit of these very high humic acid amendment levels may partially reflect the depauperate nature of the soil. An investigation by Willis and Hester (2010) reported that *P. amarum*, but not *U. paniculata*, demonstrated significantly increased growth with humic amendment levels of 5 ml m⁻², 20 ml m⁻², and 80 ml m⁻². Because neither an asymptote nor decline in growth was noted for *P. amarum*, Willis and Hester (2010) suggested that the optimal humic acid amendment level may actually be higher than their maximum application rate of 80 ml m⁻². One of the possible reasons for this is that the dune sands on which this and other dune species occur have very low clay content. The interaction of humic substances, particularly humic acids, with clay particles is a key mechanism for the retention and availability of nutrients (Jackson 1993).

Benefits of humic acid amendment were less clear in the refinement study for both *P. amarum* and *U. paniculata*. *Panicum amarum* demonstrated only slightly greater belowground biomass in the 125 ml m⁻² humic acid amendment and no real benefit in the higher humic acid amendment levels in the refinement study. Similarly, *U. paniculata* belowground biomass was increased in the 125 and 250 ml m⁻² humic acid amendment levels in the refinement study, but not the 500 ml humic acid amendment level. *Panicum amarum* and *U. paniculata* are both known to benefit significantly from fertilizer applications (Hester and Mendelsohn 1990; Willis and Hester 2010), and broadcast fertilizer application is considered a key component of the restoration approach for these species (Broome et al. 1982). Growth of both *P. amarum* and *U. paniculata* was increased by fertilizer application in the refinement study. However, no synergy of humic acid amendment with fertilizer application was detected.

Growth responses of the rear dune and swale plant species to humic acid amendment in the range-finding study were varied. *Spartina patens* displayed stepwise increases in growth with humic acid amendment up to 900 ml m⁻², whereas *P. vaginatum* showed a moderate increase in growth at 900 ml m⁻², and *D. spicata* and *B. halimifolia* largely demonstrated negative growth responses with humic acid amendment. In fact, visually evident tissue damage appeared in *D. spicata* and *B. halimifolia* at humic acid amendment levels of 300 ml m⁻² and above. Minimal stimulation of growth with any humic acid amendment was observed for all of these species, including *S. patens*, in the refinement study. However, *P. vaginatum* belowground biomass and final stem height was generally greater in the 125 ml m⁻² humic acid amendment than other humic acid treatments. *Paspalum vaginatum* final stem height also reflects enhanced growth in the 125 ml m⁻² treatments compared to other humic acid amendment levels. *Spartina patens* is known to benefit from fertilizer application (Sistani and Mays 2001), though others have reported contrasting results. For instance, Day et al. (2004) found that long-term nitrogen fertilization was detrimental to *S. patens*, likely because of increased competition from other

dune species. Also, Webb et al. (1984) detected no benefit of fertilizer addition for *S. patens* planted in a dredged sandy substrate at Galveston Bay.

Humic acid amendment benefited both salt marsh species employed in the range-finding study, although somewhat greater growth stimulation occurred in *S. alterniflora* than was noted in *A. germinans* growth. Specifically, moderate levels of humic acid amendment (100 ml m^{-2} to 900 ml m^{-2}) increased both above- and belowground biomass of *S. alterniflora* in the range-finding study. In contrast, somewhat lower levels of humic acid amendment (300 ml m^{-2}) increased belowground biomass of *A. germinans*, with minimal effect on aboveground biomass detected in the range-finding study. However, no significant stimulation of *S. alterniflora* or *A. germinans* growth was observed with any of the humic acid amendment levels (125 ml m^{-2} , 250 ml m^{-2} , 500 ml m^{-2}) employed in the refinement study. In a previous greenhouse study investigating the effects of humic acid amendment on coastal plant species, Willis and Hester (2010) reported that humic acid amendment levels of 5 ml m^{-2} , 20 ml m^{-2} , and 80 ml m^{-2} increased *S. alterniflora* belowground biomass, but not *A. germinans* above- or belowground biomass. Similarly, Naohiro et al. (2012) found that humic acid amendment did not enhance the restoration success of another mangrove species, *Rhizophora mucronata*, planted in an abandoned shrimp pond. The failure of humic acid amendment to significantly stimulate *Avicennia germinans* may reflect the woody growth form of this species (Willis and Hester 2010), which has an inherently slower growth rate than herbaceous species (Fitter and Hay 2001).

Fertilizer application significantly increased *A. germinans* growth in the refinement study, although no synergy with humic acid amendment was detected. Fertilizer has been previously demonstrated to increase growth of *A. germinans* and other mangrove species in both greenhouse (Willis and Hester 2010) and field studies (Feller et al. 2003; Naohiro et al. 2012). The intertidal nature of *S. alterniflora* and *A. germinans* habitats precludes use of fertilizer application as a restoration technique in Louisiana. However, because of its known efficacy in stimulating the growth of *S. alterniflora* and *A. germinans*, fertilizer application is a useful metric by which to evaluate novel soil amendments for these species.

The range-finding study revealed generally consistent trends of humic acid amendment across coastal plant species growth as well as species-specific responses. All of the plant species evaluated displayed visually evident tissue damage within one week of the $8,100 \text{ ml m}^{-2}$ humic acid amendment treatment being initiated. Rapid (i.e., within one week) onset of tissue damage was also noted for *D. spicata*, *S. patens*, and *B. halimifolia* at the lower concentration of $2,700 \text{ ml m}^{-2}$ humic acid amendment treatment. However, all of the species evaluated in the range-finding study showed increased growth at some low to moderate humic acid amendment level, except *B. halimifolia* and *D. spicata*. Humic substances, including humic acids, are complex polymers of hydrocarbon components that can vary widely in their composition (De Santiago et al. 2010). Among the functional groups that occur as a constituent of humic acid are aromatic compounds, the presence of which is believed to be a major mechanism in its phytotoxicity (Brunner et al. 1996; De Santiago et al. 2010). Additionally, Capasso et al. (2002) found evidence of xylem obstruction by high (>150 kilodalton) molecular weight substances, which they suggested as a mechanism for the adverse effects of humic acid-like substances on *Lycopersicon esculentum*. De Santiago et al. (2010) evaluated the effects of iron amendment in

conjunction with humic substances derived from composted cork, leonardite, and a mixture of olive husk and cotton gin waste at concentrations of 0 (control), 0.1 and 0.5 g organic carbon kg⁻¹ of soil on *Lupinus albus* growth in an iron-deficient soil. Interestingly, the leonardite-derived humic substances, the same type of source material used in the studies reported herein, displayed the greatest aromaticity compared with humic substances derived from other sources. De Santiago et al. (2010) reported that the form of iron amendment employed was of great importance in modulating the effect of the humic substance amendment. Composted cork-derived humic substances and Fe-EDDHA having significantly increased biomass, whereas the leonardite-derived humic substances and Vivianite significantly decreased biomass. Extremely high levels of humic acid amendment were specifically included in the range-finding component of this research to ascertain the point at which humic acid amendment would shift from beneficial to detrimental with these coastal plant species and substrate type. Thus, the 8,100 ml m⁻² humic acid amendment level appears to represent the endpoint beyond which only adverse effects would occur.

In conclusion, humic acid amendment does appear to enhance the growth of some coastal plant species; however, this enhancement of growth is not consistent. Nonetheless, for certain species such as *P. amarum* and *S. alterniflora*, humic acid amendment can result in enhanced plant growth. However, the application of commercial humic acid is relatively expensive in terms of product cost (\$4,100 hectare⁻¹ at a rate of 125 ml m⁻² based on a product cost of \$3.70 liter⁻¹) compared with the instigation of a broadcast fertilizer regime (878.8 kg ha⁻¹ 8-8-8 fertilizer: \$250 hectare⁻¹; 195.3 kg ha⁻¹ ammonium nitrate: \$110 hectare⁻¹). The equipment required for practical field implementation of humic acid amendment, backpack or ATV mounted sprayers, is also generally more expensive than that for broadcast fertilizer, which would typically consist of shoulder harness fertilizer spreaders. Additionally, its effectiveness in tidal systems is less predictable because of the possibility of humic acid removal through tidal action. Therefore, even though humic acid amendment does appear to benefit the growth of several important coastal plant species, careful cost-benefit analysis should be performed prior to incorporating this into a coastal restoration plan.

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CHAPTER 2. ESTABLISHMENT OF *BACCHARIS HALIMIFOLIA* IN COASTAL HABITAT USING SEED DISPERSAL

Introduction

Louisiana's coastal wetlands and barrier islands are experiencing some of the highest rates of erosion of any coastal region in the world (Penland et al. 1990). Efforts to restore these highly erosional islands often involve the dredging and deposition of nearby sediments onto the eroded portion of the island. These dredged sediments are, however, typically low in nutrients and organic matter content. Therefore, while difficult, establishing healthy vegetative cover is essential in many ways for the protection of the economic investment put into rebuilding the physical structure of these islands. Healthy vegetation helps maintain sediment stability through belowground roots, making it more difficult for over-wash events to erode away sands (Hester et al. 2005). The aboveground tissues also play a key role in promoting island integrity by trapping windblown sand and sediments, thereby keeping them in the island system.

An important species on these islands is *Baccharis halimifolia*, recognized for its habitat value. *Baccharis halimifolia* is a perennial woody shrub that can reach heights up to 5 m. In addition to its presence in barrier island swale habitat, it also exists in salt marshes and brackish swamps, illustrating its salt tolerance (Van Deelen et al. 1991). *Baccharis halimifolia* has a life history that is consistent with that of an *r* selected species as described by Pianka (1970). Mature plants can produce upward of 1.5 million seeds per year (Westman et al. 1975), a large contribution to the seed bank. Establishing stands of mature plants on these islands can help maintain island integrity during overwash and ensure seed bank development, the latter of which enables this shrub pioneer species to colonize following severe storm-induced overwash events. The establishment of woody species in Louisiana barrier island restoration projects is recognized as highly desirable and plays a role in how potential projects are ranked (CWPPRA 2008). Designs that incorporate two woody species receive the highest scores. While *Avicennia germinans* fills this role in the back-barrier salt marsh, *B. halimifolia* could potentially be incorporated into such designs for use in the swale environment.

Although several studies have investigated factors that can influence *B. halimifolia* seed germination, knowledge gaps remain. Karrfalt and Olson (1974) reported that *B. halimifolia* seeds maintained a 99% germination rate after burial in soil at a depth of 5 cm for a period of 2 years. This alludes to the need for a mechanism to initiate germination. Panetta (1979) suggested that these mechanisms are sunlight and temperature fluctuations at the time of germination. When temperature was constant, the ratio of low red/far red light was all that was needed to increase germination, with high low red/far red light ratios eliciting the greatest germination response. These experiments also found that germination in the absence of light increased in response to an increase in amplitude of temperature fluctuation. These findings suggest that optimal germination depth should be located at or near the surface due to the relatively high amount of irradiance and increased temperature fluctuations experienced there as compared to the dark, temperature-stable environment at depth.

Hydroseeding is a seed dispersal technique that has the advantage of allowing seeds to be sown onto the substrate surface while giving seeds and sediment initial moisture for germination.

With its wood pulp/cellulose matrix of hydromulch, hydroseeding is a treatment not yet widely utilized in coastal restoration, but it may deliver the desired results of enhanced establishment. Laboratory and greenhouse experiments have had mixed results with grass seed establishment via hydroseeding, the poor establishment possibly due to substrate surface microtopography and lack of subsequent moisture (Sheldon and Bradshaw 1977). Unseeded hydromulch is also used in erosion control and can hold soils in place during small to moderate rainstorms (Wohlgemuth et al 2010). Therefore, we hypothesized that hydroseeding of *B. halimifolia* would be a suitable method for establishing this species in dynamic barrier island habitats, so often characterized by shifting sand and low soil moisture.

Humic acid, a naturally occurring soil conditioner, has been shown to relieve drought stress (Sharif et al. 2002; Zhang and Schmidt 1999) and may work with hydromulch in keeping more moisture near the surface for *B. halimifolia* seed germination. Although the range-finding and refinement greenhouse studies (Chapter 1) conducted as a portion of this research indicate that *B. halimifolia* seedling growth is not enhanced by humic acid application, lower doses of humic acid may improve germination response by increasing soil moisture retention. The high molecular weight polyacids of humic acids have both indirect and direct effects on plant growth (Jackson 1993). The indirect effects of humic acid on plant growth include the ability to modify the physical and chemical components of soil. Humic substances increase the water holding capacity of soils and thus, when present in sufficient amounts, may help to alleviate drought conditions (Sauchelli 1944). Addition of humates can also alter the viscosity and specific gravity of soils (Swietochwski 1960) and reduce soil erosion by increasing sorption and the binding force of the very fine soil particles to the electrolytically charged water (Jackson 1993).

The purpose of this study is to investigate techniques to enhance the seed germination and establishment of *B. halimifolia* and improve sediment conditions for seedling growth. The optimal burial depth for high seed germination rates of *B. halimifolia* seeds was hypothesized to be at or near the sediment surface. The time of year in which the seeds are dispersed may also have an effect on germination success. Hydromulch and humic acid amendment were hypothesized to increase seed germination in sediments with low organic matter by increasing moisture-retaining qualities, which increase germination response. A series of greenhouse experiments tested these hypotheses: one that determined if there was a required dormancy period for *B. halimifolia* seed germination, a second that determined limits of sand burial depth on seed germination and emergence, and another that investigated the effects of shade and precipitation frequency on seed germination in hydromulch. The final study included interactions between hydromulch, humic acid, precipitation regime, and sediment organic matter. This information is anticipated to be valuable to coastal managers tasked with restoring a range of habitat types including woody vegetation such as *B. halimifolia* on restored barrier islands.

Materials and Methods

Monthly Germination

Atchafalaya Basin river sand (0.12 – 0.25mm grain size) similar to the fine sands in swale habitats on Louisiana's barrier islands was purchased from a commercial supplier and placed into five half-liter containers. The half-liter containers were then placed in shallow tubs of water to allow for constant moisture during seed germination. *B. halimifolia* seeds were

collected from mature plants located in the coastal Louisiana town of Delcambre during the fall of 2010. From these seeds, 0.025 g were weighed, counted, and determined to contain 222 seeds. Each experimental unit received 0.025 grams of seeds, and the total number of seeds that germinated and established was recorded. This experiment was repeated once a month in the University of Louisiana's Center for Ecology and Environmental Technology (CEET) greenhouse from December through July.

Sand Burial

The same source of sediment employed for the monthly germination study (Atchafalaya Basin river sand) was placed in 30 half-liter containers that were then placed in shallow tubs of water to allow for constant moisture during seed germination. Five burial depths were chosen (0.0, 0.5, 1.0, 2.0, 3.0 cm) with each depth being replicated six times. *Baccharis halimifolia* seeds were collected from mature plants located in the coastal Louisiana town of Delcambre during the fall of 2008; 50 seeds were used per experimental unit. All containers were placed in the CEET greenhouse during the spring of 2008. The number of seeds that germinated and emerged in each burial depth were counted at three-day intervals.

Precipitation Regime and Sediment Amendments

This study incorporates a factorial experimental design that includes three organic matter levels (0%, 5%, 30%), two precipitation regimes (average annual rainfall, half annual rainfall), two hydromulch levels (present, absent), and two humic acid levels (present, absent). All conditions were replicated four times, giving a total of 96 experimental units. Dredged sands similar to those found on barrier island restoration projects was collected and placed into individual 4.730L containers. The sand was found to have 0% organic matter; therefore, dried peat moss was added in the appropriate amounts to formulate both 5% and 30% organic matter treatments. Once all organic matter treatments were established, the containers were placed in a greenhouse and watered frequently for two weeks prior to the start of the experiment to maintain uniform saturation. Humic acid soil conditioner composed of 4% humic acid derived from brown coal from the manufacturer (3 Tier Technologies, Longwood, FL) was applied using their recommended dosage of 52.6 ml humic acid L⁻¹ to each pot. Note that this dosage was considerably lower than those used in the previous greenhouse studies to assess plant growth response to humic acid amendment. Hydromulch composed of 80% paper product and 20% wood fiber from the manufacturer Jet Spray was applied according to the manufacturer's recommended amounts and 50 *Baccharis halimifolia* seeds were placed in each container. The precipitation regime was then implemented with watering occurring twice weekly and the number of seeds which germinated was counted every 3 days.

Precipitation Frequency and Shade Treatment

In this study the effects of precipitation frequency and shade on *B. halimifolia* seed germination in hydromulch was assessed. A factorial experimental design was employed, using average annual rainfall for coastal Louisiana (1,600 mm/year) applied in two precipitation frequencies (weekly precipitation applied one day a week or divided into two applications three days apart). A shade treatment with two light levels was also used (60% light transmittance or 100% light transmittance). Each experimental unit was replicated five times to give a total of 20 experimental units. Sediment similar to that found on swale habitat located on Louisiana's barrier islands was filled to a depth of 6 cm in containers measuring 17cm x 13.5 cm x 8.5 cm.

Each experimental unit received a mixture of 68.85 cm³ hydromulch from the manufacturer Jet Spray and was combined with .1 g (740) of *B. halimifolia* seeds and placed in the University of Louisiana greenhouse located at the Center for Environmental and Ecological Technologies. Treatments were randomized and then watered according to the determined precipitation frequency and allowed to germinate in either ambient greenhouse light or under shade cloth (60% light transmittance). The numbers of seeds that germinated were determined daily for three weeks.

Results

Monthly Germination/Sand Burial

The study on the effect of storage time following seed dispersal revealed that viability of the 2010 seed cohort remained fairly constant during the post-dispersal, eight-month period (December through July), averaging 39-41% germination. The effect of burial depth on *B. halimifolia* seed germination was found to be inhibitory at depths greater than 1 cm. None of the 600 seeds present in burial treatments between 2 cm and 3 cm germinated and emerged. The depth at which germination was greatest was at the surface. In the surface burial treatment, 191 out of 300 seeds germinated and established (64%). The results show that burial at even shallow depths has a drastic effect on germination (Figure 2.1). The time required for seeds to germinate was typically about 10 days, and after 15 days no additional germination and emergence was observed.

Precipitation Frequency and Shade Treatment

Baccharis halimifolia seed germination was found to be greatest under ambient greenhouse light (Figure 2.2, bottom panel); germination under the shade cloth was significantly reduced ($F = 6.676$, $P < 0.05$). Precipitation frequency also had a significant effect on seed germination (Figure 2.3). The twice weekly precipitation frequency brought about a significantly greater germination response ($F = 6.203$, $P < 0.05$). The number of days to reach maximum germination differed among the two precipitation frequencies with the twice weekly treatment achieving maximum germination sooner (Figure 2.2, top panel).

Precipitation Regime and Sediment Amendments

The results indicate a highly significant difference in germination success between rainfall treatments ($F = 121.597$, $p < 0.01$) with full average rainfall causing an increase in germination (Figures 2.4, 2.5, 2.6). Hydromulch elicited a highly significant difference in germination response across all treatments ($F = 7.4843$, $p < 0.01$) with treatments containing hydromulch having higher percentages of germination. Hydromulch was found to have the greatest impact on germination success in treatments with 0% organic matter (Figure 2.4). Organic matter had a significant effect on germination response ($F = 3.995$, $p < 0.05$), and there was a significant interaction between rainfall and organic matter ($F = 3.138$, $p < 0.05$); experimental units with 5% and 30% organic matter had higher germination percentages than the 0% organic matter treatments (Figures 2.5, 2.6). Humic acid was found to have no significant effect on *B. halimifolia* germination across all treatment conditions ($F = .453$, $p > 0.5$).

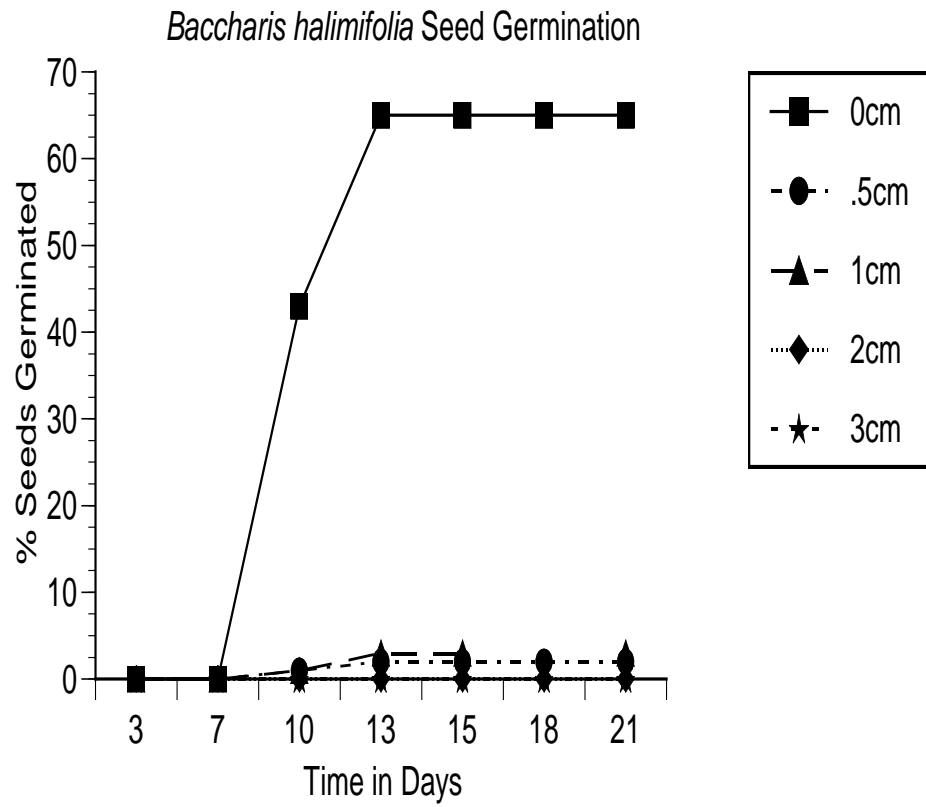


Figure 2.1 The effect of burial depth on *B. halimifolia* cumulative seed germination (mean) over time.

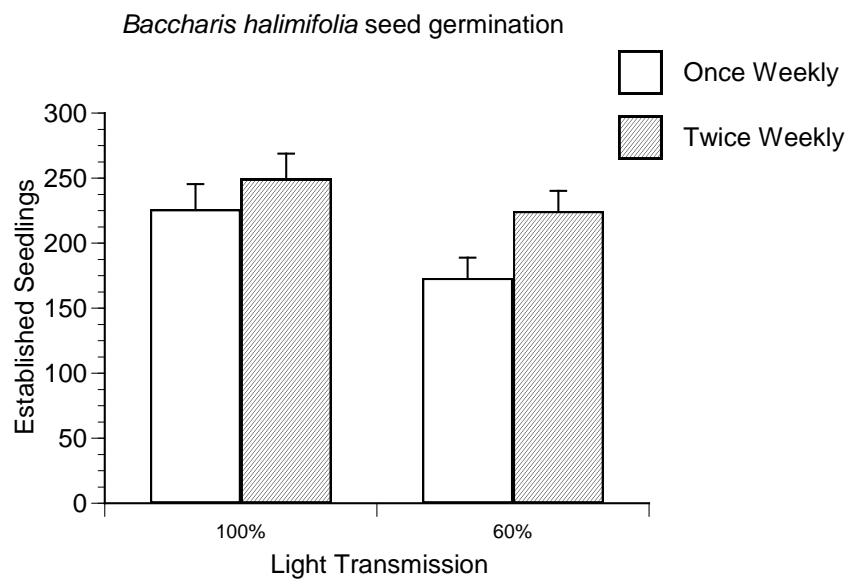
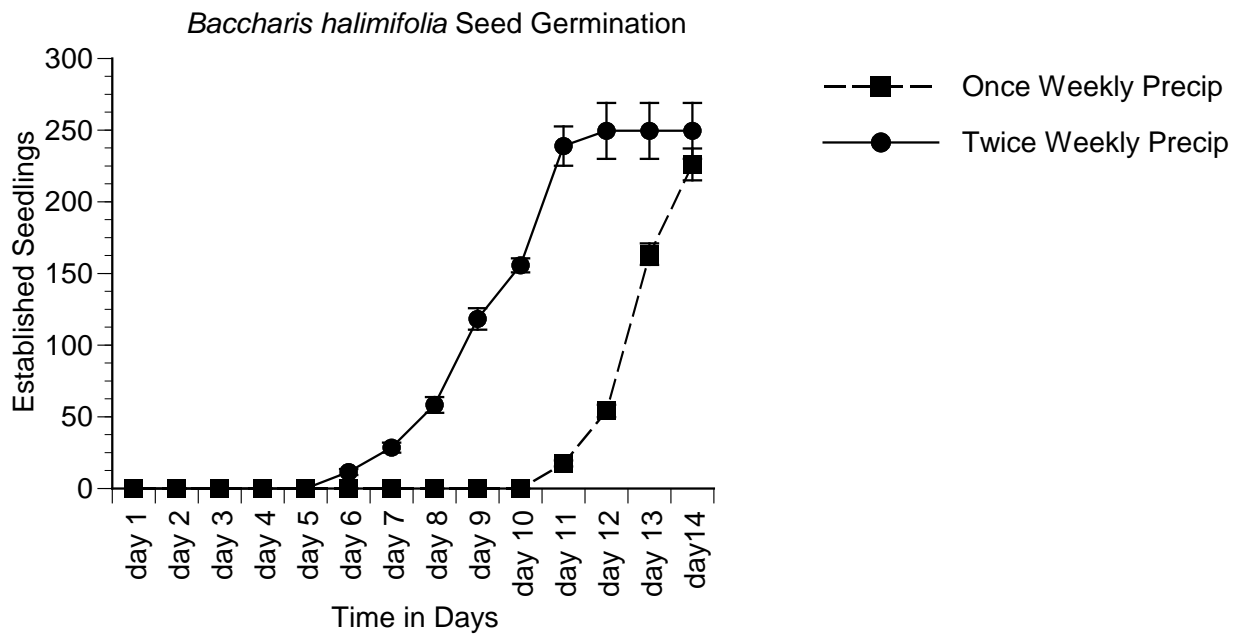


Figure 2.2 The effect of shade and precipitation frequency on *B. halimifolia* seed germination and seedling survival (top panel; mean \pm SE, $n = 5$). The effect of precipitation frequency on *B. halimifolia* cumulative seed germination under ambient greenhouse light over time (bottom panel; mean \pm SE, $n = 5$).

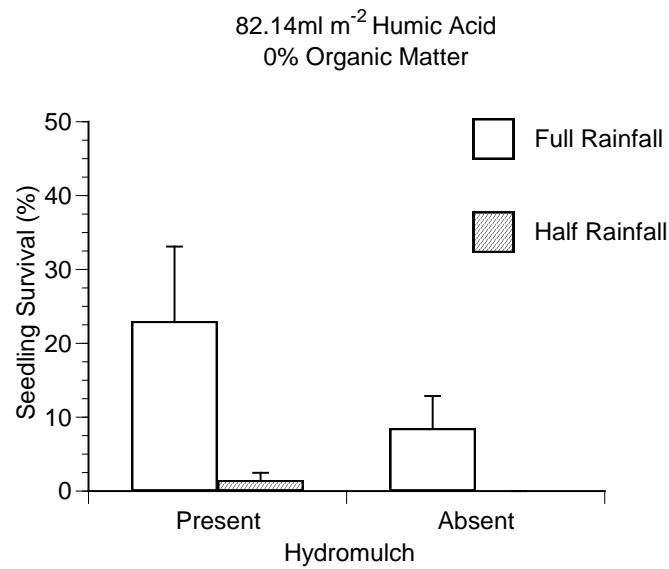
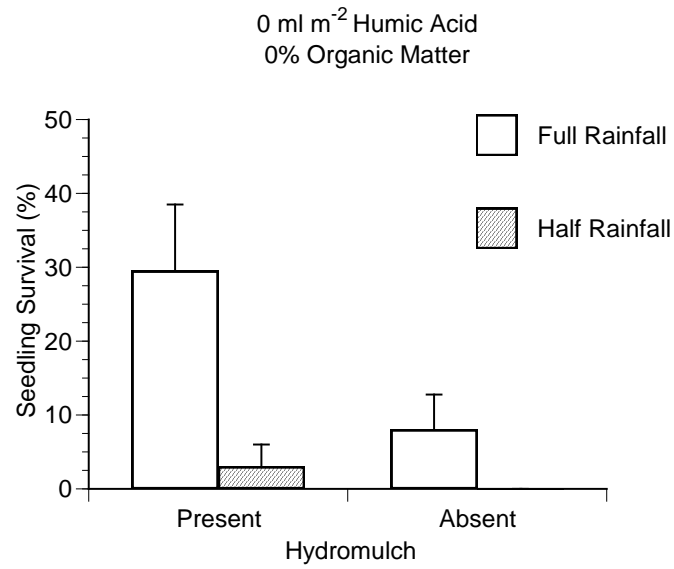


Figure 2.3 The effect of organic matter, hydromulch and precipitation regime on *Baccharis halimifolia* seed germination and seedling survival (mean \pm SE, n = 4, LSD = 13.546).

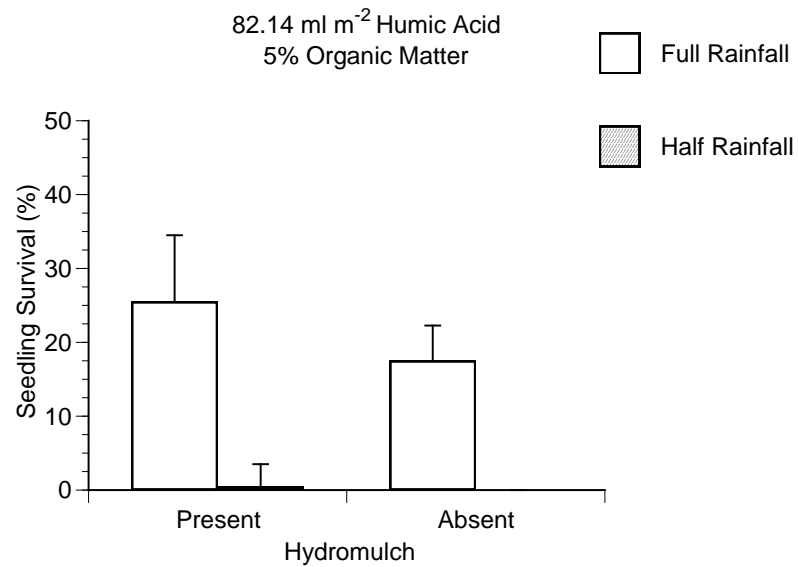
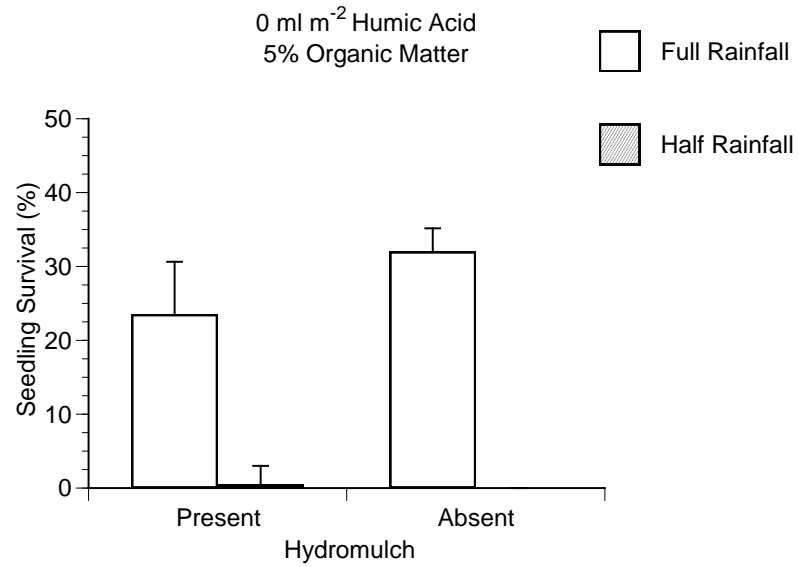


Figure 2.4 The effect of organic matter, hydromulch and precipitation regime on *Baccharis halimifolia* seed germination and seedling survival (mean \pm SE, $n = 4$, LSD = 13.546).

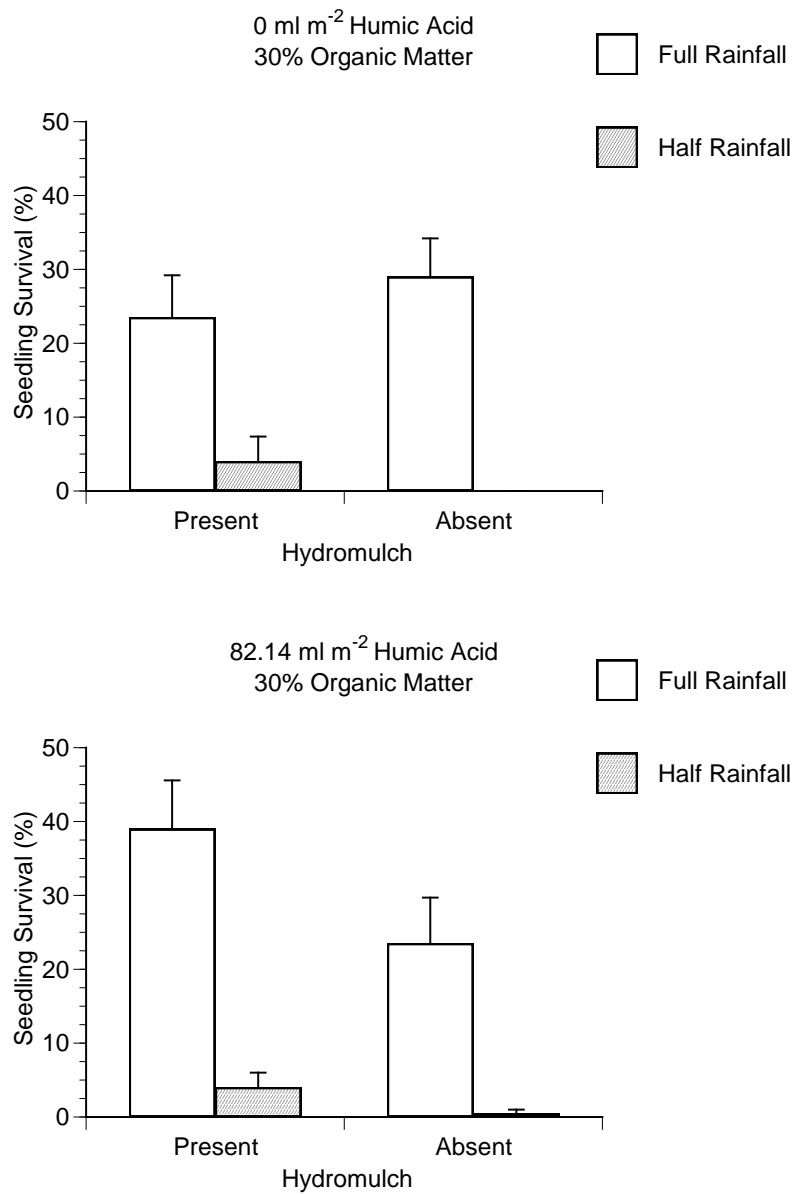


Figure 2.5 The effect of organic matter, hydromulch and precipitation regime on *Baccharis halimifolia* seed germination and seedling survival (mean \pm SE, $n = 4$, LSD = 13.546).

Discussion

The ability of *B. halimifolia* to germinate and establish on the surface of sediments alludes to its early-seral nature. The capacity of *B. halimifolia* to quickly colonize disturbed sites also suggests that organic matter may not be as important as sediment moisture in eliciting a germination response (Van Deelen 1991), although during periods of low precipitation, soil organic matter may become increasingly important by holding moisture in the soil for longer periods of time. The current experiments indicate that organic matter content did have the effect of increasing germination percentages in treatments with full annual rainfall. In the treatments simulating a drought situation, however, no significant differences in number of seeds were found germinated among the three organic matter percentages. Weather conditions are unpredictable and the amount of rainfall a given restoration project experiences is highly variable. Therefore, as indicated by the field study, the timing of hydromulch/seed application is critical.

When trying to establish *B. halimifolia* on a restoration site, an emphasis should be placed on procedures that retain water near the surface. The monthly germination experiments conducted on the coastal Louisiana 2010 seed cohort indicate the same lack of required dormancy implied by Westman (1975) and Panetta (1977) on seeds from populations growing in Queensland, Australia. This finding may allow for greater flexibility in timing seed dispersal activities at restoration sites during times of the year when temperatures are not as high and the probability of rainfall is greater. Additionally, hydromulch treatments have been shown to hold considerable moisture (nearly 10 times the moisture of bare sand), and in greenhouse studies they significantly increased germination percentages in treatments with 0 % organic matter. More studies on seedling growth in hydromulch and humic acid are needed to better assess their value in barrier island restoration. Egerova et al (2003) found that patches of *Spartina alterniflora* increased the number of *B. halimifolia* plants per site, with more *B. halimifolia* found in the larger *S. alterniflora* patches. This facilitation effect may be from the shade provided by *S. alterniflora* to the surrounding sediment surface. Thus, while data from the current shade study and from Westman 1975 indicate that *B. halimifolia* germination is higher in sunny environments, a critical balance between sunlight, sufficient soil moisture, and optimal germination temperature within patches of vegetation at a colonization site may facilitate successful germination and establishment. For example, Profit and Young (1999) reported that the mean summer temperature below a *S. alterniflora* canopy was 3.9 C° cooler than temperatures above the canopy, which may cause less evaporation from the sediment surface, resulting in adequate moisture for *B. halimifolia* seed germination. Restoration efforts should be planned for dates when the local weather forecast calls for rain events the week following seed dispersal. Incorporating this critical timing along with the potential for beneficial facilitation processes from neighboring plants may prove to be an attractive option for coastal managers to consider when trying to establish *B. halimifolia* by seed.

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CHAPTER 3. EVALUATION OF TECHNIQUES TO ENHANCE *AVICENNIA GERMINANS* RESTORATION THROUGH PROPAGULE DISPERSAL

Introduction

Black mangrove, *Avicennia germinans*, is a natural vegetative component of Louisiana's barrier island salt marsh community. It contributes to important ecosystem services, such as coastal storm surge protection and avian habitat provision (Hester et al. 2005; Visser et al. 2005; Krauss et al. 2009). As such, *A. germinans* has been used in barrier island restoration projects in Louisiana, though not with great success (Khalil and Lee 2006). Unfavorable conditions of restored back-barrier salt marsh platforms (i.e., soil texture and elevation) may limit effective *A. germinans* restoration (Fearnley 2008). The efficacy of restoration projects is likely to be improved by designs that address the physiological tolerances of *A. germinans* (Lewis 2005; Alleman and Hester 2011) and alleviate stressful conditions apparent on restored barrier islands.

The propagules of *A. germinans* may be especially effective for restoration projects due to their ease of transport and dispersal at a restoration site, though several factors affect their subsequent establishment (Krauss et al. 2008 and references therein). In particular, desiccation (Toledo et al. 2001), physical disturbance (Balke et al. 2011), and poor sediment quality (Fearnley 2008) are environmental factors that can limit *A. germinans* propagule establishment and seedling growth, particularly at restoration sites. If a mangrove or herbaceous canopy is not present to facilitate natural colonization (Lewis and Dunstan 1976; Bosire et al. 2003; McKee et al. 2007; Whigham et al. 2009), then additional amendments may be necessary to alleviate stressful conditions of bare restoration sites.

There were four objectives of our greenhouse investigation of propagule establishment. The first objective was to assess the effect of hydromulch application on *A. germinans* propagule survival and establishment. Hydromulch is a slurry of wood fibers and tackifier that is applied to the ground. Wood fiber mulch application results in improved soil moisture and reduction of soil temperature fluctuations (Gruda 2008). The second objective was to assess the effects of humic acid dosage and soaking on *A. germinans* propagule survival and establishment. The third objective was to assess the application of humic acid on propagule establishment on sediment from a created back-barrier marsh platform rather than the coarser grain size of commercially-available sand. The fourth objective was to elucidate the effect of tidal inundation on hydromulch, because propagules establish within the intertidal range, and hydromulch would be flooded.

Materials and Methods

Propagules for all three experiments were collected from the Caminada-Moreau Headland on October 20, 2010. They were brought to the Center for Ecology and Environmental Technology (CEET) at University of Louisiana in Lafayette, LA, and kept in 36 ppt artificial salt water (Instant Ocean, Spectrum, Inc.) until the start of the experiment. Water was refreshed every other day and pericarps were removed from propagules. All experiments had four propagules per pot, 15 pots per experiment, for a total of 60 propagules per experiment. Pots were placed in reservoirs of 36 ppt artificial salt water, maintained 15 cm below the surface of

the sediment throughout the experiment, and were kept at ambient greenhouse conditions. Propagules were monitored and misted with water twice weekly from the beginning of the experiment until March 15, 2011.

Hydromulch Experiment

The experimental design for this study was a three-level, one-way, completely randomized design with five replicate pots (each containing four propagules). The first level consisted of propagules lying directly on commercial sand (grain size 0.12-0.25mm), which serves as a control for both this experiment and the humic acid experiment. The second level consisted of propagules placed on the sand with 400 ml of hydromulch mixture (3 volumetric liters of dry hydromulch mixed with 15 L of water) placed over the propagules. A small portion of hydromulch that was directly over the propagules was removed so that the propagules could be located, but still remain mostly covered. The third level consisted of adding 400 ml of hydromulch to each pot of sand, and placing propagules directly on top of the hydromulch. This experiment was initiated on October 26, 2010.

Humic Acid Experiment

The experimental design for the humic acid experiment was a three-level, one-way, completely randomized design with five replicate pots (each containing four propagules). The first level consisted of propagules soaking for 24 hours in a 10% humic acid solution with deionized water, and then placed directly on sand. The second level consisted of placing propagules on the sand and then applying 250 ml m⁻² humic acid (4% humic acid derived from coal: Huma-Boost, 3 Tier Technologies, Longwood, FL) to the sand and propagules. The third level consisted of placing propagules on the sand and then applying 500 ml m⁻² humic acid to the sand and propagules. This experiment was initiated on October 26, 2010.

Created Marsh Sediment Experiment

Sediment from the CWPPRA TE-50 created marsh platform was collected from Whiskey Island, LA, on October 26, 2009, brought to CEET and placed into pots. The experimental design for this experiment was a three-level, one-way, completely randomized design with five replicate pots (each containing four propagules). The first level consisted of propagules placed on sediment with no addition of humic acid. The second level consisted of propagules placed on sediment with 250 ml m⁻² humic acid applied over propagules. The third level consisted of propagules soaked for 24 hours in a 10% humic acid solution (in deionized water) and then placed on sediment. This experiment was initiated on October 29, 2010.

All propagules were monitored for time until developmental events occurred, including survival, occurrence of fungus, extension of the radicle, the lifting of the cotyledons from the sediment surface, and the appearance of first true leaves. Time until event data were visualized and analyzed using Kaplan-Meier (product-limit) curves. Comparisons among treatments were determined with log-rank tests and 95% confidence intervals. At the end of the project, mortality and establishment were compared using an ANOVA framework (graphs not shown as Kaplan-Meier curves provide more in-depth data). All analyses were performed using JMP 9 (SAS Institute).

Effect of Inundation on Hydromulch

An experiment to elucidate the effect of inundation on hydromulch was conducted February 18-21, 2011, at the CEET greenhouse facility. Hydromulch was agitated for 15 minutes before application to 4 L pots containing sand. Three pots had 250 ml of hydromulch applied and were immediately placed in a water reservoir, where they were completely flooded. Three more pots had 250 ml of hydromulch applied and, after 72 hours, were placed in water reservoirs and flooded.

Results

Hydromulch Experiment

Hydromulch treatment had a highly significant effect on mortality ($F = 336.0$, $p < 0.01$) and establishment ($F = 82.3$, $p < 0.01$). In the control treatment (propagules on sand), 80% of propagules died on average, and an average of 15% became established. All propagules placed on top of hydromulch died. The most successful treatment was placing hydromulch over propagules; 100% of propagules survived and 85% became established, on average. Propagules that did not become established seemed to be trapped under the hydromulch.

Kaplan-Meier curves of time until event revealed that treatment had a highly significant effect on survival. The treatment of hydromulch over propagules resulted in significantly greater survivorship (Figure 3.1; top panel, $X^2 = 45.3321$, $p < 0.01$). Occurrence of fungus (Figure 3.1, bottom panel) and extension of the radicle (Figure 3.2, top panel) did not differ with treatment. Similar to survivorship, the treatment of hydromulch over propagules had a highly significant positive effect on cotyledons lifting off of the sediment (Figure 3.2, bottom panel, $X^2 = 37.9468$, $p < 0.01$), and development of first leaves (Figure 3.3, top panel, $X^2 = 24.7728$, $p < 0.01$).

Humic Acid Experiment

Humic acid treatment did not have a significant effect on mortality or establishment. While treatments were not significantly different, the highest mortality occurred in the 250 ml m⁻² humic acid treatment (95%), and the greatest establishment occurred in the soaking treatment (10%). There were no clear differences in survivorship (Figure 3.3, bottom panel), occurrence of fungus (Figure 3.4, top panel), extension of radicles (Figure 3.4, bottom panel), or lifting of cotyledons off of the substrate (Figure 3.5) between treatments.

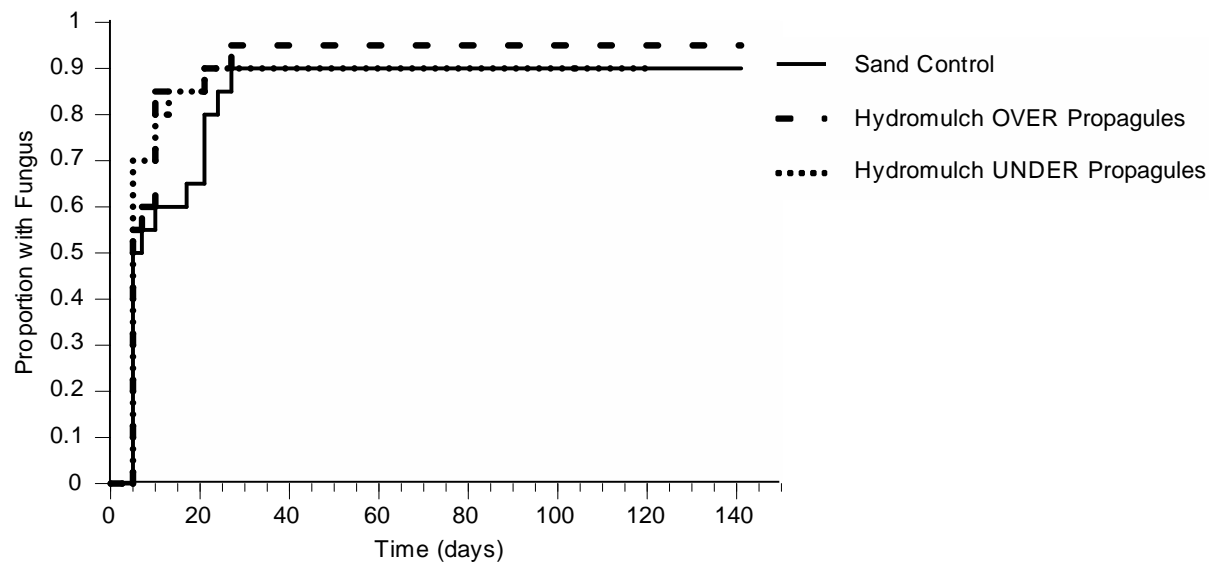
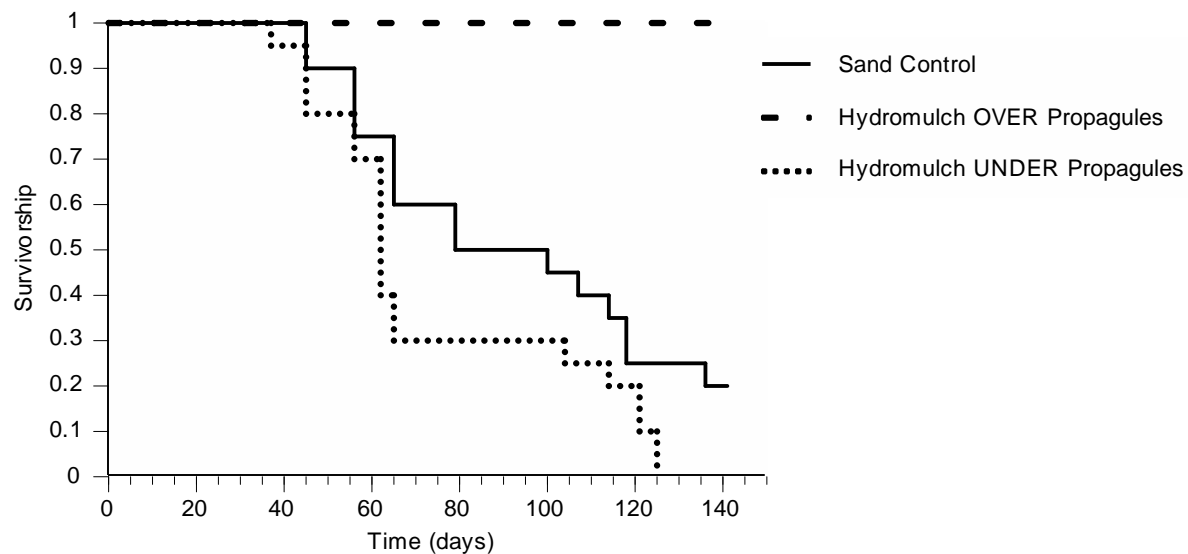


Figure 3.1 Kaplan-Meier curves of survivorship for propagules on sand, hydromulch *over* propagules, and hydromulch *under* propagules (top panel). Kaplan-Meier curves of proportion of propagules with fungus for propagules on sand, hydromulch *over* propagules, and hydromulch *under* propagules (bottom panel).

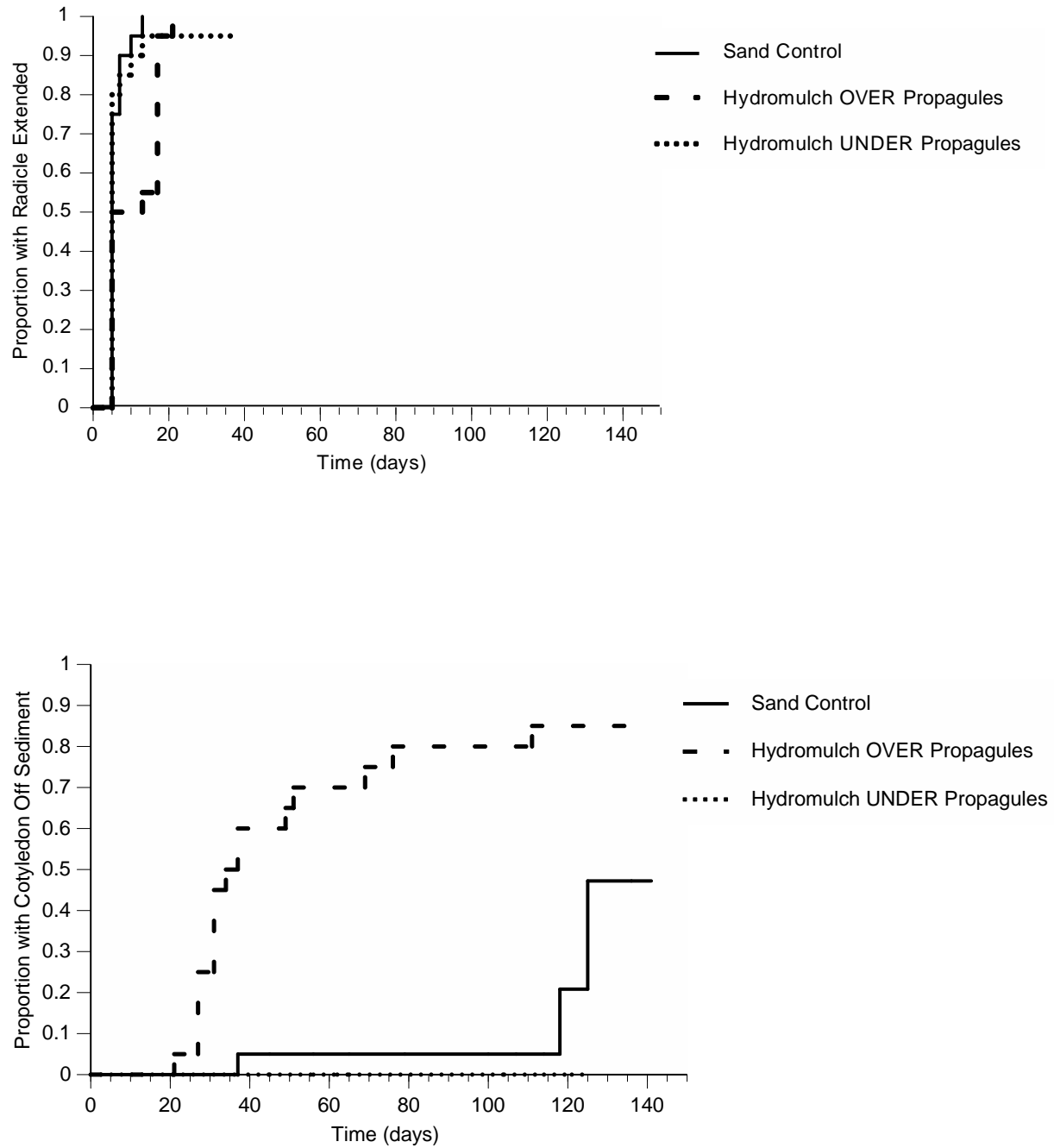


Figure 3.2 Kaplan-Meier curves of proportion of propagules with radicles extended for propagules on sand, hydromulch *over* propagules, and hydromulch *under* propagules (top panel). Kaplan-Meier curves of proportion of propagules with cotyledons lifted off of the sediment for propagules on sand, hydromulch *over* propagules, and hydromulch *under* propagules (bottom panel).

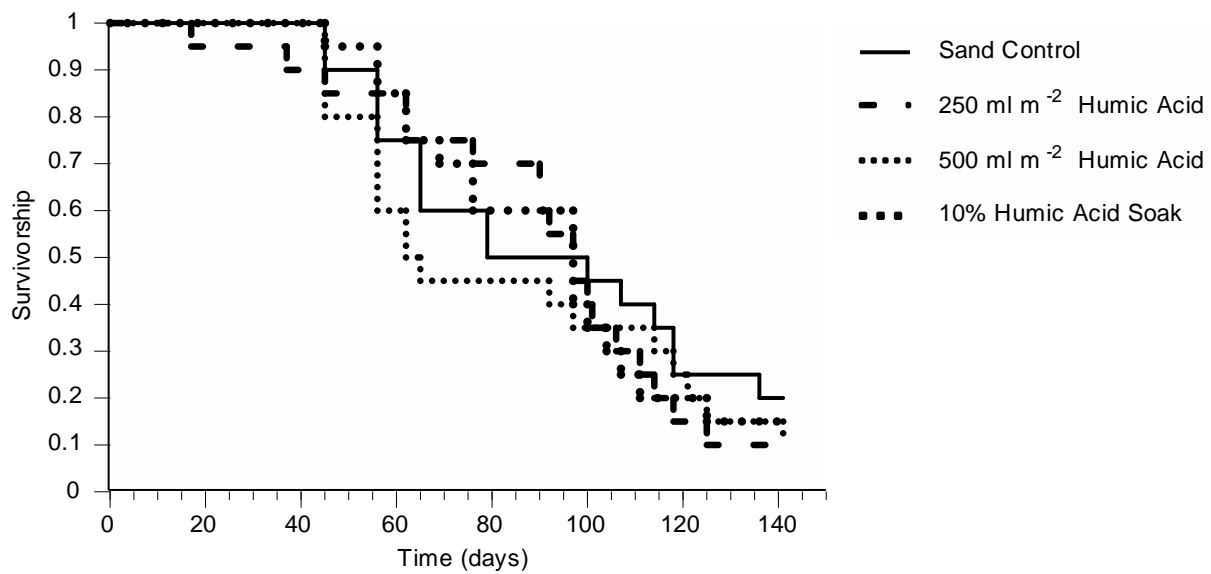
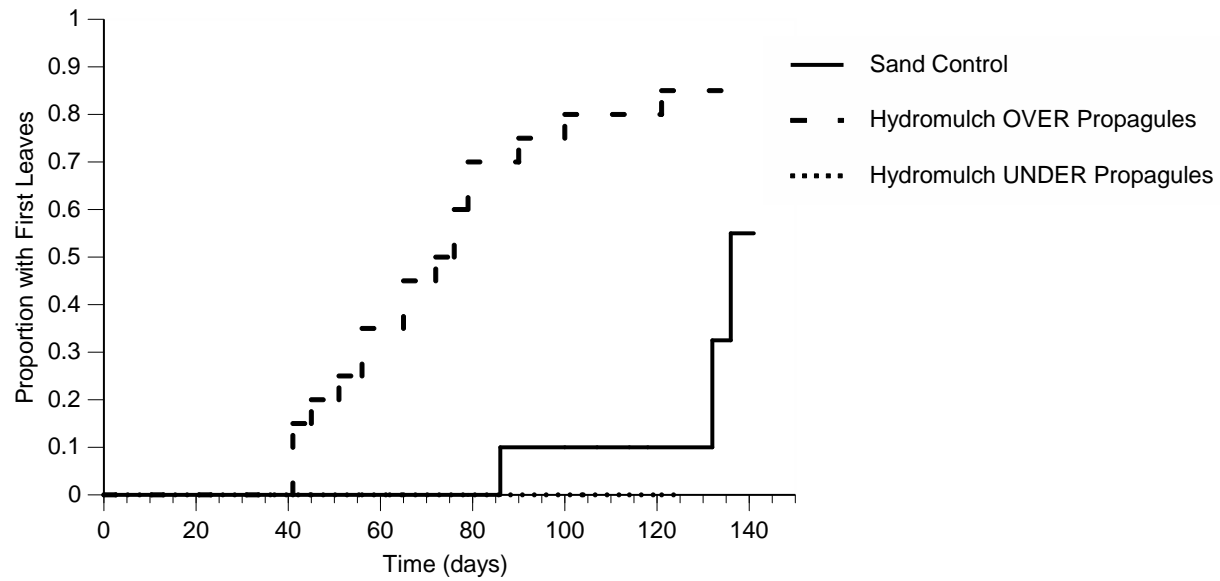


Figure 3.3 Kaplan-Meier curves of proportion of propagules with appearance of first true leaves for propagules on sand, hydromulch *over* propagules, and hydromulch *under* propagules (top panel). Kaplan-Meier curves of survivorship for propagules on sand, propagules on sand with 250 ml m⁻² humic acid added, propagules on sand with 500 ml m⁻² added, and propagules soaked in a 10% humic acid solution (bottom panel).

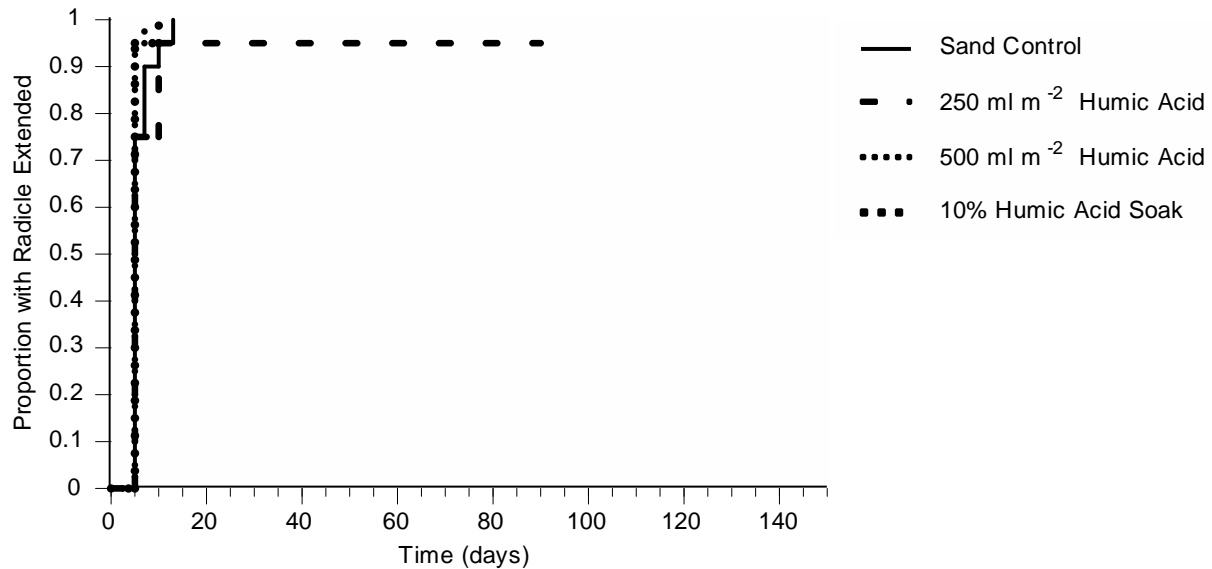
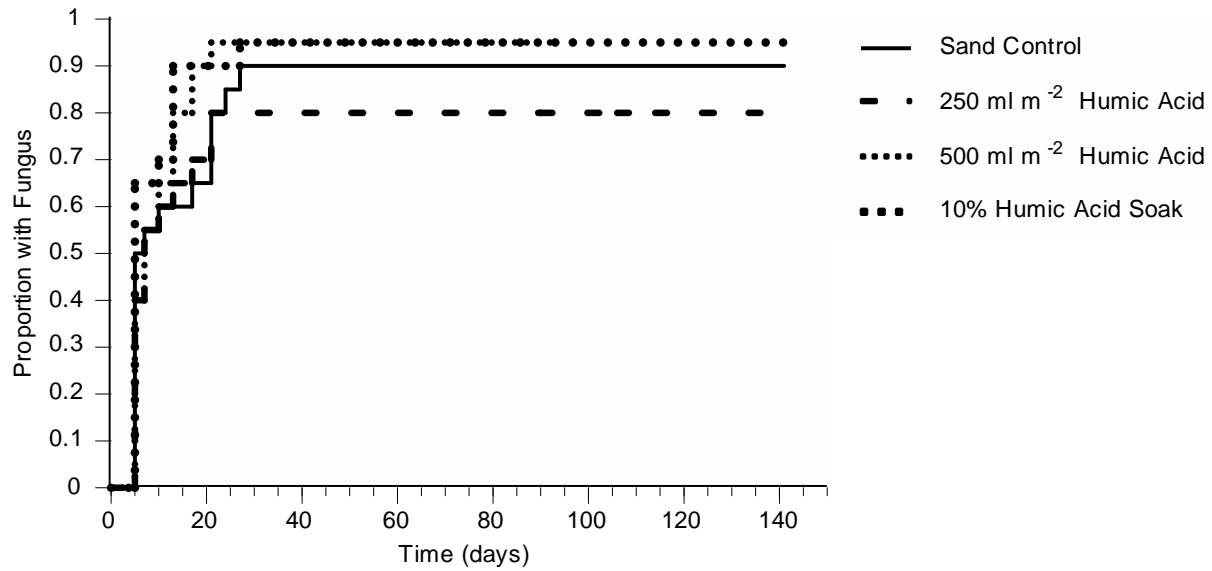


Figure 3.4 Kaplan-Meier curves of proportion of propagules with fungus for propagules on sand, propagules on sand with 250 ml m⁻² humic acid added, propagules on sand with 500 ml m⁻² added, and propagules soaked in a 10% humic acid solution (top panel). Kaplan-Meier curves of proportion of propagules with radicles extended for propagules on sand, propagules on sand with 250 ml m⁻² humic acid added, propagules on sand with 500 ml m⁻² added, and propagules soaked in a 10% humic acid solution (bottom panel).

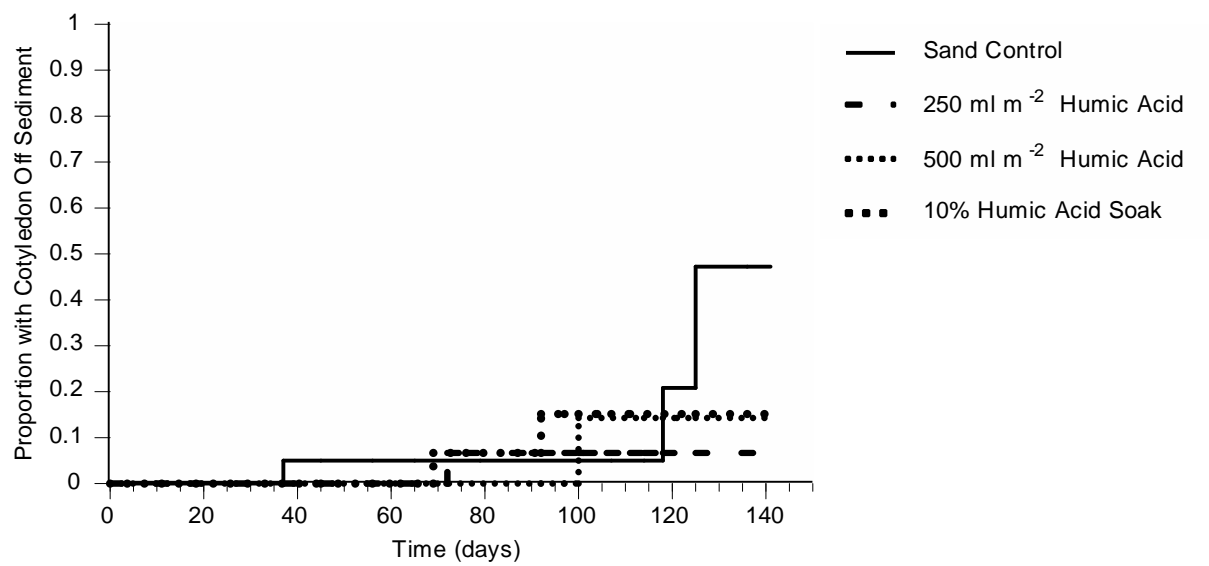


Figure 3.5 Kaplan-Meier curves of proportion of propagules with cotyledons lifted off of the sediment for propagules on sand, propagules on sand with 250 ml m⁻² humic acid added, propagules on sand with 500 ml m⁻² added, and propagules soaked in a 10% humic acid solution.

Created Marsh Sediment Experiment

All propagules died in this experiment without establishing. There was no significant effect of treatment. Kaplan-Meier curves revealed that there was a significant negative effect of humic acid application on survivorship in which survivorship was slightly lower for propagules on sediment with 250 ml m⁻² humic acid applied (Figure 3.6, top panel, $X^2 = 13.7441$, $p < 0.01$). There was no difference between treatments on the occurrence of fungus (Figure 3.6, bottom panel) or extension of the radicle for propagules (Figure 3.7).

Effect of Inundation on Hydromulch

In both treatments of immediate flooding and waiting 72 hours before flooding, hydromulch floated off the sand within five minutes of being flooded. No hydromulch remained on the sand.

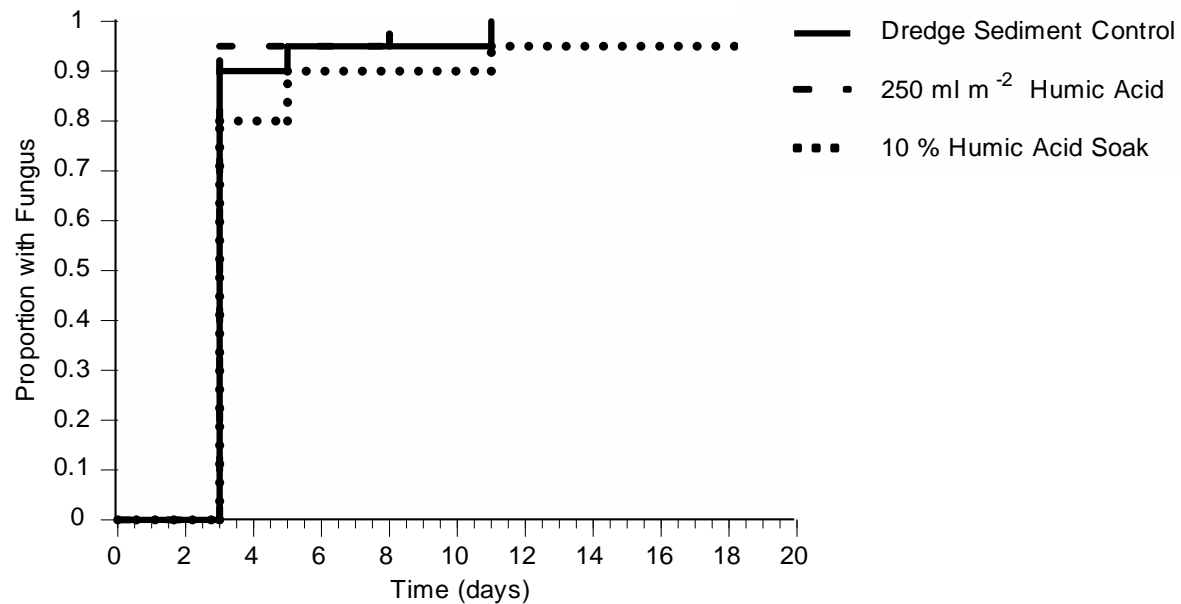
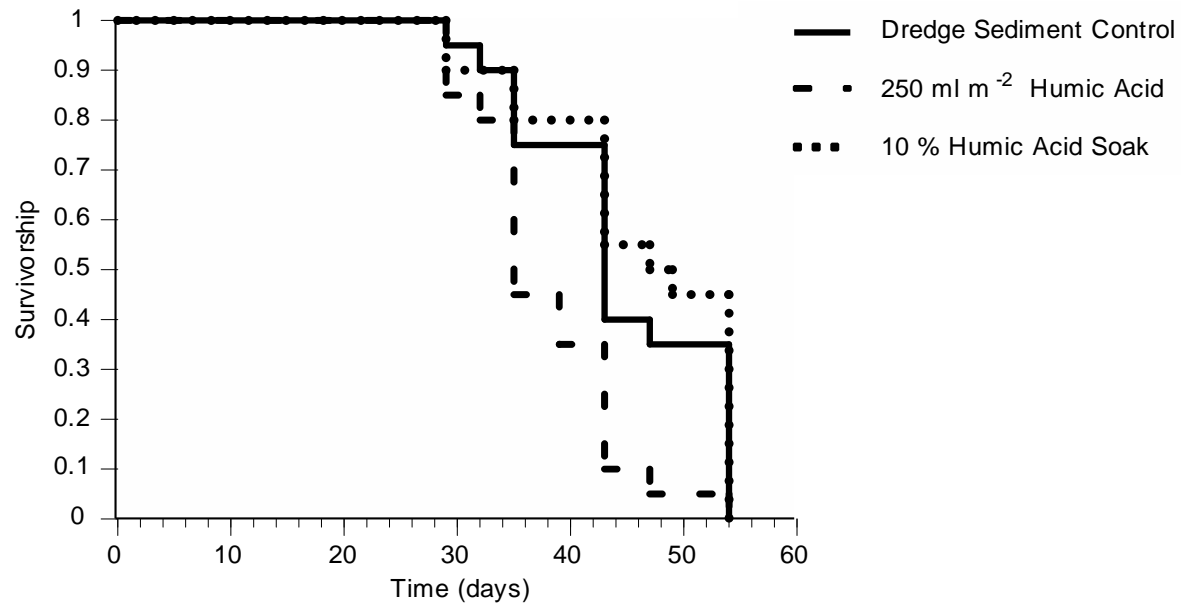


Figure 3.6 Kaplan-Meier curves of survivorship for propagules on created marsh sediment, propagules on created marsh sediment with 250 ml m⁻² humic acid added, and propagules soaked in a 10% humic acid solution and placed on created marsh sediment (top panel). Kaplan-Meier curves of proportion of propagules with fungus for propagules on created marsh sediment, propagules on created marsh sediment with 250 ml m⁻² humic acid added, and propagules soaked in a 10% humic acid solution and placed on created marsh sediment (bottom panel).

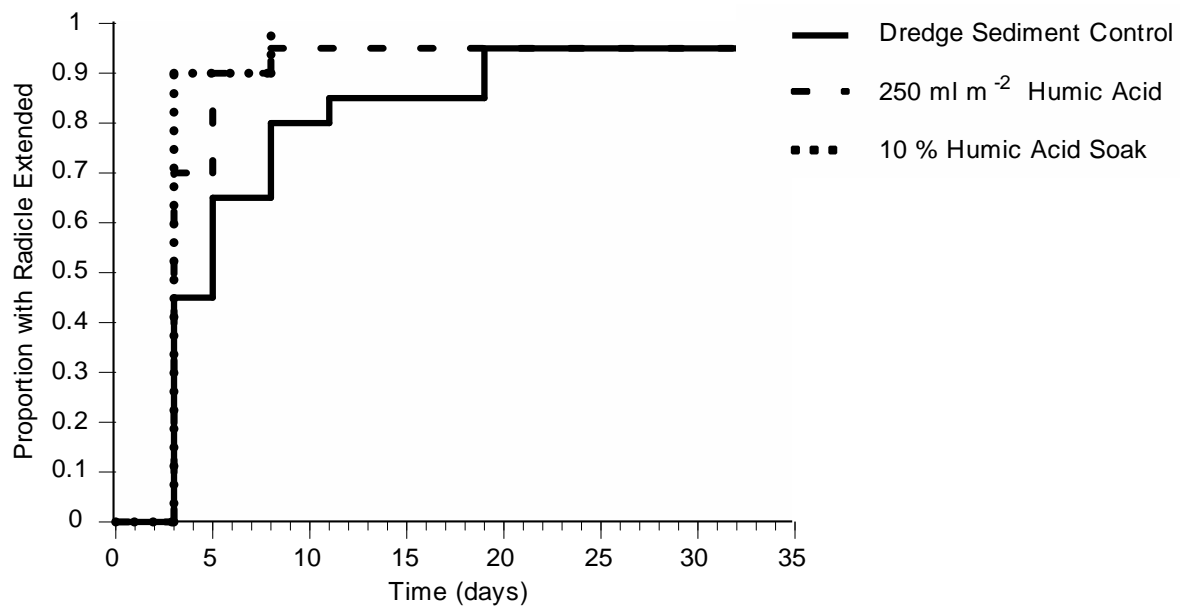


Figure 3.7 Kaplan-Meier curves of proportion of propagules with radicles extended for propagules on created marsh sediment, propagules on created marsh sediment with 250 ml m⁻² humic acid added, and propagules soaked in a 10% humic acid solution and placed on created marsh sediment.

Discussion

Hydromulch

Propagule survival and establishment were greatest with hydromulch application over propagules. This application approach likely prevented desiccation of propagules, which was the observed cause of death for propagules in this study, aside from fungal infection. In Belize, herbaceous vegetation can reduce soil temperature and salinity, resulting in greater mangrove growth than in bare areas (McKee et al. 2007). Similarly, salinity and soil temperature are lower within a Kenyan mangrove stand than in an adjacent bare area, resulting in greater recruitment of seedlings within the stand (Bosire et al. 2003). In our study, hydromulch may enable both propagules and sediment to retain moisture until propagules have established and emerge through the hydromulch layer.

Hydromulch application under propagules was not effective at improving survival and establishment. It is possible that the hydromulch layer created a physical barrier to rooting in the sediment. Observations of *Avicennia marina* distribution in southeastern Queensland suggested that propagules could establish before drying out with greater success in areas vegetated by a *Sarcocornia* sp., a low stature succulent plant with thin cover, than in areas vegetated by *Sporobolus* sp., which has a taller canopy and denser cover (Jones et al. 2004). Additionally, establishment of *A. marina* in southeastern Australia is reduced where the ground is covered by the macroalgae *Hormosira banksii* (Clarke and Myerscough 1993). In these two instances, the existing plants may hinder propagule roots from contacting the sediment, which may explain the effect of hydromulch applied under propagules.

Three important caveats exist for the use of hydromulch in salt marsh to improve *A. germinans* survival and establishment. First, the hydromulch disintegrates with flooding, and may not be effective after a high tide event. Second, excessive hydromulch cover over propagules can cause damage or death by smothering the propagules (Goforth and Williams 1984; Alleman and Hester 2011). Third, light is an important resource for *A. germinans* seedlings (McKee 1995), too much hydromulch cover may impede establishment and growth. Therefore, hydromulch application deserves further attention and development as a restoration tool, but within these considerations.

Humic Acid

Humic acid application, either percolated through sediment or as a soaking treatment, did not improve *A. germinans* establishment or survival. If humic acid were used to improve soil conditions for herbaceous salt marsh vegetation, it would not be expected to have a direct effect on *A. germinans*. Overall, humic acid application on sand did not differ from survival of propagules given the control treatment, which was only 20%. Propagules were extremely prone to fungus; however, propagules are able to survive even if they have some fungus on their cotyledons.

Created Marsh Sediment

The lack of *A. germinans* establishment on created marsh sediment demonstrates the difficulty of propagule establishment and survival in restoration field conditions. These propagules died within approximately six weeks of the study initiation, which highlights the

narrow window of establishment that exists for restoration efforts with this sediment type. We observed cracking of the sediment, likely due to the compaction and dewatering of the sediment. Interestingly, cracking of mudflats can support *A. germinans* colonization along the French Guiana coast. The mud cracks, which develop and degrade with wetting and drying cycles, trap the propagules, creating a coastal mangrove fringe (Fiot and Gratiot 2006). The propagules on sediment in our study, however, did not fall in cracks.

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CHAPTER 4. EFFECTS OF HUMIC ACID, SALINITY, AND SPECIES INTERACTIONS ON *AVICENNIA GERMINANS* AND *SPARTINA ALTERNIFLORA*

Introduction

A major component of successful restoration of back-barrier salt marsh is the establishment of the salt marsh plant community. For Louisiana barrier islands, the salt marsh plant community consists of smooth cordgrass (*Spartina alterniflora*) and black mangrove (*Avicennia germinans*) growing in association (Patterson et al. 1993; Courtemanche et al. 1999; Hester et al. 2005; Perry and Mendelssohn 2009). These two species are planted during restoration projects to stabilize the marsh sediment, that is, to reinforce or create back-barrier salt marsh (Khalil and Lee 2006). Optimizing facilitation should be considered before establishing these species together at a restoration site.

Restoration site conditions prior to vegetative establishment can be extremely stressful for plant establishment. The salt marsh platform of Whiskey Island was engineered to be at a targeted intertidal elevation within five years of construction (Green 2007), though vegetative plantings are initiated before optimal elevation is reached. In addition, characteristics of sediments at restoration sites include higher bulk density and lower soil moisture than those of natural back-barrier salt marshes (Fearnley 2008). Salt can also build up on the surface of sediments as water evaporates and sediments compact. Previously, drought has been a factor in the limited success of vegetative plantings at barrier islands (Khalil and Lee 2006). Therefore, techniques that assist in alleviating environmental and site stress, such as high salinity, may increase the success of restoration.

Humic acid is a soil amendment that may help to alleviate soil stress for plants. Low concentrations (0.05 g kg^{-1}) of humic acid application have been shown to increase available water holding capacity of Mediterranean soils due to hydrophilic groups that are part of the humic acid structure (Piccolo et al. 1996). Humic acids with low molecular size have been shown to improve plant growth, most probably through hormone-like activity (Nardi et al. 2002). In drought conditions, foliar application of humic acid increased shoot and root growth of tall fescue and shoot growth of creeping bentgrass (Zhang and Schmidt 2000). Asik et al. (2009) also reported a marginal increase in wheat (*Triticum durum*) biomass with the application of 1 g kg^{-1} under conditions of 60 mM NaCl . The application of humic acid to salt marsh species has recently been investigated. Willis and Hester (2010) reported enhanced growth of *S. alterniflora* after three months following humic acid application rates of $5\text{--}80 \text{ ml m}^{-2}$, but application did not have a similar effect on *A. germinans*. In addition, elevated salinity conditions (48 ppt) resulted in a lack of increased cumulative height for both species, which the humic acid application levels were not able to alleviate (Willis and Hester 2010).

Vegetative structure can facilitate the establishment and growth of woody species, including mangroves, in areas with stressful soil conditions. For example, density and vigor of *A. germinans* seedlings were greater in patches of *Distichlis spicata* than in bare ground at Twin Cays, Belize (McKee et al. 2007). The herbaceous canopy was effective at lowering soil temperatures and consequently reducing soil salinity at this site (McKee et al. 2007). Another halophyte, *Batis maritima*, is also associated with mangroves in Florida and Belize, and may

play a role in mangrove establishment (Whigham et al. 2009). Toledo et al. (2001) reported that *A. germinans* growth was greatest at an arid restoration site when seedlings were planted in clusters of two. Further, neighbor effects were positive for young *A. germinans* at Indian River Lagoon, FL, and neighbor effects were not expected to be negative until the mangrove stands matured (Rey 1994). The vegetative structure of *S. alterniflora* has been shown to reduce soil temperature, resulting in greater growth and survival of *Baccharis halimifolia*, a woody coastal species (Egerova et al. 2003). *Spartina alterniflora* may also protect *A. germinans* within its canopy from freeze damage (Lugo and Zucca 1977). Based on these studies, *S. alterniflora* may facilitate *A. germinans* in stressful environmental conditions.

The objective of this study was to determine whether inter- and intraspecific interactions of *A. germinans* and *S. alterniflora* are altered at moderate versus high salinity cross-classified with humic acid application treatments. Further, this study assessed the potential of humic acid to alleviate physiological stress for *A. germinans* and *S. alterniflora* at moderate and elevated salinity. Negative inter- and intraspecific interactions were predicted to be more prevalent at the moderate salinity, and positive inter- and intraspecific interactions were predicted to be more apparent at the high salinity treatment. With the addition of humic acid, it was expected that salinity stress would be ameliorated, particularly for *S. alterniflora*. Finally, biomass and cumulative height of *S. alterniflora* were predicted to be reduced relatively more than that of *A. germinans* at high salinity.

Materials and Methods

Effect of Humic Acid and Salinity on Species Interactions and Physiology

Avicennia germinans seedlings and *S. alterniflora* were obtained from a commercial nursery (Green Seasons Nursery, L.L.C.) and maintained at ambient light, temperature, and at salinity of 20 ppt (Instant Ocean, Spectrum Brands) at the Center for Ecology and Environmental Technology (CEET), at the University of Louisiana in Lafayette. An additive series design was employed to test for interspecific and intraspecific interactions between and among *A. germinans* and *S. alterniflora*. The design consisted of five vegetative treatments: one stem of *S. alterniflora*, two stems of *S. alterniflora*, one *A. germinans* seedling, two *A. germinans* seedlings, and one seedling of *A. germinans* with one stem of *S. alterniflora*. Treatments are hereafter referred to as SPAL alone, SPAL with SPAL, AVGE alone, AVEG with AVGE, and AVGE with SPAL. The additive series design (vegetation treatment) was completely cross-classified with two additional treatments: salinity and humic acid (4% humic acid derived from coal: Huma-Boost, 3 Tier Technologies, Longwood, FL) application. There were four replicates for each combination of treatments for a total of 80 experimental units, each within a 3.8 L pot containing river silt and accompanying individual water reservoir. The moderate (24 ppt) and high (48 ppt) salinity treatments were accomplished by adjusting artificial salt water in stepwise increments of 4–6 ppt per week. Humic acid (or an equivalent volume of tap water) was applied by percolation through the soil volume at the beginning of the study at two application rates, 0 ml m⁻² and 500 ml m⁻² humic acid. Water was conserved within individual reservoirs and maintained at a consistent water level 15 cm below the surface using tap water and adjusting salinity as needed. Plants were fertilized with 100 ml of 20% Hoaglands solution approximately every two months during the nongrowing season and every two weeks during the growing season. Porewater was collected from reservoirs, filtered using a 0.45 µm filter, acidified below

a pH of 2, and then submitted as samples for inductively coupled plasma optical emission spectroscopy (ICP-OES, Louisiana State University Soil Testing and Plant Analysis Laboratory) analysis to measure effects of treatments on relevant porewater elements.

For 15 months, cumulative height of both species was determined and subsequently analyzed with a repeated measures analysis of variance (Gotelli and Ellison 2004). Cumulative height of *A. germinans* was averaged in the AVGE-with-AVGE treatment to demonstrate intraspecific interactions. Prior to harvest, four replicate measurements of leaf chlorophyll content index (CCI) were performed with a chlorophyll meter (SPAD-502 Plus, Konica Minolta), and light-adapted quantum yield of photosystem II was determined using a fluorescence meter (FluorPen FP100, PSI). In order to compare chlorophyll content index values to chlorophyll concentrations in leaves, chlorophyll content index was also determined in additional leaves from the AVGE-with-SPAL treatment. Leaves were then collected, and actual chlorophyll content was determined using the methods from Biber (2007) and equations from Ritchie (2006). Xylem pressure potential of *S. alterniflora* leaves and *A. germinans* stems was determined immediately prior to biomass harvesting using a plant moisture vessel (Skye Instruments Ltd, Powys, UK). At harvest, a subsample of green leaf tissue was collected and then homogenized with a Wiley Mill and submitted to the LSU Soil Testing and Plant Analysis Laboratory for the determination of total carbon and nitrogen content following standard methods. For leaf elemental analysis, a subsample of leaf tissue was collected, dried to a constant weight, ground to pass through number 20 mesh using a Wiley Mill, and separated into two aliquots. The first aliquot was submitted to the LSU Soil Testing and Plant Analysis Laboratory for the determination of total carbon and nitrogen content following standard methods. At harvest all plants were rinsed of sediment and sorted by species into aboveground and belowground components, dried at 65°C to a constant weight, and weighed to determine biomass partitioning. Aboveground biomass of *A. germinans* was averaged in the AVGE-with-AVGE treatment to demonstrate intraspecific interactions; however, belowground biomass was not averaged. Data were analyzed with analysis of variance using JMP 9 (SAS Institute).

Results

Effect of Humic Acid and Salinity on Species Interactions and Physiology

Productivity of *A. germinans* responded to the vegetation treatment but not to the salinity and humic acid treatments. There was a highly significant effect of vegetation treatment on biomass of *A. germinans* in which aboveground biomass from the AVGE-alone treatment was greater than *A. germinans* biomass of all other vegetation treatments (Figure 4.1; $F = 7.8$, $p < 0.01$). Compared to the AVGE-alone treatment, biomass of *A. germinans* was reduced by 11% in the AVGE-with-SPAL treatment, and average biomass of *A. germinans* from the AVGE-with-AVGE treatment was reduced by 29%. Vegetation treatment also had a highly significant effect on belowground biomass, which was greater in the AVGE-with-AVGE treatment than in others (Figure 4.2; $F = 6.0$, $p < 0.01$). There was a significant effect of a three-way interaction between humic acid, salinity, and vegetation over time (Hunyh-Feldt $F = 2.6$, $p < 0.05$) and final cumulative height per plant (Figure 4.3; $F = 8.7$, $p < 0.01$) in which the height of *A. germinans* from the AVGE-alone treatment was greater than that of *A. germinans* from the AVGE-with-SPAL treatment within the 24 ppt, 500 ml m⁻² humic acid conditions.

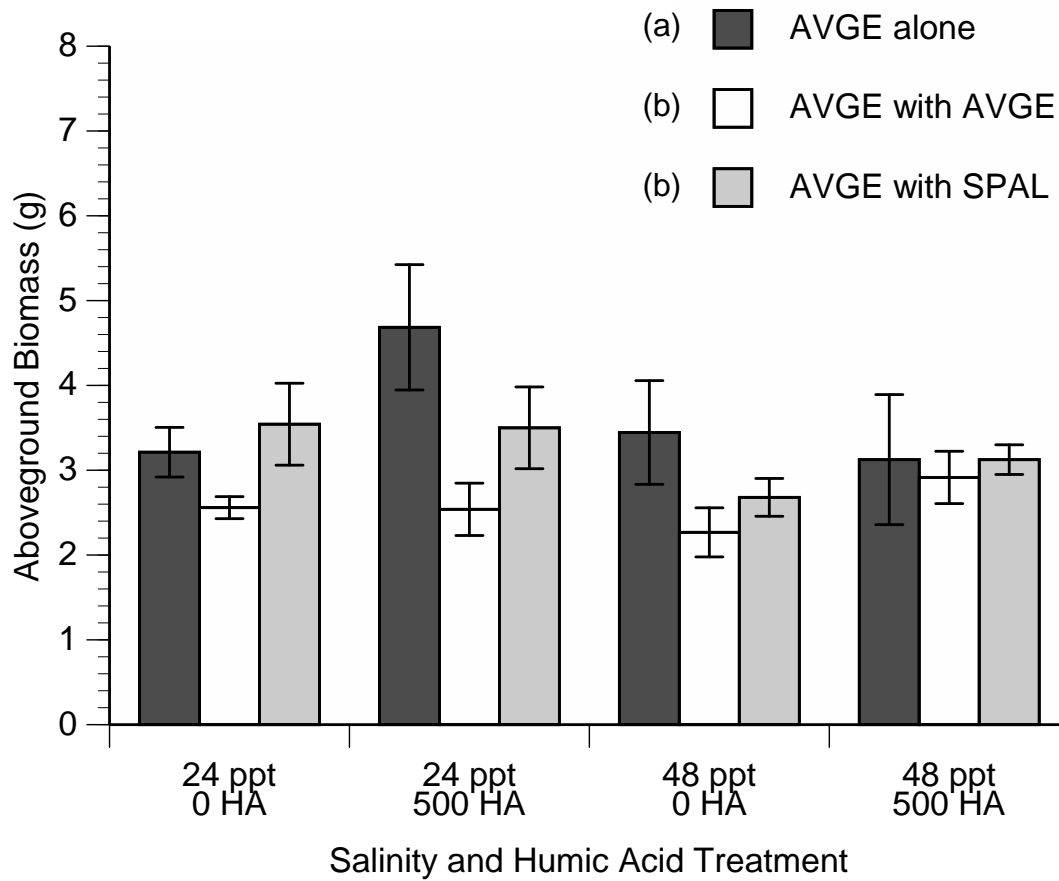


Figure 4.1 The effect of humic acid, salinity, and vegetation treatment on aboveground biomass of *Avicennia germinans* per plant (mean \pm SE, $n = 4$, LSD = 1.2).

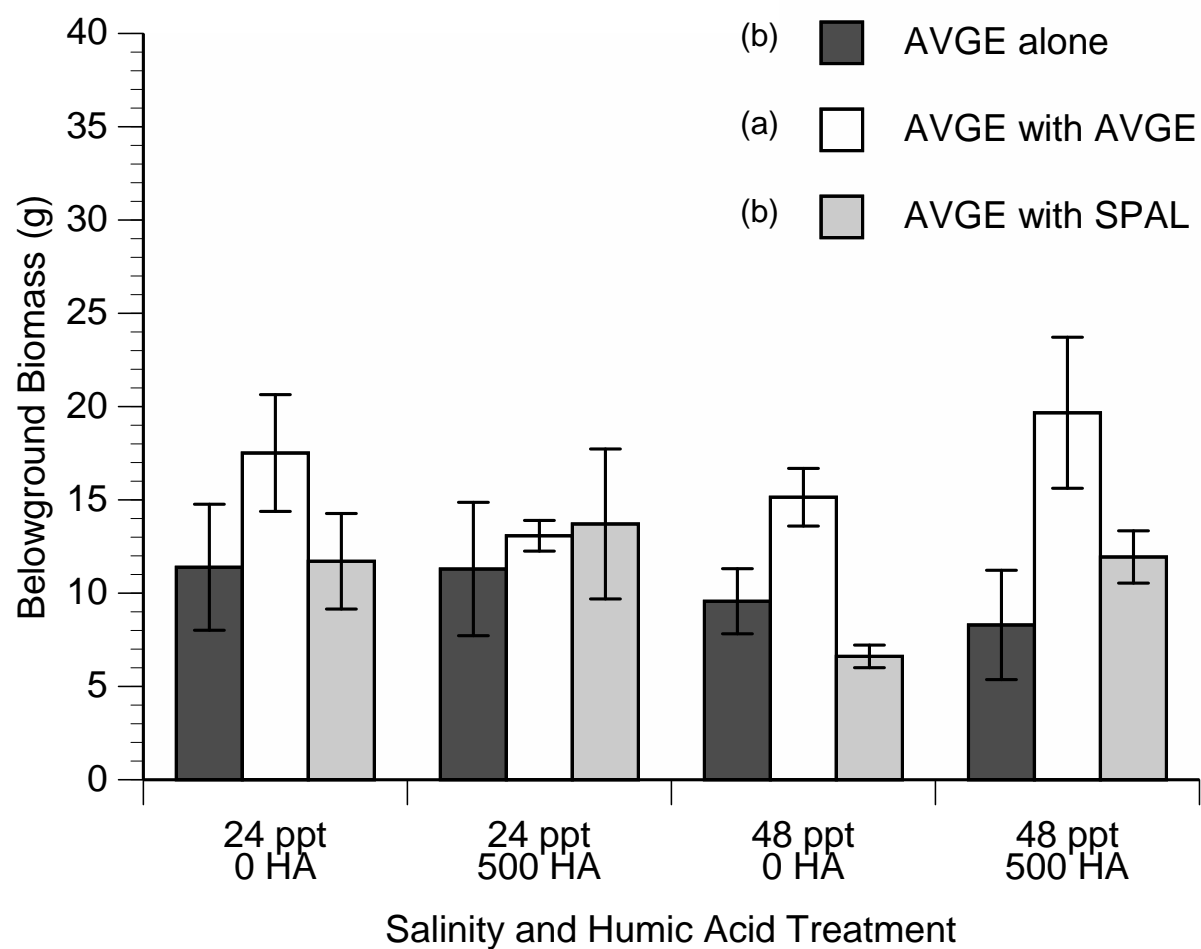


Figure 4.2 The effect of humic acid, salinity, and vegetation treatment on) belowground biomass of *Avicennia germinans* (mean \pm SE, $n = 4$, LSD = 7.9).

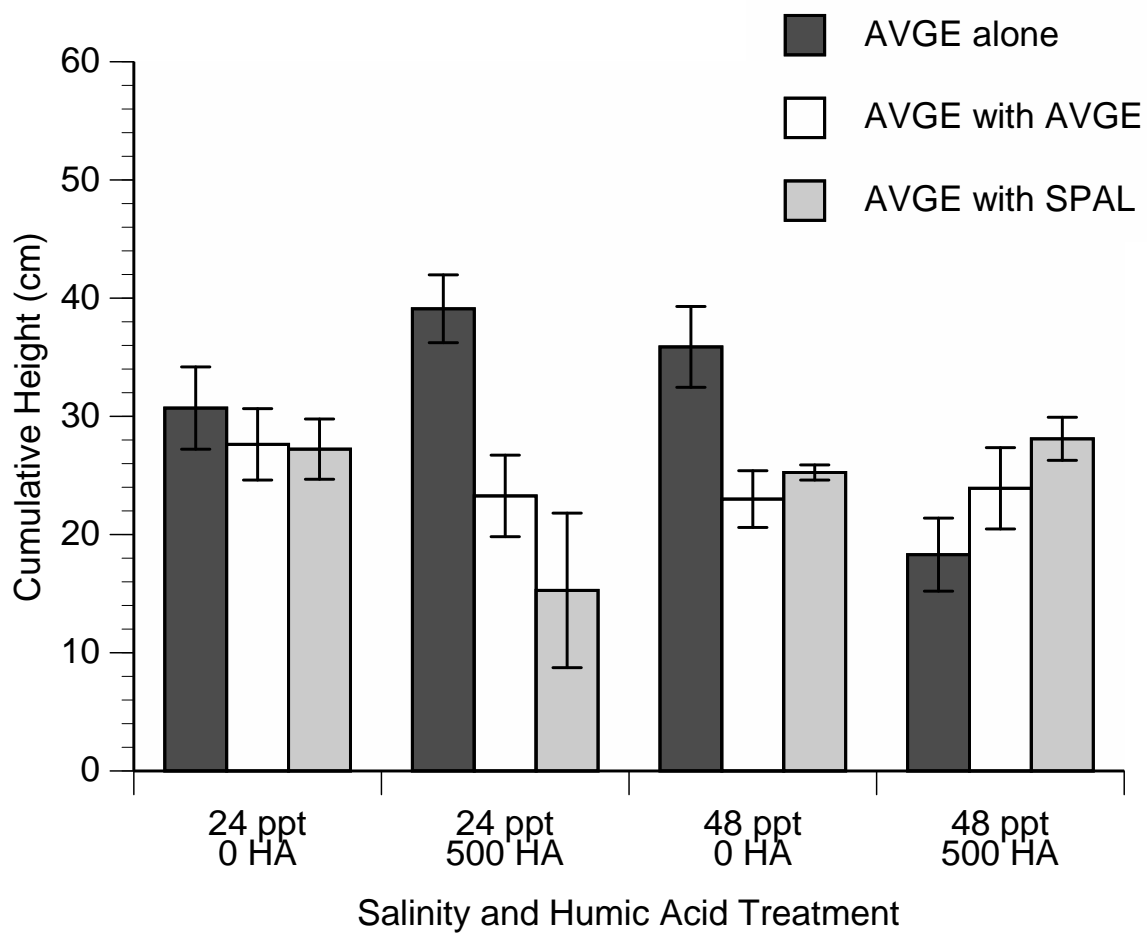


Figure 4.3 The effect of humic acid, salinity, and vegetation treatment on cumulative height of *Avicennia germinans* per plant (mean \pm SE, $n = 4$, LSD = 10.1).

The height of *A. germinans* with the AVGE-alone, 48 ppt salinity treatment was lower with the 500 ml m⁻² humic acid treatment than with the no-humic-acid treatment (Figure 4.3; $F = 9.9$, $p < 0.01$). Vegetation also had a significant effect on cumulative height per plant where the tallest *A. germinans* were those from the AVGE-alone treatment (Figure 4.3; $F = 5.2$, $p < 0.05$).

Humic acid, salinity, and vegetation treatments had highly significant effects on *S. alterniflora* growth responses. Aboveground biomass of *S. alterniflora* was reduced by 40% in the 48 ppt salinity treatment compared to the 24 ppt treatment (Figure 4.4; $F = 23.5$, $p < 0.01$). Alternately, addition of 500 ml m⁻² humic acid resulted in a 26% increase in aboveground biomass (Figure 4.4; $F = 7.5$, $p < 0.01$). The aboveground biomass of *S. alterniflora* was reduced by 37% in the AVGE-with-SPAL treatment compared to the SPAL-alone treatment. Within the SPAL-with-SPAL treatment, *S. alterniflora* aboveground biomass was only 32% greater than the biomass from the SPAL-alone treatment (Figure 4.4; vegetation effect: $F = 21.9$, $p < 0.01$). There were highly significant effects of interactions between humic acid and vegetation (Figure 4.5; $F = 2.7$, $p < 0.01$) as well as between salinity and humic acid (Figure 4.5; $F = 3.4$, $p < 0.01$) on belowground biomass of *S. alterniflora*. Specifically, the application of 500 ml m⁻² humic acid with any vegetation treatment resulted in belowground biomass similar to belowground biomass of SPAL-with-SPAL treatment without humic acid. In addition, the 24 ppt and 500 ml m⁻² humic acid treatments resulted in the greatest *S. alterniflora* belowground biomass. Over time, humic acid application had a significant effect on cumulative height (Hunyh-Feldt $F = 2.8$, $p < 0.05$), where height was greater with humic acid. In addition, both salinity (Hunyh-Feldt $F = 19.7$, $p < 0.01$) and vegetation (Hunyh-Feldt $F = 4.2$, $p < 0.01$) had highly significant effects on cumulative height over time, in which *S. alterniflora* was taller in the 24 ppt treatment and the shorter in the SPAL-with-AVGE treatment compared to the others. A significant positive effect of humic acid on cumulative height of *S. alterniflora* increased height by 28% when 500 ml m⁻² humic acid was applied (Figure 4.6; $F = 5.3$, $p < 0.05$). Salinity and vegetation treatments also had highly significant effects on cumulative height of *S. alterniflora*. Cumulative height of *S. alterniflora* stems was reduced by 54% in the 48 ppt salinity treatment compared to the 24 ppt treatment (Figure 4.6; $F = 45.1$, $p < 0.01$), and cumulative height was also significantly reduced in the AVGE-with-SPAL treatment (Figure 4.6, $F = 18.9$, $p < 0.01$).

Species, salinity, and vegetation treatments had an effect on physiological metrics, whereas humic acid did not affect physiological responses that we measured. Chlorophyll a content ($\mu\text{g Chl a ml}^{-1}$ 90% v:v acetone in distilled water) was positively correlated with chlorophyll content index (CCI) for both species (Table 4.1). The correlation was stronger for *A. germinans* ($r^2 = 0.63$, $p < 0.01$) than for *S. alterniflora* ($r^2 = 0.3384$, $p = 0.0229$). Mean CCI of *A. germinans* was 51.7 ± 1.1 and was significantly greater than the mean CCI for *S. alterniflora*, 38.5 ± 1.4 (Table 4.1; $F = 58.9$, $p < 0.01$). Quantum yield of photosystem II (QY) from light-adapted leaves was significantly larger for *S. alterniflora* than for *A. germinans* (Table 4.1; $F = 44.8$, $p < 0.01$). The elevated (48 ppt) treatment had a highly significant negative effect on QY of *S. alterniflora* and resulted in an 8.5% decrease in QY compared to the 24 ppt treatment (Table 4.1; $F = 7.3$, $p < 0.01$). The C:N ratio of *S. alterniflora* was greater than that of *A. germinans* (Table 4.1; $F = 75.1$, $p < 0.01$), and both species had reduced C:N ratios with the high salinity treatment (Table 4.1; $F = 46.5$, $p < 0.01$).

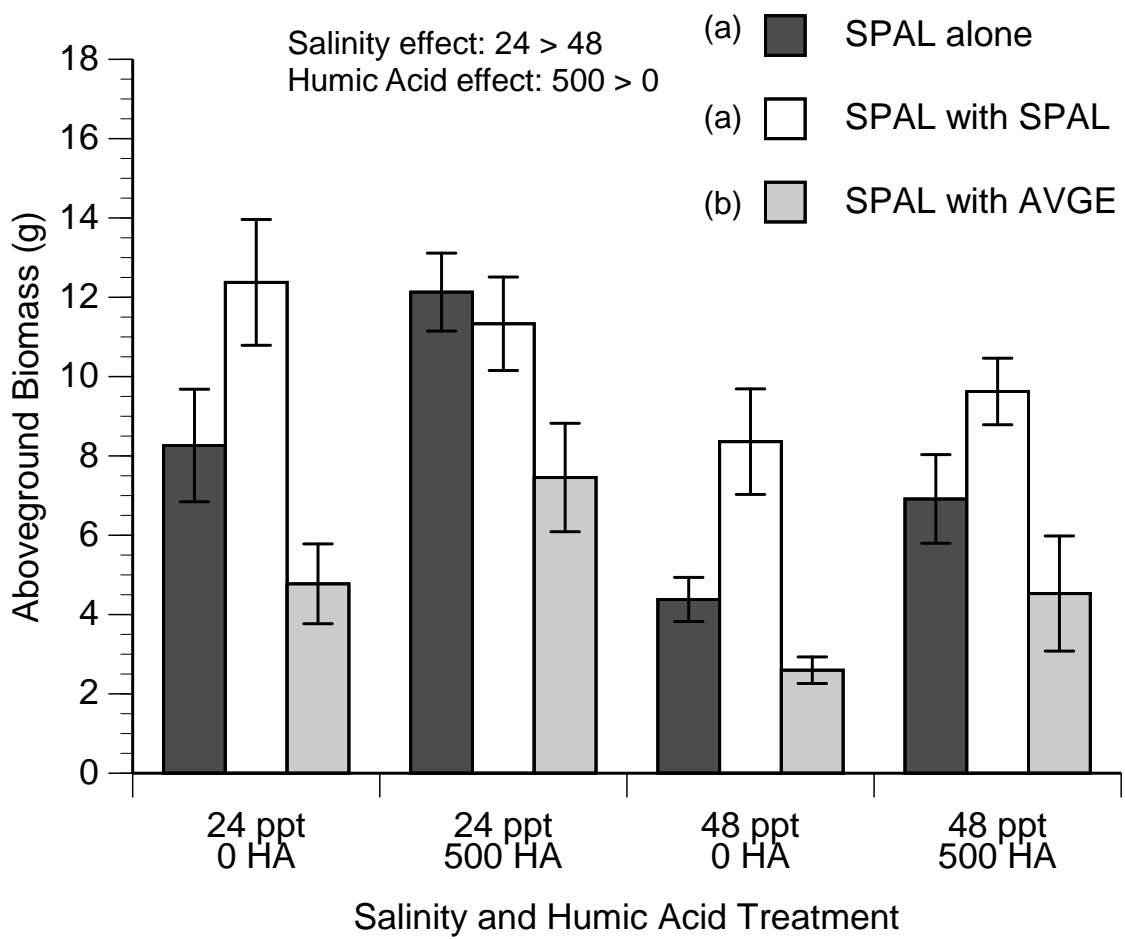


Figure 4.4 The effect of humic acid, salinity, and vegetation treatment on aboveground biomass of *Spartina alterniflora* (mean \pm SE, $n = 4$, LSD = 3.4).

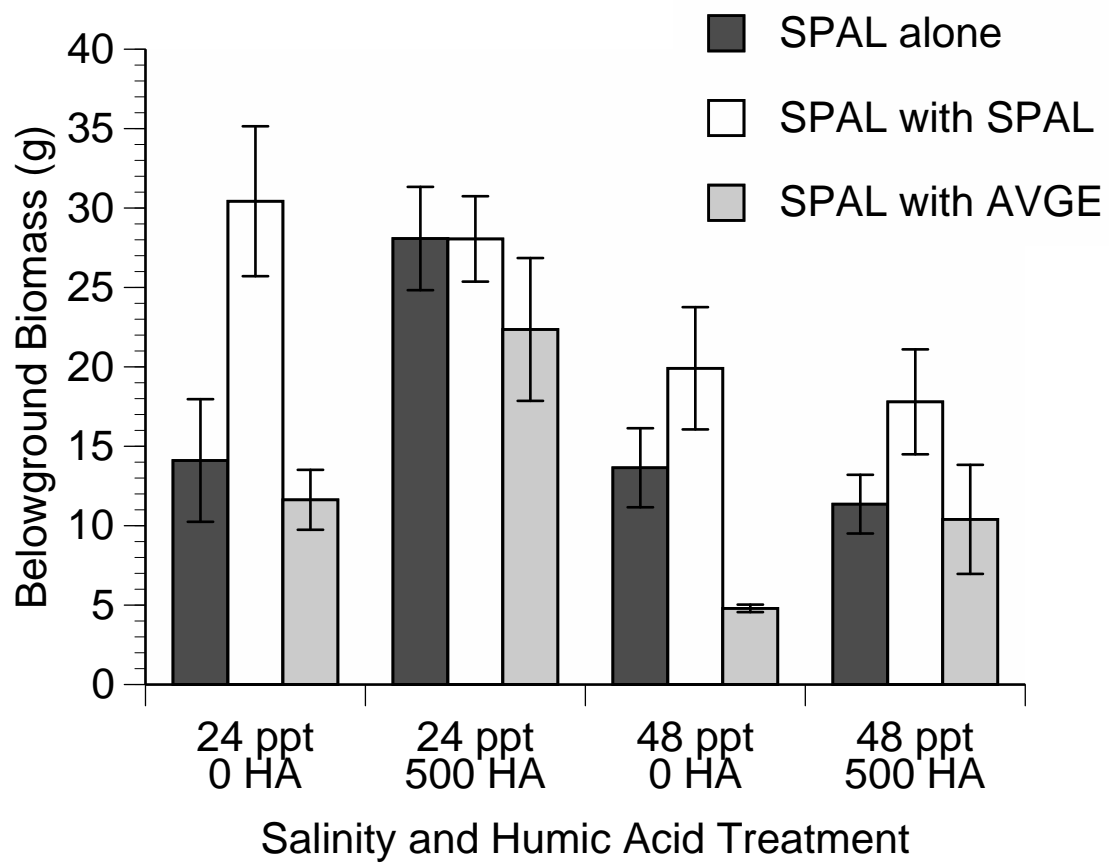


Figure 4.5 The effect of humic acid, salinity, and vegetation treatment on belowground biomass of *Spartina alterniflora* (mean \pm SE, $n = 4$, LSD = 9.3).

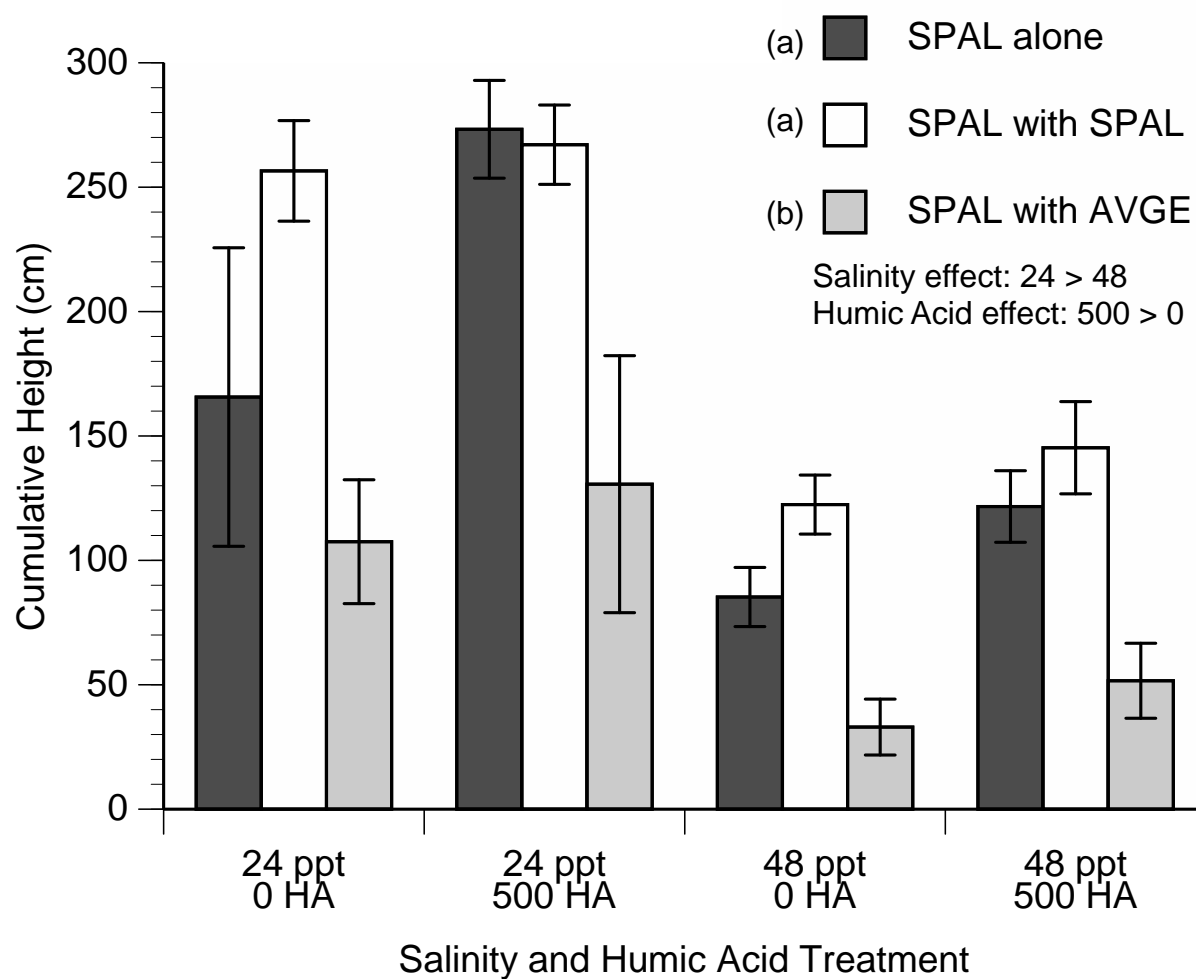


Figure 4.6 The effect of humic acid, salinity, and vegetation treatment on cumulative height of *Spartina alterniflora* (mean \pm SE, $n = 4$, $LSD = 79.1$).

Table 4.1 Foliar measurements including chlorophyll a as a function of chlorophyll content index (CCI), CCI, light-adapted quantum yield (QY), and C:N ratio for *A. germinans* and *S. alterniflora* under moderate and elevated salinity.

Measurement	Salinity (ppt)	Species	
		<i>Avicennia germinans</i>	<i>Spartina alterniflora</i>
Chl a relationship with CCI		Chl a ($\mu\text{g ml}^{-1}$) = $0.34 (\text{CCI}) - 10.63$	Chl a ($\mu\text{g ml}^{-1}$) = $0.12 (\text{CCI}) - 1.92$
CCI			
	24	$52 \pm 2^{\text{A}}$	$39 \pm 1^{\text{B}}$
	48	$52 \pm 2^{\text{A}}$	$38 \pm 3^{\text{B}}$
Light-adapted QY			
	24	$0.44 \pm 0.02^{\text{B}}$	$0.56 \pm 0.01^{\text{Aa}}$
	48	$0.43 \pm 0.02^{\text{B}}$	$0.51 \pm 0.01^{\text{Ab}}$
C:N			
	24	$41.0 \pm 0.9^{\text{Ba}}$	$50.5 \pm 1.1^{\text{Aa}}$
	48	$34.1 \pm 0.7^{\text{Bb}}$	$42.9 \pm 1.6^{\text{Ab}}$

Xylem pressure potential measurements of *S. alterniflora* from the 48 ppt salinity treatment generally reached the lowest potential measureable by the device (-4 MPa) for the leaf blade shape, and very likely had xylem pressure potential more negative than -4 MPa. In a significant interaction between species, vegetation, and salinity on xylem pressure potential, the least negative value (-1.50 ± 0.29 MPa) was recorded for *A. germinans* from the AVGE-with-SPAL and 24 ppt salinity treatment, and the most negative xylem pressure potential (-4.27 ± 0.37 MPa) recorded was from *A. germinans* in the AVGE-with-SPAL and 48 ppt salinity treatment (Figure 4.7; $F = 4.7$, $p < 0.05$). There was also a significant interaction between salinity and vegetation treatment in which xylem pressure potential was less negative for both species in the 24 ppt salinity treatment with the single (AVGE- or SPAL-alone) and interspecific (AVGE-with-SPAL) vegetation treatments compared to both species in the interspecific, 48 ppt treatment (Figure 4.7; $F = 4.5$, $p < 0.05$). Species and salinity had highly significant effects on xylem pressure potential in which values were less negative for *A. germinans* when compared to *S. alterniflora* (Figure 4.7; $F = 35.6$, $p < 0.01$), and less negative in the 24 ppt salinity treatment (Figure 4.7; $F = 13.3$, $p < 0.01$).

In a highly significant effect of salinity on porewater potassium, the elevated salinity treatment resulted in greater phosphorus (Table 4.2; $F = 7.1$, $p < 0.01$). In addition, there was a highly significant three-way interaction of salinity, vegetation, and humic acid treatments on porewater potassium (Table 4.2; $F = 4.5$, $p < 0.01$). The treatment combination of elevated salinity, no humic acid, and AVGE-with-AVGE vegetation resulted in the greatest amount of potassium, which was significantly greater than that produced by any moderate salinity treatment or elevated salinity treatments of no humic acid and SPAL-only, SPAL-with-SPAL, and 500 ml m^{-2} humic acid and SPAL-with-SPAL or AVGE-only. There was also a significant negative effect of humic acid addition on potassium (Table 4.2; $F = 5.3$, $p < 0.05$) and a highly significant positive effect of salinity on potassium (Table 4.2; $F = 696.9$, $p < 0.01$).

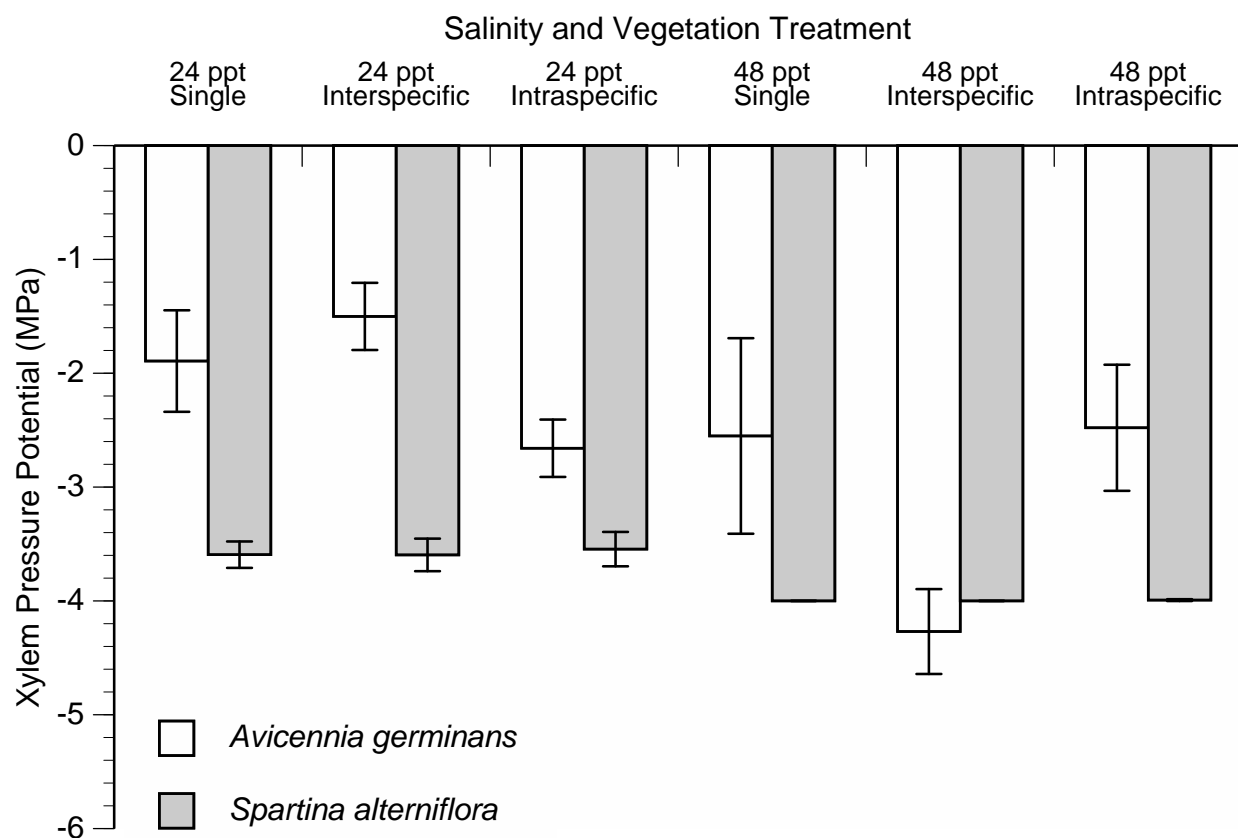


Figure 4.7 The effect of humic acid, salinity, and vegetation treatment on pressure potential of *Avicennia germinans* and *Spartina alterniflora* (mean \pm SE, $n = 4$, *Avicennia germinans* LSD = 2.3, *Spartina alterniflora* LSD = 0.6) xylem. For vegetation treatments, *Single* indicates AVGE or SPAL, *Interspecific* indicates AVGE-with-SPAL, and *Intraspecific* AVGE-with-AVGE or SPAL-with-SPAL, respective of the species.

Table 4.2 The effect of vegetation treatment, salinity, and humic acid on phosphorus and potassium.

Vegetation	Treatment		Phosphorus ($\mu\text{g g}^{-1}$)		Potassium ($\mu\text{g g}^{-1}$)	
	Salinity	Humic Acid	Mean	$\pm SE$	Mean	$\pm SE$
AVGE	24 ppt	0 ml m ⁻²	0.085	(0.033)	255.0	(28.5)
		500 ml m ⁻²	0.071	(0.021)	229.1	(34.4)
	48ppt	0 ml m ⁻²	0.101	(0.024)	770.7	927.4)
		500 ml m ⁻²	0.081	(0.030)	612.0	(54.8)
AVGE with AVGE	24 ppt	0 ml m ⁻²	0.073	(0.017)	217.3	(31.8)
		500 ml m ⁻²	0.045	(0.003)	281.0	(24.9)
	48ppt	0 ml m ⁻²	0.098	(0.015)	874.2	(77.2)
		500 ml m ⁻²	0.077	(0.008)	700.6	(40.6)
AVGE with SPAL	24 ppt	0 ml m ⁻²	0.071	(0.021)	270.9	(28.8)
		500 ml m ⁻²	0.060	(0.009)	224.3	(9.7)
	48ppt	0 ml m ⁻²	0.103	(0.012)	685.9	(28.6)
		500 ml m ⁻²	0.081	(0.005)	679.4	(55.1)
SPAL with SPAL	24 ppt	0 ml m ⁻²	0.049	(0.006)	220.1	(26.1)
		500 ml m ⁻²	0.051	(0.006)	205.4	(5.1)
	48ppt	0 ml m ⁻²	0.130	(0.032)	656.5	(24.6)
		500 ml m ⁻²	0.230	(0.116)	650.4	(36.0)
SPAL	24 ppt	0 ml m ⁻²	0.074	(0.004)	252.8	(44.8)
		500 ml m ⁻²	0.047	(0.005)	158.1	(31.8)
	48ppt	0 ml m ⁻²	0.139	(0.045)	660.1	(59.4)
		500 ml m ⁻²	0.085	(0.010)	745.7	(42.9)
LSD			0.101		109.0	

Discussion

Humic Acid Benefit for S. alterniflora

Humic acid application resulted in an increase of *S. alterniflora* growth at both moderate and high salinity treatments in our study. Willis and Hester (2010) also noted an increase in *S. alterniflora* cumulative height within three months after humic acid application rates ranging from 5 – 80 ml m⁻², whereas this study showed an increase in cumulative height at five months with a 500 ml m⁻² application rate. Further, belowground biomass of *S. alterniflora* increased with humic acid addition in this study, which is in agreement with other studies that reported enhanced root development (i.e., lateral roots and root hairs) with humic acid, probably due in part to hormone-like activity of humic acid (reviewed by Trevisan et al. 2010).

Contrary to expectations, no significant interactions occurred between salinity and humic acid on *S. alterniflora* aboveground biomass or cumulative height. There was a salinity and humic acid interaction on belowground biomass; however, humic acid addition resulted in greater belowground biomass at the moderate salinity rather than the elevated salinity. Further, the addition of humic acid did not improve water status of *S. alterniflora*, indicating that humic acid was not alleviating physiological drought symptoms caused by high salinity. These results are in agreement with the positive effect of humic acid application for wheat at control and salinity treatments, though there was no salinity and humic acid interaction (Asik et al. 2009). Additionally, Piccolo et al. (1996) suggested that the ability of humic acid to increase water-holding capacity is dependent on sediment characteristics; therefore, in conditions with high salinity, sediment characteristics may limit the benefit of humic acid.

Humic acid did not have an effect on biomass, cumulative height, or physiological parameters of *A. germinans*. Similarly, after five months, 5 – 80 ml m⁻² application rates did not result in increased height or biomass of *A. germinans* subjected to moderate (24 ppt) or elevated (48 ppt) salinity (Willis and Hester 2010). Humic acid salts have been previously investigated for their effectiveness at removing pyrene in mangrove microcosms using a 6.7% w:w addition (humic acid to sediment), which resulted in approximately 50% reduction in biomass after six months (Ke et al. 2003). High concentrations of humic acid may be toxic to mangrove species, though the humic acid application concentration of 500 ml m⁻² in this study did not result in apparent detrimental effects.

Interspecific and Intraspecific Interactions

Negative interspecific and intraspecific interactions were evident in this study. The growth of *A. germinans* seedlings was negatively affected by their being planted in pairs or with *S. alterniflora*. This result was unexpected since a previous study demonstrated improved growth when *A. germinans* was planted in pairs. In that case, though, mangrove pairs were planted with at least 1 m of open area surrounding them and sediments were very dry (Toledo et al. 2001), unlike this greenhouse study, where sediments remained saturated. The presence of an additional plant within the experimental unit may have created or intensified a resource limitation. For example, light availability is an important resource for *A. germinans*, and it has been shown to limit growth (McKee 1995a) and explain seedling density patterns in Belize (McKee 1995b). An additional plant in the experimental unit may have partially shaded seedlings in this study. Space may have also been a limiting factor, resulting in negative plant

interactions; biomass of *S. alterniflora* in the optimum salinity and humic acid conditions of 24 ppt and 500 ml m⁻² humic acid did not surpass approximately 12 g, regardless of one or two stems of *S. alterniflora* having initially been planted in an experimental unit. Further, the growth of *S. alterniflora* was reduced when planted with *A. germinans*, compared to treatments without *A. germinans*. The results of this study suggest that competitive interactions between *A. germinans* and *S. alterniflora* observed in the field (Patterson et al. 1993) are not alleviated.

Salinity Tolerance

Both *S. alterniflora* and *A. germinans* occupy the salt marsh niche on barrier islands, with *S. alterniflora* at lower elevations with greater flooding and *A. germinans* at higher elevations with greater salinity (Hester et al. 2005). This study supports that zonation pattern due to the greater salt tolerance demonstrated by *A. germinans*. Leaves of *A. germinans* had more chlorophyll, a quantum yield of photosystem II that was insensitive to salinity, and xylem pressure potential that was not as low as that of *S. alterniflora*, suggesting that *A. germinans* invests energy in resources supporting photosynthesis and adaptations to salinity stress. In the hypersaline (33–55 ppt) conditions of a mangrove scrub forest in Florida, leaf nitrogen and photosynthesis increased for *A. germinans* when it was fertilized with nitrogen, which further demonstrates the ability of this species to utilize available resources when salinity stress is high (Lovelock and Feller 2003). Leaf xylem potential was even more reduced (-5 MPa) for *A. germinans* growing in a hypersaline (~60 ppt) basin in Puerto Rico, though rates of photosynthesis for those plants were comparable to other mangrove stands (Lugo et al. 2007).

The ability of *S. alterniflora* to selectively exclude and secrete Na⁺ more so than K⁺ (Bradley and Morris 1991), in addition to the ability to increase production of proline and glycine betaine with increased salinity (Hester et al. 2001), support the survival and growth of this species in highly saline conditions, such as that of this study. However, quantum yield of photosystem II and xylem pressure potential, C:N ratio, height, and biomass were reduced for *S. alterniflora* with elevated salinity in our study. Hester et al. (2001) similarly demonstrated decreases in net CO₂ assimilation and xylem pressure potential of *S. alterniflora* with 30 ppt salinity conditions when compared to control (1 ppt) conditions. In addition, conditions of high salinity and drought have previously been shown to result in lower survival and chlorosis of leaves for *S. alterniflora* (Brown et al. 2006).

Restoration Implications

These results demonstrate a benefit of humic acid application for *S. alterniflora* within a controlled greenhouse setting. A potential way to use humic acid for restoration projects may be to incorporate humic acid while growing *S. alterniflora* transplants for restoration projects. Larger plants may increase survival at stressful restoration sites without major changes to plant tissue C:N ratios. Further, both species have positive attributes for restoration: *S. alterniflora* grows faster, supported by greater quantum yield efficiency, though *A. germinans* has greater salt tolerance. Allowing more space between individuals may alleviate the negative interaction between these species.

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FIELD STUDIES

CHAPTER 5. FIELD TRIALS OF THE UTILITY OF INCREASED PLANTING DENSITY, HUMIC ACID AMENDMENT AND FERTILIZER APPLICATION FOR ENHANCED BARRIER ISLAND PLANTING SUCCESS

Introduction

Barrier islands and headlands are valuable components of the coastal zone that contain several unique habitats, including the dune and swale, as well as back-barrier marshes (Hester et al. 2005). Through these habitats, barrier islands provide a number of important ecosystem functions, including the provision of valuable fisheries and wildlife habitat and the reduction of wave and storm energies to inland marshes and human communities (Dahl and Woodward 1977; Swilling et al. 1997; Stone and McBride 1998; Miller et al. 2001). Because of the large expenses associated with the placement of appropriate material for dune and back-barrier marsh creation, the restoration of barrier island habitats is extremely costly (Khalil et al. 2010). Subsequent to the creation of these habitats, through placement of appropriately sized sediment, the successful maintenance of the newly created habitats is predicated on the successful establishment and expansion of vegetation (Seneca and Cooper 1971; Maun and Krajnyk 1989; Mendelssohn and Hester 1991; Hester and Mendelssohn 1992). The presence of healthy vegetation adapted to these harsh environments stabilizes the recently introduced sediments through a variety of mechanisms, including sand binding and the reduction of wind and water velocity (Morton 2002). Given the high cost of sediment placement for barrier island restoration efforts and the crucial role of vegetation in retaining this material, the exploration of techniques to facilitate the rapid establishment and expansion of appropriate coastal vegetation in these habitats is warranted.

Coastal dunes are stressful environments in which a number of factors, including salt spray, shifting sands, limited water and nutrient availability, and overwash events, act to reduce vegetative establishment, abundance, and growth (Barbour et al. 1985; Hester and Mendelssohn 1992; Courtemanche et al. 2001). The exceptionally stressful nature of these environments in terms of plant establishment and expansion, particularly when compounded by the often marginal nature of dredged sand material, makes the selection of appropriate plant species during restoration planning critical (Mendelssohn and Hester 1988; Mendelssohn et al. 1991; Hester and Mendelssohn 1992). In particular, coastal dunes exhibit reduced nutrient and water availability mainly because of the low organic matter content of these sandy soils. Although these stressors present a major challenge to successful establishment of coastal vegetation (Barbour et al. 1985; Mendelssohn and Hester 1985; Hester and Mendelssohn 1990), the impact of these stressors is often reduced once plants have established through the incorporation of organic matter produced by root expansion and aboveground productivity (Stevenson and Day 1996; Dilustro and Day 1997). Thus, barrier island restoration efforts utilizing plant species capable of growing in these harsh environments have the greatest likelihood of success.

Uniola paniculata (sea oats) and *Panicum amarum* (bitter panicum) are two primary dune grasses employed to stabilize dune habitats throughout the Gulf of Mexico and southern Atlantic coasts (Dahl and Woodard 1977; Mendelssohn and Hester 1988; Mendelssohn et al. 1991; Hester and Mendelssohn 1991; Hester and Mendelssohn 1992; Miller et al. 2001; Snyder and Boss 2002; Gormally and Donovan 2010; Lonard and Judd 2011; Lonard et al. 2011). Both species

are highly adapted to the harsh conditions that occur in coastal dune environments, including shifting sands, salt spray, high salinity, and low water and nutrient availability (Wagner 1964; Miller et al. 2001). Additionally, *U. paniculata* and *P. amarum* produce extensive fibrous root systems that stabilize sand (Hester and Mendelssohn 1989; Miller et al. 2001; Lonard et al. 2011; Lonard and Judd 2011). *Uniola paniculata* and *P. amarum* are also less susceptible to damage from sand burial than some other dune species, such as *Paspalum vaginatum* (Mendelssohn et al. 1991). Additional plant species that are currently employed in the restoration of rear dune and swale habitats in Louisiana include marshhay cordgrass (*Spartina patens*), seashore paspalum (*P. vaginatum*), saltgrass (*Distichlis spicata*), seashore dropseed (*Sporobolus virginicus*), and seashore bluestem (*Schizachyrium maritimum*). Interestingly, Zong et al. (2010) characterized the capacity of several warm-season turf grasses, including *P. vaginatum* and *Cynodon dactylon*, for use in a coastal beach area of Jiangsu Province, China, using multiple indexes and found that *P. vaginatum* consistently performed the best across all indexes, whereas the performance of *C. dactylon* was consistently the lowest.

Employing higher planting densities of individual plants during vegetation establishment efforts has been shown to have both positive (Sheridan et al. 1998; Budelsky and Galatowitsch 2000) and negative effects (Brown and Rice 2000; Huddleston and Young 2004) on restoration effectiveness. In stressful environments, higher plant densities can result in intraspecific facilitation, increasing overall survivorship and growth relative to lower plant densities (Fajardo and McIntire 2011). Higher density plantings of native grasses have been shown to be effective in a California grassland restoration effort in which long-term benefits of higher native grass density and lower non-native grass density were observed (Lulow 2007). The use of higher density plantings during restoration efforts enhances the provision of ecosystem services, such as reducing erosive forces, even over short time intervals. For instance, increased planting density of vegetation has been shown to result in a concomitant increase in soil shear strength by five weeks, which was attributed to fibrous root reinforcement of soils (Loades et al. 2010). Also, the density of planting for the dune grass *Ammophila arenaria*, along with nitrogen fertilizer application, has been demonstrated to be of great importance for the rapid binding of sand (Brown and Hafenrichter 1948b). Brown and Hafenrichter (1948b) specifically found that high-density planting spacing in conjunction with 40 lbs N acre⁻¹ was an optimally efficient approach, accumulating sand to a depth of 0.28 feet *Ammophila arenaria*.

Due to the nutrient-poor content of dune sands (Tackett and Craft 2010), the use of a fertilization regime is normally incorporated into dune planting restoration projects (Van der Valk 1974; Broome et al. 1982; Mendelssohn and Hester 1988). Broadcast fertilization regimes have been used successfully for dune and swale plant restorations in states other than Louisiana (Brown and Hafenrichter 1948c; Broome et al. 1982) and in non-state-sponsored planting efforts (Mendelssohn and Hester 1988; Mendelssohn et al. 1991; Hester and Mendelssohn 1992). However, no specified fertilization regime is currently implemented in dune and swale plantings undertaken by the State of Louisiana. Importantly, the primary dune-building grasses along the coast of the Gulf of Mexico, *U. paniculata* and *P. amarum*, are both known to respond well to macronutrient fertilizer application (Broome et al. 1982). *Spartina patens*, an important plant species in swale habitats has been shown to respond favorably to nitrogen addition (Sistani and Mays 2001), although under elevated nutrient conditions it can be outcompeted in dune environments (Day et al. 2004). Micronutrient limitation is typically not a concern in dune and

swale systems (Hester and Mendelssohn 1990; Mendelssohn et al. 1991), most likely because of the micronutrients contained in salt spray (Boyce 1954; Van der Valk 1974).

The use of naturally derived products (e.g., liquid composts and humic acid extracts) to improve poor quality soils has been a field of increasing study, primarily in regard to marginal agricultural soils and horticultural applications. As described in previous chapters, humic acid, which is an operationally defined fraction of organic matter that is insoluble at a pH less than 2, but soluble at higher pH levels (Zhang et al. 2002), has been shown to be an effective soil conditioner. Soil nitrogen and phosphorus levels can be increased through the application of humic acid (Brannon and Somers 1985; Sharif et al. 2002). The assimilation of these nutrients by vegetation can also be increased (Fagbenro and Agboda 1993), very likely due to alterations in cell permeability (Valdrighi et al. 1996). Humic acids extracted from soil often have a total nitrogen content of about 4% (Sparks 2003), and as nitrogen becomes available through microbial activity, this nitrogen pool can be thought of as emulating a slow-release nitrogen fertilizer (Nisar and Mir 1989; Sharif et al. 2002). Several agricultural species, including *Hordeum vulgare* L. (barley) and *Avena sativa* L. (oat) seedlings, have shown enhanced nitrate uptake with humic acid amendment (Maggioni et al. 1987; Nisar and Mir 1989). Because of its chelating properties, humic acid can also play a substantial role in rendering micronutrients such as iron available and enhancing their uptake (Stevenson 1982; Chen and Aviad 1990; Stevenson 1991; Spark et al. 1997; Pinton et al. 1998; Pinton et al. 1999). Adani et al. (1998) investigated humic acid application on *Lycopersicon esculentum* (tomato) and found increases in N, P, and Fe uptake and biomass production.

Environmental stressors, such as physiological drought, elevated salinity, and heat stress, often disrupt metabolic processes and lead to the formation of free radicals, which in turn impact crucial cellular processes (Andivia et al. 2012). For example, several environmental stressors can disrupt photosynthetic processes, leading to the formation of radical oxygen species (Scandalios 1997; Smirnoff 1995) that cause enzyme inactivation and lipid peroxidation, thereby damaging plant cells and reducing plant health (Price and Hendry 1989; Quatacci and Navari Izzo 1992). The production of antioxidants to ameliorate impacts to metabolic activities may be one mechanism by which coastal plants species such as *S. alterniflora* thrive in coastal environments (Husband et al. 2012). Importantly, humic acid is thought to enhance the antioxidant status of plants by emulating plant-regulating compounds (Zhang and Schmidt 2000). It may thus provide additional benefit to dune and swale species in these stressful environments.

The research presented herein examines the benefits and feasibility of several plant establishment enhancement techniques, namely, increased planting density, humic acid amendment, and fertilizer application, in a large scale barrier island restoration effort. Application rates and approaches were optimized based on previous research as well as the greenhouse studies detailed in earlier chapters. In particular, the capacity of these techniques to increase live plant coverage and ameliorate harsh soil conditions over a two-year study duration are assessed. Finally, cost estimates are provided for employing the soil amendments in a field setting.

Materials and Methods

Whiskey Island Field Study Implementation

The field portion of this research was performed at Whiskey Island (latitude: 29° 02'N, longitude: 90° 49'W), a portion of the Isle Dernieres barrier island chain, in conjunction with a dune and swale planting effort by the Louisiana Office of Coastal Protection and Restoration (Figure 5.1, Images 5.2 and 5.3). This research was coordinated with the planting contractor to ensure all planting efforts occurred during the same time frame (May 2010 to mid-June 2010) and that all plants were acquired from the same source. To achieve the experimental planting densities for the three target species, blocks were selected from the contractor's planting areas of *U. paniculata*, *P. amarum*, and *S. patens* to contain a 6.1 m x 32 m low-density planting area (contractor plantings) and a 6.1 m x 32 m high-density planting area (contractor plantings supplemented by additional plantings). The high-density planting densities for *U. paniculata* and *P. amarum* were double (0.76 m centers) the current State of Louisiana planting density (1.52 m centers), whereas the planting density for *S. patens* (0.5 m centers) was triple the State of Louisiana planting density (1.52 m centers). Installation of the 2 m x 2 m monitoring plots was initiated immediately after the high-density planting treatments were completed in representative areas. Thereafter, the fertilizer regime and humic acid treatments were initiated in preselected areas within the planting density that had been randomly selected during the experimental design process. The fertilizer regime treatment consisted of an ambient level and fertilizer application at the rate described in Broome et al. (1982). Fertilizer application was accomplished using chest-mount fertilizer spreaders, with 8-8-8 applied in spring 2010 and spring 2011 at a rate of 878.4 kg ha⁻¹ and ammonium nitrate applied in summer 2010 and fall 2010 at a rate of 195.3 kg ha⁻¹. The humic acid amendment treatment consisted of 0 ml m⁻², 125 ml m⁻², and 250 ml m⁻² of 4 % active ingredient humic acid (3 Tier Technologies, Longwood, FL; Image 5.1). These levels, which are high compared to agricultural applications, were selected based on previous research by Willis and Hester (2010) as well as the results of the range-finding study. Humic acid was applied with hand-pump backpack sprayers in spring 2010 and spring 2011. Subsequent to the initiation of the experiment in late summer 2010, it was discovered that the planting contractor was utilizing fertilizer tablets (21 grams 20-10-5) for all contracted dune and swale species. To adjust for this, an amount of broadcast fertilizer equivalent to the contractor's fertilizer tablet was applied to each fertilized treatment plot during the fall 2010 treatment application and sampling trip. It was also discovered in late summer 2010 that a mixture of *Spartina spartinae* and *S. patens* rather than monospecific *S. patens* had been planted in the swale area. As there was no effective measure to resolve this as a component of the experimental design, *S. spartinae* were treated as equivalent to *S. patens* in the results and discussion of this chapter.

Whiskey Island Field Data Collection

All experimental plots were monitored immediately after planting but prior to initiation of the fertilizer and humic acid amendment treatments in spring 2010. Plots were thereafter monitored in summer 2010, fall 2010, spring 2011 and fall 2011, in each case prior to the application of fertilizer regime and humic acid amendment treatments. In all experimental plots, live and dead vegetative cover by species was visually determined to the nearest 5%. The average canopy height of the target plant species was also determined. Three to six of the newly expanded leaves of target species within a plot were collected for nutrient characterization and kept cold until returned to the laboratory. Additionally, plots were surveyed using a rotary laser



Figure 5.1 Site map for the experimental dune and swale restoration project on Whiskey Island (top panel), and the experimental restoration at the +4 foot contour on New Cut (bottom panel). For each study, the black outline represents one experimental planting block, which contained all treatments.



Image 5.1 Application of commercial humic acid on *Uniola paniculata* plantings shortly after installation in spring 2010.



Image 5.2 *Uniola paniculata* plantings one month (top panel) and three months (bottom panel) after installation.



Image 5.3 *Panicum amarum* one month (top panel) and three months (left portion of image; bottom panel) after installation at Whiskey Island dune restoration site.

level and survey values were tied to local benchmarks (TE14-SM-01) and a continuously recording water level gauge. Soil samples were collected to a depth of 15 cm using a stainless steel soil scoop, immediately bagged, and kept cold until they were transported to the lab. In spring 2010 and fall 2010, quantum yield, an indicator of photosynthetic capacity, was determined on light-adapted leaves using a FluorPen FP 100 (Photon Systems Instruments, Brno, Czech Republic).

Whiskey Island Statistical Analyses

All data were analyzed using the general linear models of JMP 9.0 (SAS Institute). Data were analyzed as a 3 plant species x 3 humic acid amendment x 2 fertilizer regime RBD with 5 blocks (total of 240 experimental plots). Repeated measures were employed where appropriate; Hunyh-Feldt corrected p values were utilized.

New Cut Field Study Implementation

An additional field evaluation of planting density and species combination, humic acid amendment, and fertilizer regime at the +4 foot contour of the New Cut portion of the Isle Dernieres barrier island chain was initiated in late September and early October of 2010 (Figure 5.1, Image 5.4). An earlier attempt (May 2010) to initiate this study was unable to be completed, as the island was restricted from any access due to the Deepwater Horizon oil spill response. The plant source for this study was the same as for the Whiskey Island planting effort (Erosion Control Services, Simmesport, LA). Five planting treatment areas, 10.7 m x 12.8 m in size were randomly assigned within each block. The low-density monospecific *S. patens* level, which is representative of the State of Louisiana's current planting density, was planted at 1.5 m centers. The high planting density for each species was triple the current State of Louisiana planting density (0.5 m). Installation of the 2 m x 2 m monitoring plots was initiated immediately after the high-density planting treatments were completed. The fertilizer regime treatment followed Broome et al. (1982) and consisted of an ambient level and 878.4 kg ha⁻¹ 8-8-8 fertilizer application, accomplished using chest-mount fertilizer spreaders in fall 2010. The humic acid amendment treatment consisted of 0 ml m⁻² and 250 ml m⁻² of 4 % active ingredient humic acid (3 Tier Technologies, Longwood, FL), applied using hand-pump backpack sprayers. The experimental design for this study consisted of a 5 species-density combination (*S. patens*–low density, *S. patens*–high density, *P. amarum*–high density, *D. spicata*–high density, *S. patens*, *P. amarum*, *D. spicata* mixed–high density) x 2 fertilizer regime (ambient, fertilized) x 2 humic acid amendment (0 ml m⁻², 250 ml m⁻²) randomized block design with 5 blocks for a total of 100 plots.

New Cut Field Data Collection

All experimental plots were monitored immediately after planting but prior to initiation of the fertilizer and humic acid amendment treatments in fall 2010. Plots were monitored again in spring 2011. In all experimental plots, live and dead vegetative cover by species was visually determined to the nearest 5%. Soil samples were collected to a depth of 15 cm using a stainless steel soil scoop, immediately bagged, and kept cold until they were transported to the laboratory. Nearly all experimental planting units had perished by the spring 2011 sampling, likely due to desiccation stress; therefore, no additional monitoring or sample collection was performed. Meteorological data, including precipitation, were downloaded for the relevant time period from the LUMCON Bay Tambour weather buoy (TAML1; latitude: 29.19N, longitude: 90.67W).



Image 5.4 Field site at New Cut portion of the Isle Dernieres one week after plant installation.

New Cut Statistical Analyses

All data were analyzed using the general linear models of JMP 9.0 (SAS Institute). Data were analyzed as a 5-species-density combination (*S. patens*–low density, *S. patens*–high density, *P. amarum*–high density, *D. spicata*–high density, *S. patens*, *P. amarum*, *D. spicata* mixed–high density) x 2 fertilizer regime (ambient, fertilized) x 2 humic acid amendment (0 ml m⁻², 250 ml m⁻²) randomized block design with 5 blocks (total of 100 experimental plots).

Plant Tissue Processing

Upon returning to the lab, leaf samples were rinsed with deionized water, dried to a constant weight at 65° C, and ground using a Wiley Mill to pass through a number 20 screen. One aliquot of dried leaf material was submitted to the LSU Soil Testing and Plant Analysis Laboratory for the determination of total nitrogen and carbon content using standard laboratory techniques. The second aliquot of leaf material was digested using trace-metal-grade hot nitric acid following the methods of Jones et al. (1991) and was then submitted to the LSU Soil Testing and Plant Analysis Laboratory for determination of elemental content using ICP-OES (EPA method 200.7).

Soil Physico-Chemical Characterization

Soil samples were collected at the conclusion of the study for the determination of soil moisture, conductivity, pH, nutrient status, and organic matter. The samples were weighed and then dried at 65° C until a constant weight was achieved and soil moisture was calculated. Dried soil samples were homogenized and an approximately 2 g subsample was combusted at 500° C for 5 hours to determine percent organic matter (Parent and Caron 1993). Additional subsamples of the homogenized soil samples were subjected to two 1:2 (w:v) extraction procedures employing deionized water and 2M KCl, respectively. One aliquot of the deionized water extract was used for the determination of pH and conductivity (Rhoades 1990). The second aliquot of deionized water extract was submitted to the LSU Soil Testing and Plant Analysis Laboratory for the determination of total phosphorus, potassium, and other relevant cations using ICP-OES (EPA method 200.7). The KCl extract was submitted to the SLU microbial testing laboratory for the determination of ammonium and nitrate-nitrite using colorimetric methods (EPA method 350.1 and 353, respectively).

Results

Vegetative Cover

A significant effect of season (Figures 5.2–5.4; $F = 136.6$, $p < 0.01$) and of season and species ($F = 42.5$, $p < 0.01$) was found for live cover, as all species expanded from the initial plantings; but *P. amarum* consistently expanded over time. Interestingly, significant interactions of season and planting density (Figures 5.2–5.4; $F = 7.3$, $p < 0.01$) as well as season, planting density, and species (Figures 5.2–5.4; $F = 2.2$, $p < 0.05$) were detected for live cover. These significant effects result from *U. paniculata* and *S. patens* demonstrating a continuing benefit of higher planting density on live cover, whereas *P. amarum* live cover rapidly became

Uniola paniculata

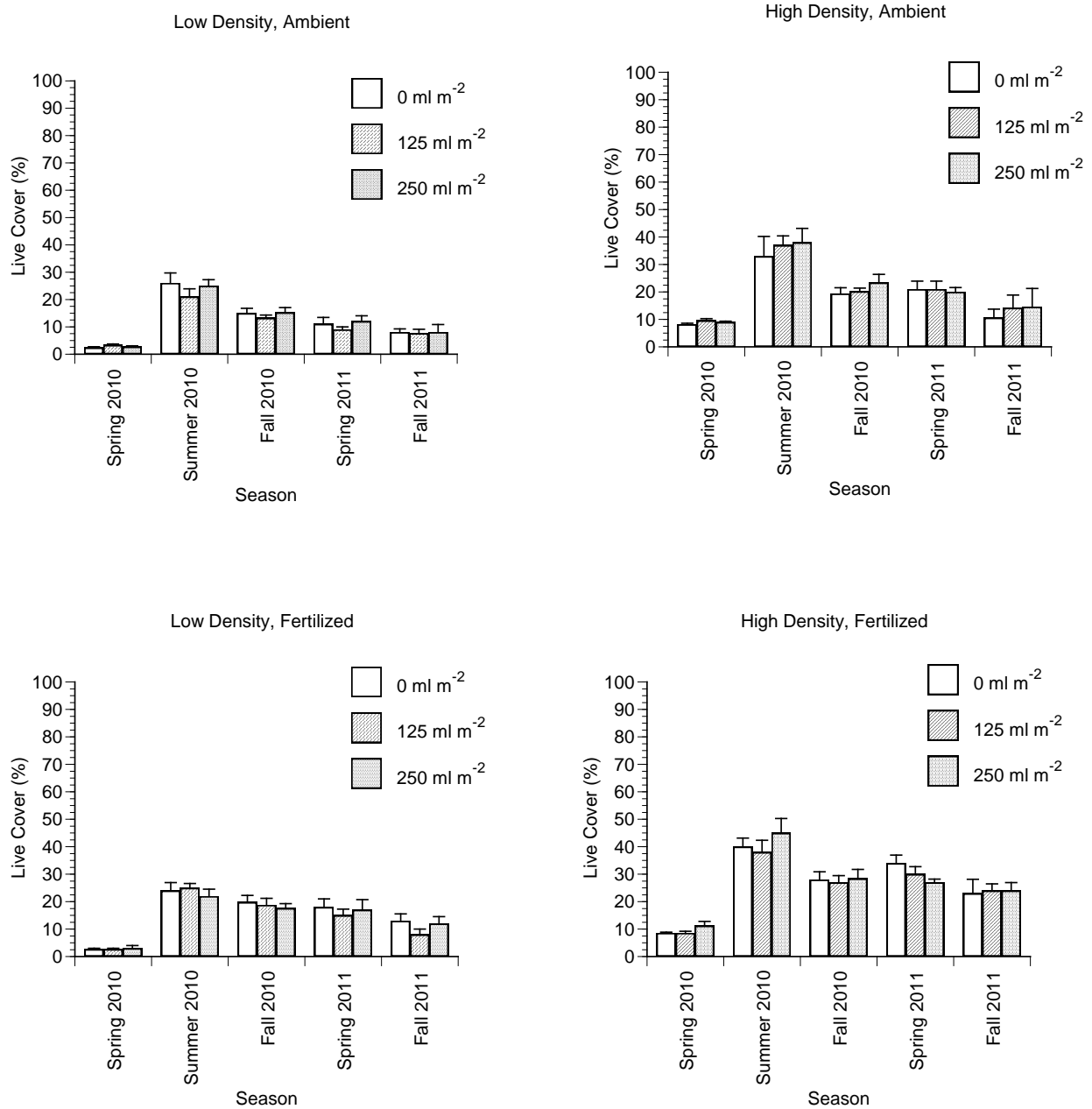


Figure 5.2 The effect of season, planting density, fertilizer regime, and humic acid amendment on *Uniola paniculata* live cover (%; mean \pm SE, n = 5, spring 2010 LSD = 2.095, summer 2010 LSD = 8.719, fall 2010 LSD = 7.625, spring 2011 LSD = 9.294, fall 2011 LSD = 12.777)

Panicum amarum

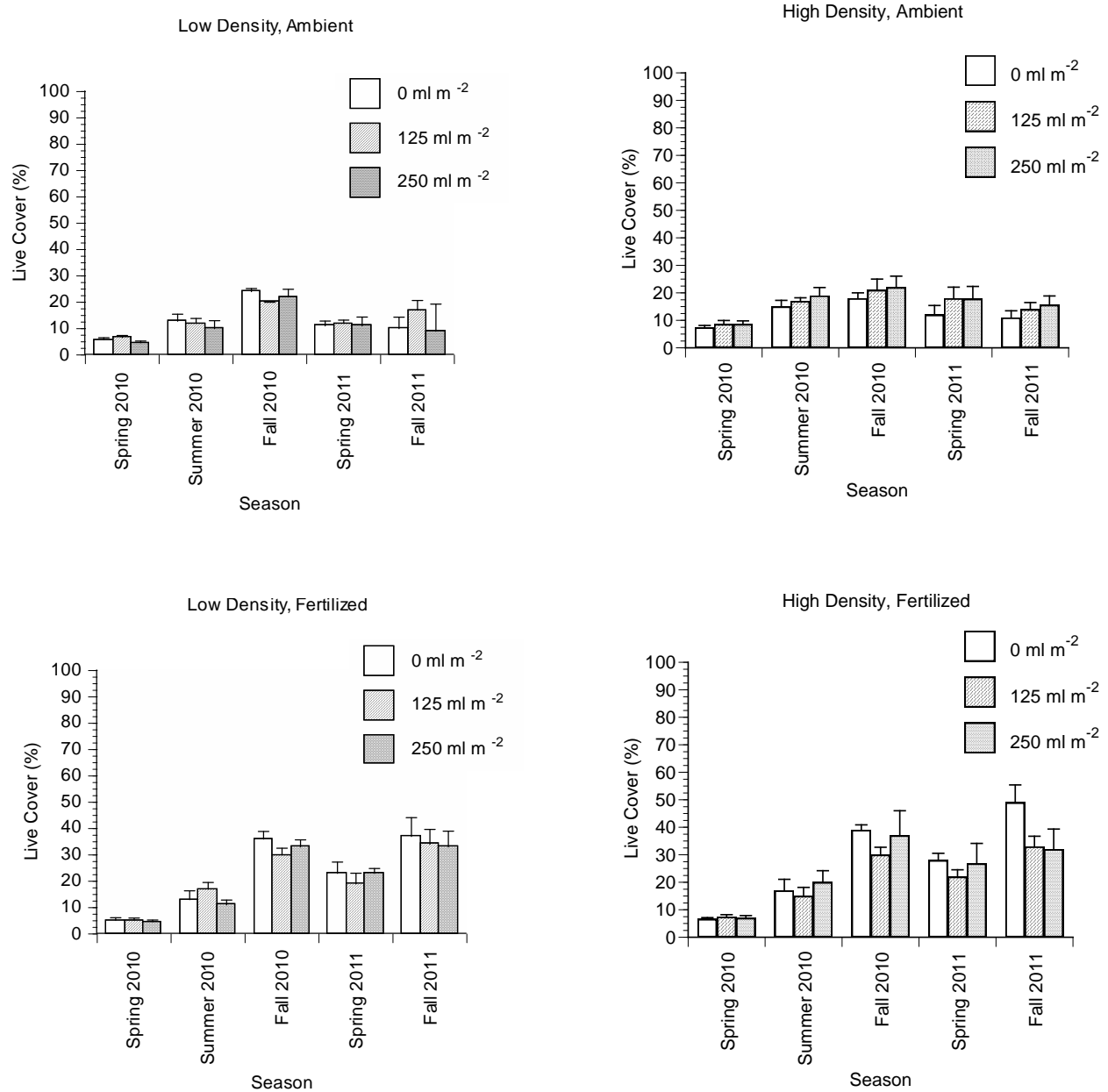


Figure 5.3 The effect of season, planting density, fertilizer regime, and humic acid amendment on *Panicum amarum* live cover (%; mean \pm SE, n = 5, spring 2010 LSD = 2.095, summer 2010 LSD = 8.719, fall 2010 LSD = 7.625, spring 2011 LSD = 9.294, fall 2011 LSD = 12.777).

Spartina patens

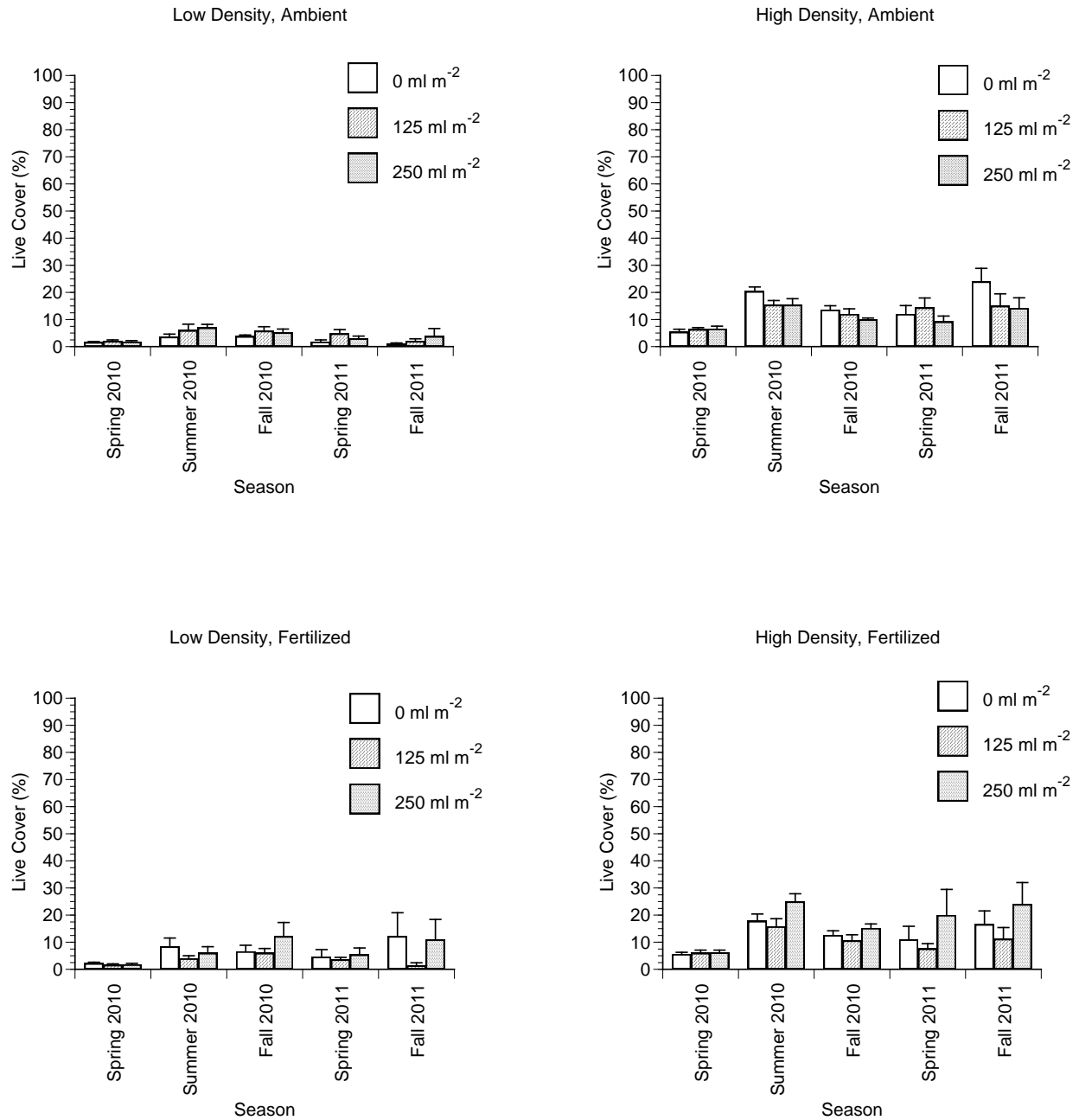


Figure 5.4 The effect of season, planting density, fertilizer regime, and humic acid amendment on *Spartina patens* live cover (%; mean \pm SE, n = 5, spring 2010 LSD = 2.095, summer 2010 LSD = 8.719, fall 2010 LSD = 7.625, spring 2011 LSD = 9.294, fall 2011 LSD = 12.777)

equivalent between the two planting densities. A significant effect of season, fertilizer regime, and species was detected for live cover, resulting from *P. amarum* and *U. paniculata* having greater expansion of live cover over time with fertilizer application than *S. patens* (Figures 5.2–5.4; $F = 7.1$, $p < 0.01$).

A significant effect of season (Figures 5.5–5.7; $F = 63.0$, $p < 0.05$) and a marginally significant effect of season and planting density (Figures 5.5–5.7; $F = 2.6$, $p < 0.1$) were found for *C. dactylon* cover, which increased after the summer 2010 sampling, especially in the low-density plots. A highly significant effect of season and species (Figures 5.5–5.7; $F = 7.8$, $p < 0.01$) was detected, as *C. dactylon* cover increased far more in *S. patens* plots than in *U. paniculata* or *P. amarum* plots by fall 2010. *Cynodon dactylon* cover was lower in *U. paniculata* high-density than in low-density plots after summer 2010, while *C. dactylon* cover was similar in high-density and low-density *P. amarum* and *S. patens* plots (Figures 5.5–5.7; $F = 2.4$, $p < 0.05$). Interestingly, a marginally significant effect of season, planting density, and humic acid amendment was found for *C. dactylon* cover, which somewhat increased in low-density plots receiving 125-ml m⁻² humic acid amendment but decreased in plots receiving 250-ml m⁻² humic acid amendment after summer 2010. A trend toward increased *C. dactylon* cover with increased humic acid amendment, however, was seen in the high-density planting treatment (Figures 5.5–5.7; $F = 1.9$, $p < 0.1$).

A highly significant effect of season and fertilizer (Figures 5.5–5.7; $F = 39.1$, $p < 0.01$) as well as a significant effect of season, planting density, and fertilizer (Figures 5.5–5.7; $F = 3.4$, $p < 0.05$) was detected for *C. dactylon* cover, as fertilized plots substantially increased *C. dactylon* cover after summer 2010, particularly in low-density plots. Importantly, a significant effect of season, humic acid amendment, and fertilizer regime was found for *C. dactylon* cover (Figures 5.5–5.7; $F = 2.1$, $p < 0.05$), where *C. dactylon* cover after summer 2010 increased with 125 ml m⁻² but decreased with 250 ml m⁻² humic acid amendment in fertilized plots; ambient fertilizer plots, however, showed consistently low *C. dactylon* cover. A significant effect of season, fertilizer, and species was detected for *C. dactylon* cover (Figures 5.5–5.7; $F = 6.7$, $p < 0.01$), resulting from *C. dactylon* cover increasing in fertilized plots for all species but being particularly elevated for *S. patens*.

A highly significant effect of season (Figures 5.8–5.10; $F = 282.2$, $p < 0.01$) as well as a highly significant interaction of season and species (Figures 5.8–5.10; $F = 15.4$, $p < 0.01$) was detected for total vegetative cover, resulting from total cover increasing for *P. amarum* and *S. patens* treatments through time but leveling off more rapidly for *U. paniculata*. Significant interactions of season and planting density (Figures 5.8–5.10; $F = 2.7$, $p < 0.05$), as well as season, species, and planting density (Figures 5.8–5.10; $F = 2.2$, $p < 0.05$) were detected, driven by the increase in total vegetative cover for high-density *S. patens* plots, with less dramatic increases seen for *U. paniculata* and *P. amarum* plots. The addition of fertilizer increased total vegetative cover over time (Figures 5.8–5.10; $F = 88.8$, $p < 0.01$), except for *U. paniculata* plots in fall 2011 (Figures 5.8–5.10; $F = 3.0$, $p < 0.01$), possibly due to impacts from Tropical Storm Lee. Of great interest is a significant interaction of season, fertilizer regime, and humic acid amendment (Figures 5.8–5.10; $F = 4.1$, $p < 0.01$), where the humic acid amendment level of 250 ml m⁻²



Image 5.5 Image of the *Uniola paniculata* planting area in fall 2011 subsequent to the passing of Tropical Storm Lee showing different treatment areas of planting density (**A**: low density; fertilizer **B**: low density; no fertilizer, **C**: high density; no fertilizer, **D**: high density; fertilizer). Note the extent and health of *Uniola paniculata* planted at high density with fertilizer application.

Uniola paniculata

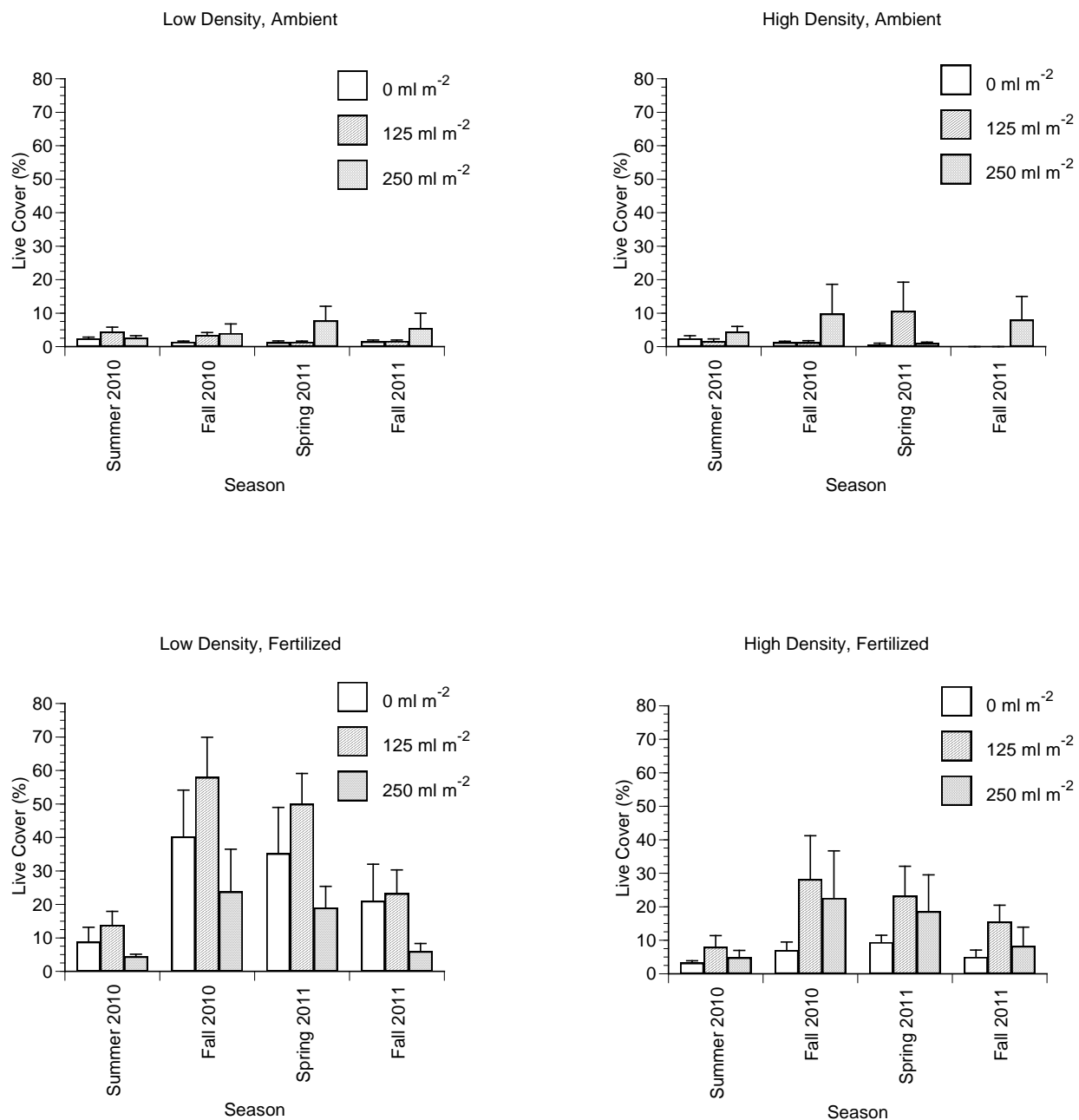


Figure 5.5 The effect of season, planting density, fertilizer regime, and humic acid amendment on *Cynodon dactylon* live cover in *Uniola paniculata* experimental plots (%; mean \pm SE, $n = 5$, summer 2010 LSD = 11.812, fall 2010 LSD = 24.807, spring 2011 LSD = 21.800, fall 2011 LSD = 14.655)

Panicum amarum

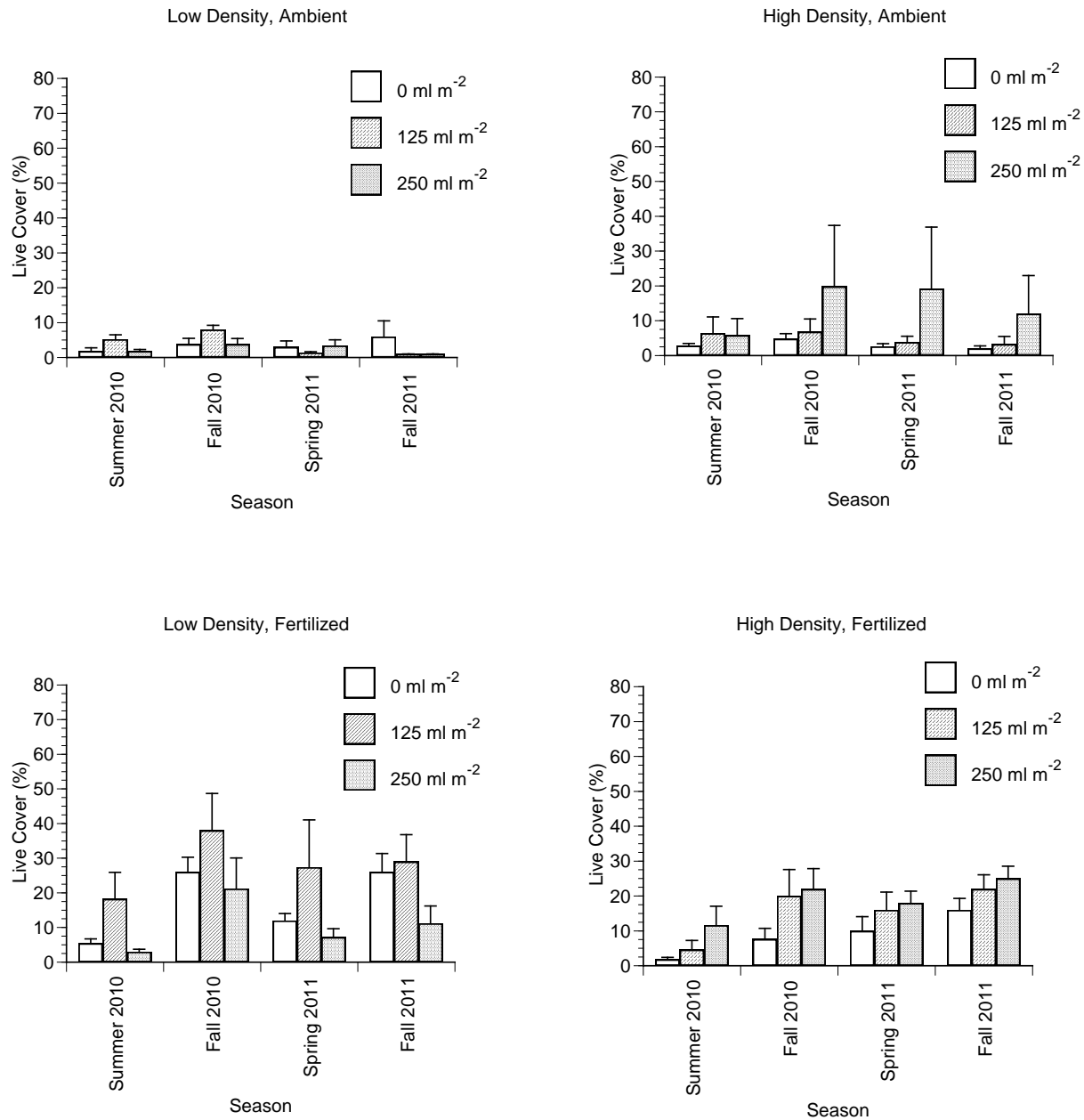


Figure 5.6 The effect of season, planting density, fertilizer regime, and humic acid amendment on *Cynodon dactylon* live cover in *Panicum amarum* experimental plots (%; mean \pm SE, $n = 5$, summer 2010 LSD = 11.812, fall 2010 LSD = 24.807, spring 2011 LSD = 21.800, fall 2011 LSD = 14.655)

Spartina patens

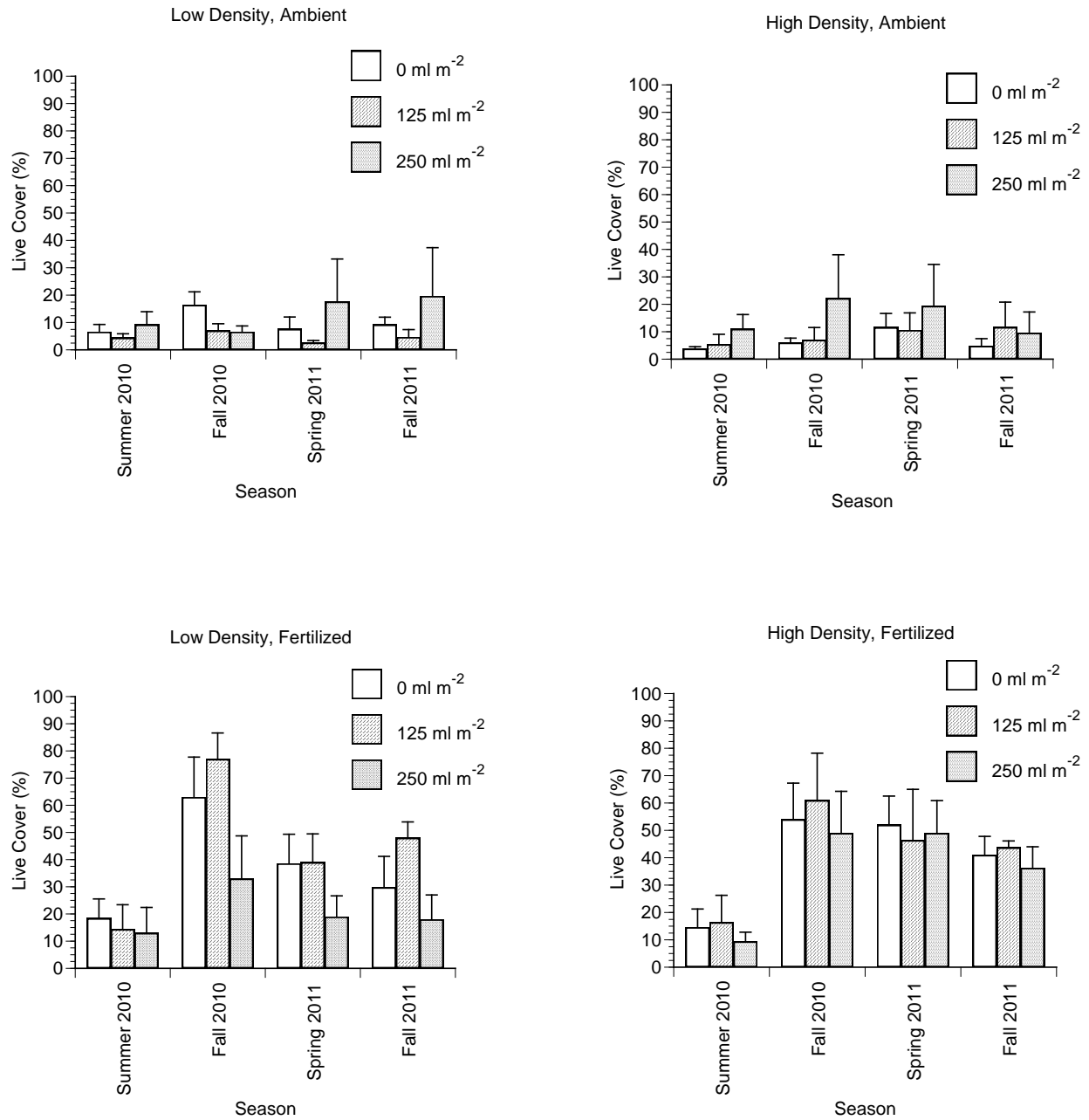


Figure 5.7 The effect of season, planting density, fertilizer regime, and humic acid amendment on *Cynodon dactylon* live cover in *Spartina patens* experimental plots (%; mean \pm SE, $n = 5$, summer 2010 LSD = 11.812, fall 2010 LSD = 24.807, spring 2011 LSD = 21.800, fall 2011 LSD = 14.655)

Uniola paniculata

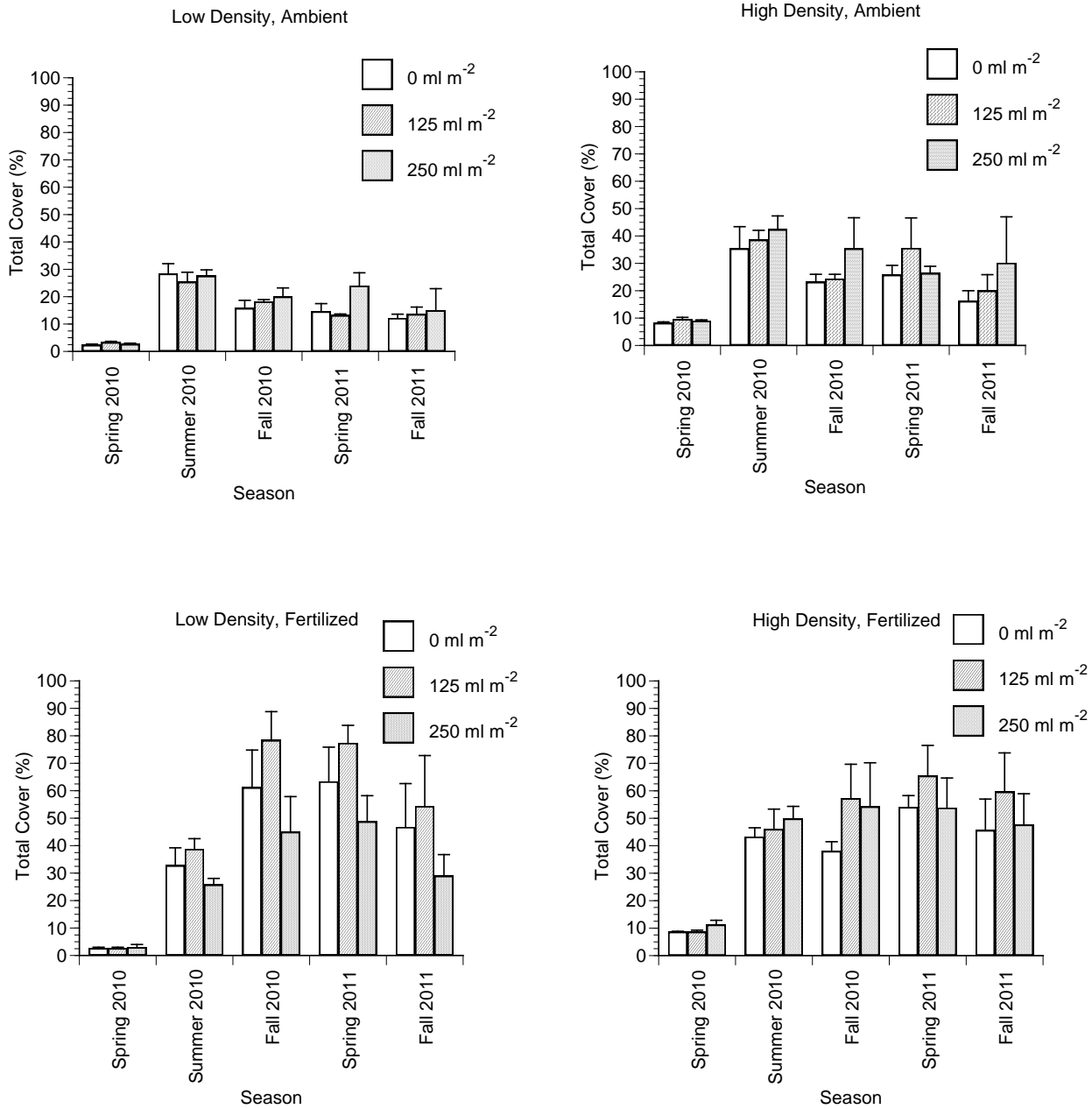


Figure 5.8 The effect of season, planting density, fertilizer regime, and humic acid amendment on total cover in *Uniola paniculata* experimental plots (%; mean \pm SE, n = 5, spring 2010 LSD = 2.095, summer 2010 LSD = 13.972, fall 2010 LSD = 23.544, spring 2011 LSD = 21.542, fall 2011 LSD = 22.400).

Panicum amarum

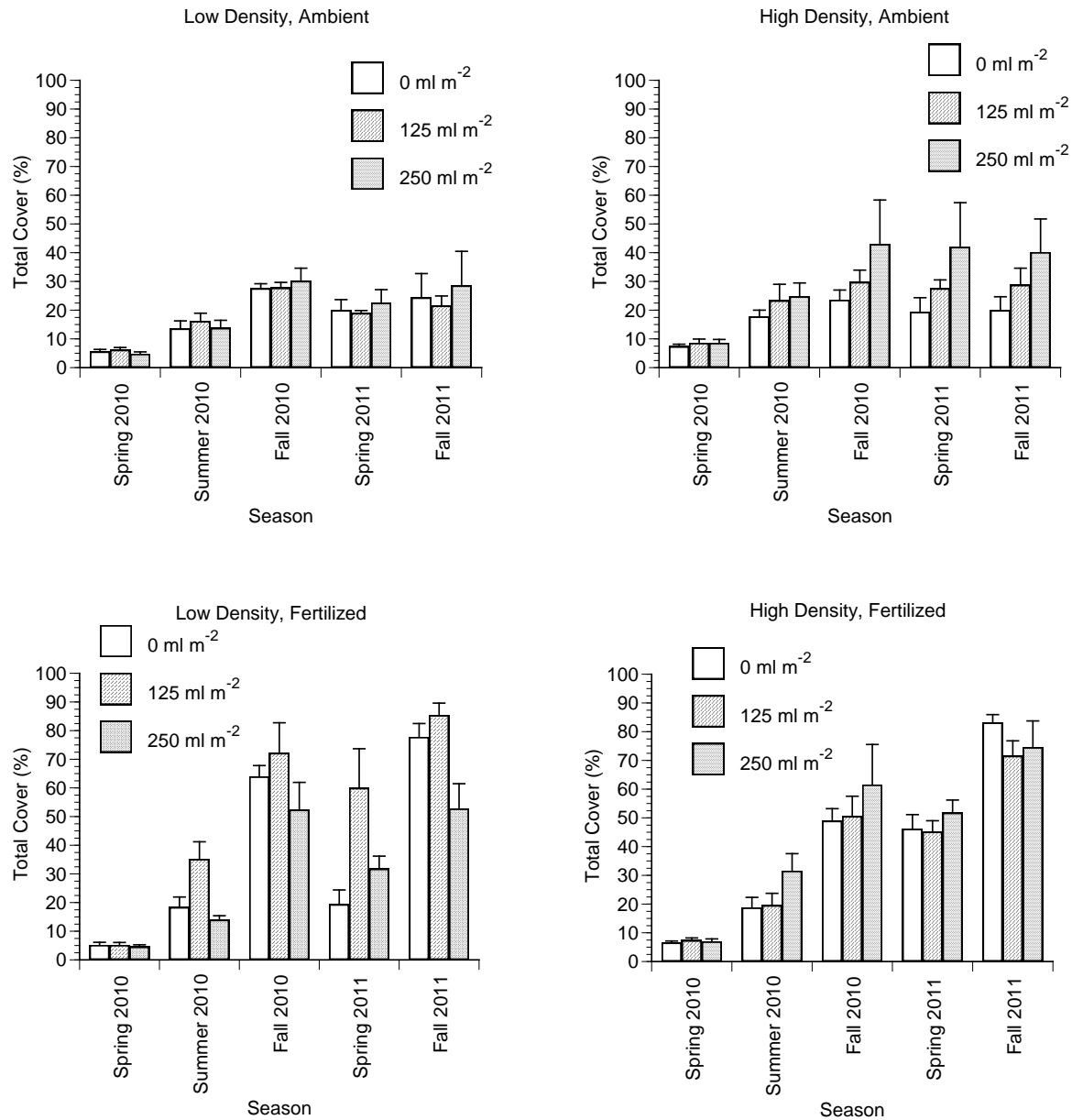


Figure 5.9 The effect of season, planting density, fertilizer regime, and humic acid amendment on total cover in *Panicum amarum* experimental plots (%; mean \pm SE, n = 5, spring 2010 LSD = 2.095, summer 2010 LSD = 13.972, fall 2010 LSD = 23.544, spring 2011 LSD = 21.542, fall 2011 LSD = 22.400).

Spartina patens

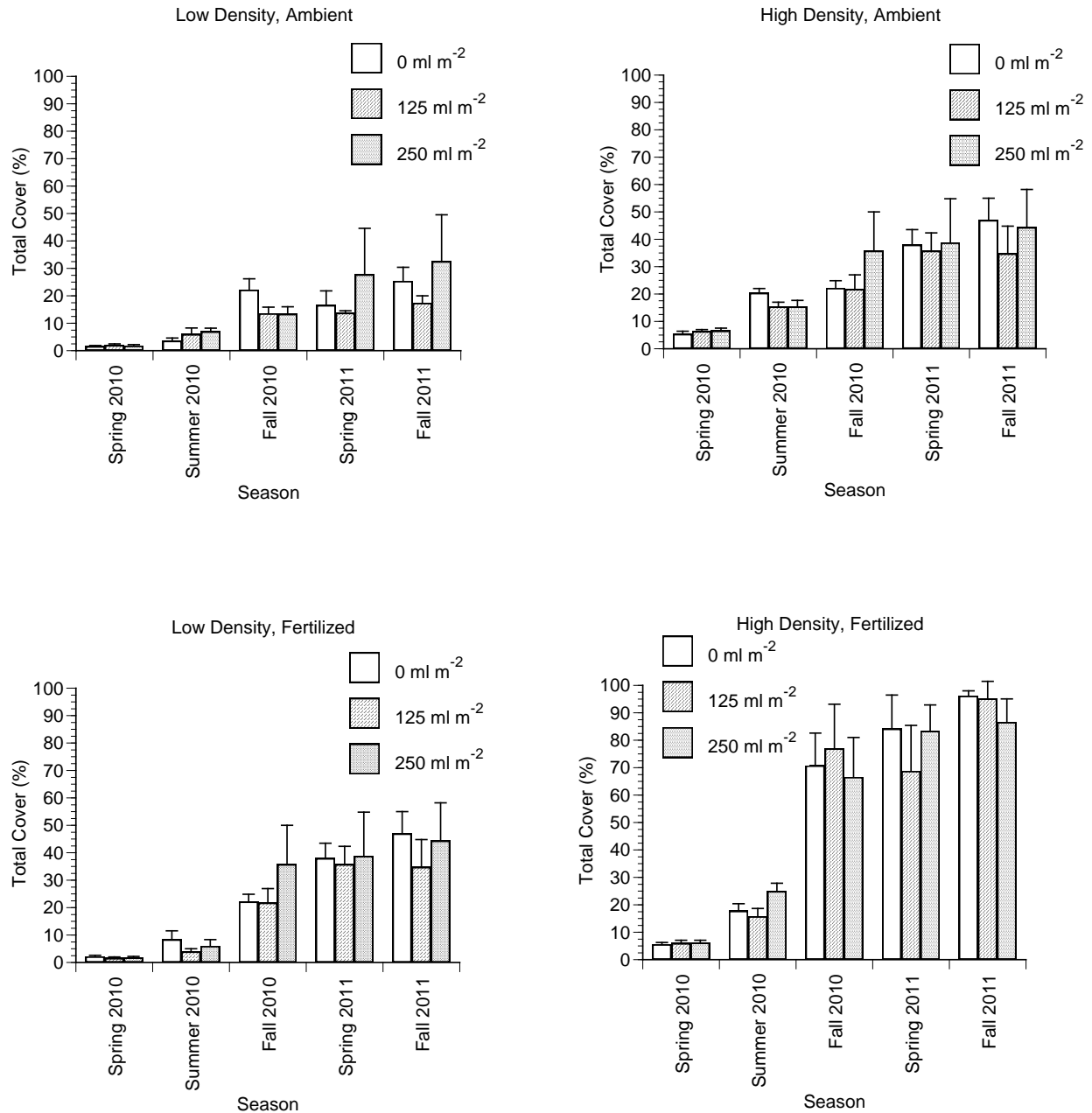


Figure 5.10 The effect of season, planting density, fertilizer regime, and humic acid amendment on total cover in *Spartina patens* experimental plots (%; mean \pm SE, $n = 5$, spring 2010 LSD = 2.095, summer 2010 LSD = 13.972, fall 2010 LSD = 23.544, spring 2011 LSD = 21.542, fall 2011 LSD = 22.400).

decreased total vegetative cover under fertilized conditions but increased total vegetative cover under ambient conditions.

Panicum amarum Belowground Biomass

A highly significant effect of season (Figure 5.11; $F = 119.8$, $p < 0.01$) as well as a significant interaction of season and planting density (Figure 5.11; $F = 2.9$, $P < 0.05$) was detected for *P. amarum* belowground biomass. A highly significant increase of *P. amarum* belowground biomass in fertilized treatments was detected at the conclusion of the study (Figure 5.11; $F = 10.8$, $p < 0.01$). No other significant main effects or interactions were detected for *P. amarum* belowground biomass.

Average Stem Height and Quantum Yield

A highly significant effect of season (Figures 5.12–5.14; $F = 233.2$, $p < 0.01$) and interaction of season and species (Figures 5.12–5.14; $F = 70.2$, $p < 0.01$) was found, resulting from *P. amarum* and *S. patens* average stem height generally increasing over the sampling seasons while *U. paniculata* average stem height was similar from season to season. Highly significant interactions of season and planting density (Figures 5.12–5.14; $F = 5.3$, $p < 0.01$) as well as season, species, and planting density were found (Figures 5.12–5.14; $F = 4.4$, $p < 0.01$), stemming from a substantial decrease in low-density *S. patens* average stem height in fall 2012 while *P. amarum* average stem height was actually at its highest. Highly significant interactions of season and fertilizer regime (Figures 5.12–5.14; $F = 17.5$, $p < 0.01$) as well as season, species, and fertilizer regime (Figures 5.12–5.14; $F = 5.8$, $p < 0.01$) were found, with fertilizer application increasing *U. paniculata* average stem height over time, while *P. amarum* demonstrated no similar stimulation. A highly significant effect of season (Figures 5.15–5.17; $F = 61.9$, $p < 0.01$), and a significant interaction of planting density and season (Figures 5.15–5.17; $F = 4.3$, $p < 0.05$) for quantum yield were detected in which quantum yield was lower in fall 2010 than in summer 2010 and fall 2011, particularly for the high-density planting treatment. Also, a highly significant interaction of season and species was detected (Figures 5.15–5.17; $F = 11.6$, $p < 0.01$) in which *S. patens* demonstrated the highest quantum yield in summer and fall 2010, but *U. paniculata* demonstrated the highest quantum yield in spring 2011.

Plot Elevation

A highly significant effect of season (Figures 5.18–5.20; $F = 159.9$, $p < 0.01$) was detected for plot elevation, resulting from decreases in elevation in spring and fall of 2011. Importantly the spring and fall 2011 loss of elevation was ameliorated by the high planting density, generating an interaction (Figures 5.18–5.20; $F = 2.4$, $p < 0.1$). A significant interaction of season and species was detected, stemming from *U. paniculata* plots increasing elevation slightly in fall 2011 compared to spring 2011, whereas *P. amarum* and *S. patens* continued to decrease in fall 2011 (Figures 5.18–5.20; $F = 17.4$, $p < 0.01$).

Leaf Tissue Chemistry

A highly significant effect of season (Tables A5.1–A5.6; $F = 432.5$, $p < 0.01$) as well as a highly significant interaction of season and species (Tables A5.1–A5.6; $F = 54.6$, $p < 0.01$) was detected for plant tissue nitrogen. These significant effects reflect a lowering of plant tissue nitrogen through time, with *P. amarum* demonstrating higher tissue nitrogen levels in the summer 2010 and spring 2011 samplings; whereas *U. paniculata* decreased only after the

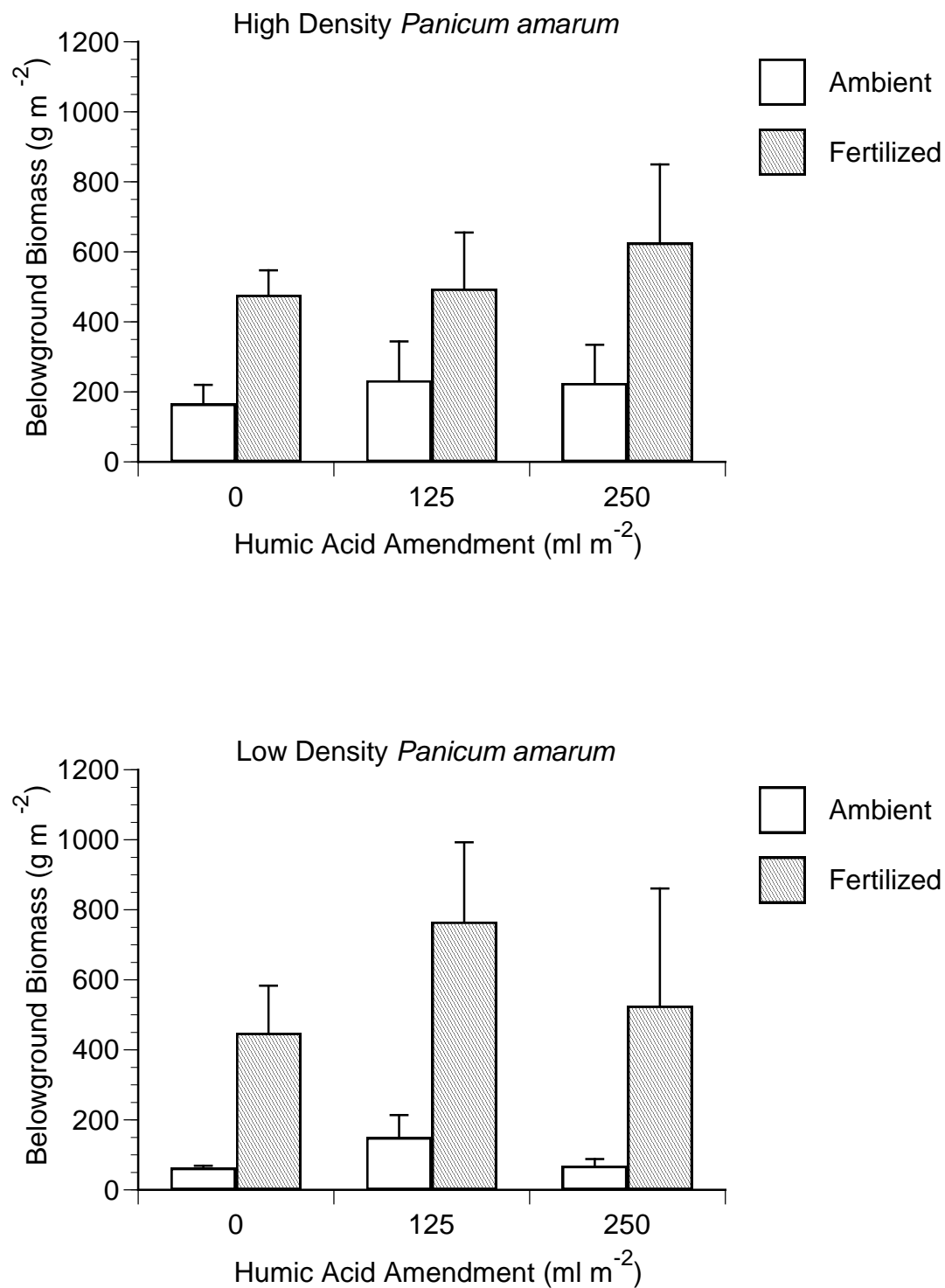


Figure 5.11 The effect of planting density, fertilizer addition, and humic acid amendment on fall 2011 *P. amarum* belowground biomass (%; mean \pm SE, $n = 5$, LSD = 1206.4).

Uniola paniculata

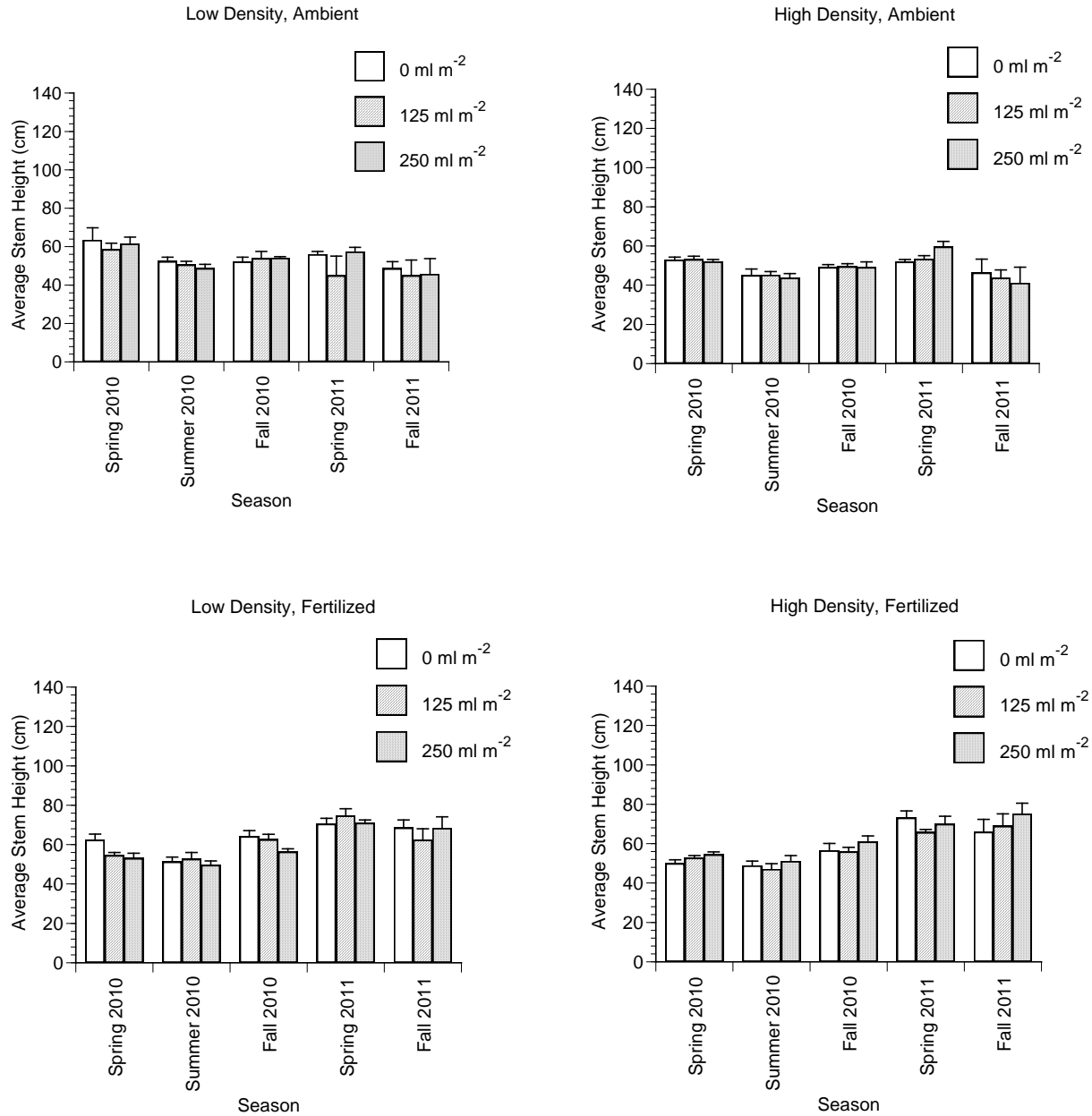


Figure 5.12 The effect of season, planting density, fertilizer regime, and humic acid amendment on *Uniola paniculata* average stem height (cm; mean \pm SE, $n = 5$, Sp 2010 LSD = 7.812, Su 2010 LSD = 13.907, Fa 2010 LSD = 8.773, Sp 2011 LSD = 21.542, Fa 2011 LSD = 17.388).

Panicum amarum

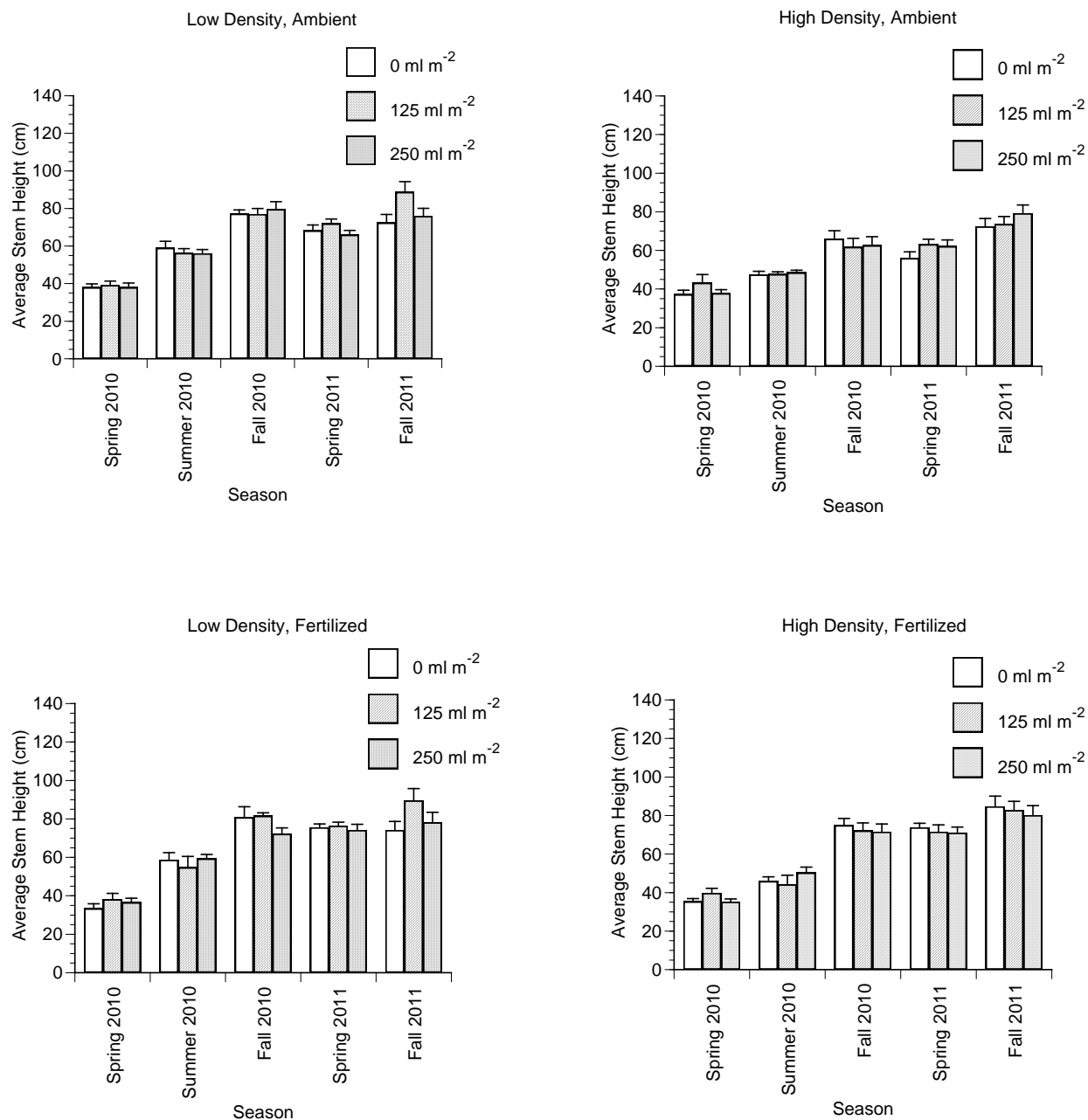


Figure 5.13 The effect of season, planting density, fertilizer regime, and humic acid amendment on *Panicum amarum* average stem height (cm; mean \pm SE, $n = 5$, Sp 2010 LSD = 7.812, Su 2010 LSD = 13.907, Fa 2010 LSD = 8.773, Sp 2011 LSD = 21.542, Fa 2011 LSD = 17.388).

Spartina patens

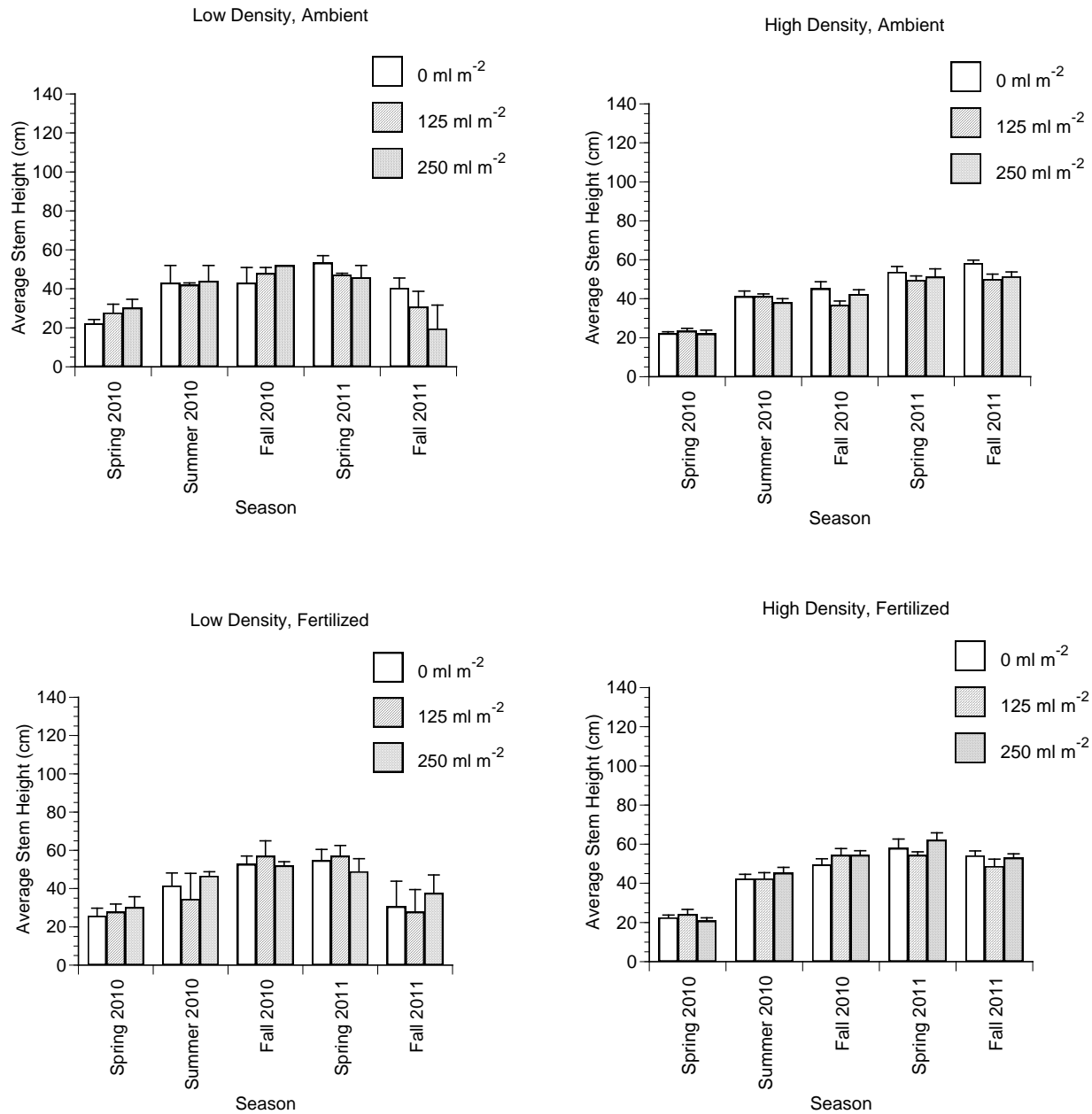


Figure 5.14 The effect of season, planting density, fertilizer regime, and humic acid amendment on *Spartina patens* average stem height (cm; mean \pm SE, $n = 5$, Sp 2010 LSD = 7.812, Su 2010 LSD = 13.907, Fa 2010 LSD = 8.773, Sp 2011 LSD = 21.542, Fa 2011 LSD = 17.388).

Uniola paniculata

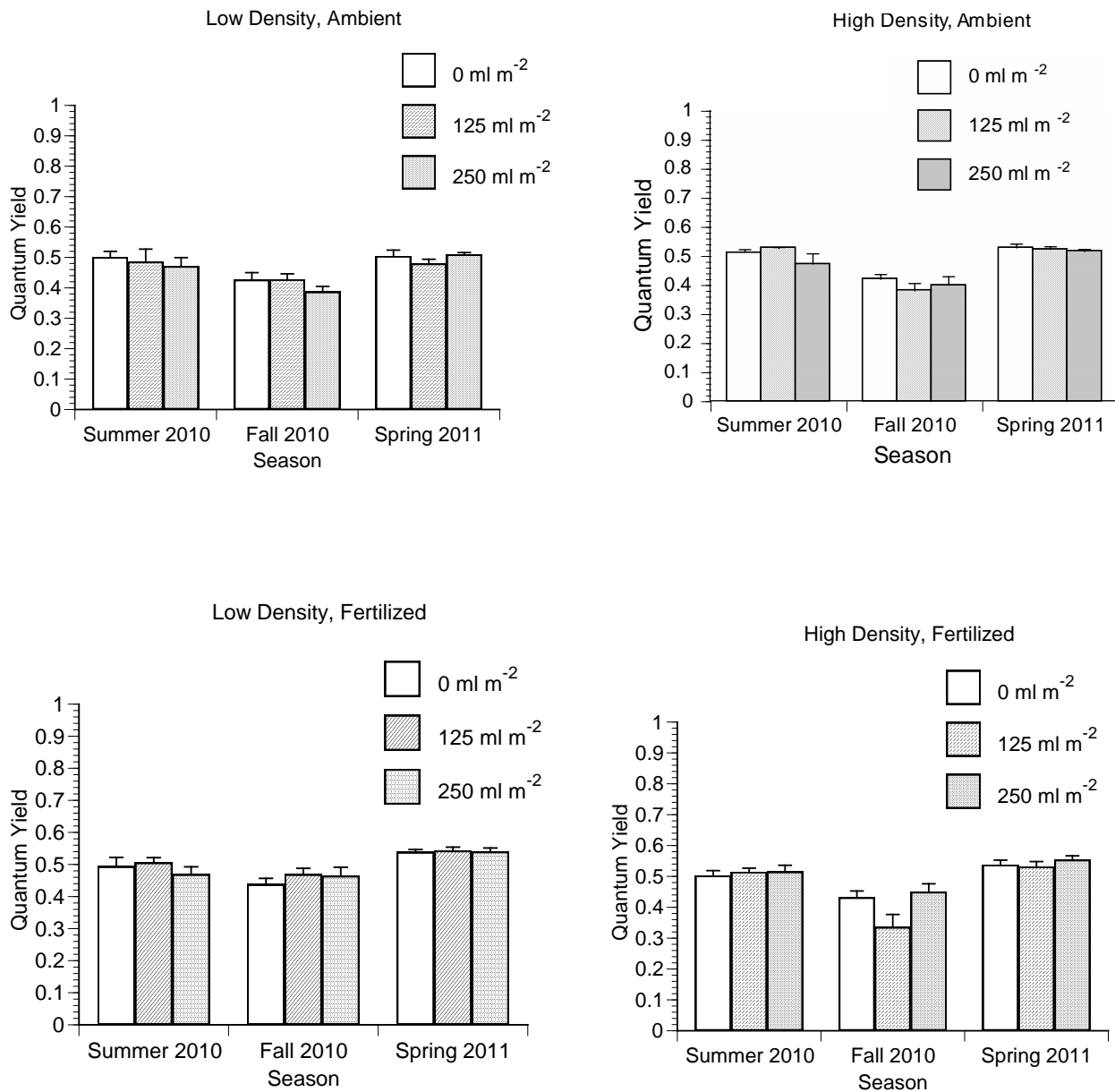


Figure 5.15 The effect of season, planting density, fertilizer regime, and humic acid amendment on *Uniola paniculata* quantum yield (mean \pm SE, n = 5, Su 2010 LSD = 0.115, Fa 2010 LSD = 0.132, Sp 2011 LSD = 0.0659).

Panicum amarum

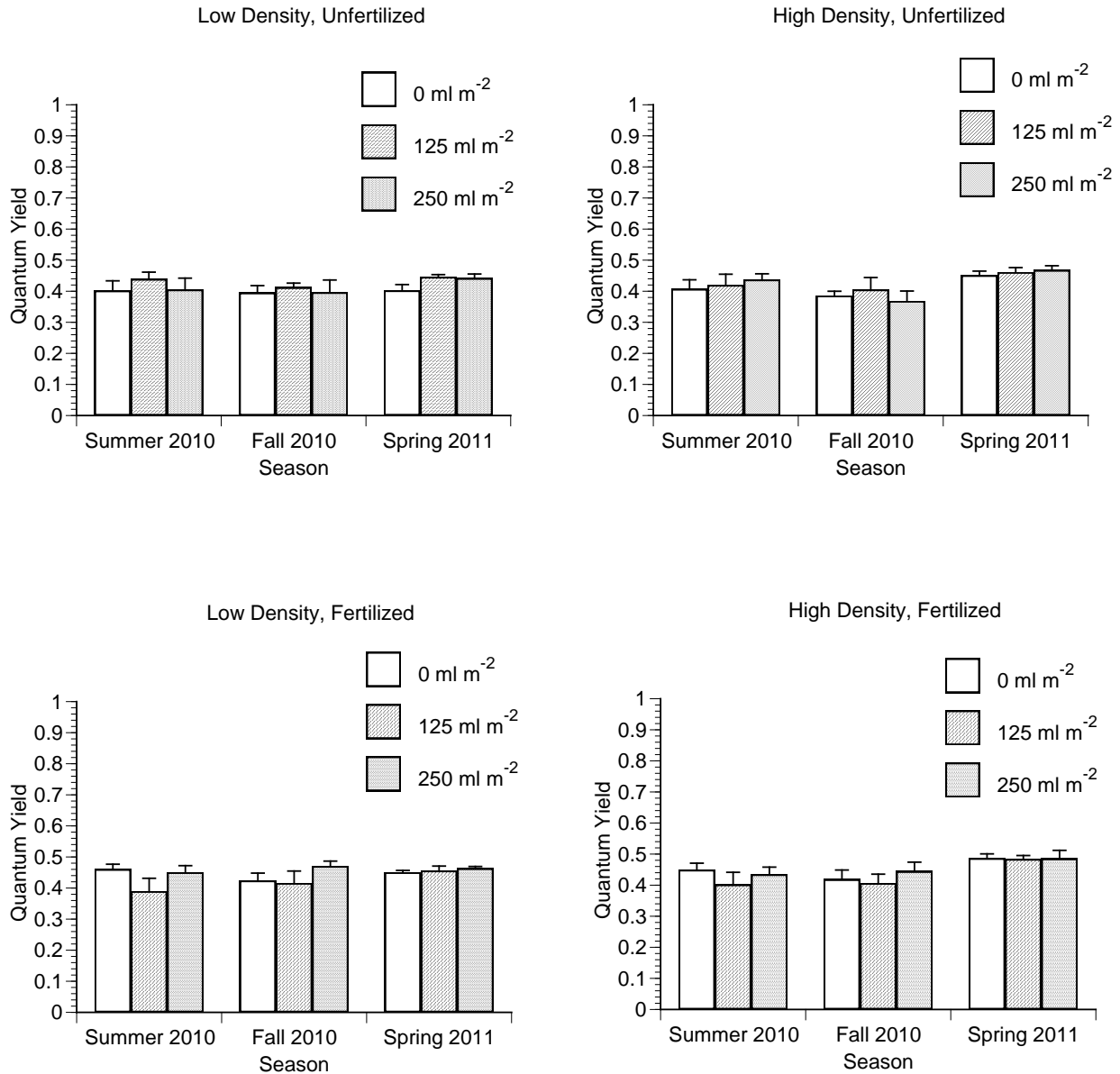


Figure 5.16 The effect of season, planting density, fertilizer regime, and humic acid amendment on *Panicum amarum* quantum yield (mean \pm SE, n = 5, Su 2010 LSD = 0.115, Fa 2010 LSD = 0.132, Sp 2011 LSD = 0.0659).

Spartina patens

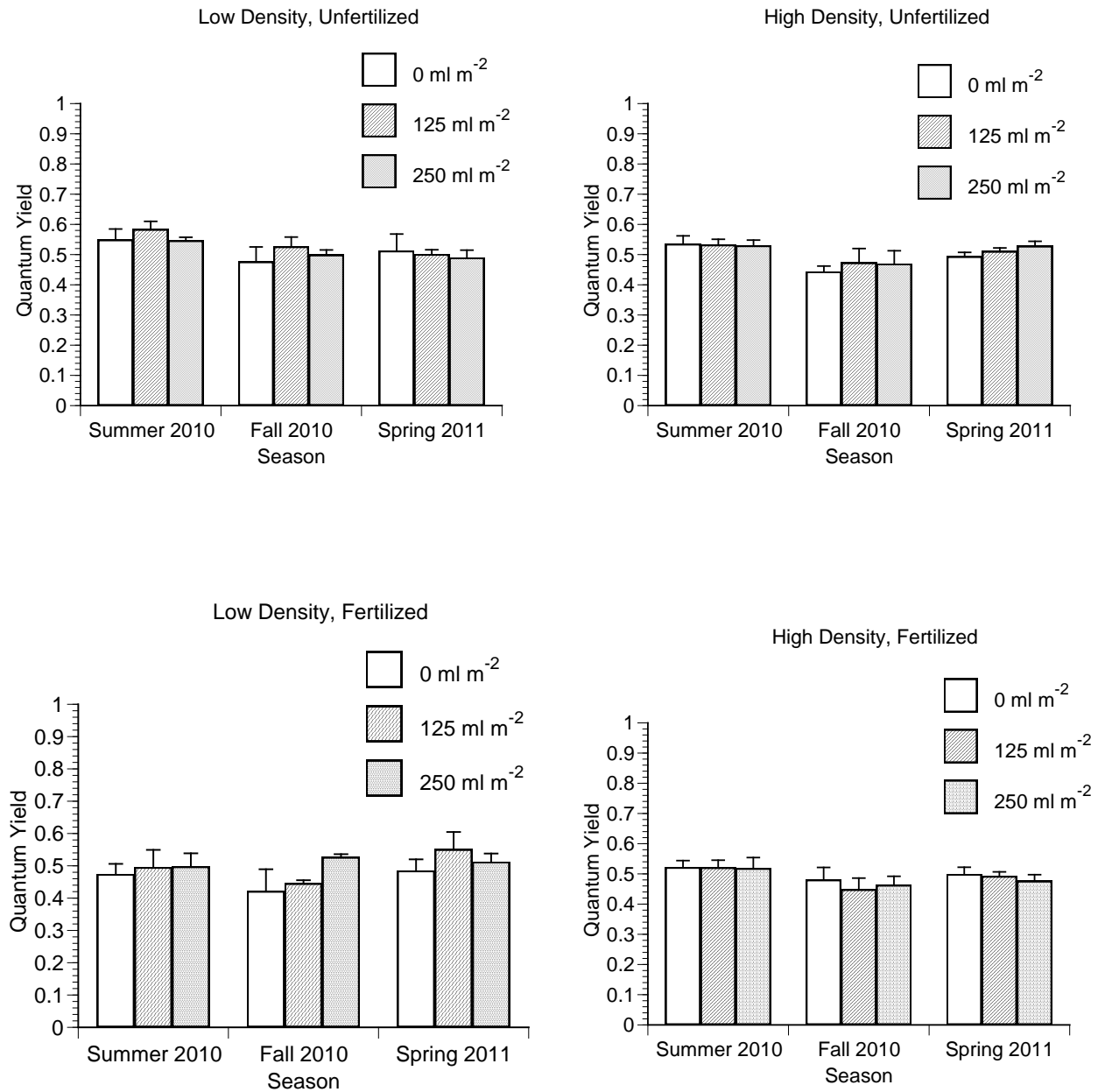


Figure 5.17 The effect of season, planting density, fertilizer regime, and humic acid amendment on *Spartina patens* quantum yield (mean \pm SE, n = 5, Su 2010 LSD = 0.115, Fa 2010 LSD = 0.132, Sp 2011 LSD = 0.0659).

Uniola paniculata

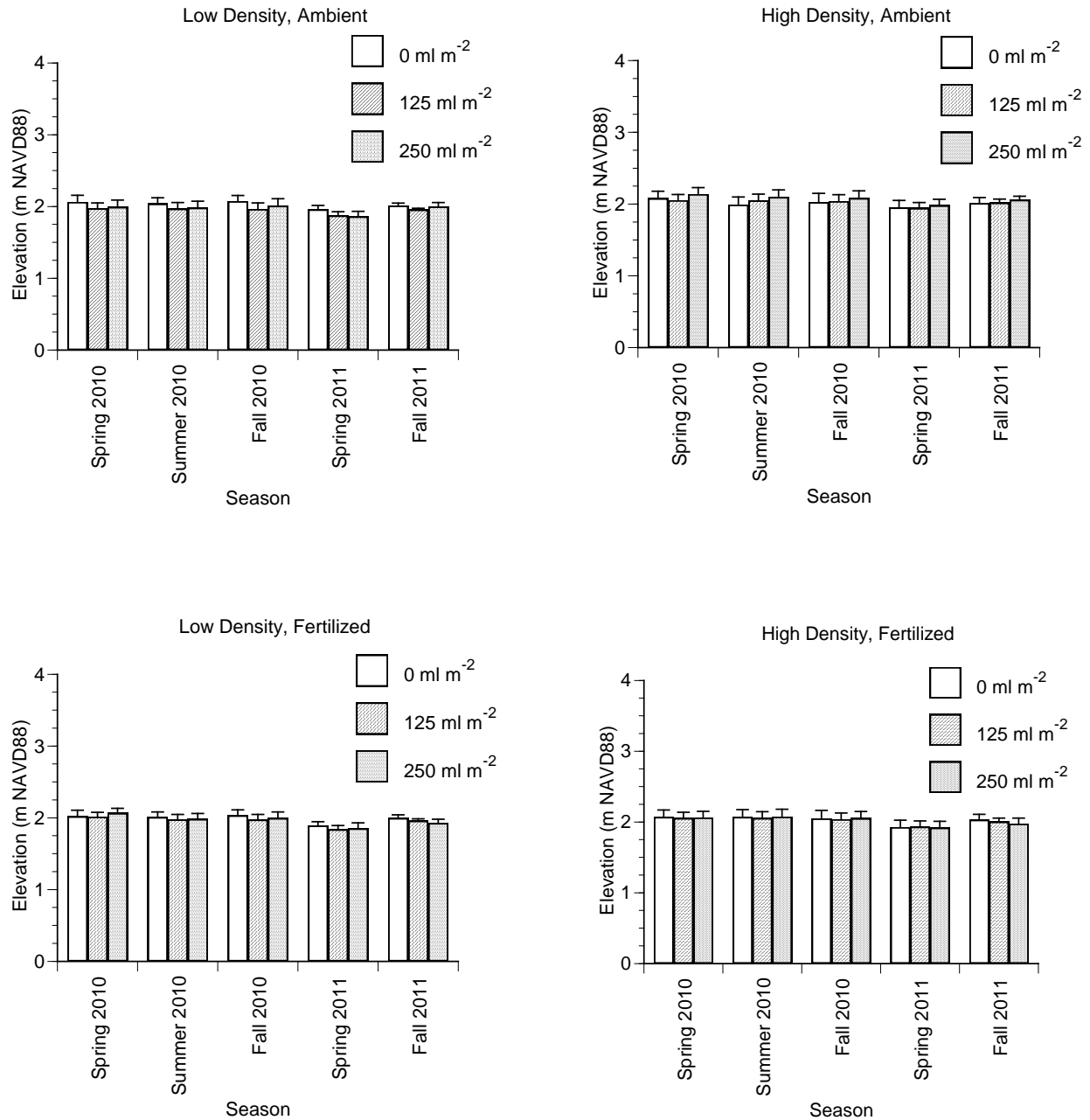


Figure 5.18 The effect of season, planting density, fertilizer regime, and humic acid amendment on *Uniola paniculata* plot elevation (m; mean \pm SE, n = 5, Sp 2010 LSD = 0.297, Su 2010 LSD = 0.250, Fa 2010 LSD = 0.254, Sp 2011 LSD = 0.229, Fa 2011 LSD = 0.200).

Panicum amarum

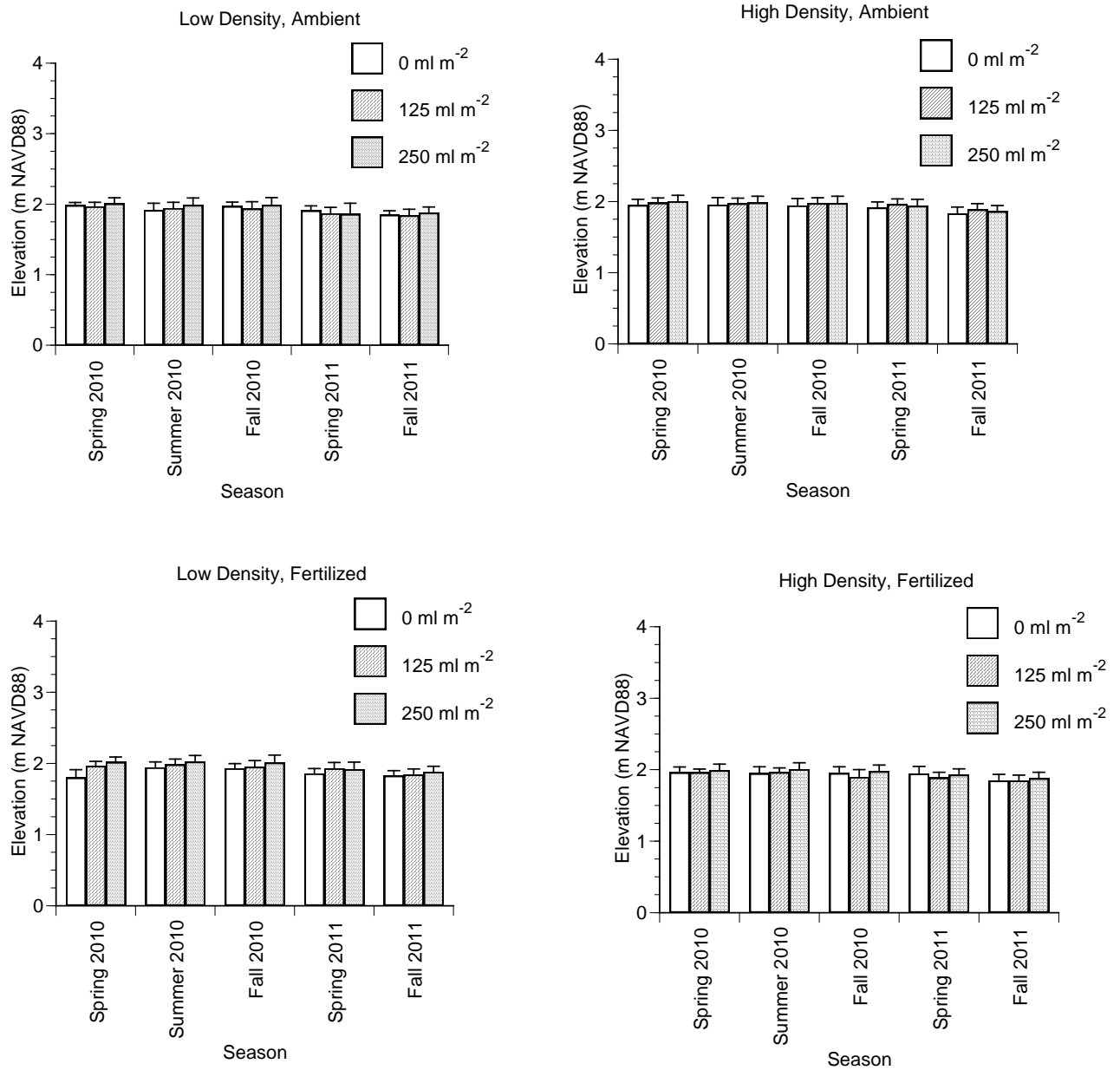


Figure 5.19 The effect of season, planting density, fertilizer regime, and humic acid amendment on *Panicum amarum* plot elevation (m; mean \pm SE, n = 5, Sp 2010 LSD = 0.297, Su 2010 LSD = 0.250, Fa 2010 LSD = 0.254, Sp 2011 LSD = 0.229, Fa 2011 LSD = 0.200).

Spartina patens

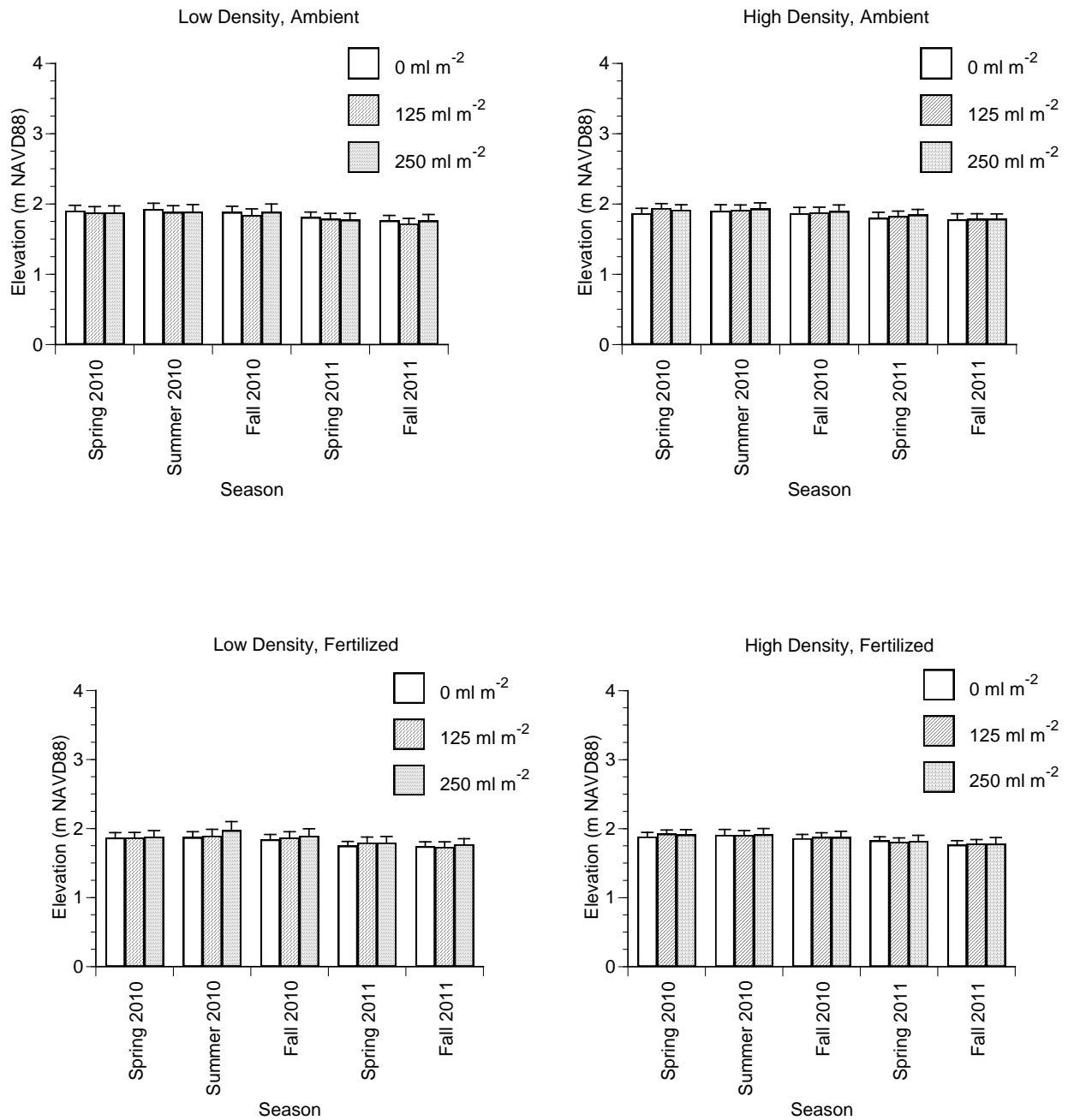


Figure 5.20 The effect of season, planting density, fertilizer regime, and humic acid amendment on *Spartina patens* plot elevation (m; mean \pm SE, n = 5, Sp 2010 LSD = 0.297, Su 2010 LSD = 0.250, Fa 2010 LSD = 0.254, Sp 2011 LSD = 0.229, Fa 2011 LSD = 0.200).

summer 2010 sampling. Highly significant interactions of season and planting density (Tables A5.1–A5.6; $F = 5.0$, $p < 0.01$) as well as season, species, and planting density (Tables A5.1–A5.6; $F = 7.2$, $p < 0.01$) were detected, resulting from *S. patens* leaf tissue nitrogen not showing the consistent reduction in fall 2010 as the other species. Highly significant interactions of season and fertilizer regime (Tables A5.1–A5.6; $F = 9.9$, $p < 0.01$) as well as season, species, and fertilizer regime (Tables A5.1–A5.6; $F = 3.6$, $p < 0.01$) were detected for leaf nitrogen. These significant effects result from fertilized plots having greater leaf tissue nitrogen than unfertilized plots for all species in summer and fall of 2010, but in spring of 2011, *S. patens* leaf tissue nitrogen was lower in fertilized plots than unfertilized plots, but *U. paniculata* and *P. amarum* continued to have higher leaf tissue nitrogen in fertilized plots.

A highly significant effect of season (Tables A5.1–A5.6; $F = 103.9$, $p < 0.01$) as well as a highly significant interaction of season and species (Tables A5.1–A5.6; $F = 136.9$, $p < 0.01$) was detected for plant tissue phosphorus, resulting from a substantial jump in the phosphorus concentrations of *U. paniculata* leaf tissue in fall 2011 compared with *P. amarum* and *S. patens*. Similarly, a highly significant effect of season (Tables A5.1–A5.6; $F = 85.4$, $p < 0.01$) and interaction of season and species (Tables A5.1–A5.6; $F = 116.1$, $p < 0.01$) was detected for plant tissue potassium, but in this case it was the result of *P. amarum* leaf tissue having elevated potassium levels at the beginning of the study (summer 2010) and then dropping to levels similar to those exhibited by *U. paniculata* and *S. patens*.

Soil Physico-Chemical Characterizations

A highly significant effect of season (Tables A5.7–A5.21; $F = 313.1$, $p < 0.01$) and interaction of season and species (Tables A5.7–A5.21; $F = 27.9$, $p < 0.01$) was detected for soil pH, driven by soil pH becoming less basic over time, except for *S. patens* plots, which remained basic until fall 2011. No other main effects or interactions regarding soil pH were detected. Soil conductivity was higher for *S. patens* plots than for *U. paniculata* and *P. amarum* plots at the beginning of the study, but all plots showed a substantial reduction in soil conductivity in fall 2011, leading to a highly significant effect of season (Tables A5.7–A5.21; $F = 5.7$, $p < 0.01$) and interaction of season and species (Tables A5.7–A5.21; $F = 3.4$, $p < 0.01$). Season and the interaction of season and species had highly significant effects on soil moisture (Tables A5.7–A5.21; $F = 50.8$, $p < 0.01$; $F = 22.3$, $p < 0.01$, respectively), with soil moisture being lowest in spring 2011, especially for *U. paniculata*. A marginally significant interaction of season and planting density was found (Tables A5.7–A5.21; $F = 2.3$, $p < 0.1$), in which high-density planting had lower soil moisture except in fall 2010.

Soil ammonium varied quite significantly by season (Tables A5.7–A5.21; $F = 13.5$, $p < 0.01$), but no other significant main effects or interactions were detected. A highly significant effect of season (Tables A5.7–A5.21; $F = 6.6$, $p < 0.01$) and an interaction of season and species (Tables A5.7–A5.21; $F = 2.3$, $p < 0.05$) on nitrate-nitrite was detected, with nitrate-nitrite tending to increase in fall 2011 for all species but *S. patens*. No other main effects or interactions regarding nitrate-nitrite were detected. Soil phosphorus was higher in fall 2010 for *U. paniculata* and *P. amarum* than other seasons, but higher for *S. patens* in summer 2010 than other seasons, leading to a highly significant effect of season (Tables A5.7–A5.21; $F = 162.2$, $p < 0.01$) as well as interaction of season and species (Tables A5.7–A5.21; $F = 47.0$, $p < 0.01$). No other main

effects or interactions regarding phosphorus were detected. A highly significant effect of season ($F = 8.3$, $p < 0.01$) and a significant interaction of season and species (Tables A5.7-A5.21; $F = 2.2$, $p < 0.05$) on soil potassium was detected, with soil potassium being higher for *P. amarum* and *S. patens* plots than for *U. paniculata* plots in all sampling seasons except fall 2011, at which time all plots became equivalent. Interestingly, a marginally significant interaction of season, species, humic acid amendment, and fertilizer on soil potassium was noted (Tables A5.7–A5.21; $F = 1.6$, $p < 0.1$) in unfertilized *S. patens* plots, with soil potassium increasing in fall 2010 and spring 2011 with humic acid amendment.

New Cut

A highly significant effect of time (Figures 5.21–5.25; $F = 7.8$, $p < 0.01$) and a highly significant interaction of time and species-planting density (Figures 5.21–5.25; $F = 9.0$, $p < 0.01$) were detected for total cover, where total cover in the high density-*S. patens* treatment was greater in spring 2011 than in fall 2010, but the low density-*S. patens* treatment was equivocal in fall 2010 and spring 2011. No other significant effects were detected for total cover. Soil pH was significantly less basic in spring 2011 than in fall 2010 (Tables A5.22–A5.26; $F = 41.0$, $p < 0.01$). No other significant effects were detected for soil pH. Soil conductivity was significantly lower in spring 2011 than in fall 2010 (Tables A5.22–A5.26; $F = 65.9$, $p < 0.01$). No other significant effects were detected for soil conductivity. Soil organic matter was significantly lower in spring 2011 than in fall 2010 (Tables A5.22–A5.26; $F = 25.6$, $p < 0.01$), particularly in the high-density-*D. spicata* and low-density-*S. patens* treatments ($F = 2.6$, $p < 0.05$). No other significant effects were detected for soil organic matter. Soil ammonium was significantly higher in spring 2011 than fall 2010 (Tables A5.22–A5.26; $F = 8.4$, $p < 0.01$). No other significant effects were detected for soil ammonium. No significant effects of time, species-planting density, fertilizer regime, or humic acid amendment, or the interaction thereof, were detected for soil nitrate-nitrite or phosphorus concentrations. Investigation of the meteorological data acquired from the Bay Tambour weather buoy (Figure 5.26) indicates minimal precipitation ($< 5\text{mm}$) following the New Cut planting effort.

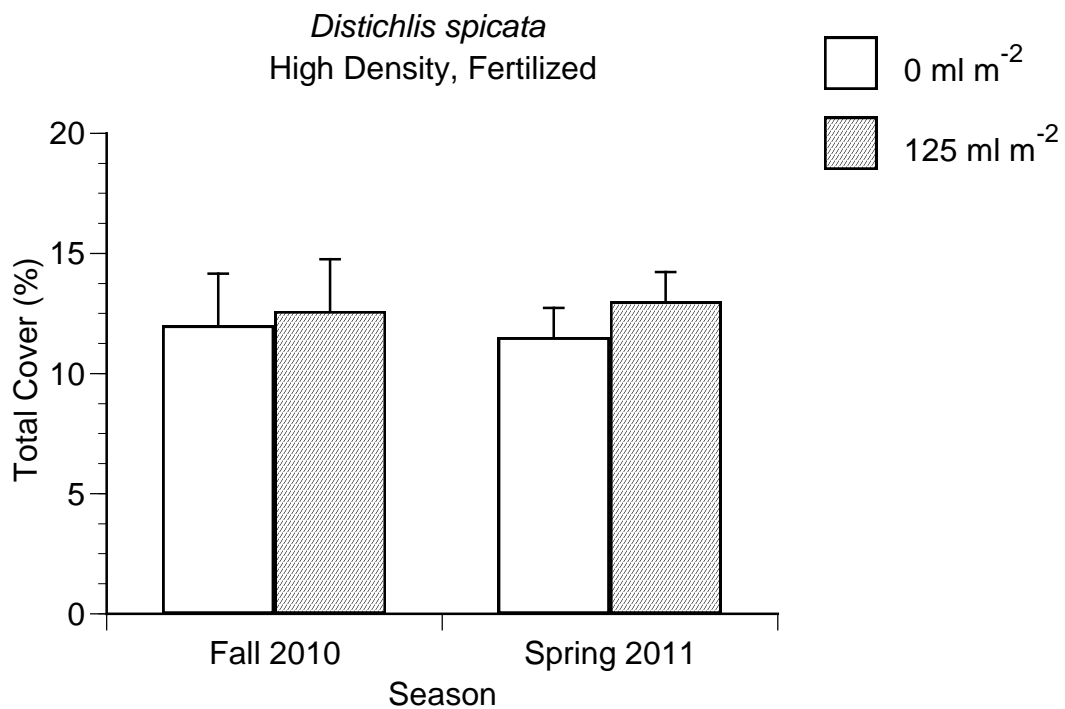
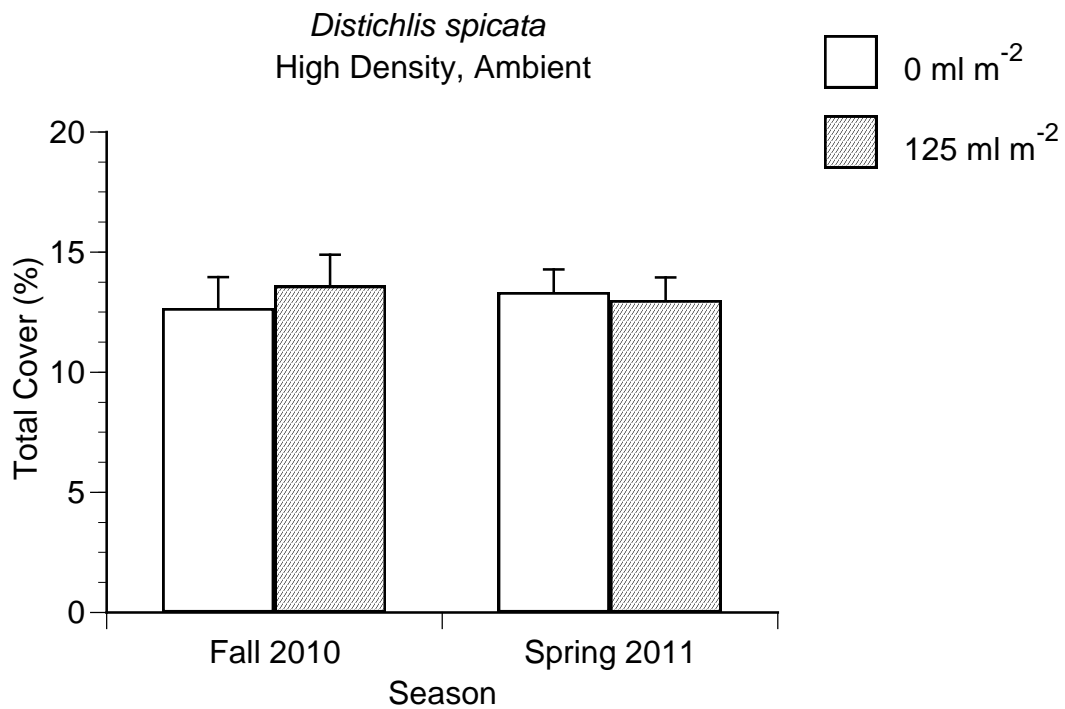


Figure 5.21 The effect of season, planting density, fertilizer regime, and humic acid amendment on *Distichlis spicata* total cover (mean \pm SE, $n = 5$, fall 2010 LSD = 3.63, spring 2011 LSD = 2.41).

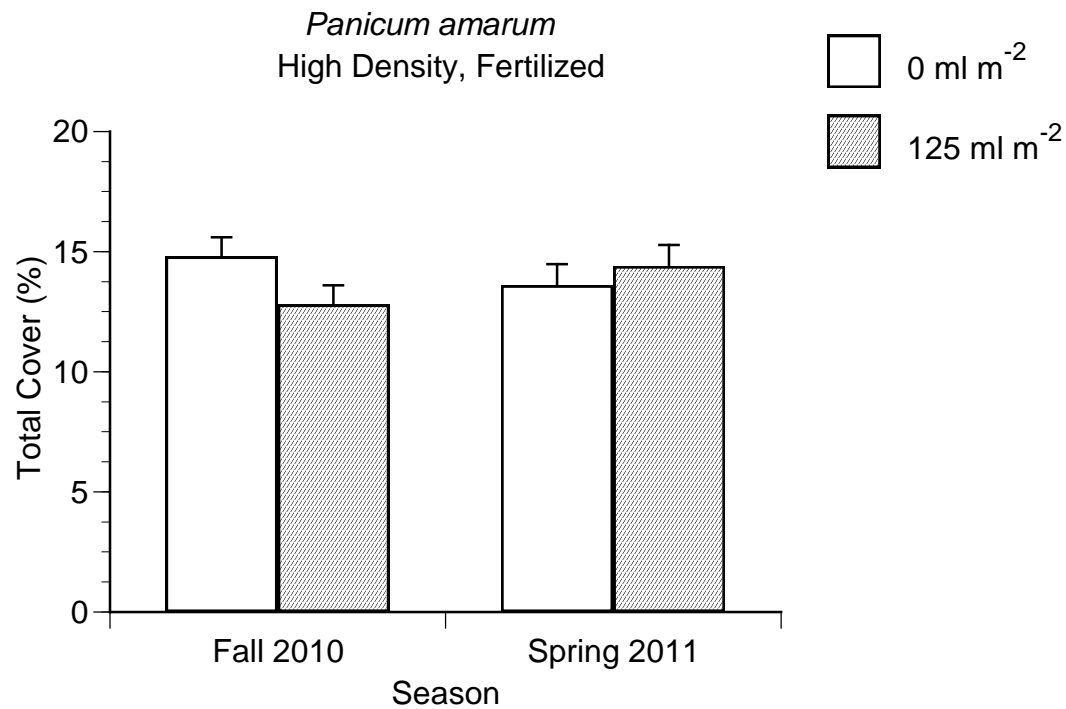
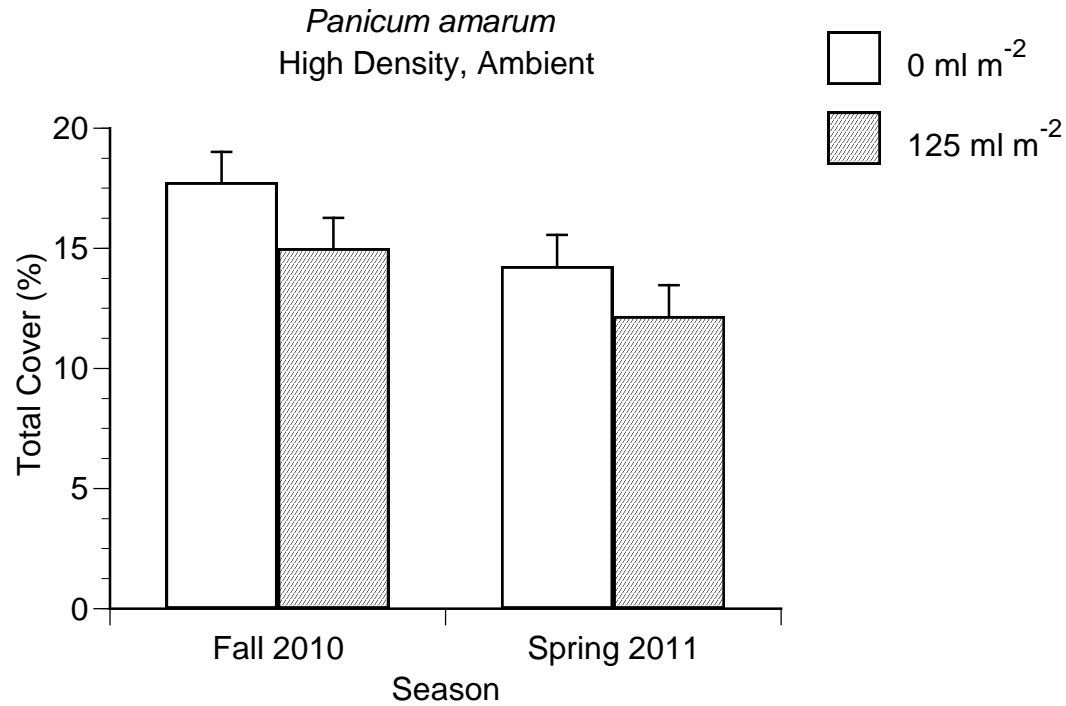


Figure 5.22 The effect of season, planting density, fertilizer regime, and humic acid amendment on *Panicum amarum* total cover (mean \pm SE, $n = 5$, fall 2010 LSD = 3.63, spring 2011 LSD = 2.41).

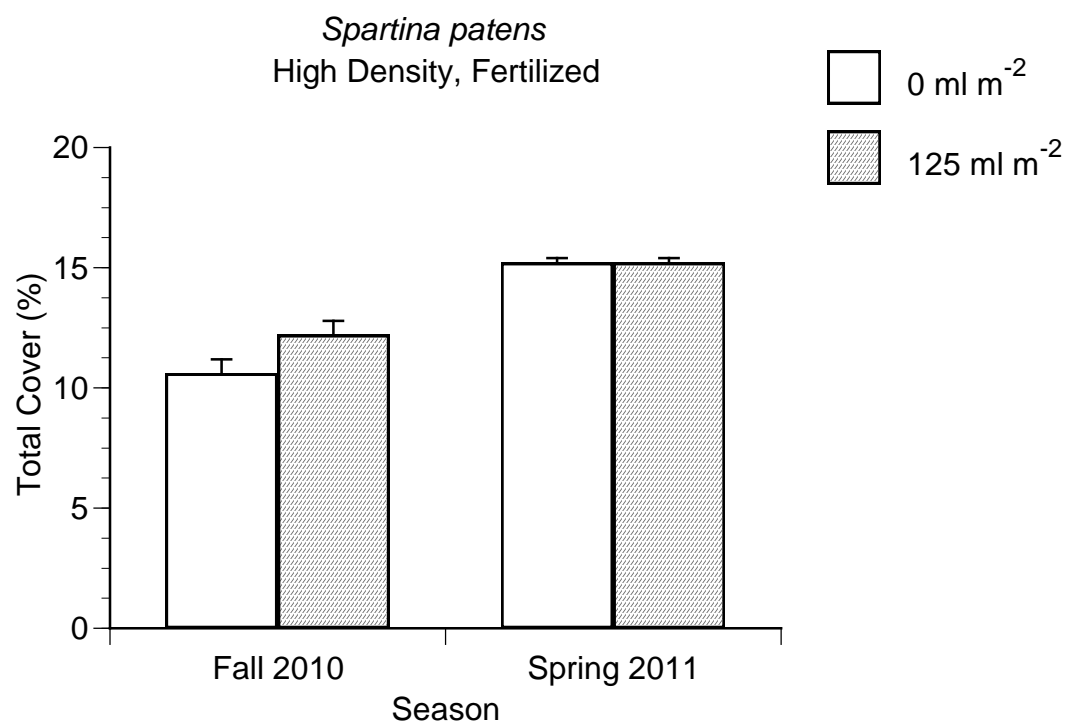
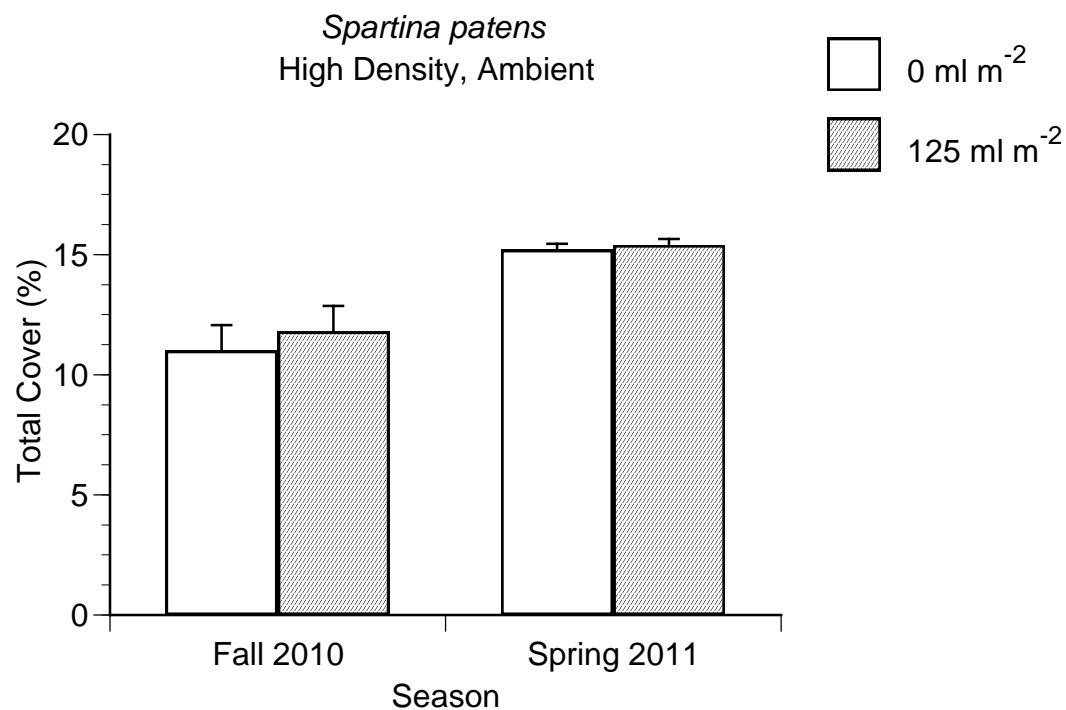


Figure 5.23 The effect of season, planting density, fertilizer regime, and humic acid amendment on *Spartina patens* total cover (mean \pm SE, $n = 5$, fall 2010 LSD = 3.63, spring 2011 LSD = 2.41).

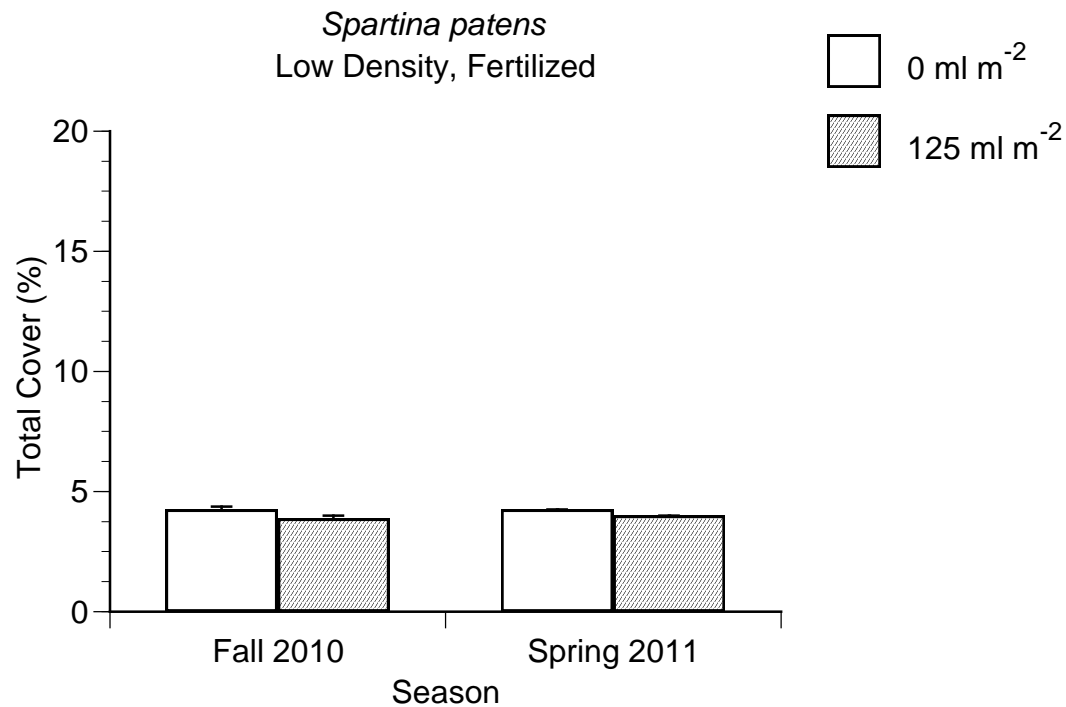
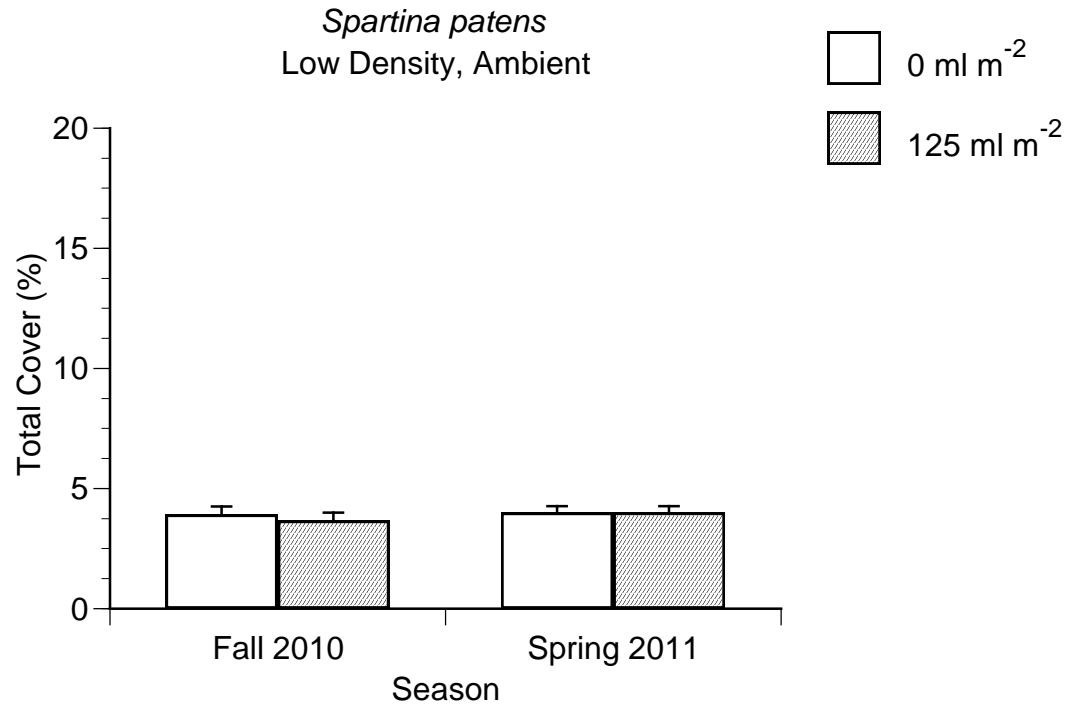


Figure 5.24 The effect of season, planting density, fertilizer regime, and humic acid amendment on *Spartina patens* total cover (mean \pm SE, $n = 5$, fall 2010 LSD = 3.63, spring 2011 LSU = 2.41).

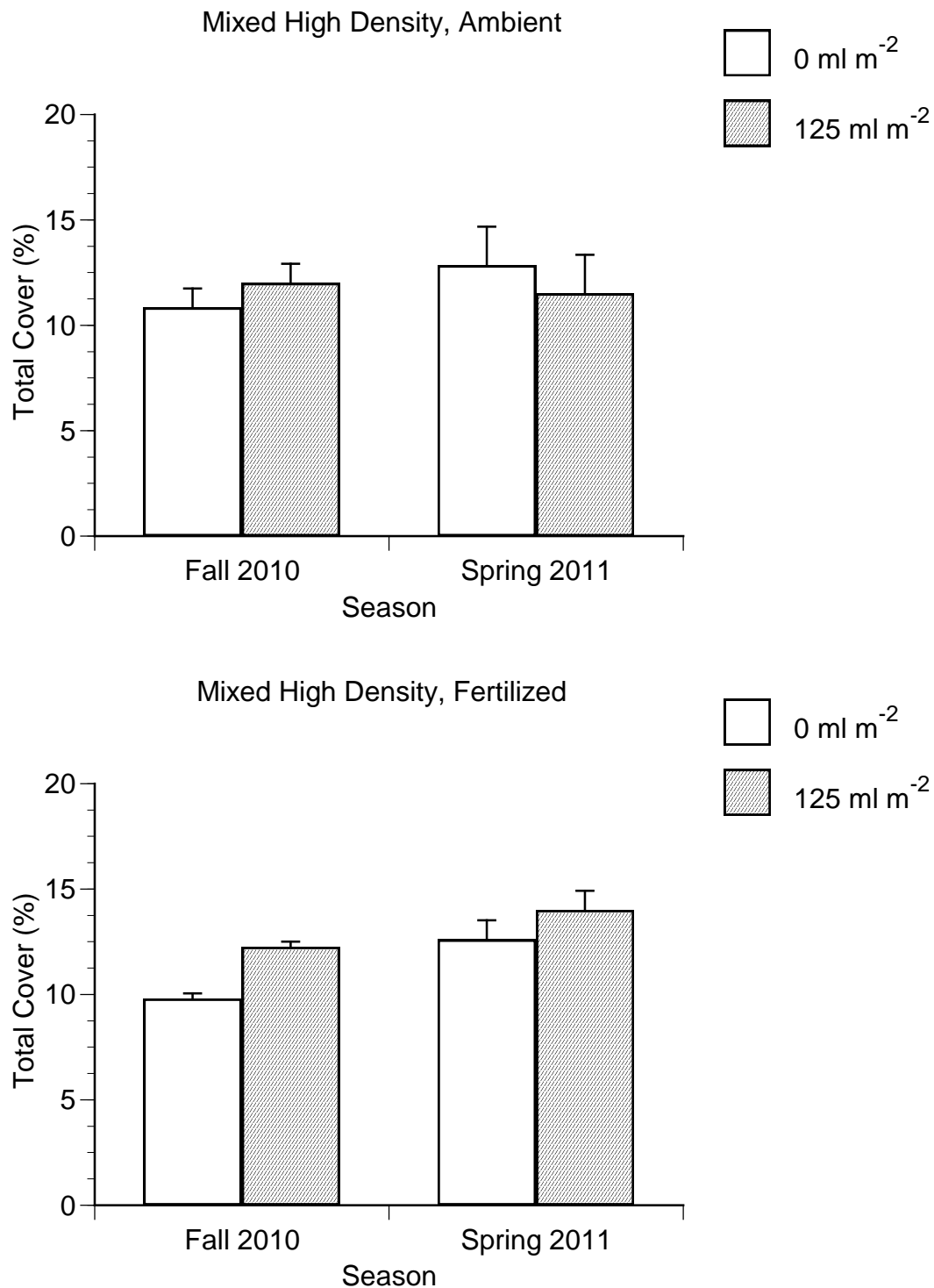


Figure 5.25 The effect of season, planting density, fertilizer regime, and humic acid amendment on mixed plot *Distichlis spicata*, *Spartina patens*, *Panicum amarum* total cover (mean \pm SE, $n = 5$, fall 2011 LSD = 3.63, spring 2011 LSD = 2.41).

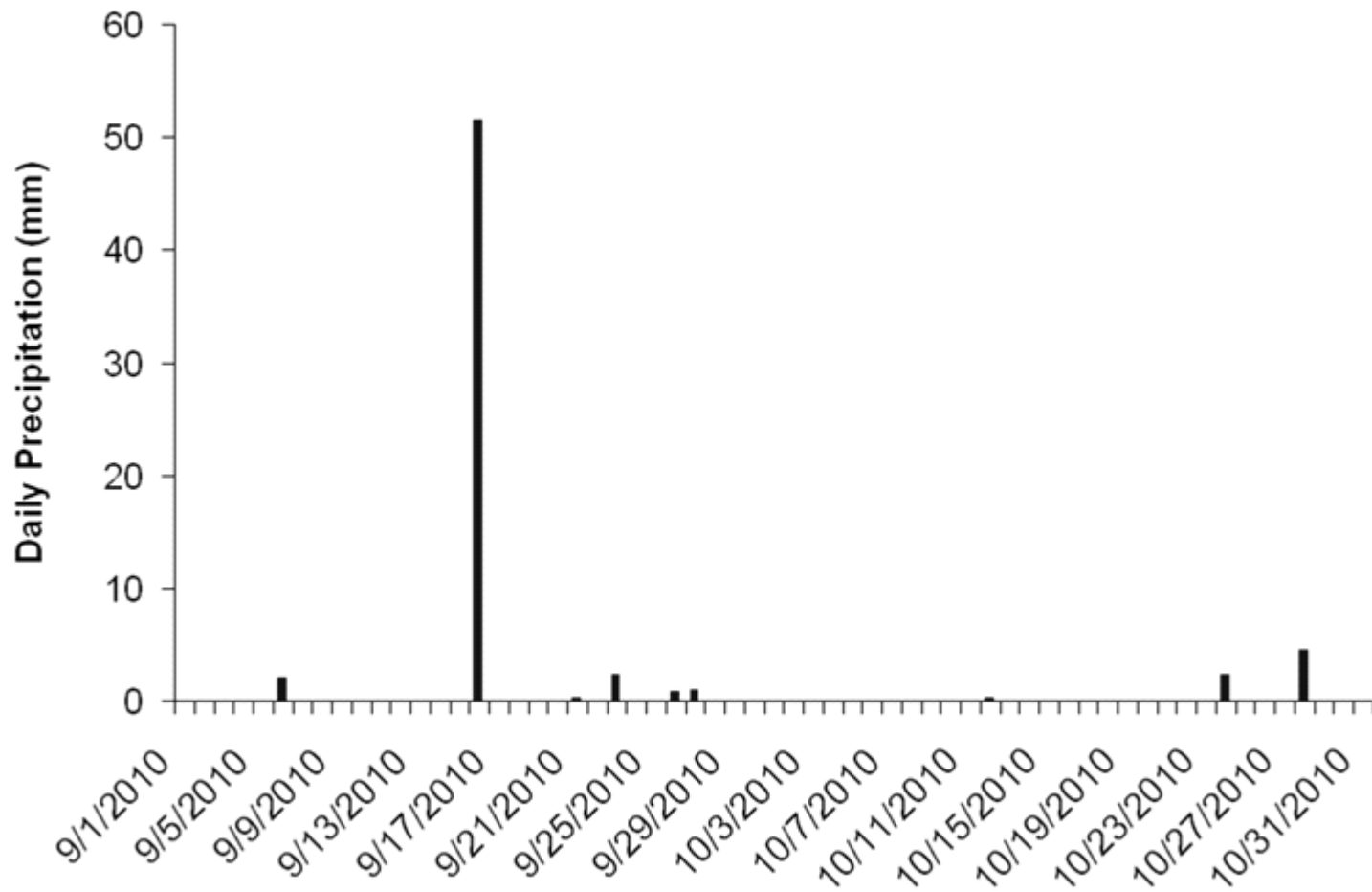


Figure 5.26 Daily precipitation amounts at the Bay Tambour meteorological station, representative of the New Cut planting site.

Discussion

This field study has revealed important information regarding the restoration of barrier island plant communities on the Louisiana coast. Specifically, a lasting benefit of higher density plantings in terms of increased vegetative cover and sand accretion for some dune and swale species was found. Also, the utility of a regular broadcast fertilizer regime as a key component of successful barrier island plantings was demonstrated. Less benefit was found for humic acid amendment treatments than was anticipated, very likely due to the unusually harsh growing conditions during the study period. Also, the sandy nature of the dune and swale soils at the field site may have prevented the retention of humic acid in the substrate, further limiting the effect of this amendment. Further, the long-term implications of including non-target plant species such as *C. dactylon* (Bermuda grass) in the restoration of Louisiana barrier island dunes and swales was revealed by this study. These results were apparent despite substantial perturbations regarding the planned implementation of this study due to the Deepwater Horizon oil spill and an anomalously extended period of warm, dry conditions.

Importantly, the high-density planting approach appears to result in a long-term benefit for *U. paniculata* in dune habitat and *S. patens* in swale habitat, as the high-density planting demonstrated much higher live cover of these species throughout both years of the study. Higher densities of individual plants of either the same or different species, particularly in stressful environments, can improve overall plant growth by ameliorating stressful soil conditions (Brooker and Callaghan 1998). Similar facilitation has been noted for seedlings in the dune environments of the southeastern U.S., where protection by existing vegetation from sand burial allowed greater establishment than in barren areas (Franks 2003). It should be emphasized that this benefit of essentially doubling live cover for *U. paniculata* and *S. patens* was maintained even when comparing fertilized treatments through the fall 2011 sampling, indicating that this benefit of increased live cover cannot be achieved by using lower density plantings with increased fertilizer application. However, *P. amarum* did not exhibit a sustained increase in live cover in the high-density planting treatment when compared with the low-density planting treatment, suggesting that there would be no substantial benefit in employing high density plantings with this species. *Panicum amarum* is generally considered to be a rapid colonizer of dune habitats in Louisiana (Hester and Mendelssohn 1990), and thus it was able to spread into open areas within plots more rapidly than *U. paniculata*.

Of great interest is that plot elevations were found to decrease less in 2011 for the high-density planting treatment than for the low-density planting treatment for both *U. paniculata* and *S. patens*, suggesting that the high-density plantings increased sand retention rates during tropical storm events. There is a trend toward increased plot elevation in the high-density plantings for *U. paniculata* and *S. patens* that was discernible in the first sampling period and became more apparent in later sampling periods. It should be noted that plot elevations during the first sampling period were determined after the completion of planting efforts as well as all other monitoring and sampling efforts. Thus, any increase in plot elevation represents an actual effect of the high-density planting rather than an effect of high-density planting location, which was randomized. This benefit of higher planting density on sand accumulation in dune habitats was also reported by Brown and Hafenrichter (1948a), who found that a higher planting density of

the dune grass *Ammophila arenaria* significantly increased sand dune volume subsequent to dune restoration projects in Oregon.

Importantly, live cover of *C. dactylon* was generally reduced in high-density plantings compared with low-density plantings for all three target species. This suggests that the high-density planting approach may confer an advantage for establishing preferred dune grass species when other, less desirable species may already be present. Quantum yield, which is an indicator of the photosynthetic capacity of a leaf and therefore a useful indicator of physiological stress, was determined in summer and fall of 2010, with the higher planting density treatment tending to display more optimal values. These quantum yield results indicate that the physiological stress experienced by individual plants was somewhat ameliorated by higher planting density—results that have been noted for other species (Fajardo and McIntire 2011). Average stem height was either similar between high-density and low-density planting treatments or increased in high-density plantings throughout the study, indicating that individual plant growth was not reduced by the higher density of planting units. Concentrations of extractable soil macro-nutrients (nitrogen, phosphorus, and potassium) were not reduced in the high-density planting treatments when compared with the low-density planting treatments for *U. paniculata* and *P. amarum*. That macro-nutrient resources are not being depleted to an extent that inhibits plant growth is important, given the scarcity of nutrients in dune ecosystems (Broome et al. 1982; Hester et al. 2005). This is corroborated by tissue macro-nutrient concentrations for all species, which were not substantially altered by planting density. Soil organic matter was not affected by planting density, indicating that although plant production was greater in the high planting density treatment, as can be surmised by the increased live cover with equivalent average canopy height, productivity was not translated into increased soil organic matter content within the time span of this study.

Fertilizer application increased live cover of all species, particularly *P. amarum*. Interestingly, *U. paniculata* and *S. patens* live cover was less dramatically increased than is typically reported for these species in other studies (Broome et al. 1982; Webb et al. 1980; Hester and Mendelssohn 1990; Willis and Hester 2010). However, this apparently reduced benefit of fertilizer application may be attributed to several factors. The grower unexpectedly incorporated fertilizer pellets directly into the planting furrows during the installation of all planting units. As a result, all plants, other than the additional plants installed by CPEL volunteers between grower planted units, received some level of fertilizer application at the initiation of the study. This is evident in examining *U. paniculata* average canopy height, which was increased by fertilizer application in the spring 2011 and fall 2011, but not during the 2010 sampling periods. Additionally, live cover of *C. dactylon* within all target species planting areas was greatly increased with fertilizer application starting in summer 2010 and in all sampling periods thereafter. Therefore, soil nutrients introduced by the fertilizer application may have been scavenged by *C. dactylon* before they could be acquired by the target planting species, as *C. dactylon* is known to respond quite favorably to fertilization (Burton and De Vane 1952; Webb et al. 1980). Although, *P. amarum* and *S. patens* do not display substantially increased average canopy height with fertilizer application, for *S. patens* this may reflect unfavorable soil conditions that limited the ability of plants to utilize available nutrients for growth. It is also important to note that this study occurred during a time of unusually low precipitation for coastal Louisiana; thus soil moisture rather than soil nutrient concentration may have been the limiting

resource for plant growth during this time period. Fertilizer application did not significantly alter soil pH or increase soil conductivity, indicating that fertilizer application at this rate did not exacerbate soil conditions. Interestingly, extractable soil ammonium, nitrate-nitrite, and potassium were not significantly different with fertilizer application. This is very likely a result of the substantial increase of *C. dactylon* presence, which appeared able to rapidly acquire and utilize the applied soil nutrients, as has been noted in other studies (Webb et al. 1980). This expansion of *C. dactylon*, which may limit sand transport where it has established, probably prevented a significant effect of fertilizer application on plot elevation from being discerned.

Humic acid amendment at the levels employed in this field study did not increase live cover or average canopy height for *U. paniculata*, *P. amarum*, or *S. patens*. These results are consistent with the associated greenhouse refinement study (Chapter 1), in which no effect of humic acid amendment was found for dosages of 125, 250, or 500 ml m⁻² for these species. However, this is in sharp contrast to the associated greenhouse range-finding study (Chapter 1) findings where *P. amarum*, and *S. patens* demonstrated substantial increases in live aboveground and belowground biomass with humic acid amendment levels of 100 to 900 ml m⁻². Although in the greenhouse range-finding study minimal stimulation of aboveground growth with humic acid amendment was found with *U. paniculata*, belowground biomass was significantly increased with 100 to 900 ml m⁻² humic acid. In a previous greenhouse study, Willis and Hester (2010) found a clear benefit of 5, 20, and 80 ml m⁻² humic acid amendment regarding cumulative stem height, aboveground, belowground, and total biomass for *P. amarum*, but found no increase in *U. paniculata* cumulative stem height, aboveground, belowground, or total biomass with humic acid amendment of 5, 20, and 80 ml m⁻². Increased aboveground live biomass for *P. amarum* was also found for humic acid amendment dosages of 20, 40, and 80 ml m⁻² in another greenhouse study examining humic acid amendment and vegetative fragments (Willis and Hester 2008).

No significant effect of humic acid amendment was found for extractable soil ammonium, nitrate-nitrite, phosphorus, or potassium. One mechanism by which humic acid amendment improves soil quality is that through this infusion of organic matter, the cation exchange capacity of soils is increased, which in turn allows for greater nutrient retention and exchange (Jackson 1993). The lack of significant differences between humic acid treatments may reflect the utilization of these nutrients by the plants prior to sampling. Given the nutrient-deficient nature of dune systems, and the time between samplings, it is quite possible that any available nutrients were acquired by the plantings, and nutrient levels were reduced to ambient levels. This is further corroborated by the lack of significant differences between fertilizer treatments, suggesting that nutrient levels are drawn down to ambient levels relatively quickly.

Soil organic matter was not significantly increased with humic acid amendment. Because of the relatively small amount of humic acid introduced into plots (125 or 250 ml m⁻²), compared to the scale of the soil organic matter determination technique (loss on ignition; %), it was not anticipated that humic acid amendment would directly affect soil organic matter content in a measurable fashion, but may affect belowground biomass productivity. Based on other studies available in the peer-reviewed literature (Willis and Hester 2008; 2010), as well as the greenhouse studies conducted as a portion of this research effort (Chapter 1), it was expected that belowground biomass may be increased. Because of the destructive nature of belowground biomass sampling, this metric was not included in the overall project design. However, because

of the robustness of the *P. amarum* plantings, we decided to collect belowground biomass cores during the final field sampling. A clear trend toward increased *P. amarum* belowground biomass was found in the field study with the 125 ml m⁻² humic acid amendment level in low density plantings. Thus, it is possible that an increase in soil organic matter would be detectable over a longer (decadal) study duration. Importantly, humic acid amendment did not substantially alter soil pH or conductivity in a negative fashion.

Humic acid amendment (4% active ingredient) can be implemented during planting efforts in dune and swale environments at a cost of approximately \$4,100 hectare⁻¹ at a rate of 125 ml m⁻² based on a product cost of \$3.70 liter⁻¹. Note that this does not include the one-time costs of a sprayer device to apply the humic acid, which is necessary for large-scale applications. Current costs for ATV mountable power sprayers range from \$200 to \$500. A broadcast fertilizer regime can be implemented at a rate of 878.8 kg ha⁻¹ of 8-8-8 fertilizer and 195.3 kg ha⁻¹ ammonium nitrate for approximately \$250 hectare⁻¹ and \$110 hectare⁻¹ per application, respectively. The costs of increased planting densities are simply a multiplier of current planting density costs, although the larger plant number may result in a lower cost per plant.

Given the value of the rapid establishment and expansion of key dune-building species, such as high-density plantings of *U. paniculata* to retaining scarce sand resources in Louisiana dune systems, the high-density planting approach should be considered as an additional technique when designing planting approaches using *U. paniculata*. However, the cost of employing a similar high-density planting approach for *P. amarum* is not warranted, when the additional planting cost for marginally increased plant cover is considered. The use of a broadcast fertilizer regime has been shown to be effective in increasing vegetative coverage with a relatively low cost. Humic acid amendment was not as effective as broadcast fertilizer in enhancing vegetative coverage, and both the product and the application equipment are more expensive. Thus, although humic acid amendment may be appropriate for consideration in certain restoration scenarios, these results do not indicate that it should be included as a component of typical restoration efforts. The inclusion of *C. dactylon* seeding into restoration efforts and its subsequent presence at restoration sites may bear further discussion in the restoration community, as its presence was associated with a reduced coverage of target dune and swale species.

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CHAPTER 6. FIELD STUDY ASSESSING THE ESTABLISHMENT OF *BACCHARIS HALIMIFOLIA* THROUGH HYDROMULCH-ASSISTED SEED DISPERSAL

Introduction

Barrier island restoration sites benefit from the quick establishment of healthy vegetation. Belowground root structures bind sediments in the desired location, preventing relocation during overwash events (Hester et al. 2005). Aboveground vegetative structures are equally important in sediment stability by serving to trap windblown sands, allowing them to stay in the barrier island system. *Baccharis halimifolia* is a rapidly growing woody shrub that is not currently used in barrier island restoration, though it may be a good candidate for inclusion in restoration efforts in the swale environment. *Baccharis halimifolia* can reach 5 m in height and displays good salt tolerance as indicated by its presence in salt marsh and swale habitats. (Van Deelen 1991). *Baccharis halimifolia* is one of the few woody species occurring on Louisiana barrier islands, and its presence provides an additional dimension of habitat beyond that of nearby plants. The establishment of woody species in Louisiana barrier island restoration projects is recognized as highly desirable and plays a role in how potential projects are ranked (CWPPRA 2008). The highest scores are given to designs that incorporate two woody species.

Baccharis halimifolia can survive periodic flooding and drought events. Its existence in coastal habitats is indicative of its salt tolerance. Young et al. (1994) found *B. halimifolia* growing in salinity of 2 – 5 g L⁻¹ and depth to groundwater from 15 to 25 cm. Studies performed with irrigation solutions of up to 12 ppt salinity had no effect on *B. halimifolia* growth (Graves 2003). *Baccharis halimifolia* can grow over a wide range of soil pH. Values between pH 5 and 9 with a mean of 7.2 +/- 0.3 have been observed along the Gulf Coast (Westman 1975). Mature *B. halimifolia* plants can produce up to 1.5 million seeds, and seedlings start seed production as early as the second growing season (Westman 1975). Seeds are wind- and water-dispersed and can be relocated several miles away from the parent colony. Germination has been found to occur within a month of seed set, which indicates little or no required dormancy period. Once germination requirements are met, seeds typically take 7–16 days to germinate (Westman 1975). The goal of these field trials was to investigate the feasibility of establishing *B. halimifolia* by seed on a newly created restoration site using the information gathered from the previously described greenhouse studies.

The previously conducted greenhouse studies revealed that *B. halimifolia* seed germination is optimal when seeds are kept at the sediment surface. Greenhouse experiments with hydromulch also indicate that *B. halimifolia* seed germination is increased when hydromulch is used in sediments with low organic matter. The reservation of soil moisture near the surface is critical for germination. Hydromulch, a mixture of wood/cellulose fibers and water, has the advantages of moisture retention and the ability to hold seeds in the desired location within a given restoration site. However, greenhouse studies have shown that a subsequent rainfall event is necessary for reliable *B. halimifolia* seed germination, even when using hydromulch. Two field experiments were conducted. The first experiment was initiated within the swale habitat of the Whiskey Island TE-50 *S. patens* plantings (2010) and monitored the success of *B. halimifolia* seed germination in hydromulch. The second experiment was initiated

in April 2011 within natural stands of *S. patens* with the added application of a simulated rainfall event. *B. halimifolia* seed germination within hydromulch was monitored.

Methods and Materials

Hydromulch Field Study 2010

The 2010 hydromulch *Baccharis halimifolia* field study was initiated on October 15, 2010. The factorial experimental design consisted of 1 m² plots within the 125 ml m⁻² treatment of the TE-50 *S. patens* plantings (2010) on Whiskey Island (Figure 6.1). Treatments included two *S. patens* density levels (high, low) and two fertilizer levels (fertilized, ambient), and each treatment was replicated five times. The high planting density for *S. patens* (0.5 m centers) was triple the low planting density (1.52 m centers). Fertilizer application was accomplished using chest-mount fertilizer spreaders, with 8-8-8 applied in spring 2010 and spring 2011 at a rate of 878.4 kg ha⁻¹ and with ammonium nitrate applied in summer 2010 and fall 2010 at a rate of 195.3 kg ha⁻¹. *Baccharis halimifolia* seeds for this study were collected from mother plants growing in Louisiana's coastal zone during the fall of 2009. Each experimental plot was sprayed with 5.68 liters of hydromulch slurry containing 30,000 *B. halimifolia* seeds per plot. *Baccharis halimifolia* seed germination within each plot was then monitored on October 27, 2010.

Hydromulch Field Study 2011

A field study was conducted in April 2011 testing the establishment of *B. halimifolia* from application of hydromulch and *B. halimifolia* seed using information gathered from the first field study and the greenhouse experiments. A field site was selected on a previously restored portion of Whiskey Island but beyond the recently planted area. Plots 1 m² in size were established in a factorial experimental design employing two *S. patens* canopy cover treatments (high 20–35% cover, low 5–15% cover) and two precipitation regimes (no additional precipitation and a one-time application of 30.8 mm, the weekly average precipitation for coastal Louisiana, which was applied three days after hydroseeding). Soil samples were collected to a depth of 2 cm using a stainless steel soil scoop, immediately bagged, and kept cold until they were transported to the lab. The soil samples were weighed and then dried at 65° C until a constant weight was achieved and soil moisture was calculated. A 1:2 ratio of dry soil to deionized water was used for the determination of pH and conductivity. *B. halimifolia* seeds for this study were collected from mother plants growing in Louisiana's coastal zone during the fall of 2010. Each treatment plot received 5.68 liters of hydromulch slurry containing 30,000 *B. halimifolia* seeds in a total of 32 experimental plots. Two *S. patens* canopy cover (high, low) control plots, which received no hydromulch or *B. halimifolia* seed, were also established within each block to test for natural *B. halimifolia* recruitment. Three days after the start of the experiment, the treatment plots that required additional precipitation were watered with 30 liters per plot of water.

The *B. halimifolia* germination success within plots was determined 10 days later. The plots were revisited in June and October of 2011 for visual estimates of germination success. Precipitation data were collected from a weather buoy in Tambour Bay for the 2010 study and the Terrebonne Bay gauge for the precipitation estimates during the 2011 study.



Figure 6.1 Site map for experimental *Bacharris halimifolia* restoration project on Whiskey Island. Black outlines represent experimental *B. halimifolia* establishment blocks, with each block containing all treatments.

Results

Hydromulch Field Study 2010

The 2010 field study was monitored on October 27, 2010. No *B. halimifolia* seed germination was detected. Rain data from the Tambour Bay rain gauge indicated that the area received no rainfall during the month of October 2010. (Figure 5.25)

Hydromulch Field Study 2011

Initial soil pH and conductivity for the second study are listed in Table 6.1. Initial soil moisture was determined to be highly significant between cover treatments (Figure 6.1; $F = 10$, $p < 0.01$). Monitoring of the field study 10 days after setup indicated no *B. halimifolia* germination. A second monitoring took place in June and a third in October both of which indicated no *B. halimifolia* establishment. Observations from the second monitoring trip determined that some of the hydromulch plots were covered by 0.5cm of sand. Precipitation data from the Terrebonne rain gauge indicated 30 days of no rain following the start date of this experiment (Figure. 6.2)

Table 6.1 The effect of *Spartina patens* canopy cover on soil pH and conductivity (mean +/- SE, n = 8).

	Low <i>S. patens</i> cover	High <i>S. patens</i> cover
pH	8.87	8.44
$\pm SE$	0.10	0.09
Conductivity	1148.10	409.19
$\mu S\ cm^{-1}; \pm SE$	213.46	104.61

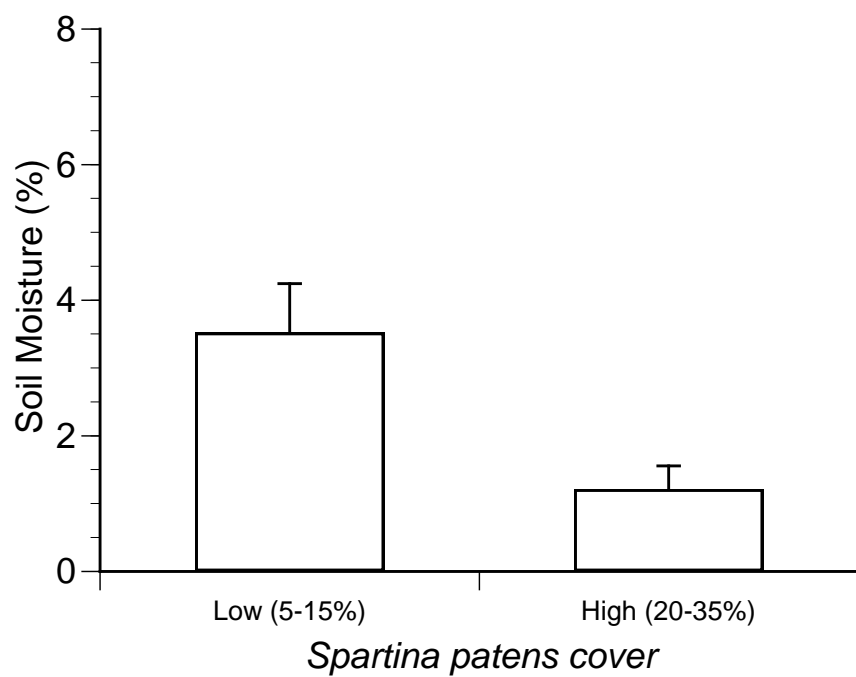


Figure 6.2 The effect of *Spartina patens* canopy cover on soil moisture (% mean \pm SE, n = 8, LSD = 1.601).

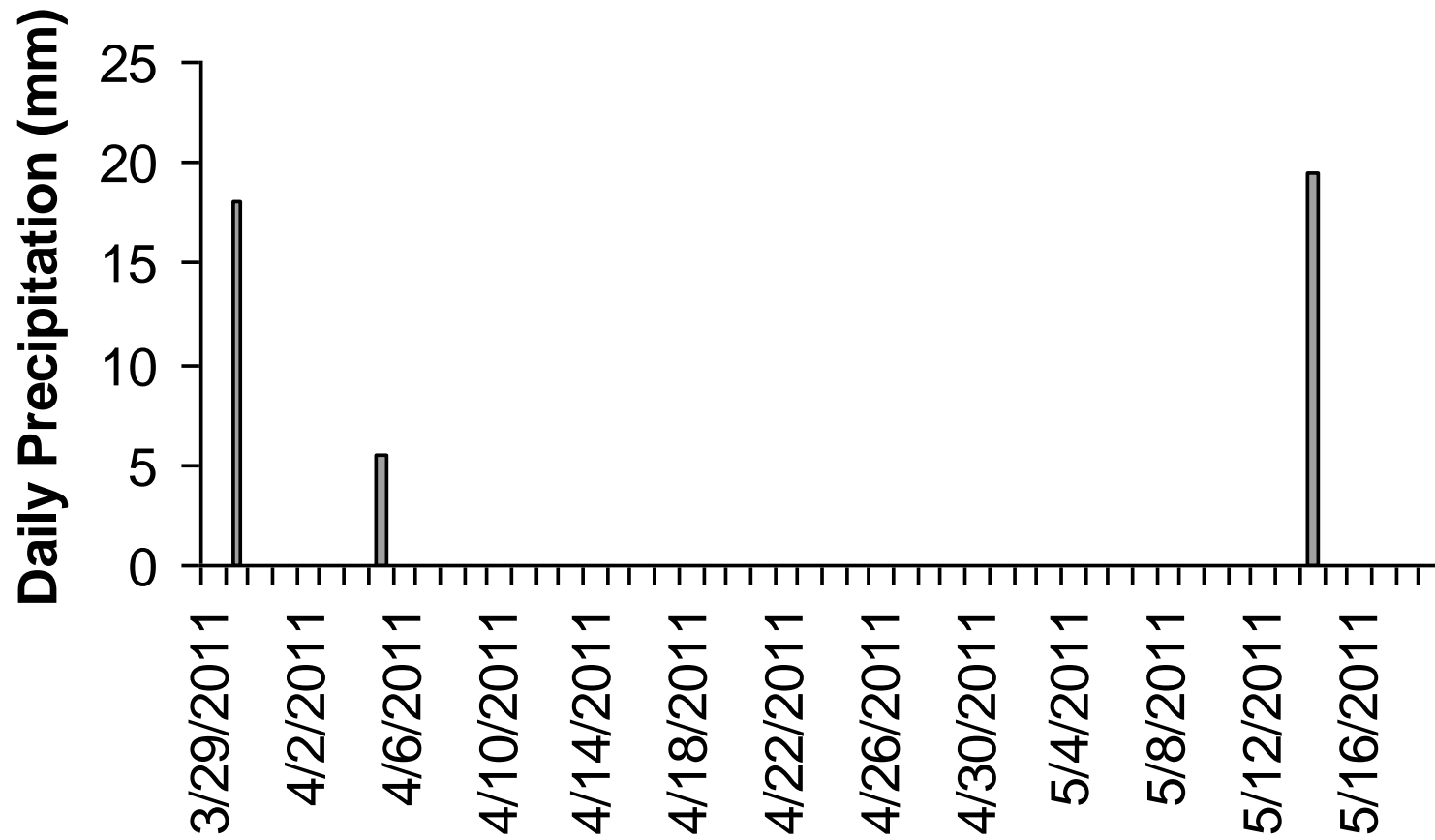


Figure 6.3. Daily precipitation amounts at the Terrebonne Bay meteorological station, representative of the 2011 Whiskey Island *Baccharis halimifolia* hydroseeding setup and first monitor.

Discussion

Contrary to our expectations, hydromulch was not successful in promoting germination of *B. halimifolia* at the Whiskey Island field sites in 2010 or 2011. Several factors contribute to the success or failure of *B. halimifolia* germination, and the results of these studies highlight some of those factors.

Soil Moisture

Soil moisture is critical for seed germination to take place on the porous sands of dune and swale barrier island restoration projects. Liu et al (2011) found that *Eremosparton songorium* needed a minimum of 2.0% soil water content to achieve successful seed germination and establishment. Our results indicate an initial soil moisture content between 1% and 3% with greater soil moisture occurring in the low-density *S. patens* cover treatment than in the high-density cover treatment. While a study done by Egerova et al. (2003), indicating the facilitation effects of *S. alterniflora* on *B. halimifolia* seedling survival may be from soil moisture positively associated with vegetative cover, the opposite was true at the Whiskey Island study site. Soil at this site is primarily sand, which has little water-holding capacity. Based on field measurements on Whiskey Island of *S. patens* swale habitat, soil moisture was low and ranged between approximately 1% and 10% (Chapter 5), demonstrating the stressful conditions in this habitat. Plants in the swale habitat, including *S. patens*, may be limited by water in this arid location, and a higher density of plants may put a greater demand on water due to greater transpiration. For example, the soil moisture measured in Chapter 5 for *S. patens* plots during the summer of 2010 was higher in low-density plots than in high-density plots (Table A5.18)

Precipitation

In the Whiskey Island swale, freshwater delivery is accomplished solely by rainfall events. In both years, natural precipitation did not occur between the project setup and monitoring, and did not occur for at least an additional four weeks following monitoring. Dry conditions may have also contributed to the amount of windblown sand as well. Lack of rainfall, or drought conditions, can negatively impact restoration success. For example, vegetative plantings of *S. patens*, *Panicum amarum*, *Spartina alterniflora*, and *Avicennia germinans* had low transplant success, most likely due to drought conditions (Khalil and Lee 2006). In addition, plant species composition shifts can occur due to drought-induced salinity changes (Visser et al 2002), causing increased competition in the target species. In our study, the simulated rainfall event did not result in germination of *B. halimifolia*. Germination of *B. halimifolia* was expected after 7 to 10 days, based on the previous research conducted with hydromulch in the greenhouse studies (Chapter 2). However, in both 2010 and 2011, germination had not occurred within 10 days at the field sites. This may be due to the greenhouse germination studies having slower moisture evaporation rates from the surface of the hydromulch due to the ideal humid conditions of the greenhouse. Light also is a critical factor for germination of *B. halimifolia* (Panetta 1979), and sand burial greater than 0.5 cm inhibits germination (Chapter 2). Site visits in the following spring and fall revealed that the hydromulch had been partially buried by sand or possibly removed altogether. Sand burial or removal could have inhibited germination within the plots.

Future Suggestions

Baccharis halimifolia establishment by hydroseeding on sandy sediments should take place when local weather conditions are predicted to include a period of rain, and when germination is induced before windblown sand has a chance to completely cover the hydromulch layer, inhibiting germination.

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CHAPTER 7. ENHANCEMENT OF *AVICENNIA GERMINANS* RESTORATION AT THE WHISKEY ISLAND CREATED SALT MARSH PLATFORM

Introduction

Mangrove restoration is one way in which we can respond to and compensate for the effects of climate change that are likely to decrease mangrove coverage (Gilman et al. 2008). Mangrove restoration to date has been attempted in mostly tropical latitudes where mangroves often occur extensively, and lessons from these attempts are available to guide future restoration attempts (Kairo et al. 2001; Toledo et al. 2001; Lewis 2005; Gilman and Ellison 2007). Unlike tropical latitudes, southern Louisiana is restricted to a single species, black mangrove (*Avicennia germinans* (L.) L.) for mangrove restoration. Further, black mangrove in southern Louisiana interacts with herbaceous, *Spartina alterniflora*-dominated salt marsh rather than traditional forested mangrove coastal ecosystems. Nonetheless, tropical mangrove restoration attempts can provide a foundation for guiding mangrove restoration in Louisiana. This foundation can be strengthened by implementing succession- and facilitation-based restoration theory that may be useful in guiding salt marsh plant community restoration (Zedler 2000; Gedan and Silliman 2009). This approach could be especially beneficial at restored barrier island salt marsh platforms, which are initially devoid of vegetation after sediment is pumped to the target elevation.

Louisiana barrier islands are a naturally dynamic component of the Mississippi River delta complex, forming from the flanking barriers of erosional headlands following abandonment and submergence of an active delta. Barrier islands are reworked by storms, relative sea-level rise, and island rollover, ultimately being reduced to sand-rich shoals over time (Penland et al. 1988). Although barrier island land building and loss is dynamic in nature, the rate of barrier island erosion (1855–2002) is exceedingly high (Penland et al. 2005) and a cause for concern because barrier islands are highly desired coastal features that provide important storm protection for coasts and habitat for organisms (Stone and McBride 1998; Visser et al. 2005). Inevitably, the dynamic nature of barrier islands poses a challenge for restoration because restoration goals often aim to repair or re-establish historic and/or natural communities. Simenstad et al. (2006) recommended setting restoration goals that include re-establishing ecological functions and services rather than re-creating a historical state. Barrier island restoration provides an open canvas for demonstrating how restoration of ecological processes and functions can proceed successfully.

Considerations for successful mangrove restoration often focus on different scales of environmental gradients that affect establishment and ultimate distribution of mangroves (Duke et al. 1998; Krauss et al. 2008). Hydrology of the area must be appropriate for the species used in the restoration project (Turner and Lewis 1997; Field 1998; Lewis 2005). For example, black mangrove seedlings have greater growth rates at approximately 15 cm–30 cm above the water table; therefore, knowledge of the mean water level in combination with elevation of planting could facilitate restoration (Alleman and Hester 2011a). The mangrove restoration area should also be a low-energy intertidal area with fine-grain sediment (Field 1998). The decision to plant mangrove seedlings, hand-disperse propagules, or allow for natural regeneration depends on the extent of mangrove populations in the greater project area. Provisions can also be made to

ameliorate stressful conditions, particularly for bare areas often present in newly restored barrier islands. For example, prior establishment of *S. alterniflora* in back-barrier marshes can stabilize sediment, provide shade that reduces sediment temperature and the development of hyper-saline conditions, reduce water energy, and add organic matter to the substrate (Lewis and Dunstan 1976; Bruno 2000). Essential plant nutrients may also be low in bare areas, presenting the need for soil amendments. Comparison of the restoration area to a reference (natural) area can provide a means for assessing the success of meeting the original project goals and objectives (Field 1998; Lewis 2005).

Specific restoration techniques investigated in this research aimed to further improve environmental conditions for *A. germinans* propagules. The first technique was combining the facilitating mechanisms of transplanted *S. alterniflora* with biodegradable fencing. The fencing was expected to reduce water energy, provide shade, lower soil salinity, be completely biodegradable in approximately five years or less, and maintain propagules at the targeted elevation of the restoration area. In addition to fences, humic acid, a naturally occurring component of soil organic matter that is a complex polymer insoluble at a pH less than 2, was investigated for its restoration enhancement potential (Zhang et al. 2002). Humic acid is considered a biostimulant and has been shown to stimulate plant growth and resistance to stress (Schmidt et al. 2003). Specifically, the complex structure of humic acid creates hormone-like activity, and as a result, increased amounts of antioxidants are produced when plants are grown under a drought stress, reducing the potential damage by radical oxygen species (Zhang and Schmidt 2000; Zhang and Ervin 2004). Importantly, humic acid can form complexes with necessary plant nutrients, particularly iron and zinc, which makes them more available to plants for uptake and stimulates growth (Mackowiak et al 2001; Chen et al. 2004; Sanchez et al. 2005). Plant responses vary considerably with the application of humic acid; some of the variability is attributed to the molecular size of the humic acid molecule as well as the pH (Piccolo et al. 1992; Nardi et al. 2002). Humic acid also occurs naturally in mangrove ecosystems; and for a Micronesian mangrove stand, humic acid concentrations varied by mangrove species and were positively correlated with root production (Gleason and Ewel 2002).

The potential value of humic acid in establishing plants in coastal restoration projects has recently begun to be investigated (Willis and Hester 2008; Willis and Hester 2010). Recent research on the application of humic acid to *S. alterniflora* and *A. germinans* identified a slight positive growth trend for *S. alterniflora* as the level of humic acid increased, particularly in belowground biomass, though the effect was not apparent for *A. germinans*. This result may have been due to the humic acid concentrations used ($0 - 80 \text{ ml m}^{-2}$) or the length of the study (6 months) (Willis and Hester 2010). When *S. alterniflora* and *A. germinans* are exposed to salinity stress, higher concentrations of humic acid may improve *S. alterniflora* significantly more than *A. germinans* growth due to the salinity tolerance mechanisms of *A. germinans* (Ball et al. 1984; Suarez et al. 1998). Greenhouse studies undertaken to inform this research effort (Chapter 1) suggest that both species benefit from low (300 ml m^{-2}) applications of humic acid amendment, but that *S. alterniflora* continues to benefit up to moderate (900 ml m^{-2}) dosages.

Objectives and Hypotheses

The objective of this study was to evaluate the potential for enhancing the establishment of *A. germinans* propagules on a constructed back-barrier salt marsh platform under factorial

conditions of humic acid dosage and propagule establishment techniques. We predicted establishment technique would have a strong effect on *A. germinans* survival and establishment due to the acceleration of succession by introducing a large cohort of target species propagules and mimicking succession by providing salt marsh structure through fencing and *S. alterniflora* transplants. Specifically, we expected the lowest levels of survival and establishment within bare plots and the greatest levels of survival within plots containing fencing, *S. alterniflora* transplants, and hand-dispersed propagules. Plots with only hand-dispersed propagules were expected to have moderate levels of survival and establishment. We also hypothesized that humic acid would have a positive effect on *A. germinans* survival and establishment due to the amelioration of environmental stress for both species. After one year, we hypothesized that plots with greater vegetative cover would have sediment characteristics that were more similar to adjacent salt marsh reference plots, indicating the potential for restoration of salt marsh function. The results of this research will advance the knowledge base for successful *A. germinans* restoration, particularly at its latitudinal range limit, which is currently expanding (Perry and Mendelssohn 2009) and will demonstrate the potential for humic acid as a salt marsh soil amendment.

Materials and Methods

We tested black mangrove establishment techniques on Whiskey Island, part of the Isles Dernieres barrier island chain along the southern Louisiana coast (latitude N29° 02', longitude W90° 49'). The marsh platform was constructed as part of the TE-50 restoration project and is approximately 300 acres, surrounded by existing salt marsh where reference plots were located. Independently from our research, approximately 50,000 plugs of *S. alterniflora* were transplanted by a commercial contractor along the existing berm as part of the restoration of this site. The majority of the aboveground tissue from the plugs died, but many rhizomes survived and produced stems the following spring (personal observation). Initially, 5,000 *A. germinans* seedlings were to be planted by the contractor at the site as well, however, poor winter growing conditions prevented this from occurring. Therefore, the project area was nearly devoid of live vegetation.

Plot elevation was guided by several sources of preliminary data: a SET benchmark provided by DNR was used to calibrate all sediment surface elevation measurements, water-level gauge data collected from April 2010 through October 2010 recorded elevations of live *A. germinans* and *S. alterniflora* close to the project area, and data collected on black mangroves on the Caminada-Moreau Headland helped to inform us about previous establishment elevations for *A. germinans* (Alleman and Hester 2011b).

In October 2010, we established five replicate blocks from west to east of three humic acid levels (0, 125 , and 250 ml m⁻², 4% humic acid derived from coal: Huma-Boost, 3 Tier Technologies, Longwood, FL). We used three propagule establishment techniques—no propagules dispersed, propagules hand-dispersed, and propagules hand-dispersed among *S. alterniflora*-enhanced fencing—parallel to and south of a tidal creek that was forming as a result of the construction design. Each fencing treatment area consisted of five biodegradable, coconut fiber fences in the shape of chevrons, with 1.2 m sides held down with wooden stakes and open at a 106° angle. We planted 10 plugs of *S. alterniflora* (approximately 50 stems) in a bowling

pin arrangement starting from the inner apex of the fence. In fall 2010, we hand-dispersed 250 *A. germinans* propagules into the propagules that were hand-dispersed among the *S. alterniflora*–enhanced fencing treatment areas (approximately 50 per sampling plot), for a total of 7,500 dispersed propagules. Because fences were staggered, with three fences parallel to the creek and two more fences slightly farther away from the creek, we also established a low-elevation plot and high-elevation plot for each combination of humic acid/vegetation type treatments. We had a total of 90 experimental plots.

We sampled all vegetation, *A. germinans* survival, and establishment in high and low elevation 4-m² sampling plots located within each treatment area. Specifically, plots were monitored for live and dead cover in fall 2010, spring 2011 and fall 2011. Transplanted smooth cordgrass stems were counted in fall 2010, late fall 2010, spring 2011, and fall 2011. Stem height was measured in fall 2010, spring 2011, and fall 2011. Propagules were added in fall 2010 and counted in late fall 2010, spring 2011, and fall 2011. In fall 2011, live propagules from the 2011 cohort were naturally present and counted. We also collected green leaf tissue from *S. alterniflora* in the plots with fences in fall 2011, which was dried to a constant weight, ground to pass through number 20 mesh and submitted to the Louisiana State University Soil Testing and Plant Analysis Laboratory for the determination of leaf total carbon and nitrogen.

In fall 2010, spring 2011, and fall 2011, we collected 5.0 cm diameter cores to a depth of 15 cm, which were then dried for the determination of soil bulk density and moisture. Dried cores were then homogenized and a subsample was used to determine soil organic matter (Parent and Caron 1993). Soil scoops were also collected to a depth of 15 cm for soil chemical characterization. Subsamples of the homogenized soil samples were subjected to two 1:2 (w:v) extraction procedures employing deionized water and 2M KCl, respectively. One aliquot of the deionized water extract was used for the determination of pH and conductivity (Rhoades 1990). The second aliquot of deionized water extract was submitted to the LSU Soil Testing and Plant Analysis Laboratory for the determination of total phosphorus, potassium, and other relevant cations through the use of ICP-OES (EPA method 200.7). The KCl extract was submitted to the Southeastern Louisiana University Microbial Testing Laboratory for determining ammonium and nitrate-nitrite using colorimetric methods (EPA method 350.1 and 353, respectively).

At five adjacent reference plots that had been previously restored in 2000, we sampled vegetation and soil as described above, and also measured soil redox potential at depths of 1 and 15 cm. We analyzed our split-plot experimental design using a general linear model in which block was considered a random effect and elevation was nested within block (Gotelli & Ellison, 2004). Tukey's HSD (Honestly Significant Difference) tests and *post hoc* contrasts were used when treatment levels were significant. All analyses were performed using JMP 9 (SAS Institute).



Figure 7.1 Site map for experimental *Avicennia germinans* restoration project in back-barrier marsh habitat of Whiskey Island. Black outlines represent experimental *A. germinans* establishment blocks, with each block containing all treatments. White triangles represent adjacent reference marsh habitat.

Results

Vegetation

In plots where *S. alterniflora* had been transplanted (those with fences), the average height of *S. alterniflora* stems was not significantly different among treatments in fall 2010, spring 2011, or fall 2011 (Figure 7.2). There were no significant differences in stem number in late fall 2010, spring 2011, or fall 2011 (Figure 7.3). The propagule establishment technique had a highly significant effect on total live cover in fall 2010 (Figure 7.4; $F = 3578.1$, $p < 0.01$), spring 2011 (Figure 7.5; $F = 45.7$, $p < 0.01$), and fall 2011 (Figure 7.6; $F = 27.2$, $p < 0.01$) in which plots with fences (and transplanted *S. alterniflora* stems) had greater live cover than other plots that were bare or had propagules added to them. Total dead cover (Figures 7.7–7.9) was not significantly different among treatments in fall 2010. In spring 2011, though, propagule establishment technique had a highly significant effect on dead cover in plots with fences, which had greater total dead cover (Figures 7.7–7.9; $F = 103.7$, $p < 0.01$). The same highly significant effect of propagule establishment technique was evident in fall 2011 (Figures 7.7–7.9; $F = 30.3$, $p < 0.01$).

Propagule establishment technique also had a highly significant effect on retaining propagules within plots. Fences retained more propagules (38) than propagules-added plots (18) or bare plots (0) (Figures 7.10–7.12, $F = 251.0$, $p < 0.01$). There was no effect of treatment on propagules in spring 2011, and all propagules found were dead. By the time monitoring occurred in fall 2011, a new cohort of propagules had naturally been dispersed into the plots. An interaction between humic acid and establishment technique had a marginally significant effect on the total number of propagules in plots in fall 2011 (Figures 7.10–7.12; $F = 2.4$, $p < 0.1$). More propagules were located within fence plots with the 250 ml m^{-2} humic acid application (2.7 ± 0.8 propagules) than in bare or fence plots with no humic acid or in bare plots with 250 ml m^{-2} humic acid (< 1 propagule). There was also a significant main effect of humic acid on total number of propagules found in plots; the greatest number of propagules was found in the 250 ml m^{-2} humic acid plots (Figures 7.10–7.12; $F = 4.0$, $p < 0.05$).

There was a highly significant interaction between elevation and propagule establishment technique on live propagules present. Low-elevation plots with fences had the greatest number of live propagules found in late fall 2010 (12), followed by high elevation plots with fences (6) (Figures 7.13–7.15, $F = 84.8$, $p < 0.01$). Bare and propagule-added plots had only 0–1 live propagules by late fall 2010. By fall 2011, propagule establishment technique had a marginally significant effect on the number of live propagules present, wherein the greatest number of live propagules tended to be found in propagule-added plots (Figures 7.13–15; $F = 2.9$, $p < 0.1$). Humic acid also had a significant effect on live propagules and more were found in plots with the 250 ml m^{-2} humic acid treatment (Figures 7.13–15; $F = 4.1$, $p < 0.05$).

Reference plots had approximately 50% total cover and a canopy height greater than 50 cm (Table A7.1). Sediments were moderately reduced, as expected for salt marsh (Table A7.1).

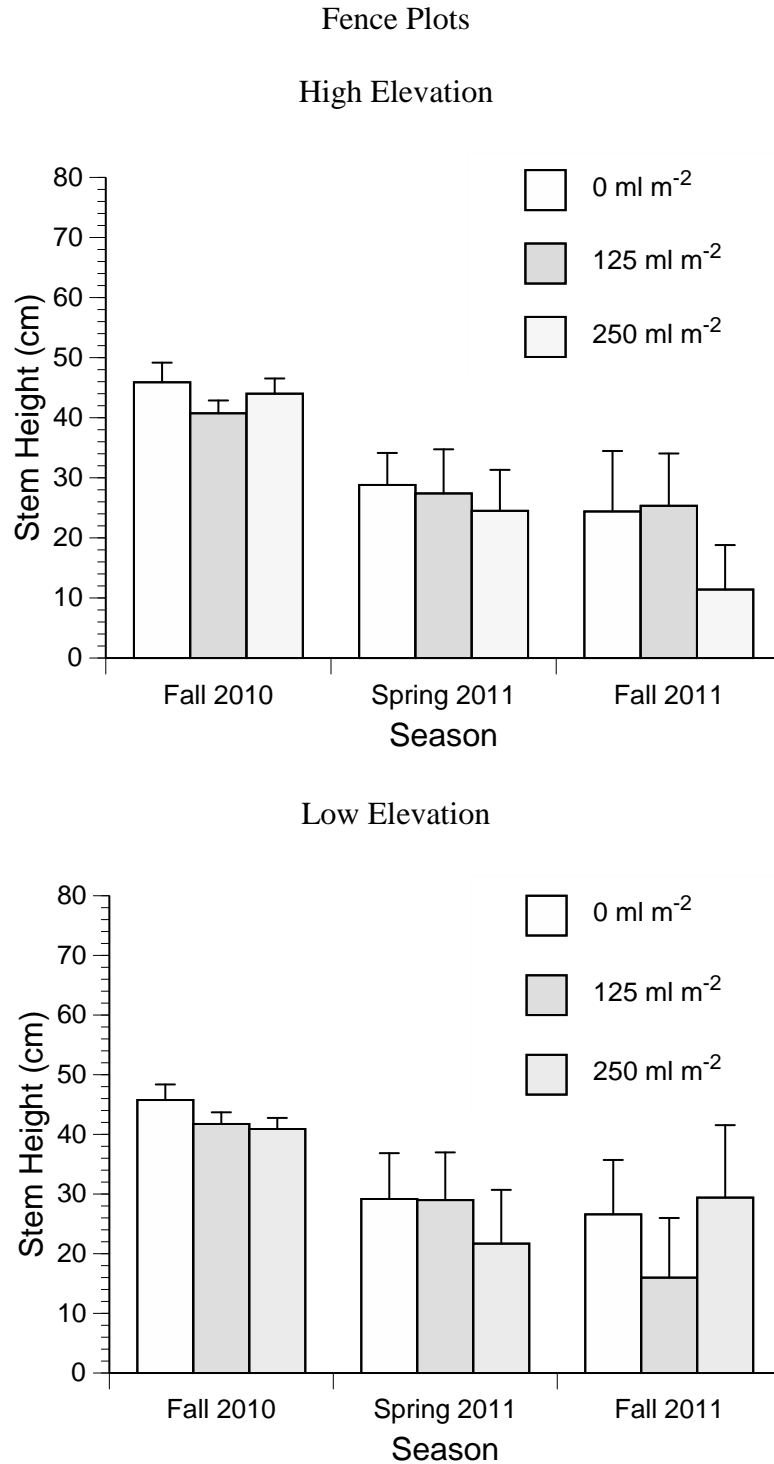


Figure 7.2 The effect of humic acid amendment on the average height of live transplanted *Spartina alterniflora* stems seasonally for fence plots at high (top panel: means \pm SE, $n = 5$, fall 2010 LSD = 7.1, spring 2011 LSD = 21.7, fall 2011 LSD = 28.3) and low elevations (bottom panel: means \pm SE, $n = 5$, fall 2010 LSD = 7.1, spring 2011 LSD = 21.7, fall 2011 LSD = 28.3).

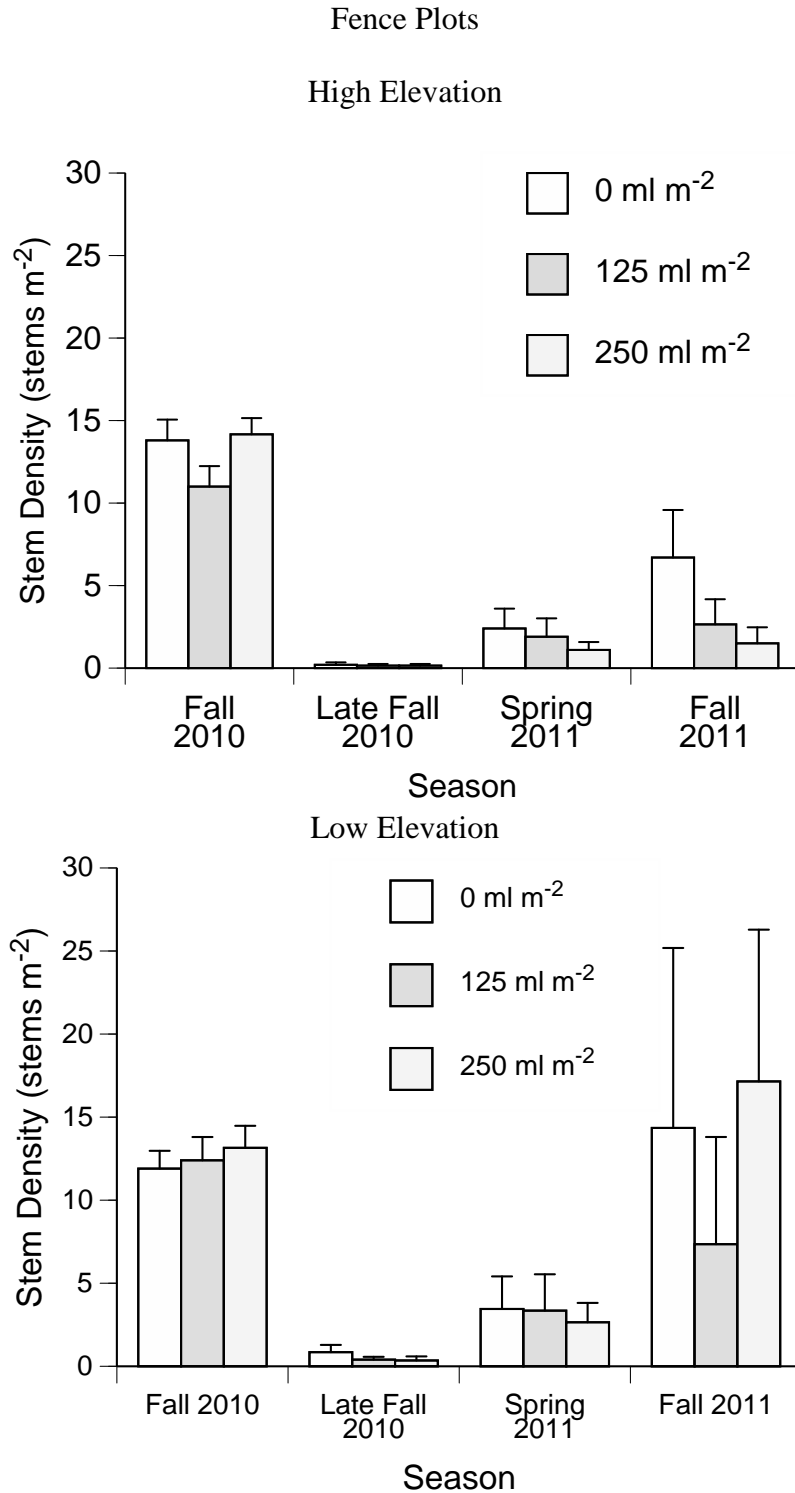


Figure 7.3 The effect of humic acid amendment on stem density of live transplanted *Spartina alterniflora* stems seasonally for fence plots at high (top panel: means \pm SE, $n = 5$, fall 2010 LSD = 5.4, late fall 2010 LSD = 0.07, spring 2011 LSD = 4.3, fall 2011 LSD = 28.3) and low elevations (bottom panel: means \pm SE, $n = 5$, fall 2010 LSD = 5.4, late fall 2010 LSD = 0.07, spring 2011 LSD = 4.3, fall 2011 LSD = 28.3).

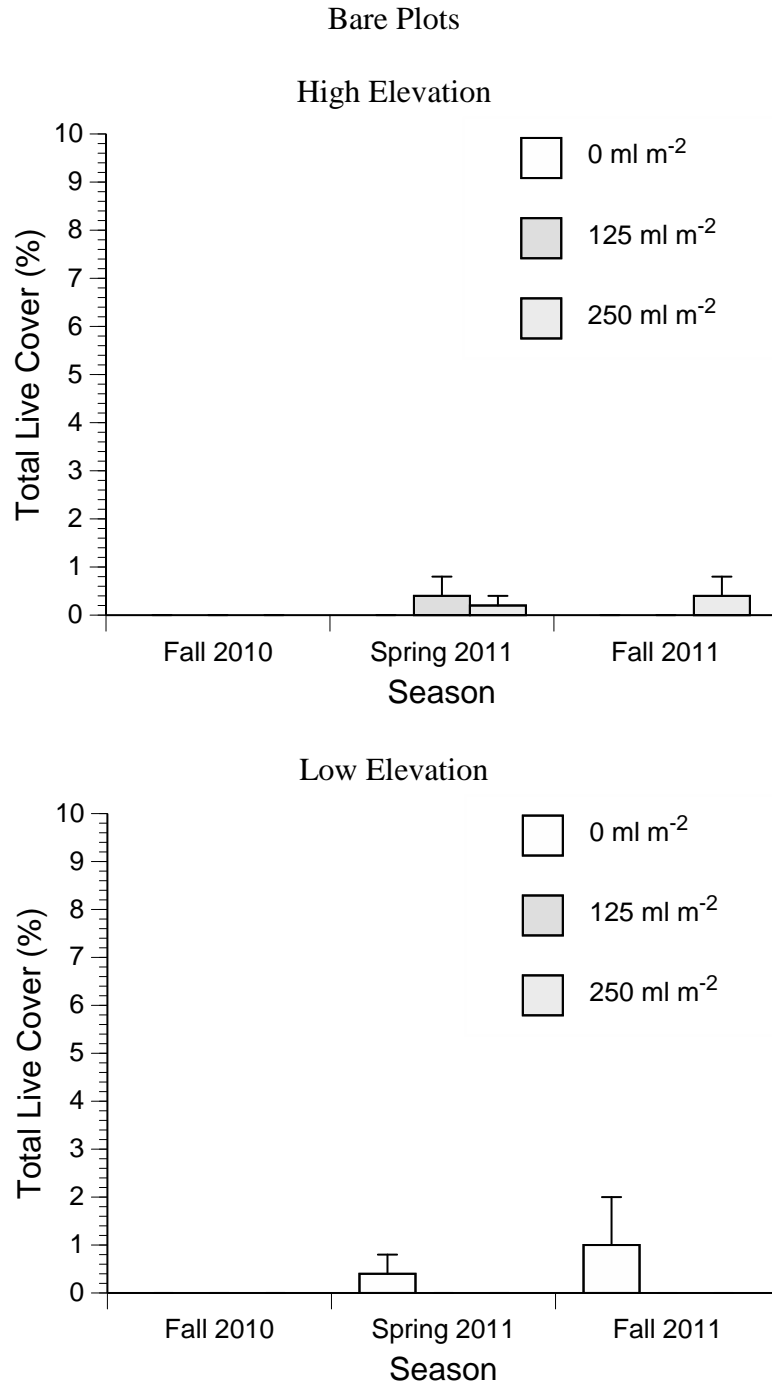


Figure 7.4 The effect of humic acid amendment on total live cover seasonally for bare plots at high (top panel: means \pm SE, $n = 5$, fall 2010 LSD = 0.3, spring 2011 LSD = 1.1, fall 2011 LSD = 2.3) and low elevations (bottom panel: means \pm SE, $n = 5$, fall 2010 LSD = 0.3, spring 2011 LSD = 1.1, fall 2011 LSD = 2.3).

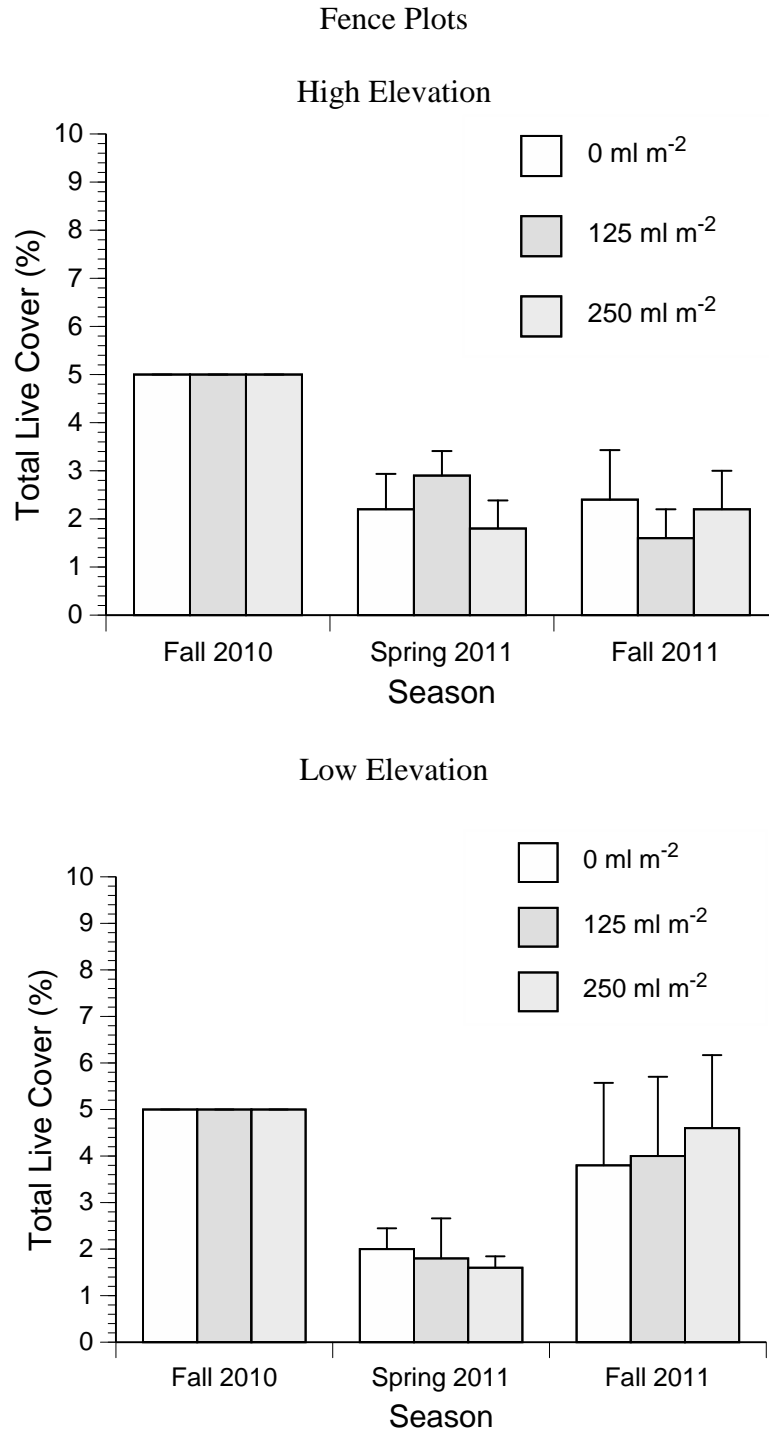


Figure 7.5 The effect of humic acid amendment on total live cover seasonally for fence plots at high (top panel: means \pm SE, $n = 5$, fall 2010 LSD = 0.3, spring 2011 LSD = 1.1, fall 2011 LSD = 2.3) and low elevations (bottom panel; means \pm SE, $n = 5$, fall 2010 LSD = 0.3, spring 2011 LSD = 1.1, fall 2011 LSD = 2.3).

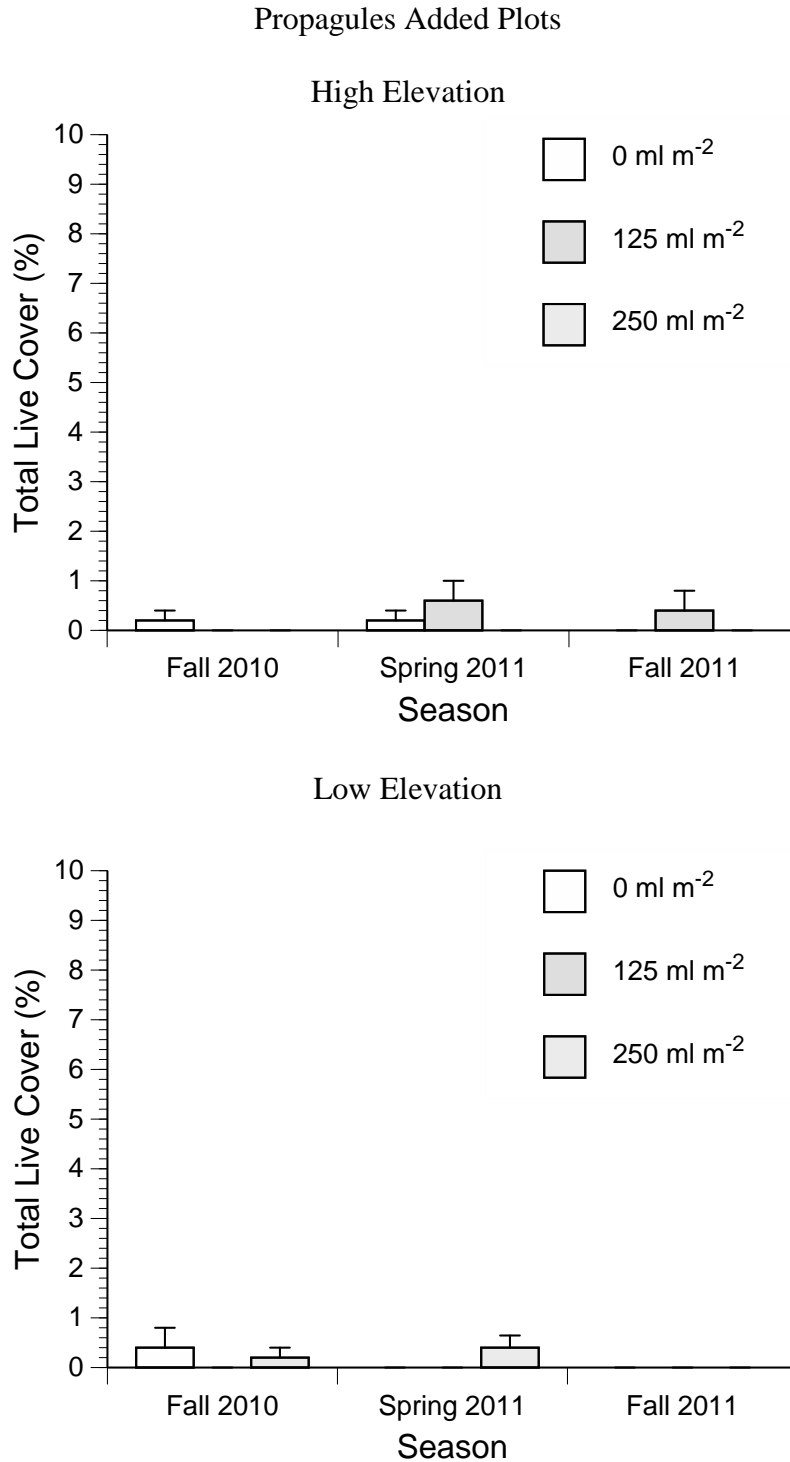


Figure 7.6 The effect of humic acid amendment on total live cover seasonally for propagules added plots at high (top panel: means \pm SE, $n = 5$, fall 2010 LSD = 0.3, spring 2011 LSD = 1.1, fall 2011 LSD = 2.3) and low elevations (bottom panel: means \pm SE, $n = 5$, fall 2010 LSD = 0.3, spring 2011 LSD = 1.1, fall 2011 LSD = 2.3).

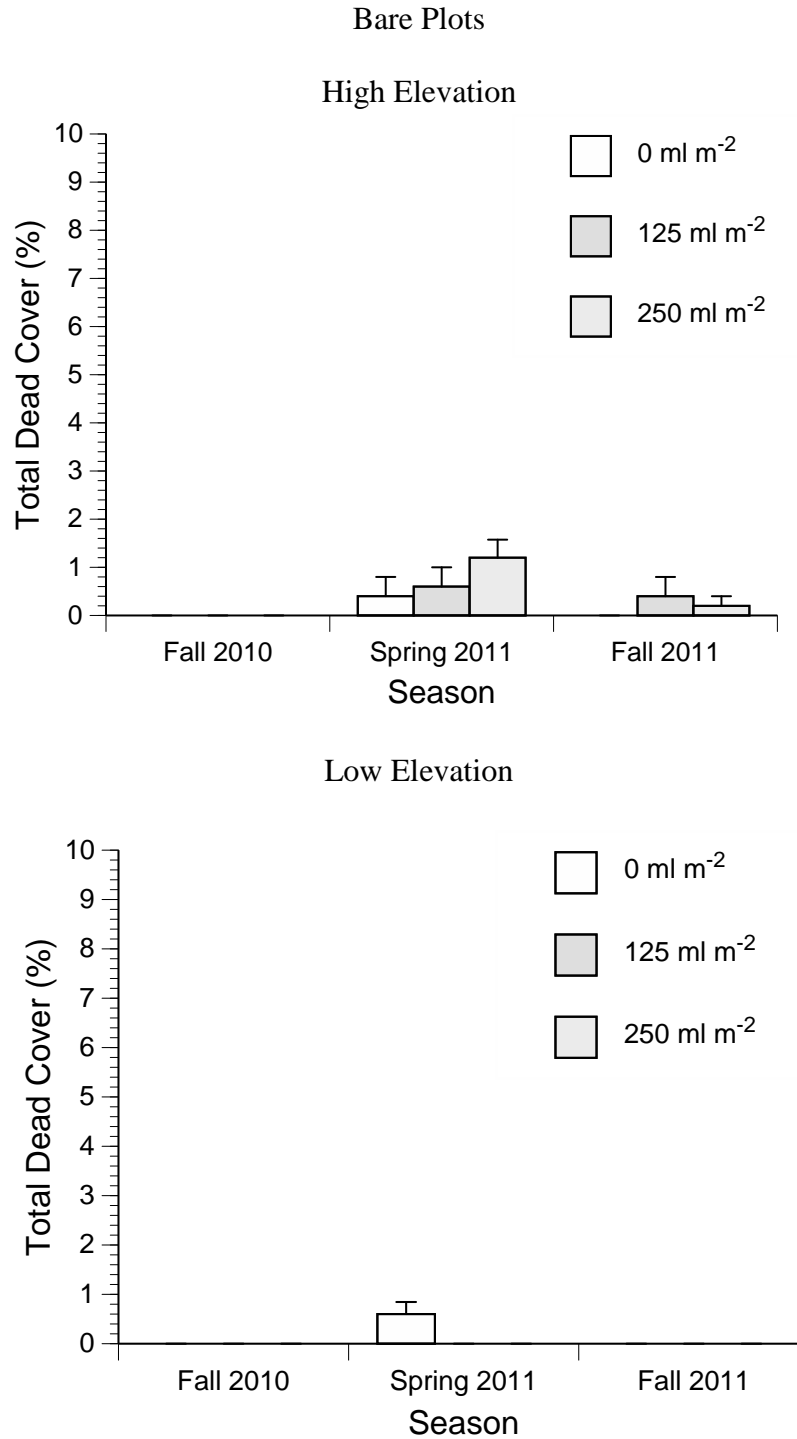


Figure 7.7 The effect of humic acid amendment on total dead cover seasonally for bare plots at high (top panel: means \pm SE, $n = 5$, fall 2010 LSD = 0.3, spring 2011 LSD = 1.4, fall 2011 LSD = 1.5) and low elevations (bottom panel: means \pm SE, $n = 5$, fall 2010 LSD = 0.3, spring 2011 LSD = 1.4, fall 2011 LSD = 1.5).

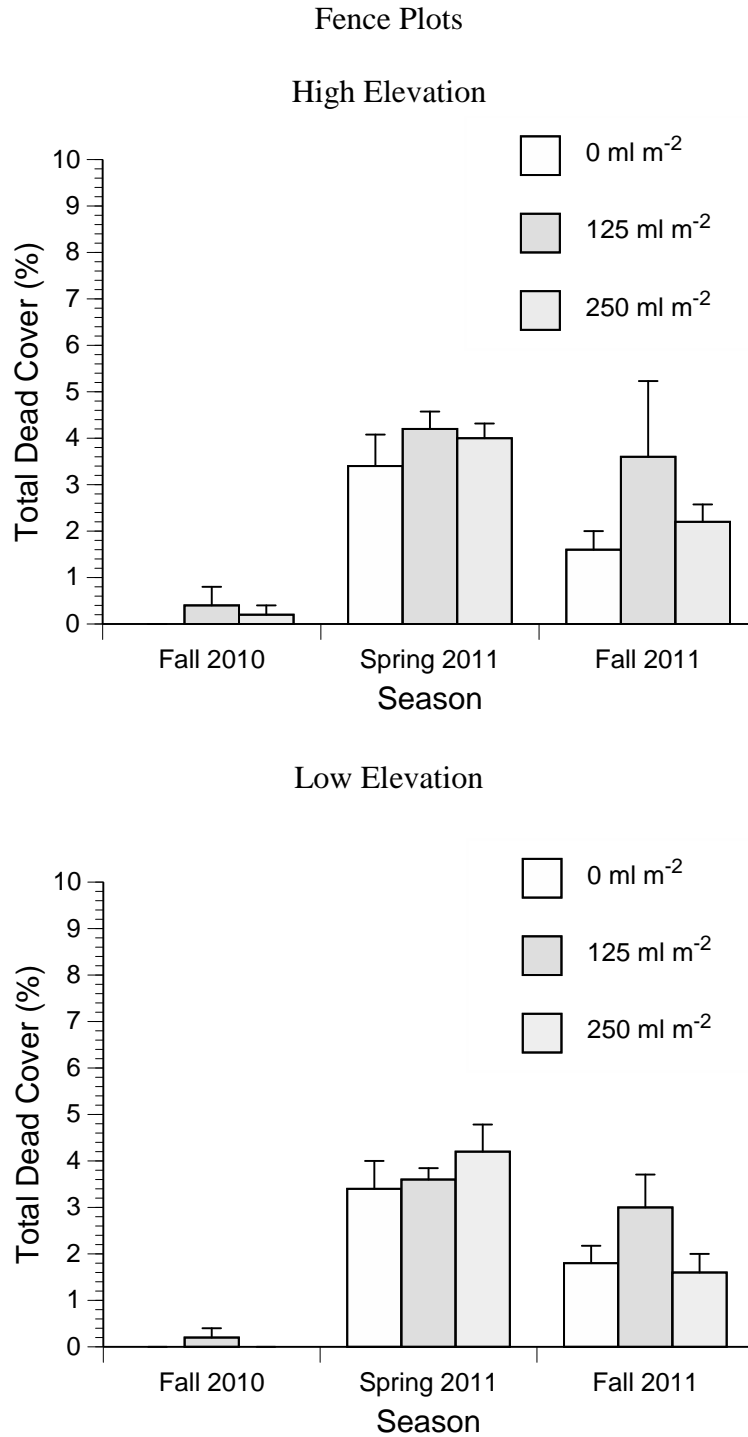


Figure 7.8 The effect of humic acid amendment on total dead cover seasonally for fence plots at high (top panel: means \pm SE, $n = 5$, fall 2010 LSD = 0.3, spring 2011 LSD = 1.4, fall 2011 LSD = 1.5) and low elevations (bottom panel: means \pm SE, $n = 5$, fall 2010 LSD = 0.3, spring 2011 LSD = 1.4, fall 2011 LSD = 1.5).

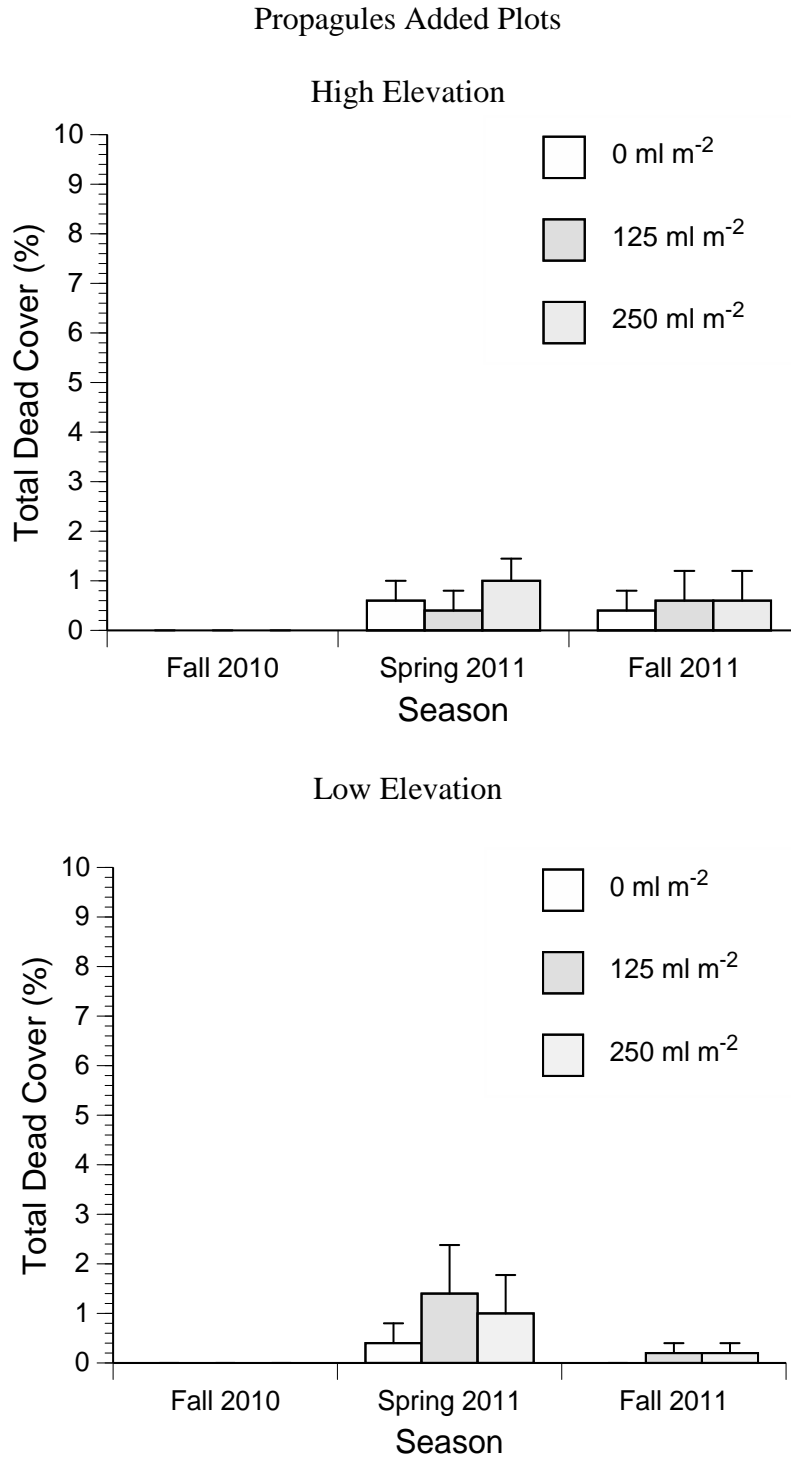


Figure 7.9 The effect of humic acid amendment on total dead cover seasonally for propagules added plots at high (top panel: means \pm SE, $n = 5$, fall 2010 LSD = 0.3, spring 2011 LSD = 1.4, fall 2011 LSD = 1.5) and low elevations (bottom panel: means \pm SE, $n = 5$, fall 2010 LSD = 0.3, spring 2011 LSD = 1.4, fall 2011 LSD = 1.5).

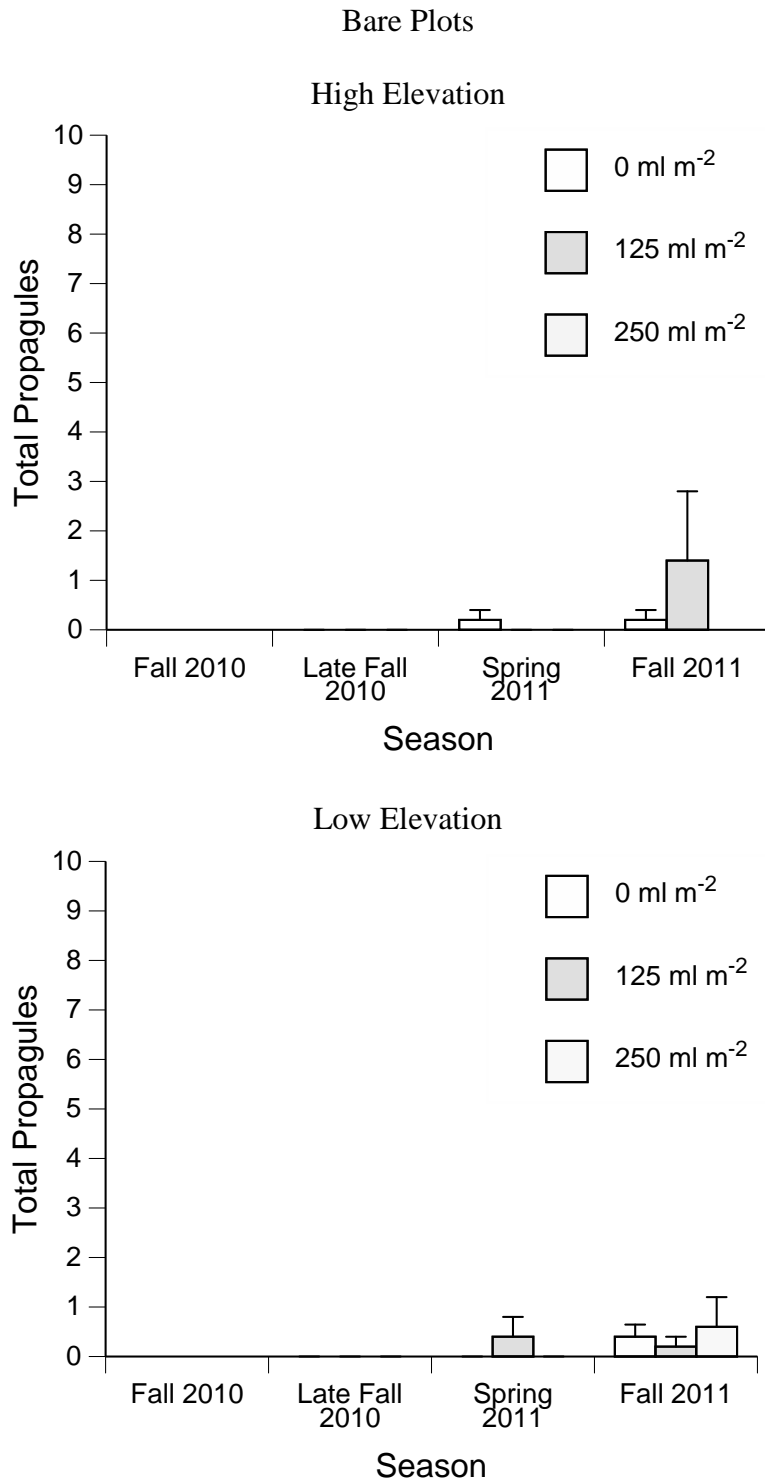


Figure 7.10 The effect of humic acid amendment on total propagules seasonally for bare plots at high (top panel: means \pm SE, $n = 5$, late fall 2010 LSD = 9.4, spring 2011 LSD = 2.1, fall 2011 LSD = 2.0) and low elevations (bottom panel: means \pm SE, $n = 5$, late fall 2010 LSD = 9.4, spring 2011 LSD = 2.1, fall 2011 LSD = 2.0).

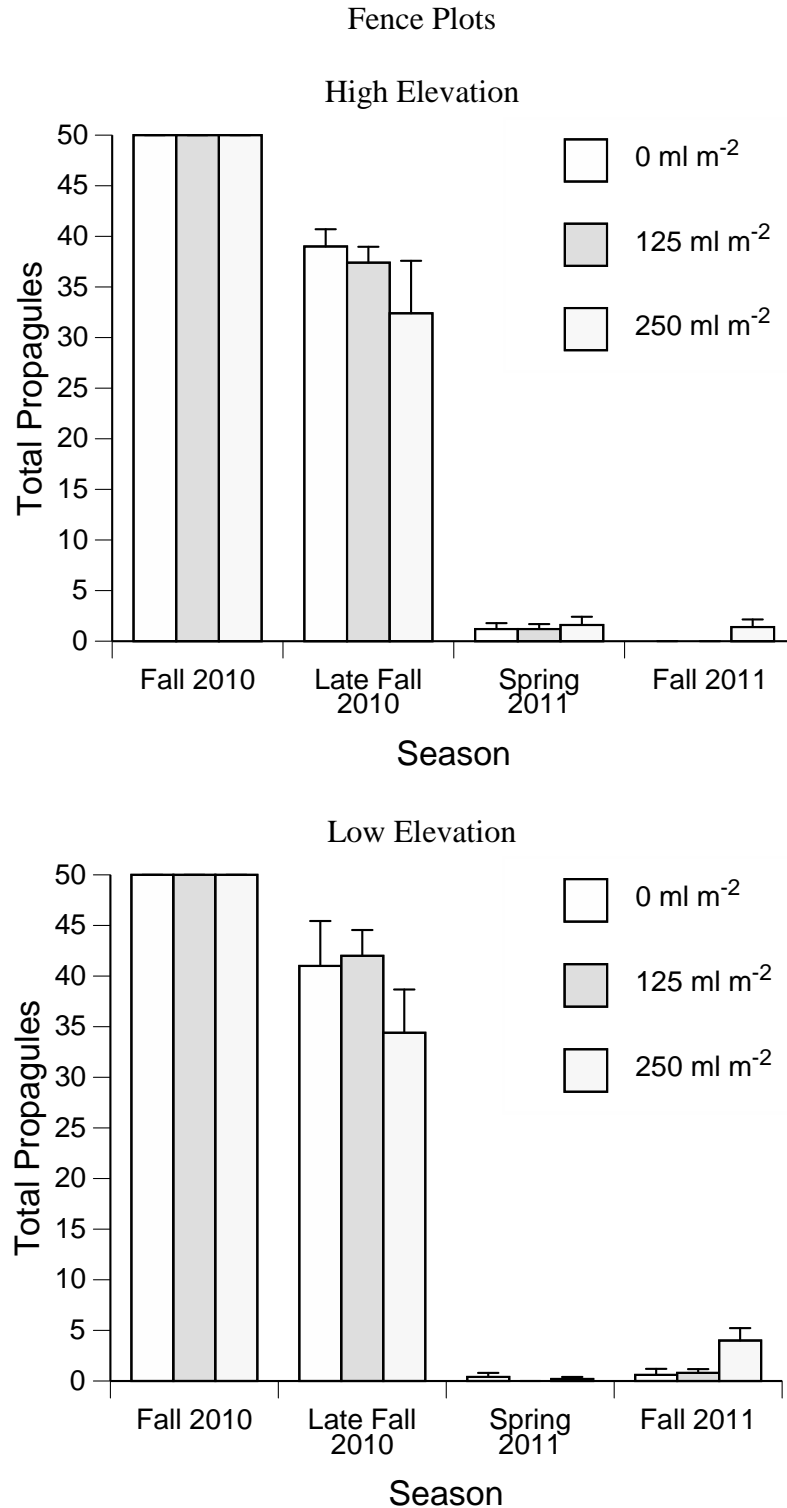


Figure 7.11 The effect of humic acid amendment on total propagules seasonally for fence plots at high (top panel: means \pm SE, $n = 5$, late fall 2010 LSD = 9.4, spring 2011 LSD = 2.1, fall 2011 LSD = 2.0) and low elevations (bottom panel; means \pm SE, $n = 5$, late fall 2010 LSD = 9.4, spring 2011 LSD = 2.1, fall 2011 LSD = 2.0).

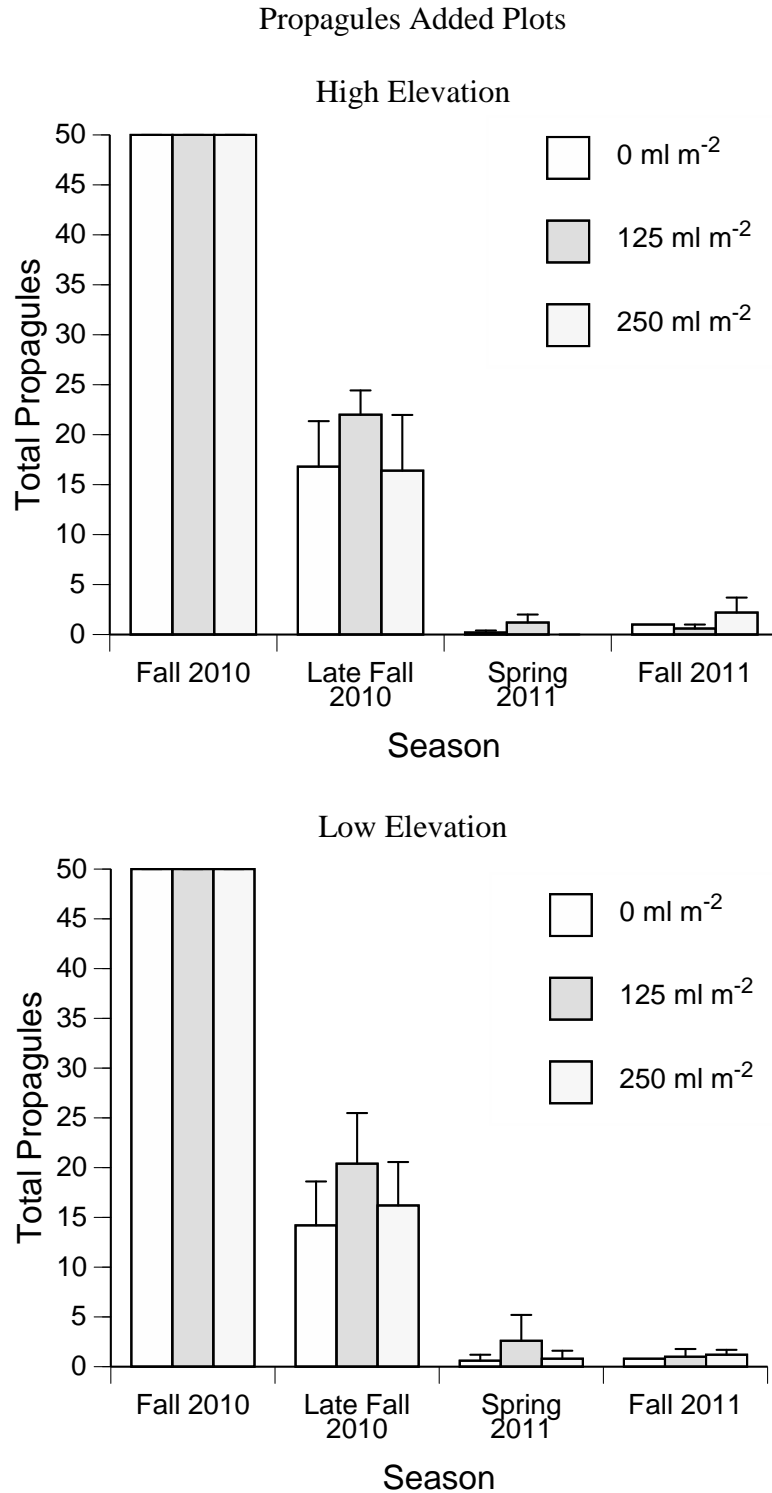


Figure 7.12 The effect of humic acid amendment on total propagules seasonally for propagules added plots at high (top panel: means \pm SE, $n = 5$, late fall 2010 LSD = 9.4, spring 2011 LSD = 2.1, fall 2011 LSD = 2.0) and low elevations (bottom panel: means \pm SE, $n = 5$, late fall 2010 LSD = 9.4, spring 2011 LSD = 2.1, fall 2011 LSD = 2.0).

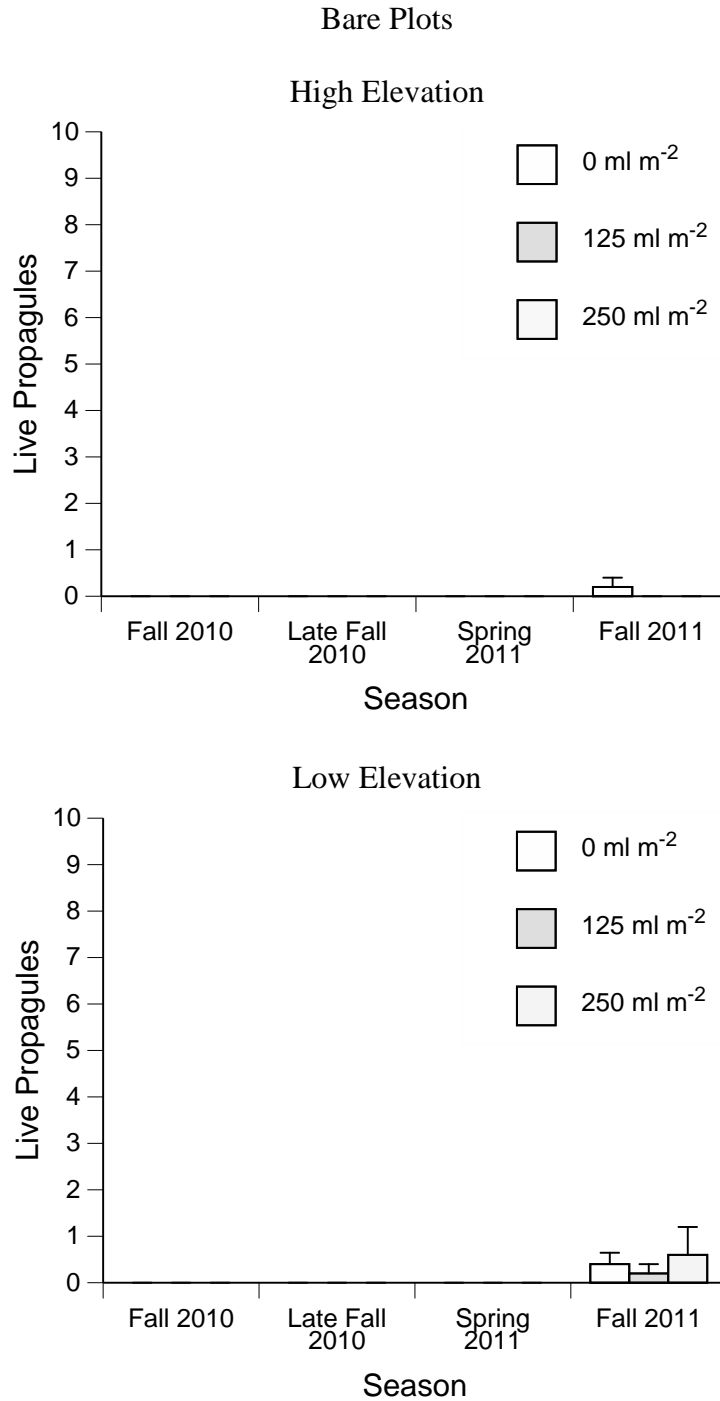


Figure 7.13 The effect of humic acid amendment on live propagules seasonally for bare plots at high (top panel: means \pm SE, $n = 5$, late fall 2010 LSD = 4.1, spring 2011 LSD = 0, fall 2011 LSD = 1.8) and low elevations (bottom panel: means \pm SE, $n = 5$, late fall 2010 LSD = 4.1, spring 2011 LSD = 0, fall 2011 LSD = 1.8).

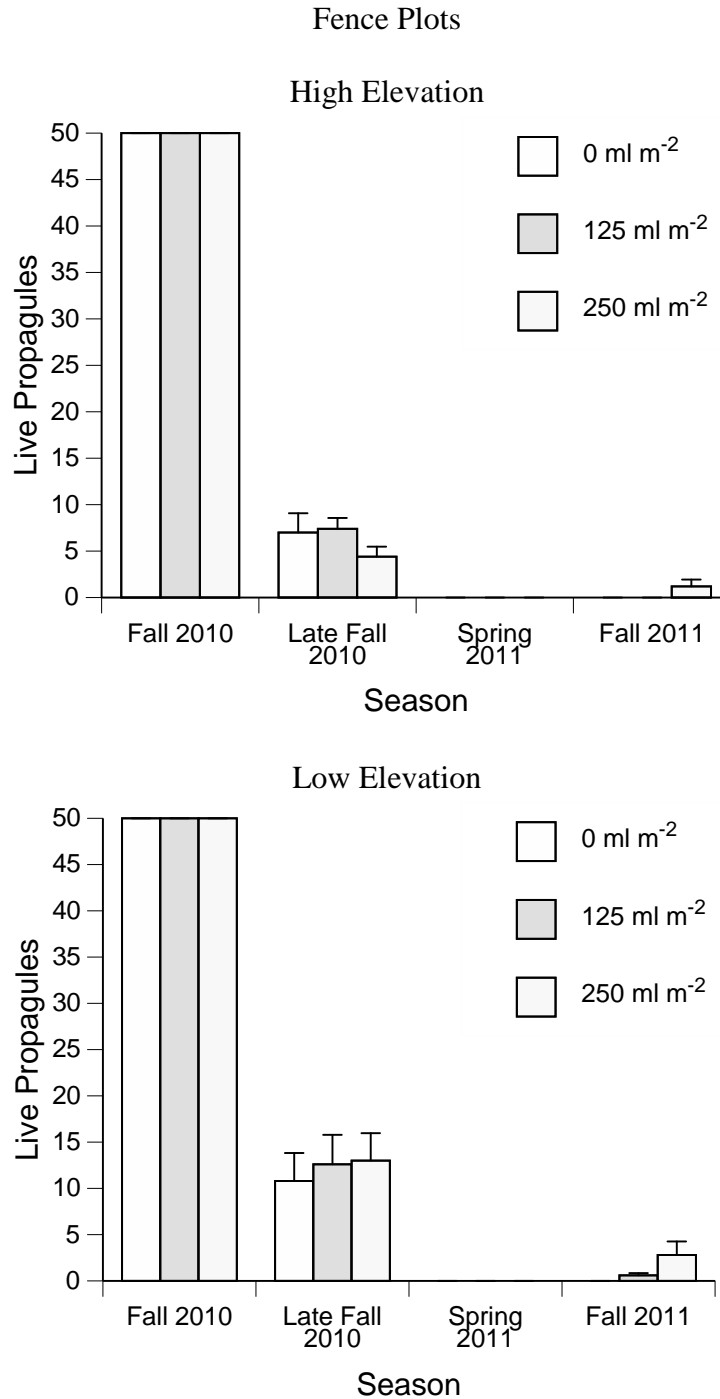


Figure 7.14 The effect of humic acid amendment on live propagules seasonally for fence plots at high (top panel: means \pm SE, $n = 5$, late fall 2010 LSD = 4.1, spring 2011 LSD = 0, fall 2011 LSD = 1.8) and low elevations (bottom panel: means \pm SE, $n = 5$, late fall 2010 LSD = 4.1, spring 2011 LSD = 0, fall 2011 LSD = 1.8).

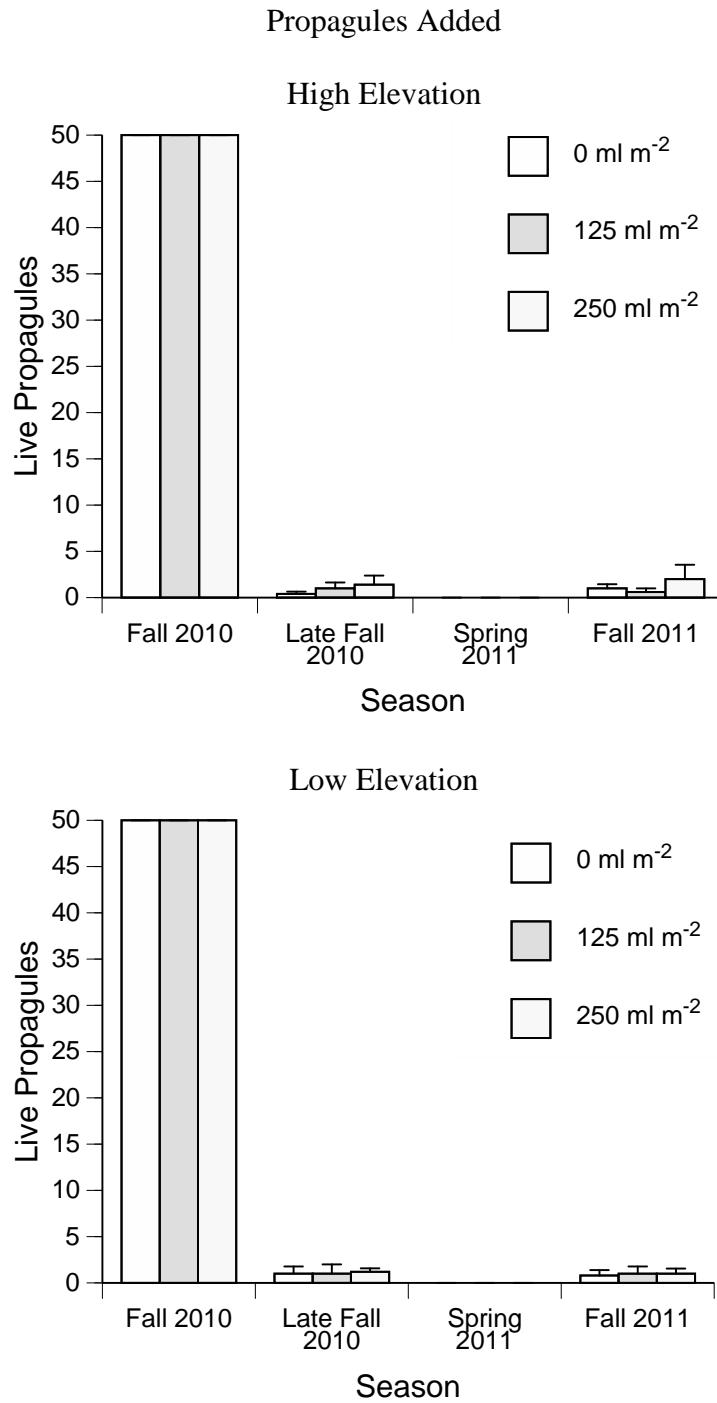


Figure 7.15 The effect of humic acid amendment on live propagules seasonally for propagules added plots at high (top panel: means \pm SE, $n = 5$, late fall 2010 LSD = 4.1, spring 2011 LSD = 0, fall 2011 LSD = 1.8) and low elevations (bottom panel: means \pm SE, $n = 5$, late fall 2010 LSD = 4.1, spring 2011 LSD = 0, fall 2011 LSD = 1.8).

The C:N ratio of *S. alterniflora* collected in fall 2011 was not significantly different among treatments and was on average, 24.7 ± 1.0 (Figure 7.16).

One established *A. germinans* seedling was found in fall 2011 within a fence, adjacent to a plot that had 125 ml m^{-2} humic acid applied to it, located in block 2, and an elevation of 0.646 m NAVD 88. The seedling was 20 cm tall and had 14 leaves.

Soil

Plot elevations were initially 0.718–0.885 m NAVD88 in fall 2010; they decreased to 0.604–0.719 m NAVD88 in fall 2011. Elevation of plots (Figures 7.17–7.19) in fall 2010 was significantly greater in high elevation plots, as planned (Figures 7.17–7.19; *post hoc* contrast, $F = 20.8$, $p < 0.01$). By spring 2011, humic acid and vegetation type had significant interaction where propagule plots without humic acid were at the lowest elevation (Figures 7.16–7.18; $F = 3.2$, $p < 0.05$). The highly significant effect of elevation was the same in spring 2011 as it was in fall 2010 (Figures 7.17–7.19; $F = 24.0$, $p < 0.01$) and in fall 2011 (Figures 7.17–7.19; $F = 23.2$, $p < 0.01$).

Soil pH in fall 2010 was not significantly different among humic acid, vegetation type, or elevation treatments (Table A7.2). A marginally significant effect of humic acid on soil pH was evident in spring 2011 (Table A7.3). The 125 ml m^{-2} treatment had a higher pH than the 0 ml m^{-2} or 250 ml m^{-2} humic acid treatments (Table A7.3; $F = 4.0$, $p < 0.1$). In fall 2011, humic acid had a significant effect on pH in which pH was lower in the 250 ml m^{-2} plots than in plots without humic acid (Table A7.4, Tukey's HSD, $F = 4.5$, $p < 0.05$). There were no significant effects of treatments on conductivity and salinity in fall 2010, spring 2011, or fall 2011 (Tables A7.2–A7.4).

There were no significant effects of treatments on soil moisture in fall 2010 (Table A7.2). By spring 2011, there were significant interactions between vegetation type and humic acid (Table A7.3; $F = 3.4$, $p < 0.05$) and between vegetation type and elevation (Table A7.3; $F = 3.5$, $p < 0.05$) on soil moisture. Compared to other plots, the plots with no humic acid and propagules added had high moisture, as did low-elevation plots with fences. Humic acid (Table A7.3; $F = 3.5$, $p < 0.05$) and elevation (Table A7.3; $F = 3.3$, $p < 0.05$) also had significant effects on soil moisture where moisture was higher in low elevation plots and plots without humic acid, compared to others. By fall 2011, there were no significant differences among treatments (Table A7.4). There were no significant effects of treatment on bulk density in fall 2010, spring 2011, or fall 2011 (Tables A7.2–A7.4). There were no significant differences of organic matter among treatments in fall 2010 (Table A7.2). In spring 2011, humic acid had a significant effect on organic matter, which was greatest in the plots without humic acid (Table A7.3; $F = 4.0$, $p < 0.05$). These effects were marginally significant in fall 2011, when organic matter was greater in low elevation plots (Table A7.4; $F = 2.0$, $p < 0.1$, Table A7.4).

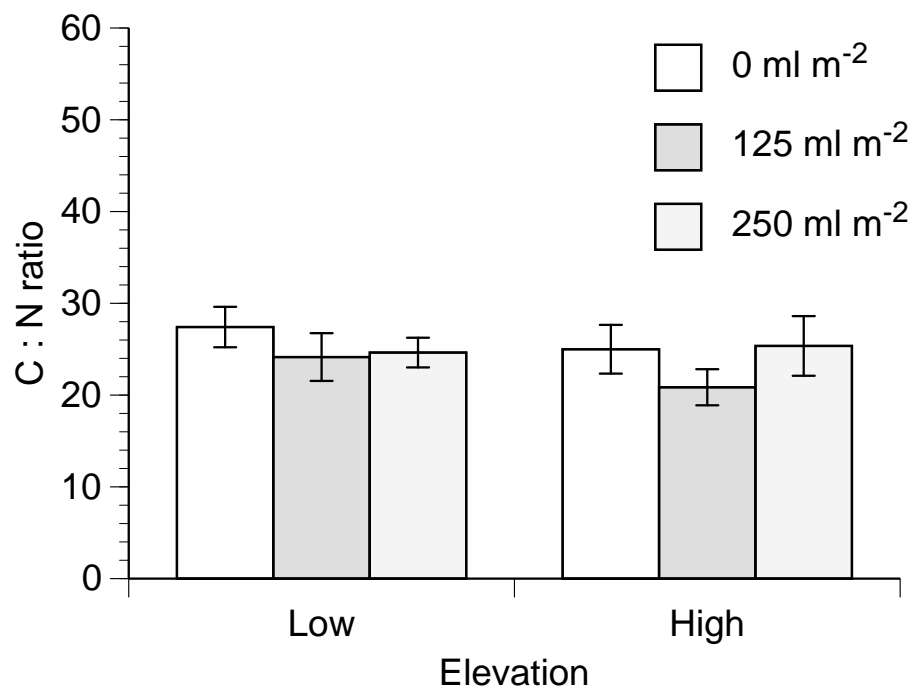


Figure 7.16 The effect of humic acid amendment on C:N ratio of *Spartina alterniflora* leaf tissue collected in fall 2011 (means \pm SE, $n = 5$, LSD = 7.5).

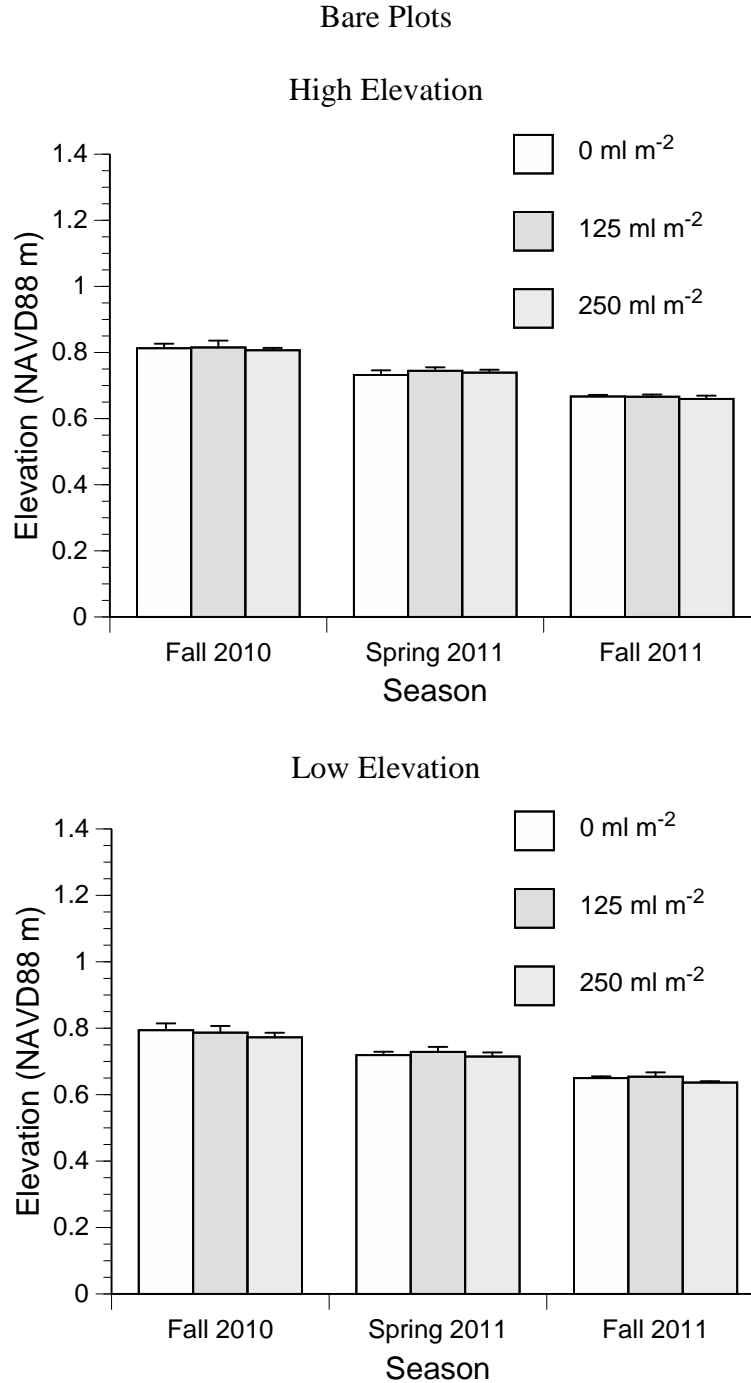


Figure 7.17 The effect of humic acid amendment on elevation seasonally for bare plots at high (top panel: means \pm SE, $n = 5$, fall 2010 LSD = 0.05, spring 2011 LSD = 0.03, fall 2011 LSD = 0.03) and low elevations (bottom panel: means \pm SE, $n = 5$, fall 2010 LSD = 0.05, spring 2011 LSD = 0.03, fall 2011 LSD = 0.03).

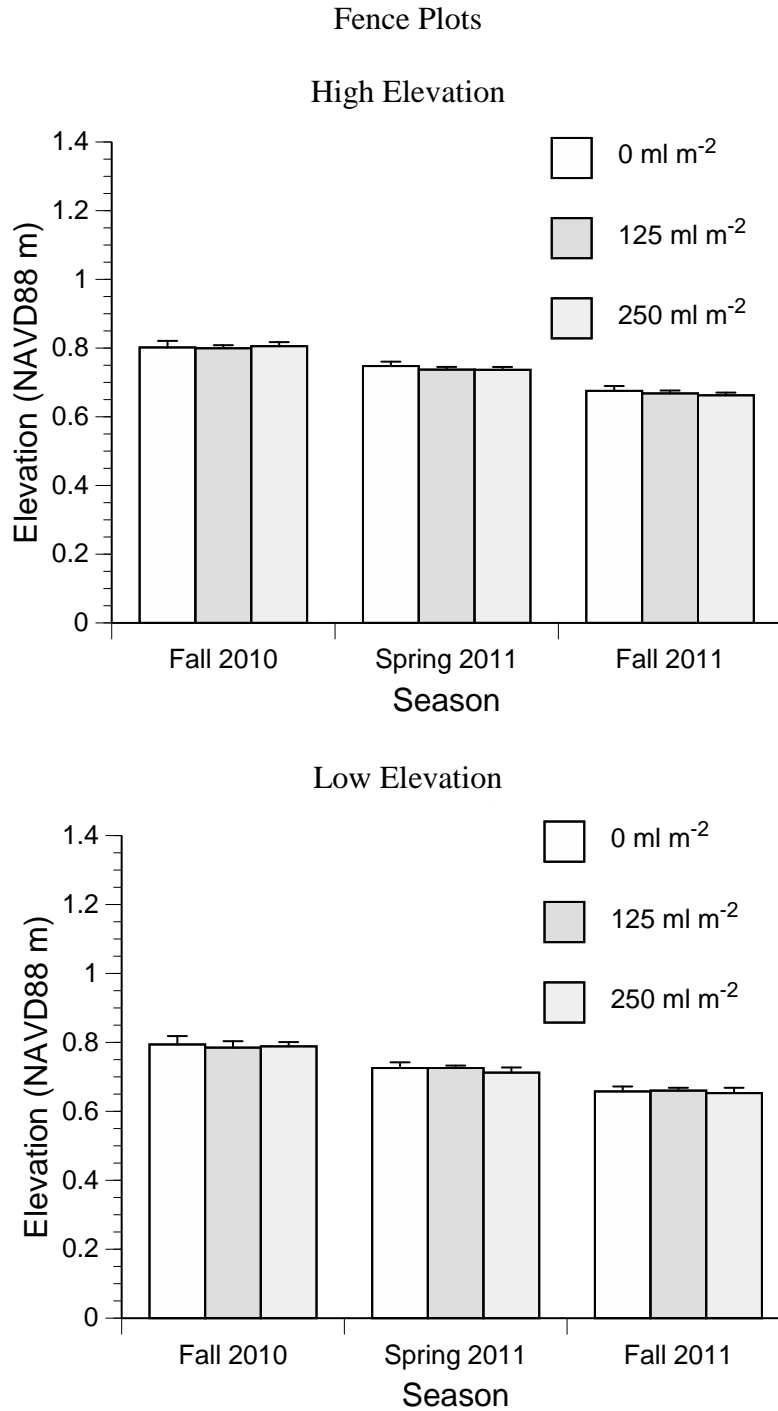


Figure 7.18 The effect of humic acid amendment on elevation seasonally for fence plots at high (top panel: means \pm SE, $n = 5$, fall 2010 LSD = 0.05, spring 2011 LSD = 0.03, fall 2011 LSD = 0.03) and low elevations (bottom panel; means \pm SE, $n = 5$, fall 2010 LSD = 0.05, spring 2011 LSD = 0.03, fall 2011 LSD = 0.03).

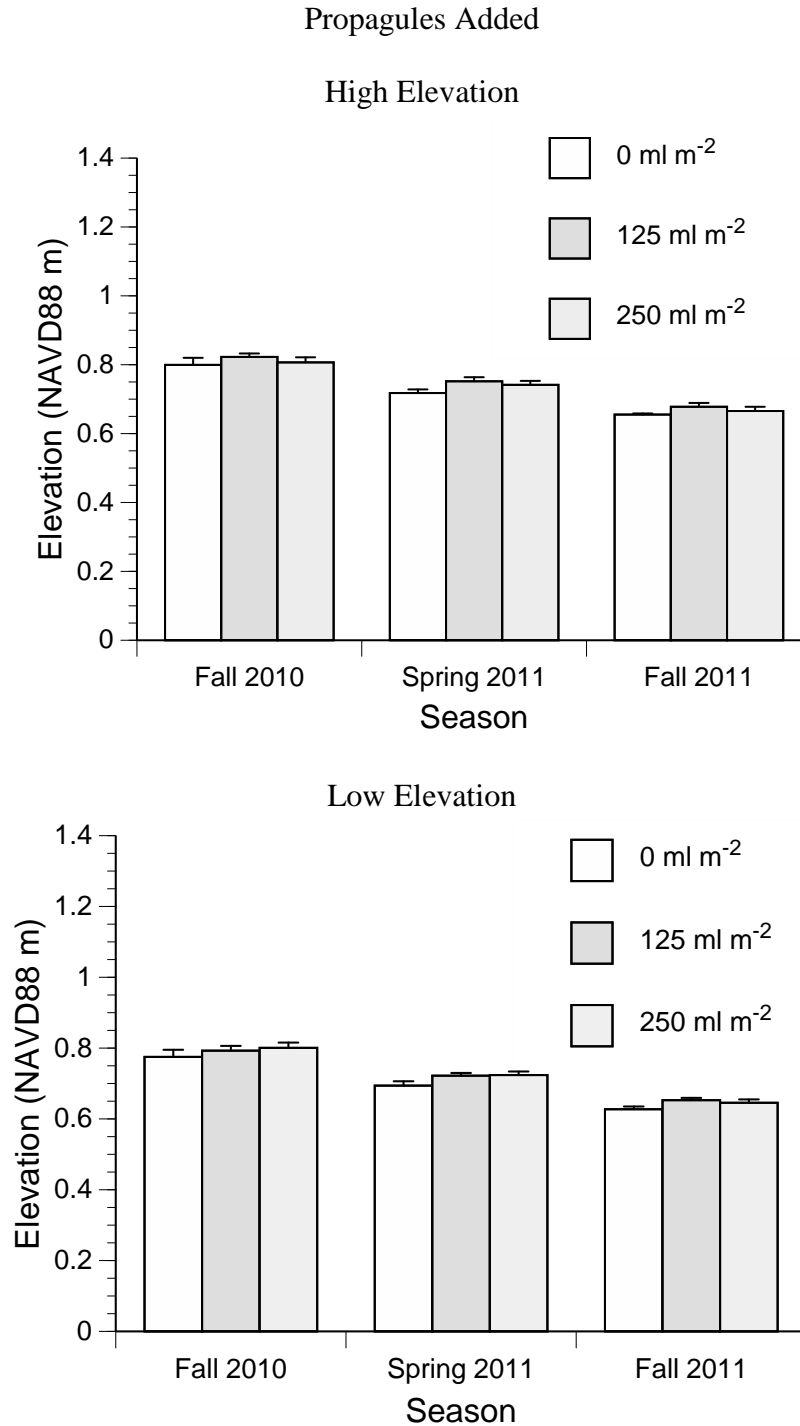


Figure 7.19 The effect of humic acid amendment on elevation seasonally for propagules added plots at high (top panel: means \pm SE, $n = 5$, fall 2010 LSD = 0.05, spring 2011 LSD = 0.03, fall 2011 LSD = 0.03) and low elevations (bottom panel: means \pm SE, $n = 5$, fall 2010 LSD = 0.05, spring 2011 LSD = 0.03, fall 2011 LSD = 0.03).

Ammonium values were not initially different among treatments in fall 2010 or spring 2011 (Tables A7.5–A7.6). However, by fall 2011, there was a highly significant effect of humic acid on ammonium in which values were greater in the 250 ml m⁻² humic acid treatment ($1.547 \pm 0.197 \mu\text{g g}^{-1}$) than in the 125 ml m⁻² humic acid treatment ($1.002 \pm 0.180 \mu\text{g g}^{-1}$), with the control treatment in-between ($1.248 \pm 0.181 \mu\text{g g}^{-1}$) (Table A7.7; Tukey's HSD, $F = 5.5$, $p < 0.01$). Nitrate-nitrite values were not significantly different among treatments or block in fall 2010 or fall 2011 (Tables A7.5 and A7.7). There was a significant effect of propagule establishment technique observed in spring 2011 in which nitrate-nitrite values were greater in the bare plots than in the propagules-added plots (Table A7.6; Tukey's HSD, $F = 3.2$, $p < 0.05$). Mean nitrate-nitrite values were $0.190 \pm 0.018 \mu\text{g g}^{-1}$ for fall 2010, $0.044 \pm 0.004 \mu\text{g g}^{-1}$, and $0.173 \pm 0.014 \mu\text{g g}^{-1}$ for fall 2011. There were no significant differences in phosphorus among treatments in fall 2010, spring 2011, or fall 2011 (Tables A7.5–A7.7). Means of phosphorus were $0.321 \pm 0.007 \mu\text{g g}^{-1}$, $0.344 \pm 0.003 \mu\text{g g}^{-1}$, and $0.079 \pm 0.004 \mu\text{g g}^{-1}$ in fall 2010, spring 2011, and fall 2011, respectively. Potassium values were not significantly different in fall 2010, spring 2011, or fall 2011; and means of potassium were $138.7 \pm 4.1 \mu\text{g g}^{-1}$, $159.6 \pm 2.9 \mu\text{g g}^{-1}$, $93.9 \pm 2.3 \mu\text{g g}^{-1}$, respectively (Tables A7.5–A7.7).

Hydrology

Between October 2010 and November 2010, all plots were flooded only once, and the average water level during this period dropped by approximately 8.5 cm compared to the average water level from April 2010 to October 2010 (Table A7.8). Between November 2010 and April 2011, marsh platform sediments had compacted, resulting in a decrease in plot elevations by approximately 10.8 cm. Following this initial drop in elevation, plot elevation decreased by another 3.1–3.4 cm by October 2011 (Table A7.8). During the first week of September 2011, Tropical Storm Lee passed to the west of the Isles Dernieres and increased the water level by approximately 80.3 cm above the April 2011 to October 2011 average. The tidal creek forming parallel to the north of the study appeared to be the major conduit for incoming and outgoing tidal water and possibly propagules from the 2011 cohort. The water flowing into the creek went from west to east, and it would have reached block 1 (starting with plot 1) first and block 5 last (ending with plot 90).

Cost Estimate of Restoration Techniques

Cost estimates were based on labor (@ \$10 per hour) and materials bought to implement a restoration technique on a 4-m² area. Costs do not include access to donor site or transportation to the restoration site. The cost of labor and materials needed for collection, preparation, and dispersal of 3,750 *A. germinans* propagules was approximately \$235. The cost per 4-m² plot for the propagules added technique was \$3.13. The cost of labor and materials needed for collection, preparation, and dispersal of 3,750 *A. germinans* propagules, 75 fences, and 750 *S. alterniflora* transplants was approximately \$4,635. The cost per 4-m² plot for the fences enhanced with vegetation and propagules-added technique was \$61.80. The fences and transplanted *S. alterniflora* has lasted at least two years; therefore, the cost per area could be greatly reduced over time.

Discussion

Benefit of Structure for Restoration

The components of the restoration design that added structure to the bare salt marsh platform showed some benefit for meeting restoration objectives. First, the biodegradable fences physically retained more propagules during the first month of the study (75% retained within plots with fences vs. 35% retained within plots without fences). Other studies have documented propagule trapping extensively by both biotic (Lewis and Dunstan 1976; Stevens et al. 2006; McKee et al. 2007) and abiotic structures (Proisy et al. 2009). Structure in the environment in the form of crab mounds (Minchinton 2001), pneumatophores (McKee 1995), and the herbaceous (McKee et al. 2007) or mangrove (Bosire et al. 2003) canopy can also improve mangrove growth by alleviating soil stress (e.g., reduced conditions, soil temperature, and moisture). In our study, fences did not produce differences in soil moisture or salinity, but they still produced a positive effect on propagule survival (18% survival with fences vs. 2% survival without fences for plots with propagules added) for the first month of the study. A year after the study was initiated, fences did not prevent propagules from stranding. In fact, the only mangrove to establish during the study did so within a fenced plot. Therefore, adding structure to the restoration site may be a technique worth further development.

Lack of Humic Acid Response at Whiskey Island

There was no apparent benefit of humic acid for *S. alterniflora* or *A. germinans* at the restoration site. Even though we were able to demonstrate a benefit of humic acid for *S. alterniflora* when grown in controlled, greenhouse conditions (Chapter 4), we did not get an effect in the field. This could have been due to a combination of high *S. alterniflora* transplant mortality within a month and to potential removal of humic acid from sediments over a longer period of time by processes of estuarine mixing (Fox 1983). At a mangrove restoration site in Thailand, humic acid was also used without success to increase organic carbon content in sediments. After three years there were no significant differences in tree height between control trees and trees with the humic acid application at the Thailand restoration site (application rate not provided, Naohiro et al., 2012). The lack of effect of humic acid on propagule survival may be due to the additional benefit of maternal reserves provided by the large cotyledons. Taken together, these data support the concept that *A. germinans* may be insensitive to the effects of humic acid as a stress ameliorator in stressful environmental conditions (Willis and Hester 2010).

Hydrologic Challenges to Restoration

Establishing or reintroducing hydrologic connections is a critical factor for mangrove restoration (Turner and Lewis 1997; Field 1998; Ellison 2000; Kairo et al. 2001; Lewis 2005), and this holds true for subtropical mangrove restoration in a back-barrier salt marsh. Ecological functions of restored mangroves such as carbon and nutrient cycling as well as faunal use depend on establishment of appropriate hydrologic conditions (McKee and Faulkner 2000; Bosire et al. 2008). Efforts to restore mangroves in locations where they will not be exposed to appropriate hydrology can lead to large-scale restoration failure (Primavera and Esteban 2008). Hydrology was also critical for the successional element of our restoration design, since the poor survival of contracted transplants prevented the establishment of healthy *S. alterniflora* marsh, within which we were to conduct our propagule establishment experiments. A successfully vegetated area

would be expected to create a positive feedback by improving local site conditions that favor further colonization and natural recruit survival (Bosire et al. 2003; McKee et al. 2007).

There were hydrologic challenges to establishing our experimental planting at this back-barrier salt marsh restoration site. One important consideration is identifying the appropriate range of elevation in which to establish the species of interest, *A. germinans*, and to do so within a time frame that would prevent desiccation of propagules. Alleman and Hester (2011b) reported establishment elevations for *A. germinans* seedlings less than 12 months old of 0.199 m NAVD88 and elevations of older *A. germinans* at 0.235 m NAVD88 near Port Fourchon, Louisiana, resulting in flooding frequencies of approximately 15%–33%. The created back-barrier marsh platform elevation of this project (0.762 m NAVD88) was engineered to decrease in elevation so that it would be intertidal (0 – 0.488 m NAVD 88) within five years after creation (Green 2007). Naturally established *A. germinans* on Whiskey Island were located at elevations of 0.64–0.84 m NAVD88, which demonstrates some of the hydrologic variability of this coastal area. Furthermore, propagules must quickly develop root systems to prevent being dislodged by hydrodynamic and sediment-dynamic forces within a short time frame (Balke et al. 2011). If there is little or no previous establishment of the species to guide placement of propagules or transplants, then a combination of historic hydrologic data, tidal cycle, and weather patterns may be used to guide placement.

The addition of fencing structures enhanced with *S. alterniflora* transplants has potential for improving black mangrove establishment at the salt marsh platform. Ensuring correct hydrology for vegetative restoration of the microtidal Louisiana coast is difficult because there is a small margin for error in which to hit a dynamic hydrologic target for establishing species within a small range of intertidal elevations. Therefore, we recommend devoting effort and resources to fully understanding the hydrology of the system, and to considering expected future changes in hydrology due to factors such as sediment compaction and relative sea-level rise. If black mangrove restoration efforts can proceed in phases, these phases should account for short-term, dynamic changes in hydrology (driven primarily by dewatering and compaction of the fill material) that could affect goals like establishment success. Restoration goals and expected trajectories of development should also reflect the ephemeral nature of barrier island habitats. Sediment characteristics that develop slowly may never approach coastal salt marsh values. Application of humic acid to marsh sediments exposed to tidal exchange does not seem to result in a growth benefit to *S. alterniflora*, and there is no obvious benefit of humic acid to *A. germinans* propagule establishment, survival, or seedling growth. Finally, this knowledge can further guide restoration planning in a manner that may result in a more complex ecosystem.

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SUMMARY

The transient nature of Louisiana's barrier islands, in conjunction with the sand-deficient environmental setting, results in substantial challenges to the restoration of these important coastal features. These restoration projects are expensive, in part due to the high costs associated with the transportation of sand and sediments to nourish the various habitats composing barrier island systems. The successful establishment of plant species that are adapted to these environments and capable of stabilizing the limited sediment resources introduced by restoration efforts are therefore paramount. This collection of research studies has assessed a number of novel restoration approaches and provided insights into optimal rehabilitation of coastal habitats.

Humic acid is an operationally defined component of soil's organic matter and has been studied extensively for the enhancement of marginal soils in agricultural and horticultural applications. Previously, a small number of greenhouse studies alluded to the potential benefit of employing humic acid amendment in coastal plant restoration efforts. However, at the time of this writing, there are no comprehensive greenhouse experiments or large-scale field studies available in the peer-reviewed literature to inform restoration managers of the utility of this soil amendment in coastal restoration scenarios. To resolve existing data gaps, the efficacy of humic acid amendment was determined in a series of greenhouse studies and large field experiments. In particular, an understanding of the effective humic acid amendment dosage range, that is, the ranges of dosages at which humic acid amendment promotes growth versus impairs growth was sought. Additionally, elucidation of the interplay between humic acid amendment and fertilization regime was undertaken.

The humic acid range-finding greenhouse study conducted as an initial component of this overall research effort revealed interspecific variation in the response of coastal plant species to this soil amendment. Several coastal plant species including *Uniola paniculata*, *Panicum amarum*, *Spartina patens*, *Spartina alterniflora*, and *Avicennia germinans* demonstrated improved growth, particularly in belowground biomass, with low to moderate levels of humic acid amendment. However, *Baccharis halimifolia* and *Distichlis spicata* were not found to benefit from any level of humic acid amendment. Interestingly, the highest level of humic acid amendment tested ($8,100 \text{ ml m}^{-2}$) detrimentally affected the growth of all species in the range-finding study, suggesting that only negative effects would occur above this dosage. A second study, examining the benefit of humic acid amendment in conjunction with fertilizer, did not reveal as clear a benefit of humic acid amendment on plant growth, nor was any synergy obvious between fertilizer addition and humic acid amendment. This study was allowed to run for a much longer period of time than the range-finding study (10 months versus 2 months), so there may have been sufficient time for plants that did not receive humic acid amendment to attain the same extent of growth as those that did receive the soil conditioner.

A field investigation was conducted to assess the efficacy of several techniques intended to enhance the success of coastal restoration planting. The techniques assessed included increased planting density, humic acid amendment, and fertilizer regime. Humic acid amendment dosages (125 ml m^{-2} ; 250 ml m^{-2}) were based on the greenhouse investigations as well as previous laboratory research. Increased planting density treatments consisted of doubling the planting density for *U. paniculata* and *P. amarum* and tripling planting density for *S. patens*.

The broadcast fertilizer regime utilized for this study was based on standard dune and swale restoration projects in the southeastern U.S. (spring addition of 8-8-8 fertilizer at a rate of 878.8 kg ha⁻¹, summer and fall addition of ammonium nitrate fertilizer at a rate of 195.3 kg ha⁻¹). Importantly, the higher planting density of *U. paniculata* and *S. patens* resulted in a long-term benefit of increased plant coverage. *Panicum amarum*, however, demonstrated no potential for sustained benefit from increased planting density, as the low planting density rapidly attained equivalent coverage to the high density planting treatment. Therefore, higher planting densities are recommended for *U. paniculata* and *S. patens* to increase the success of barrier island dune and swale plantings, but do not appear to be necessary for *P. amarum* success.

Broadcast fertilizer increased vegetative coverage of all three species. Humic acid amendment demonstrated little benefit to aboveground growth, probably due to the minimal precipitation during the study, the lack of soil components to retain the applied humic acid, and the extent to which all plants received initial fertilizer application. However, it is quite likely that the belowground biomass of target dune and swale species, an extremely important component of these habitat types, could have been increased by humic acid amendment. This benefit of humic acid amendment on belowground biomass was noted in the range-finding study conducted as a portion of this overall research project and has been widely reported in the peer-reviewed literature for agricultural and horticultural species. Implementation of a broadcast fertilizer regime is recommended to increase the success of *U. paniculata*, *P. amarum*, and *S. patens* plantings.

This study yielded multiple beneficial findings in spite of delayed plantings due to slow-growing nursery stock, the Deepwater Horizon Oil Spill, and two years of unusually low precipitation that impacted plant growth after initiation of the study. An interesting finding of this research is that expansion of previously seeded *Cynodon dactylon* may actually limit the establishment and expansion of target species, particularly *S. patens*. *Cynodon dactylon* is known to respond quite favorably to nutrient augmentation and also may have limited the availability of the applied fertilizer to target species. Because of its lawn-like growth form, it is also possible that *C. dactylon* is interfering with dune formation by restricting the movement of sand to sand fences and dune-building plant species, such as *U. paniculata* and *P. amarum*. Further investigation into the efficacy of *C. dactylon* use in barrier island restoration is warranted.

The importance of using woody vegetation in barrier island restoration efforts, including those of swale habitats, has recently become recognized. *Baccharis halimifolia* is a highly appropriate species for incorporating into the swale component of Louisiana barrier island restoration projects. This project provided key information regarding the seed-based restoration ecology of *B. halimifolia* for Louisiana barrier islands through a series of greenhouse studies and several field trials. The Louisiana *B. halimifolia* populations assessed were determined to have no required dormancy period for seeds and were capable of germinating immediately after being harvested without any pre-treatment having been employed. This is important as it suggests that commercial nurseries would be able to rapidly prepare large numbers of *B. halimifolia* seeds for a restoration project without time-consuming pre-treatments being performed. *Baccharis halimifolia* seeds demonstrated optimal germination (64%) when they were placed at the soil surface. However, greatly reduced *B. halimifolia* seed germination (< 3%) was demonstrated

when seeds were buried by greater than 0.5 cm of sand. *Baccharis halimifolia* seed germination was also substantially reduced by shading. These findings reveal the need to locate seed-based *B. halimifolia* restoration efforts in environments where they are largely shielded from sand burial but receive sufficient light to optimally germinate. In a greenhouse investigation, the use of a commercially available hydromulch increased *B. halimifolia* seed germination in treatments containing low soil organic matter, but drought conditions reduced germination across all organic matter and hydromulch treatments. The critical sensitivity of *B. halimifolia* seed germination to low soil moisture was further confirmed by several field trials in which *B. halimifolia* seed and hydromulch were applied. When precipitation did not occur within 3 days, no surviving seedlings were detectable.

Back-barrier salt marshes are well known to be valuable habitats, much like their mainland analogues. *Spartina alterniflora* and *A. germinans* are common constituents of Louisiana back-barrier salt marshes, and *S. alterniflora* plants and *A. germinans* seedlings are currently included in back-barrier salt marsh planting efforts. Greenhouse studies investigated *A. germinans* propagule establishment techniques and humic acid application on *A. germinans* and *S. alterniflora* species interactions at moderate and high salinity. Due to the improved survival and establishment of propagules placed under hydromulch in the greenhouse study, *A. germinans* propagules may benefit from a very thin non-smothering layer of hydromulch in the upper intertidal range. Interestingly, humic acid amendment (500 ml m⁻²) results in increased *S. alterniflora* biomass and cumulative height, but did not affect leaf tissue nitrogen. In addition, *A. germinans* seedlings have a very high salinity tolerance, which supports their use at constructed salt marsh platforms. Field assessments of propagule establishment techniques (propagule dispersal, *S. alterniflora*-enhanced biodegradable fencing) designed to enable propagule-based restoration of *A. germinans* were conducted. The installation of biodegradable fencing enhanced propagule retention and survival for an additional month when compared to retention and survival in bare plots, thereby possibly providing another means to allow propagules to be retained in targeted areas.

APPENDIX

Table A1.1 The effect of humic acid amendment on *Uniola paniculata* soil pH, conductivity, moisture, organic matter, ammonium, nitrate-nitrite, and phosphorus (mean +/- SE, n = 5). pH LSD = 0.45, conductivity LSD = 156.95, organic matter LSD = 0.34, moisture LSD = 0.51, ammonium LSD = 1.15, nitrate-nitrite LSD = 0.44, phosphorus LSD = 0.40, potassium LSD = 7.199

Metric	0 ml m ⁻²	100 ml m ⁻²	300 ml m ⁻²	900 ml m ⁻²	2700 ml m ⁻²	8100 ml m ⁻²
pH	6.568	6.49	6.312	6.724	6.564	8.028
(+/- SE)	(0.22)	(0.08)	(0.07)	(0.20)	(0.14)	(0.17)
Conductivity, $\mu\text{S cm}^{-1}$	162.60	223.68	261.00	284.32	334.62	429.18
(+/- SE)	(17.98)	(55.05)	(35.14)	(70.24)	(38.26)	(92.30)
Moisture, %	0.14	0.13	0.21	0.13	0.37	2.19
(+/- SE)	(0.01)	(0.02)	(0.06)	(0.02)	(0.08)	(0.46)
Organic matter, %	0.37	0.43	0.42	0.34	0.43	0.37
(+/- SE)	(0.03)	(0.01)	(0.07)	(0.02)	(0.10)	(0.01)
Ammonium, $\mu\text{g g}^{-1}$	0.39	0.32	1.28	0.62	0.40	1.63
(+/- SE)	(0.16)	(0.10)	(0.99)	(0.18)	(0.09)	(0.89)
Nitrate-Nitrite, $\mu\text{g g}^{-1}$	1.19	0.63	0.24	0.32	0.81	0.43
(+/- SE)	(0.36)	(0.13)	(0.06)	(0.05)	(0.29)	(0.08)
Phosphorus, $\mu\text{g g}^{-1}$	0.648	0.793	0.584	0.734	0.422	0.635
(+/- SE)	(0.223)	(0.312)	(0.275)	(0.311)	(0.315)	(0.318)

Table A1.2 The effect of humic acid amendment on *Panicum amarum* soil pH, conductivity, moisture, organic matter, ammonium, nitrate-nitrite, and phosphorus (mean +/- SE, n = 5). pH LSD = 0.45, conductivity LSD = 156.95, organic matter LSD = 0.34, moisture LSD = 0.51, ammonium LSD = 1.15, nitrate-nitrite LSD = 0.44, phosphorus LSD = 0.40, potassium LSD = 7.199

Metric	0 ml m ⁻²	100 ml m ⁻²	300 ml m ⁻²	900 ml m ⁻²	2700 ml m ⁻²	8100 ml m ⁻²
pH	6.756	6.926	6.902	7.118	7.036	8.216
(+/- SE)	(0.10)	(0.09)	(0.08)	(0.11)	(0.14)	(0.04)
Conductivity, $\mu\text{S cm}^{-1}$	168.82	221.74	313.82	230.86	236.34	448.38
(+/- SE)	(46.34)	(29.05)	(37.77)	(30.06)	(33.70)	(62.02)
Moisture, %	0.50	0.25	0.27	0.17	0.46	1.28
(+/- SE)	(0.36)	(0.09)	(0.05)	(0.03)	(0.19)	(0.20)
Organic matter, %	0.40	0.41	0.44	0.42	0.44	0.41
(+/- SE)	(0.04)	(0.03)	(0.08)	(0.04)	(0.03)	(0.03)
Ammonium, $\mu\text{g g}^{-1}$	1.19	1.19	1.21	1.30	0.85	0.98
(+/- SE)	(0.25)	(0.11)	(0.23)	(0.02)	(0.21)	(0.26)
Nitrate-Nitrite, $\mu\text{g g}^{-1}$	0.73	0.64	0.54	0.51	0.51	0.44
(+/- SE)	(0.09)	(0.13)	(0.11)	(0.00)	(0.05)	(0.04)
Phosphorus, $\mu\text{g g}^{-1}$	0.895	0.567	0.626	0.495	0.731	0.684
(+/- SE)	(0.311)	(0.267)	(0.211)	(0.248)	(0.312)	(0.271)

Table A1.3 The effect of humic acid amendment on *Distichlis spicata* soil pH, conductivity, moisture, organic matter, ammonium, nitrate-nitrite, and phosphorus (mean +/- SE, n = 5). pH LSD = 0.45, conductivity LSD = 156.95, organic matter LSD = 0.34, moisture LSD = 0.51, ammonium LSD = 1.15, nitrate-nitrite LSD = 0.44, phosphorus LSD = 0.40, potassium LSD = 7.199

Metric	0 ml m ⁻²	100 ml m ⁻²	300 ml m ⁻²	900 ml m ⁻²	2700 ml m ⁻²	8100 ml m ⁻²
pH	7.554	7.058	7.226	6.856	7.456	7.798
(+/- SE)	(0.07)	(0.12)	(0.17)	(0.18)	(0.31)	(0.12)
Conductivity, $\mu\text{S cm}^{-1}$	504.92	299.04	346.10	334.52	307.46	676.26
(+/- SE)	(76.00)	(49.64)	(35.84)	(61.21)	(39.68)	(106.22)
Moisture, %	0.18	0.14	0.16	0.19	0.90	0.85
(+/- SE)	(0.02)	(0.01)	(0.02)	(0.05)	(0.29)	(0.18)
Organic matter, %	0.45	0.33	0.82	0.45	0.40	0.37
(+/- SE)	(0.05)	(0.02)	(0.38)	(0.05)	(0.03)	(0.02)
Ammonium, $\mu\text{g g}^{-1}$	0.98	1.89	1.01	1.18	2.06	1.85
(+/- SE)	(0.32)	(0.53)	(0.31)	(0.28)	(0.56)	(0.62)
Nitrate-Nitrite, $\mu\text{g g}^{-1}$	0.72	0.53	0.36	0.38	0.37	0.66
(+/- SE)	(0.30)	(0.14)	(0.04)	(0.08)	(0.08)	(0.26)
Phosphorus, $\mu\text{g g}^{-1}$	0.480	0.674	0.687	0.613	0.722	0.802
(+/- SE)	(0.259)	(0.231)	(0.216)	(0.182)	(0.263)	(0.289)

Table A1.4 The effect of humic acid amendment on *Paspalum vaginatum* soil pH, conductivity, moisture, organic matter, ammonium, nitrate-nitrite, and phosphorus (mean +/- SE, n = 5). pH LSD = .45, conductivity LSD = 156.95, organic matter LSD = 0.34, moisture LSD = 0.51, ammonium LSD = 1.15, nitrate-nitrite LSD = 0.44, phosphorus LSD = 0.40, potassium LSD = 7.199

Metric	0 ml m ⁻²	100 ml m ⁻²	300 ml m ⁻²	900 ml m ⁻²	2700 ml m ⁻²	8100 ml m ⁻²
pH	6.668	6.296	6.304	6.52	6.614	8.076
(+/- SE)	(0.15)	(0.03)	(0.17)	(0.18)	(0.210)	(0.04)
Conductivity, $\mu\text{S cm}^{-1}$	130.86	118.88	201.46	164.60	161.50	251.56
(+/- SE)	(31.71)	(18.25)	(44.03)	(36.52)	(14.32)	(33.48)
Moisture, %	0.12	0.14	0.12	0.07	0.14	0.56
(+/- SE)	(0.02)	(0.02)	(0.02)	(0.03)	(0.02)	(0.17)
Organic matter, %	0.38	0.40	0.39	0.43	0.34	0.49
(+/- SE)	(0.06)	(0.02)	(0.04)	(0.05)	(0.02)	(0.03)
Ammonium, $\mu\text{g g}^{-1}$	1.16	0.61	1.02	0.55	0.34	0.70
(+/- SE)	(0.48)	(0.37)	(0.21)	(0.21)	(0.15)	(0.20)
Nitrate-Nitrite, $\mu\text{g g}^{-1}$	0.53	0.54	0.56	0.34	0.50	0.57
(+/- SE)	(0.15)	(0.02)	(0.20)	(0.08)	(0.06)	(0.20)
Phosphorus, $\mu\text{g g}^{-1}$	0.590	0.604	0.768	0.645	0.802	0.621
(+/- SE)	(0.187)	(0.173)	(0.216)	(0.220)	(0.214)	(0.248)

Table A1.5 The effect of humic acid amendment on *Spartina patens* soil pH, conductivity, moisture, organic matter, ammonium, nitrate-nitrite, and phosphorus (mean +/- SE, n = 5). pH LSD = 0.45, conductivity LSD = 156.95, organic matter LSD = 0.34, moisture LSD = 0.51, ammonium LSD = 1.15, nitrate-nitrite LSD = 0.44, phosphorus LSD = 0.40, potassium LSD = 7.199

Metric	0 ml m ⁻²	100 ml m ⁻²	300 ml m ⁻²	900 ml m ⁻²	2700 ml m ⁻²	8100 ml m ⁻²
pH	7.598	7.5025	7.61	7.4975	7.48	7.034
(+/- SE)	(0.21)	(0.25)	(0.22)	(0.11)	(0.27)	(0.19)
Conductivity, $\mu\text{S cm}^{-1}$	339.14	296.00	229.50	230.92	504.50	203.86
(+/- SE)	(91.84)	(45.87)	(51.62)	(14.98)	(135.56)	(41.22)
Moisture, %	0.20	0.26	0.26	0.41	0.65	0.10
(+/- SE)	(0.11)	(0.08)	(0.170)	(0.20)	(0.24)	(0.01)
Organic matter, %	0.33	0.36	0.34	0.37	0.37	1.01
(+/- SE)	(0.01)	(0.06)	(0.02)	(0.05)	(0.04)	(0.57)
Ammonium, $\mu\text{g g}^{-1}$	0.45	0.44	0.42	0.45	0.28	0.37
(+/- SE)	(0.06)	(0.17)	(0.06)	(0.04)	(0.06)	(0.06)
Nitrate-Nitrite, $\mu\text{g g}^{-1}$	1.58	4.48	2.05	1.34	2.63	3.02
(+/- SE)	(0.21)	(0.48)	(0.74)	(0.27)	(0.38)	(0.75)
Phosphorus, $\mu\text{g g}^{-1}$	0.504	0.657	0.583	0.789	0.673	0.853
(+/- SE)	(0.172)	(0.194)	(0.241)	(0.285)	(0.202)	(0.361)

Table A1.6 The effect of humic acid amendment on *Baccharis halimifolia* soil pH, conductivity, moisture, organic matter, ammonium, nitrate-nitrite, and phosphorus (mean \pm SE, n = 5). pH LSD = 0.45, conductivity LSD = 156.95, organic matter LSD = 0.34, moisture LSD = 0.51, ammonium LSD = 1.15, nitrate-nitrite LSD = 0.44, phosphorus LSD = 0.40, potassium LSD = 7.199

Metric	0 ml m ⁻²	100 ml m ⁻²	300 ml m ⁻²	900 ml m ⁻²	2700 ml m ⁻²	8100 ml m ⁻²
pH	7.096	7.012	7.056	7.768	7.05	8.53
(\pm SE)	(0.04)	(0.21)	(0.23)	(0.21)	(0.11)	(0.03)
Conductivity, $\mu\text{S cm}^{-1}$	225.06	218.08	136.80	198.40	273.58	357.48
(\pm SE)	(44.11)	(72.99)	(22.38)	(46.33)	(50.34)	(75.21)
Moisture, %	0.81	0.99	0.81	1.64	1.99	2.09
(\pm SE)	(0.31)	(0.28)	(0.18)	(0.45)	(0.22)	(0.13)
Organic matter, %	0.39	0.34	0.51	0.38	0.37	0.38
(\pm SE)	(0.02)	(0.03)	(0.11)	(0.02)	(0.01)	(0.01)
Ammonium, $\mu\text{g g}^{-1}$	0.49	0.58	0.48	0.44	0.44	0.41
(\pm SE)	(0.20)	(0.10)	(0.11)	(0.06)	(0.06)	(0.10)
Nitrate-Nitrite, $\mu\text{g g}^{-1}$	0.88	1.26	1.63	1.90	1.14	1.12
(\pm SE)	(0.27)	(0.22)	(0.41)	(0.60)	(0.19)	(0.12)
Phosphorus, $\mu\text{g g}^{-1}$	0.710	0.620	0.423	0.504	0.721	0.979
(\pm SE)	(0.127)	(0.103)	(0.036)	(0.201)	(0.151)	(0.152)

Table A1.7 The effect of humic acid amendment on *Spartina alterniflora* soil pH, conductivity, moisture, organic matter, ammonium, nitrate-nitrite, and phosphorus (mean +/- SE, n = 5). pH LSD = 0.30, conductivity LSD = 2772.4, organic matter LSD = 0.59, moisture LSD = 1.76, ammonium LSD = 1.29, nitrate-nitrite LSD = 0.41, phosphorus LSD = 0.365, potassium LSD = 20.069

Metric	0 ml m ⁻²	100 ml m ⁻²	300 ml m ⁻²	900 ml m ⁻²	2700 ml m ⁻²	8100 ml m ⁻²
pH	7.908	7.872	7.788	7.79	7.416	7.938
(+/- SE)	(0.11)	(0.17)	(90.12)	(0.05)	(0.09)	(0.15)
Conductivity, $\mu\text{S cm}^{-1}$	6.59	917.45	915.48	634.00	1245.26	714.49
(+/- SE)	(0.47)	(910.64)	(908.63)	(627.25)	(838.77)	(706.13)
Moisture, %	13.64	13.45	13.48	13.39	12.85	10.94
(+/- SE)	(0.37)	(0.24)	(0.32)	(0.29)	(0.38)	(1.21)
Organic matter, %	0.66	0.90	0.66	0.69	0.75	0.61
(+/- SE)	(0.06)	(0.12)	(0.07)	(0.11)	(0.14)	(0.08)
Ammonium, $\mu\text{g g}^{-1}$	1.18	1.15	1.15	1.20	1.05	1.19
(+/- SE)	(0.13)	(0.16)	(0.16)	(0.11)	(0.16)	(0.12)
Nitrate-Nitrite, $\mu\text{g g}^{-1}$	2.66	2.55	2.55	2.72	2.20	2.68
(+/- SE)	(0.44)	(0.55)	(0.55)	(0.38)	(0.56)	(0.43)
Phosphorus, $\mu\text{g g}^{-1}$	0.601	0.738	0.721	0.645	0.658	0.549
(+/- SE)	(0.172)	(0.031)	(0.276)	(0.746)	(0.311)	(0.607)

Table A1.8 The effect of humic acid amendment on *Avicennia germinans* soil pH, conductivity, moisture, organic matter, ammonium, nitrate-nitrite, and phosphorus (mean +/- SE, n = 5). pH LSD = 0.30, conductivity LSD = 2772.4, organic matter LSD = 0.59, moisture LSD = 1.76, ammonium LSD = 1.29, nitrate-nitrite LSD = 0.41, phosphorus LSD = 0.365, potassium LSD = 20.069

Metric	0 ml m ⁻²	100 ml m ⁻²	300 ml m ⁻²	900 ml m ⁻²	2700 ml m ⁻²	8100 ml m ⁻²
pH	7.61	7.57	7.69	7.48	7.53	7.96
(+/- SE)	(0.08)	(0.12)	(0.14)	(0.03)	(0.05)	(0.06)
Conductivity, $\mu\text{S cm}^{-1}$	9.17	9.98	9.25	9.24	11.39	11.76
(+/- SE)	(1.07)	(0.51)	(0.76)	(1.25)	(1.87)	(0.79)
Moisture, %	15.26	14.93	16.44	15.81	15.24	14.71
(+/- SE)	(0.92)	(0.46)	(0.58)	(0.64)	(0.56)	(0.72)
Organic matter, %	1.45	0.77	0.60	0.98	0.81	1.06
(+/- SE)	(0.65)	(0.12)	(0.08)	(0.08)	(0.09)	(0.08)
Ammonium, $\mu\text{g g}^{-1}$	1.39	1.48	1.31	1.24	1.31	1.31
(+/- SE)	(0.07)	(0.17)	(0.00)	(0.07)	(0.00)	(0.00)
Nitrate-Nitrite, $\mu\text{g g}^{-1}$	3.35	3.67	3.10	2.84	3.10	3.10
(+/- SE)	(0.26)	(0.58)	(0.00)	(0.26)	(0.00)	(0.00)
Phosphorus, $\mu\text{g g}^{-1}$	0.780	0.678	0.716	0.734	0.523	0.873
(+/- SE)	(0.196)	(0.234)	(0.266)	(0.232)	(0.231)	(0.316)

Table A1.9 The effect of fertilizer regime and humic acid amendment on *Uniola paniculata* and *Panicum amarum* leaf tissue nitrogen, phosphorus, and potassium content (mean +/- SE, n = 4). nitrogen LSD = 0.227, phosphorus LSD = 71.04, potassium LSD = 1586.8

Metric	0 ml m ⁻²		125 ml m ⁻²		250 ml m ⁻²		500 ml m ⁻²	
	Low Fertilizer	High Fertilizer	Low Fertilizer	High Fertilizer	Low Fertilizer	High Fertilizer	Low Fertilizer	High Fertilizer
<i>Uniola paniculata</i>								
Nitrogen, % (+/- SE)	0.747 (0.017)	0.739 (0.027)	0.825 (0.033)	0.841 (0.045)	0.770 (0.043)	0.710 (0.045)	0.691 (0.032)	0.716 (0.014)
Phosphorus, µg g ⁻¹ (+/- SE)	100.5 (25.5)	108.5 (9.8)	84.4 (13.1)	135.9 (14.0)	53.1 (18.0)	156.8 (9.8)	63.2 (5.4)	140.8 (18.1)
Potassium, µg g ⁻¹ (+/- SE)	952.0 (80.6)	738.1 (14.1)	920.6 (51.5)	935.9 (70.2)	718.2 (241.7)	832.4 (24.2)	825.6 (18.8)	791.9 (38.3)
<i>Panicum amarum</i>								
Nitrogen, % (+/- SE)	0.742 (0.028)	0.688 (0.022)	0.667 (0.017)	0.695 (0.022)	0.648 (0.013)	0.667 (0.052)	0.718 (0.038)	0.644 (0.027)
Phosphorus, µg g ⁻¹ (+/- SE)	121.9 (4.5)	130.2 (7.4)	57.5 (33.6)	94.4 (32.0)	52.9 (31.0)	51.6 (30.0)	102.4 (34.2)	90.7 (31.7)
Potassium, µg g ⁻¹ (+/- SE)	1541.5 (6.3)	1461.1 (74.7)	710.7 (414.8)	1096.6 (367.7)	827.8 (479.2)	714.8 (418.5)	1272.9 (426.9)	1052.9 (351.1)

Table A1.10 The effect of fertilizer regime and humic acid amendment on *Distichlis spicata* and *Paspalum vaginatum* leaf tissue nitrogen, phosphorus, and potassium content (mean +/- SE, n = 4). nitrogen LSD = 0.227, phosphorus LSD = 71.04, potassium LSD = 1586.8

Metric	0 ml m ⁻²		125 ml m ⁻²		250 ml m ⁻²		500 ml m ⁻²	
	Low Fertilizer	High Fertilizer	Low Fertilizer	High Fertilizer	Low Fertilizer	High Fertilizer	Low Fertilizer	High Fertilizer
<i>Distichlis spicata</i>								
Nitrogen, %	0.991	1.373	0.992	1.217	0.923	1.260	1.057	1.209
(+/- SE)	(0.054)	(0.183)	(0.016)	(0.069)	(0.050)	(0.103)	(0.062)	(0.050)
Phosphorus, µg g ⁻¹	115.2	224.6	106.8	142.9	108.4	150.4	113.0	142.5
(+/- SE)	(16.3)	(17.7)	(4.9)	(12.4)	(12.1)	(11.9)	(6.2)	(4.7)
Potassium, µg g ⁻¹	724.3	1258.5	693.4	896.4	899.7	855.5	859.5	881.5
(+/- SE)	(82.6)	(341.6)	(67.7)	(117.4)	(235.6)	(93.9)	(71.8)	(47.9)
<i>Paspalum vaginatum</i>								
Nitrogen, %	0.803	0.675	0.741	0.715	0.755	0.724	0.709	0.753
(+/- SE)	(0.074)	(0.061)	(0.038)	(0.014)	(0.033)	(0.040)	(0.030)	(0.057)
Phosphorus, µg g ⁻¹	127.0	109.6	91.4	158.9	114.8	357.1	130.5	247.8
(+/- SE)	(17.0)	(46.4)	(7.4)	(12.1)	(9.5)	(73.4)	(26.6)	(85.7)
Potassium, µg g ⁻¹	892.2	678.5	1077.2	1414.0	1316.4	1008.0	1172.5	670.6
(+/- SE)	(129.0)	(280.6)	(138.5)	(78.0)	(246.8)	(125.4)	(255.5)	(206.8)

Table A1.11 The effect of fertilizer regime and humic acid amendment on, *Spartina patens* and *Spartina alterniflora* leaf tissue nitrogen, phosphorus, and potassium content (mean +/- SE, n = 4). nitrogen LSD = 0.227, phosphorus LSD = 71.04, potassium LSD = 1586.8

Metric	0 ml m ⁻²		125 ml m ⁻²		250 ml m ⁻²		500 ml m ⁻²	
	Low Fertilizer	High Fertilizer	Low Fertilizer	High Fertilizer	Low Fertilizer	High Fertilizer	Low Fertilizer	High Fertilizer
<i>Spartina patens</i>								
Nitrogen, %	0.810	0.745	0.700	0.703	0.736	0.695	0.778	0.688
(+/- SE)	(0.071)	(0.046)	(0.056)	(0.026)	(0.057)	(0.017)	(0.115)	(0.021)
Phosphorus, µg g ⁻¹	125.3	236.8	157.0	261.6	112.5	254.3	130.4	249.1
(+/- SE)	(12.6)	(42.0)	(29.6)	(19.6)	(34.9)	(39.4)	(18.8)	(48.9)
Potassium, µg g ⁻¹	575.0	591.1	452.8	738.7	478.0	685.4	679.9	589.7
(+/- SE)	(107.4)	(135.3)	(53.9)	(77.6)	(150.0)	(102.6)	(198.0)	(86.0)
<i>Spartina alterniflora</i>								
Nitrogen, %	1.082	0.852	0.979	1.021	1.018	0.945	0.910	0.972
(+/- SE)	(0.033)	(0.040)	(0.072)	(0.030)	(0.076)	(0.025)	(0.019)	(0.033)
Phosphorus, µg g ⁻¹	293.8	276.3	306.0	312.2	292.6	303.1	253.4	317.9
(+/- SE)	(19.0)	(13.7)	(12.5)	(20.4)	(9.8)	(23.7)	(14.0)	(23.0)
Potassium, µg g ⁻¹	1551.5	1090.8	1392.4	1483.5	1354.9	1225.4	1322.8	1428.0
(+/- SE)	(60.2)	(100.3)	(116.5)	(97.5)	(46.4)	(52.3)	(66.1)	(99.5)

Table A1.12 The effect of fertilizer regime and humic acid amendment on *Baccharis halimifolia* and *Avicennia germinans* leaf tissue nitrogen, phosphorus, and potassium content (mean +/- SE, n = 4). nitrogen LSD = 0.227, phosphorus LSD = 71.04, potassium LSD = 1586.8

Metric	0 ml m ⁻²		125 ml m ⁻²		250 ml m ⁻²		500 ml m ⁻²	
	Low Fertilizer	High Fertilizer	Low Fertilizer	High Fertilizer	Low Fertilizer	High Fertilizer	Low Fertilizer	High Fertilizer
<i>Baccharis halimifolia</i>								
Nitrogen, %	1.006	0.983	1.011	0.995	1.160	0.985	0.972	0.814
(+/- SE)	(0.080)	(0.094)	(0.112)	(0.097)	(0.185)	(0.119)	(0.104)	(0.047)
Phosphorus, µg g ⁻¹	59.7	118.4	116.5	100.9	117.9	94.8	110.1	63.9
(+/- SE)	(10.8)	(16.1)	(8.9)	(33.5)	(15.2)	(11.0)	(10.9)	(29.9)
Potassium, µg g ⁻¹	937.1	920.4	1369.2	369.9	1022.9	1195.4	1897.1	1137.1
(+/- SE)	(415.0)	(332.0)	(740.2)	(21.5)	(91.5)	(428.5)	(1312.6)	(419.1)
<i>Avicennia germinans</i>								
Nitrogen, %	1.560	1.588	1.562	1.496	1.303	1.503	1.462	1.488
(+/- SE)	(0.085)	(0.093)	(0.060)	(0.066)	(0.247)	(0.047)	(0.054)	(0.042)
Phosphorus, µg g ⁻¹	93.3	100	134.3	117.9	111.9	101.7	132.9	131.9
(+/- SE)	(32.3)	(18.3)	(4.8)	(4.7)	(19.8)	(18.6)	(9.1)	(3.7)
Potassium, µg g ⁻¹	1905.7	1811.1	3964.6	1463	1691	1093.1	1606.4	3467.1
(+/- SE)	(760.3)	(123.3)	(2943.7)	(138.6)	(665.1)	(116.2)	(458.7)	(2356.8)

Table A1.13 The effect of fertilizer regime and humic acid amendment on *Uniola paniculata* soil pH, conductivity, moisture, organic matter, ammonium, nitrate-nitrite, and phosphorus (mean +/- SE, n = 4). pH LSD = 0.69, conductivity LSD = 56.53, moisture LSD = 4.19, organic matter LSD = 0.43, ammonium LSD = 0.941, nitrate-nitrite LSD = 0.329, phosphorus LSD = 1.012

Metric	0 ml m ⁻²		125 ml m ⁻²		250 ml m ⁻²		500 ml m ⁻²	
	Low Fertilizer	High Fertilizer	Low Fertilizer	High Fertilizer	Low Fertilizer	High Fertilizer	Low Fertilizer	High Fertilizer
pH	6.83	7.09	6.91	6.80	7.51	7.30	6.98	6.69
(+/- SE)	(0.26)	(0.56)	(0.30)	(0.28)	(0.26)	(0.38)	(0.39)	(0.31)
Conductivity, $\mu\text{S cm}^{-1}$	66.33	66.03	55.13	54.15	50.65	41.70	61.88	73.00
(+/- SE)	(26.43)	(23.06)	(18.62)	(20.93)	(8.17)	(9.68)	(16.99)	(31.17)
Moisture, %	8.60	8.37	9.79	2.88	11.95	5.43	9.29	8.52
(+/- SE)	(1.40)	(2.28)	(2.03)	(0.51)	(1.48)	(1.38)	(2.01)	(0.86)
Organic matter, %	0.26	0.35	0.37	0.35	0.29	0.32	0.39	0.46
(+/- SE)	(0.05)	(0.09)	(0.03)	(0.03)	(0.06)	(0.05)	(0.04)	(0.08)
Ammonium, $\mu\text{g g}^{-1}$	0.188	0.198	0.065	0.158	0.155	0.128	0.172	0.131
(+/- SE)	(0.053)	(0.049)	(0.015)	(0.038)	(0.060)	(0.031)	(0.055)	(0.050)
Nitrate-Nitrite, $\mu\text{g g}^{-1}$	0.289	0.251	0.275	0.301	0.277	0.419	0.27	0.264
(+/- SE)	(0.034)	(0.024)	(0.036)	(0.07)	(0.073)	(0.221)	(0.037)	(0.015)
Phosphorus, $\mu\text{g g}^{-1}$	0.266	0.339	0.226	0.318	0.320	0.403	0.183	0.564
(+/- SE)	(0.117)	(0.082)	(0.043)	(0.158)	(0.183)	(0.135)	(0.082)	(0.091)

Table A1.14 The effect of fertilizer regime and humic acid amendment on *Panicum amarum* soil pH, conductivity, moisture, organic matter, ammonium, nitrate-nitrite, and phosphorus (mean +/- SE, n = 4). pH LSD = 0.69, conductivity LSD = 56.53, moisture LSD = 4.19, organic matter LSD = 0.43, ammonium LSD = 0.941, nitrate-nitrite LSD = 0.329, phosphorus LSD = 1.012

Metric	0 ml m ⁻²		125 ml m ⁻²		250 ml m ⁻²		500 ml m ⁻²	
	Low Fertilizer	High Fertilizer	Low Fertilizer	High Fertilizer	Low Fertilizer	High Fertilizer	Low Fertilizer	High Fertilizer
pH	6.84	6.45	6.42	6.75	6.77	6.76	6.47	6.75
(+/- SE)	(0.12)	(0.10)	(0.05)	(0.13)	(0.15)	(0.29)	(0.22)	(0.17)
Conductivity, $\mu\text{S cm}^{-1}$	59.85	50.83	42.08	71.73	37.15	45.65	23.33	44.35
(+/- SE)	(21.08)	(31.49)	(7.61)	(38.09)	(5.08)	(5.14)	(7.39)	(13.82)
Moisture, %	9.98	4.85	8.13	7.62	8.04	5.65	8.58	5.54
(+/- SE)	(0.31)	(0.92)	(1.13)	(1.05)	(0.95)	(1.41)	(1.16)	(0.65)
Organic matter, %	1.34	0.41	0.40	0.38	0.31	0.32	0.43	0.42
(+/- SE)	(0.70)	(0.06)	(0.05)	(0.04)	(0.02)	(0.07)	(0.020)	(0.09)
Ammonium, $\mu\text{g g}^{-1}$	0.188	0.198	0.065	0.158	0.155	0.128	0.172	0.131
(+/- SE)	(0.053)	(0.049)	(0.015)	(0.038)	(0.060)	(0.031)	(0.055)	(0.050)
Nitrate-Nitrite, $\mu\text{g g}^{-1}$	0.349	0.298	0.276	0.404	0.297	0.264	0.280	0.222
(+/- SE)	(0.049)	(0.056)	(0.028)	(0.089)	(0.000)	(0.035)	(0.026)	(0.020)
Phosphorus, $\mu\text{g g}^{-1}$	0.419	0.179	0.1390	0.194	0.311	0.182	0.392	0.180
(+/- SE)	(0.125)	(0.079)	(0.044)	(0.084)	(0.229)	(0.066)	(0.238)	(0.074)

Table A1.15 The effect of fertilizer regime and humic acid amendment on *Distichlis spicata* soil pH, conductivity, moisture, organic matter, ammonium, nitrate-nitrite, and phosphorus (mean +/- SE, n = 4). pH LSD = 0.69, conductivity LSD = 56.53, moisture LSD = 4.19, organic matter LSD = 0.43, ammonium LSD = 0.941, nitrate-nitrite LSD = 0.329, phosphorus LSD = 1.012

Metric	0 ml m ⁻²		125 ml m ⁻²		250 ml m ⁻²		500 ml m ⁻²	
	Low Fertilizer	High Fertilizer	Low Fertilizer	High Fertilizer	Low Fertilizer	High Fertilizer	Low Fertilizer	High Fertilizer
pH	7.01	6.67	7.10	6.85	6.67	6.37	6.68	6.58
(+/- SE)	(0.32)	(0.17)	(0.38)	(0.16)	(0.18)	(0.16)	(0.13)	(0.08)
Conductivity, $\mu\text{S cm}^{-1}$	37.50	51.35	102.48	44.33	46.43	33.58	77.95	71.18
(+/- SE)	(4.27)	(19.36)	(38.56)	(13.58)	(7.53)	(7.69)	(24.38)	(34.20)
Moisture, %	5.13	5.05	5.92	2.84	5.60	2.23	4.59	2.54
(+/- SE)	(0.89)	(2.20)	(0.87)	(0.94)	(0.89)	(0.62)	(0.84)	(0.53)
Organic matter, %	0.85	0.32	0.44	0.32	0.46	0.42	0.41	0.42
(+/- SE)	(0.34)	(0.04)	(0.07)	(0.04)	(0.09)	(0.09)	(0.03)	(0.05)
Ammonium, $\mu\text{g g}^{-1}$	0.111	0.175	0.091	0.284	0.087	0.177	0.145	0.077
(+/- SE)	(0.056)	(0.047)	(0.0350)	(0.110)	(0.037)	(0.071)	(0.058)	(0.027)
Nitrate-Nitrite, $\mu\text{g g}^{-1}$	0.481	0.686	0.243	0.888	0.37	0.248	0.26	0.317
(+/- SE)	(0.169)	(0.328)	(0.02)	(0.356)	(0.161)	(0.018)	(0.057)	(0.015)
Phosphorus, $\mu\text{g g}^{-1}$	0.500	0.110	0.378	0.315	1.123	0.797	0.572	0.245
(+/- SE)	(0.172)	(0.031)	(0.006)	(0.130)	(0.251)	(0.251)	(0.224)	(0.037)

Table A1.16 The effect of fertilizer regime and humic acid amendment on *Paspalum vaginatum* soil pH, conductivity, moisture, organic matter, ammonium, nitrate-nitrite, and phosphorus (mean +/- SE, n = 4). pH LSD = 0.69, conductivity LSD = 56.53, moisture LSD = 4.19, organic matter LSD = 0.43, ammonium LSD = 0.941, nitrate-nitrite LSD = 0.329, phosphorus LSD = 1.012

Metric	0 ml m ⁻²		125 ml m ⁻²		250 ml m ⁻²		500 ml m ⁻²	
	Low Fertilizer	High Fertilizer	Low Fertilizer	High Fertilizer	Low Fertilizer	High Fertilizer	Low Fertilizer	High Fertilizer
pH	7.47	6.99	6.81	6.83	7.27	7.37	7.01	7.27
(+/- SE)	(0.13)	(0.20)	(0.14)	(0.16)	(0.35)	(0.14)	(0.22)	(0.31)
Conductivity, $\mu\text{S cm}^{-1}$	39.90	47.55	30.80	62.48	22.97	29.40	25.95	30.23
(+/- SE)	(14.84)	(17.20)	(4.94)	(27.80)	(5.74)	(4.78)	(1.91)	(2.51)
Moisture, %	10.05	5.91	8.17	8.41	8.67	8.62	8.60	6.40
(+/- SE)	(4.41)	(0.67)	(0.36)	(0.75)	(1.19)	(0.41)	(0.25)	(0.87)
Organic matter, %	0.36	0.69	0.42	0.51	0.49	0.35	0.33	0.38
(+/- SE)	(0.03)	(0.30)	(0.04)	(0.02)	(0.11)	(0.05)	(0.03)	(0.03)
Ammonium, $\mu\text{g g}^{-1}$	0.725	0.271	0.39	0.488	2.345	0.277	0.1	0.494
(+/- SE)	(0.383)	(0.12)	(0.126)	(0.091)	(1.397)	(0.124)	(0)	(0.131)
Nitrate-Nitrite, $\mu\text{g g}^{-1}$	0.316	0.261	0.481	0.245	0.258	0.314	0.336	0.282
(+/- SE)	(0.035)	(0.05)	(0.142)	(0.055)	(0.034)	(0.032)	(0.039)	(0.026)
Phosphorus, $\mu\text{g g}^{-1}$	0.204	0.239	0.28	0.166	0.234	0.168	0.3260	0.257
(+/- SE)	(0.013)	(0.029)	(0.647)	(0.056)	(0.044)	(0.057)	(0.083)	(0.05)

Table A1.17 The effect of fertilizer regime and humic acid amendment on *Spartina patens* soil pH, conductivity, moisture, organic matter, ammonium, nitrate-nitrite, and phosphorus (mean +/- SE, n = 4). pH LSD = 0.69, conductivity LSD = 56.53, moisture LSD = 4.19, organic matter LSD = 0.43, ammonium LSD = 0.941, nitrate-nitrite LSD = 0.329, phosphorus LSD = 1.012

Metric	0 ml m ⁻²		125 ml m ⁻²		250 ml m ⁻²		500 ml m ⁻²	
	Low Fertilizer	High Fertilizer	Low Fertilizer	High Fertilizer	Low Fertilizer	High Fertilizer	Low Fertilizer	High Fertilizer
pH	6.51	7.14	7.19	6.62	7.47	7.33	6.93	7.24
(+/- SE)	(0.10)	(0.36)	(0.35)	(0.22)	(0.58)	(0.24)	(0.21)	(0.06)
Conductivity, $\mu\text{S cm}^{-1}$	92.58	44.33	64.03	84.65	67.83	91.23	77.03	49.98
(+/- SE)	(38.13)	(9.30)	(14.98)	(26.77)	(4.11)	(28.75)	(27.50)	(7.56)
Moisture, %	7.98	4.36	7.44	3.72	8.26	5.09	8.23	6.32
(+/- SE)	(0.86)	(0.65)	(1.05)	(0.80)	(1.11)	(0.69)	(0.32)	(0.63)
Organic matter, %	0.90	0.34	0.49	0.38	0.34	0.42	0.44	0.47
(+/- SE)	(0.54)	(0.02)	(0.03)	(0.02)	(0.05)	(0.03)	(0.10)	(0.03)
Ammonium, $\mu\text{g g}^{-1}$	0.555	0.611	0.454	0.771	0.554	0.982	0.454	1.096
(+/- SE)	(0.129)	(0.281)	(0.132)	(0.361)	(0.194)	(0.319)	(0.208)	(0.331)
Nitrate-Nitrite, $\mu\text{g g}^{-1}$	0.223	0.224	0.308	0.265	0.429	0.235	0.224	0.256
(+/- SE)	(0.026)	(0.025)	(0.062)	(0.072)	(0.217)	(0.068)	(0.087)	(0.082)
Phosphorus, $\mu\text{g g}^{-1}$	0.500	0.110	0.378	0.315	1.123	0.797	0.572	0.245
(+/- SE)	(0.172)	(0.031)	(0.006)	(0.130)	(0.251)	(0.251)	(0.224)	(0.037)

Table A1.18 The effect of fertilizer regime and humic acid amendment on *Baccharis halimifolia* soil pH, conductivity, moisture, organic matter, ammonium, nitrate-nitrite, and phosphorus (mean +/- SE, n = 4). pH LSD = 0.69, conductivity LSD = 56.53, moisture LSD = 4.19, organic matter LSD = 0.43, ammonium LSD = 0.941, nitrate-nitrite LSD = 0.329, phosphorus LSD = 1.012

Metric	0 ml m ⁻²		125 ml m ⁻²		250 ml m ⁻²		500 ml m ⁻²	
	Low Fertilizer	High Fertilizer	Low Fertilizer	High Fertilizer	Low Fertilizer	High Fertilizer	Low Fertilizer	High Fertilizer
pH	6.95	6.28	6.91	6.77	6.56	6.72	6.75	6.55
(+/- SE)	(0.25)	(0.16)	(0.17)	(0.26)	(0.08)	(0.15)	(0.17)	(0.09)
Conductivity, $\mu\text{S cm}^{-1}$	63.50	100.18	57.43	44.53	75.85	96.83	95.38	114.25
(+/- SE)	(19.63)	(9.46)	(13.74)	(8.95)	(35.81)	(32.34)	(10.52)	(17.17)
Moisture, %	12.89	7.72	15.97	10.23	15.14	9.73	13.92	11.73
(+/- SE)	(2.47)	(2.60)	(1.05)	(3.26)	(1.72)	(2.43)	(1.56)	(2.34)
Organic matter, %	0.35	0.45	0.36	0.38	0.47	0.30	0.36	0.43
(+/- SE)	(0.103)	(0.047)	(0.029)	(0.085)	(0.083)	(0.088)	(0.030)	(0.036)
Ammonium, $\mu\text{g g}^{-1}$	0.279	0.373	0.251	0.494	0.287	0.685	0.665	0.356
(+/- SE)	(0.054)	(0.126)	(0.119)	(0.182)	(0.067)	(0.388)	(0.409)	(0.137)
Nitrate-Nitrite, $\mu\text{g g}^{-1}$	0.268	0.319	0.224	0.283	0.35	0.239	0.374	0.213
(+/- SE)	(0.044)	(0.09)	(0.046)	(0.053)	(0.047)	(0.035)	(0.08)	(0.052)
Phosphorus, $\mu\text{g g}^{-1}$	0.201	0.329	0.187	0.496	0.302	0.12	0.245	0.3
(+/- SE)	(0.061)	(0.099)	(0.026)	(0.472)	(0.138)	(0.048)	(0.109)	(0.108)

Table A1.19 The effect of fertilizer regime and humic acid amendment on *Spartina alterniflora* soil pH, conductivity, moisture, organic matter, ammonium, nitrate-nitrite, and phosphorus (mean +/- SE, n = 4). pH LSD = 0.55, conductivity LSD = 2305.3, moisture LSD = 3.41, organic matter LSD = 0.24, ammonium LSD = 0.789, nitrate-nitrite LSD = 0.215, phosphorus LSD = .9130

Metric	0 ml m ⁻²		125 ml m ⁻²		250 ml m ⁻²		500 ml m ⁻²	
	Low Fertilizer	High Fertilizer	Low Fertilizer	High Fertilizer	Low Fertilizer	High Fertilizer	Low Fertilizer	High Fertilizer
pH	6.41	6.81	6.38	6.65	6.40	6.52	6.55	6.84
(+/- SE)	(0.12)	(0.22)	(0.10)	(0.10)	(0.14)	(0.14)	(0.24)	(0.21)
Conductivity, $\mu\text{S cm}^{-1}$	3.43	2.59	3.66	2.99	4.07	4.20	3.42	3.32
(+/- SE)	(1.30)	(0.09)	(0.84)	(0.32)	(0.99)	(1.23)	(0.81)	(1.16)
Moisture, %	20.90	20.38	22.25	22.08	21.33	22.23	20.74	21.96
(+/- SE)	(0.40)	(0.94)	(0.72)	(0.17)	(0.14)	(0.39)	(0.16)	(0.50)
Organic matter, %	0.40	0.47	0.77	0.47	0.72	0.55	0.44	0.79
(+/- SE)	(0.05)	(0.02)	(0.15)	(0.10)	(0.12)	(0.00)	(0.05)	(0.17)
Ammonium, $\mu\text{g g}^{-1}$	0.845	0.387	0.535	0.695	0.437	0.79	0.775	0.94
(+/- SE)	(0.248)	(0.101)	(0.17)	(0.176)	(0.162)	(0.047)	(0.113)	(0.327)
Nitrate-Nitrite, $\mu\text{g g}^{-1}$	0.308	0.09	0.085	0.084	0.086	0.1	0.155	0.101
(+/- SE)	(0.134)	(0.006)	(0.003)	(0.016)	(0.016)	(0.006)	(0.059)	(0.009)
Phosphorus, $\mu\text{g g}^{-1}$	0.126	0.225	0.174	0.103	0.212	0.099	0.163	0.093
(+/- SE)	(0.041)	(0.083)	(0.048)	(0.024)	(0.017)	(0.039)	(0.062)	(0.0140)

Table A1.20 The effect of fertilizer regime and humic acid amendment on *Avicennia germinans* soil pH, conductivity, moisture, organic matter, ammonium, nitrate-nitrite, and phosphorus (mean +/- SE, n = 4). pH LSD = 0.55, conductivity LSD = 2305.3, moisture LSD = 3.41, organic matter LSD = 0.24, ammonium LSD = 0.789, nitrate-nitrite LSD = 0.215, phosphorus LSD = .9130

Metric	0 ml m ⁻²		125 ml m ⁻²		250 ml m ⁻²		500 ml m ⁻²	
	Low Fertilizer	High Fertilizer	Low Fertilizer	High Fertilizer	Low Fertilizer	High Fertilizer	Low Fertilizer	High Fertilizer
pH	6.56	6.40	6.68	6.59	6.25	6.47	6.58	6.66
(+/- SE)	(0.17)	(0.30)	(0.22)	(0.12)	(0.28)	(0.23)	(0.12)	(0.21)
Conductivity, $\mu\text{S cm}^{-1}$	2.64	3.02	2.04	3.91	2.95	2.81	4.66	4.94
(+/- SE)	(0.30)	(0.43)	(0.17)	(0.76)	(0.24)	(0.27)	(1.18)	(0.52)
Moisture, %	19.97	20.66	19.35	20.00	19.86	19.80	19.94	21.88
(+/- SE)	(0.37)	(0.33)	(0.34)	(0.32)	(0.10)	(0.53)	(0.20)	(1.21)
Organic matter, %	0.52	0.46	0.50	0.53	0.51	0.57	0.60	0.50
(+/- SE)	(0.07)	(0.02)	(0.04)	(0.07)	(0.02)	(0.11)	(0.03)	(0.10)
Ammonium, $\mu\text{g g}^{-1}$	0.439	0.457	0.457	0.501	0.422	0.451	0.469	0.441
(+/- SE)	(0.041)	(0.059)	(0.030)	(0.081)	(0.054)	(0.048)	(0.058)	(0.073)
Nitrate-Nitrite, $\mu\text{g g}^{-1}$	0.142	0.141	0.141	0.137	0.144	0.141	0.14	0.142
(+/- SE)	(0.003)	(0.005)	(0.002)	(0.007)	(0.004)	(0.004)	(0.005)	(0.006)
Phosphorus, $\mu\text{g g}^{-1}$	0.115	0.235	0.156	0.205	0.226	0.244	0.158	0.162
(+/- SE)	(0.057)	(0.051)	(0.057)	(0.037)	(0.061)	(0.059)	(0.068)	(0.042)

Table A5.1 The effect of planting density, fertilizer regime, and humic acid amendment on summer 2010 and fall 2010 *U. paniculata* tissue nitrogen, phosphorus, and potassium (mean +/- SE, n = 5; Su 2010 nitrogen LSD = 0.24, phosphorus LSD = 5.36, potassium LSD = 45.88; Fa 2010 nitrogen LSD = 0.20, phosphorus LSD = 2.51, potassium LSD = 22.27).

<i>Uniola paniculata</i>	Low Density						High Density					
	Ambient			Fertilized			Ambient			Fertilized		
Summer 2010												
Metric	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²
Nitrogen	1.16	1.12	1.11	1.31	1.19	1.20	0.91	1.19	1.12	1.22	0.98	1.19
%; ± SE	(0.02)	(0.13)	(0.04)	(0.06)	(0.04)	(0.11)	(0.10)	(0.09)	(0.10)	(0.10)	(0.09)	(0.09)
Phosphorus	604.2	509	641.2	468.7	445.6	380.7	581.2	488.3	447.1	502.5	631.1	566.0
µg g ⁻¹ ; ± SE	(126.1)	(135.7)	(122.8)	(179.7)	(121.6)	(123.1)	(162.8)	(179.0)	(139.1)	(160.9)	(138.4)	(142.1)
Potassium	6693.2	4814.6	4582.6	4236.4	4993.2	3370.8	4198.2	4395	4304.8	5446.8	5014.8	6829.6
µg g ⁻¹ ; ± SE	(2302.1)	(953.8)	(525.0)	(1845.8)	(2015.4)	(868.0)	(1896.2)	(1992.4)	(1876.1)	(2222.0)	(1468.1)	(1864.7)
Fall 2010												
Nitrogen	0.78	1.15	0.76	1.00	0.92	0.94	0.74	0.97	0.79	1.02	0.79	0.89
%; ± SE	(0.04)	(0.08)	(0.02)	(0.10)	(0.05)	(0.09)	(0.04)	(0.07)	(0.07)	(0.11)	(0.09)	(0.11)
Phosphorus	743.4	982.1	908.0	758.9	671.2	770.3	796.4	926	773.6	719.1	781.4	715
µg g ⁻¹ ; ± SE	(43.7)	(110.6)	(59.2)	(76.5)	(47.4)	(59.7)	(62.9)	(117.0)	(69.3)	(71.8)	(16.5)	(90.1)
Potassium	6634.6	6040.2	5312.8	7526.4	6461	5635.8	6355.3	5821.1	5856.2	6843.8	6386.1	6884.0
µg g ⁻¹ ; ± SE	(1731.9)	(453.3)	(362.8)	(498.1)	(1285.7)	(723.3)	(906.7)	(1218.5)	(1081.1)	(1465.3)	(803.6)	(1185.8)

Table A5.2 The effect of planting density, fertilizer regime, and humic acid amendment on spring 2011 and fall 2011 *U. paniculata* tissue nitrogen, phosphorus, and potassium (mean \pm SE, n = 5; Sp 2011 nitrogen LSD = 0.18, phosphorus LSD = 2.36, potassium LSD = 25.88; Fa 2011 nitrogen LSD = 0.25, phosphorus LSD = 6.32, potassium LSD = 14.14).

<i>Uniola paniculata</i>		Low Density						High Density				
		Ambient		Fertilized				Ambient		Fertilized		
Spring 2011												
Metric	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²
Nitrogen	0.80	0.94	0.82	0.96	0.80	0.95	0.89	0.91	0.87	0.98	0.86	0.96
%; ± SE	(0.03)	(0.04)	(0.03)	(0.06)	(0.04)	(0.05)	(0.04)	(0.03)	(0.02)	(0.05)	(0.02)	(0.03)
Phosphorus	642.3	566.6	698.9	660.8	596.5	636.3	504.2	593	487.4	616.6	582.3	498
µg g ⁻¹ ; ± SE	(57.4)	(131.8)	(66.0)	(77.8)	(37.2)	(43.3)	(117.2)	(160.3)	(125.6)	(48.5)	(44.5)	(59.0)
Potassium	6889.0	5529.4	7937.2	8991.6	6982.6	8333.2	5768.0	6065.6	5485.4	9224	8107.4	6583.8
µg g ⁻¹ ; ± SE	(1143.5)	(1168.5)	(694.2)	(1452.8)	(1036.5)	(730.4)	(1409.3)	(1627.8)	(1831.8)	(1616.5)	(1129.8)	(1671.7)
Fall 2011												
Nitrogen	0.89	0.84	0.89	0.87	0.83	0.89	0.84	0.83	0.88	0.83	0.77	0.82
%; ± SE	(0.12)	(0.10)	(0.17)	(0.11)	(0.15)	(0.09)	(0.15)	(0.10)	(0.10)	(0.12)	(0.10)	(0.06)
Phosphorus	1852.3	1666.6	1644.1	1909.3	1774.7	1852.4	2002.2	1916.8	2519.8	1923.8	1685.2	2674.1
µg g ⁻¹ ; ± SE	(225.9)	(242.5)	(512.30)	(241.1)	(310.9)	(427.4)	(216.7)	(330.7)	(268.2)	(297.9)	(227.4)	(365.0)
Potassium	5272.4	4999.2	3464.4	4453.2	4137.2	4010.6	4702.8	4885.6	3994	4225.8	4709.4	4164.4
µg g ⁻¹ ; ± SE	(673.8)	(401.0)	(949.7)	(527.0)	(331.6)	(300.7)	(530.4)	(725)	(243.7)	(364.9)	(460.2)	(428.3)

Table A5.3 The effect of planting density, fertilizer regime, and humic acid amendment on summer 2010 and fall 2010 *P. amarum* tissue nitrogen, phosphorus, and potassium (mean \pm SE, n = 5; Su 2010 nitrogen LSD = 0.24, phosphorus LSD = 5.36, potassium LSD = 45.88; Fa 2010 nitrogen LSD = 0.20, phosphorus LSD = 2.51, potassium LSD = 22.27)

Panicum amarum	Low Density						High Density					
	Ambient			Fertilized			Ambient			Fertilized		
Summer 2010												
Metric	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²
Nitrogen	1.35	1.43	1.37	1.39	1.31	1.43	1.28	1.31	1.33	1.42	1.29	1.33
%; ± SE	(0.05)	(0.07)	(0.03)	(0.07)	(0.07)	(0.02)	(0.04)	(0.06)	(0.05)	(0.08)	(0.11)	(0.11)
Phosphorus	14.87	18.32	10.04	14.08	16.46	12.9	11.5	12.57	11.5	15.16	13.09	13.26
µg g ⁻¹ ; ± SE	(2.54)	(3.29)	(2.69)	(3.49)	(3.34)	(2.39)	(2.01)	(2.49)	(0.77)	(2.21)	(2.71)	(3.1)
Potassium	121.48	165.04	140.22	130.75	119.02	132.56	148.84	124.18	179.17	173.93	107.65	125.88
µg g ⁻¹ ; ± SE	(9.34)	(20.96)	(24.56)	(23.68)	(4.84)	(19.4)	(22.71)	(19.31)	(25.36)	(31.81)	(25)	(17.33)
Fall 2010												
Nitrogen	0.97	1.00	0.96	0.96	1.01	1.02	0.97	1.05	0.99	1.08	0.92	1.01
%; ± SE	(0.03)	(0.06)	(0.03)	(0.05)	(0.07)	(0.04)	(0.04)	(0.03)	(0.07)	(0.07)	(0.09)	(0.08)
Phosphorus	11.17	12.12	11.66	11.92	11.22	10.29	11.45	11.29	10.97	10.15	10.09	8.78
µg g ⁻¹ ; ± SE	(0.92)	(0.58)	(0.76)	(0.77)	(0.83)	(0.62)	(0.73)	(1.19)	(1.06)	(0.33)	(0.66)	(1.26)
Potassium	94.93	95.35	99.39	99.98	97.25	88.57	95.4	99.75	91.85	93.62	91.85	90.8
µg g ⁻¹ ; ± SE	(3.9)	(3.17)	(3.52)	(3.98)	(4.31)	(10.37)	(4.15)	(3.46)	(4.01)	(3.06)	(4.3)	(1.86)

Table A5.4 The effect of planting density, fertilizer regime, and humic acid amendment on spring 2011 and fall 2011 *P. amarum* tissue nitrogen, phosphorus, and potassium (mean \pm SE, n = 5; Sp 2011 nitrogen LSD = 0.18, phosphorus LSD = 2.36, potassium LSD = 25.88; Fa 2011 nitrogen LSD = 0.25, phosphorus LSD = 6.32, potassium LSD = 14.14).

Panicum amarum	Low Density						High Density					
	Ambient			Fertilized			Ambient			Fertilized		
Spring 2011												
Metric	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²
Nitrogen	1.21	1.33	1.30	1.28	1.23	1.23	1.22	1.31	1.22	1.26	1.18	1.25
%; ± SE	(0.04)	(0.04)	(0.02)	(0.04)	(0.03)	(0.03)	(0.04)	(0.04)	(0.04)	(0.03)	(0.04)	(0.07)
Phosphorus	2.61	1.95	1.07	2.44	4.07	2.53	2.34	1.2	2.05	1.5	2.01	2.69
µg g ⁻¹ ; ± SE	(0.98)	(0.63)	(0.3)	(0.78)	(0.77)	(0.9)	(1.07)	(0.57)	(0.73)	(0.27)	(0.51)	(0.62)
Potassium	19.16	16.39	12.39	21.72	19.51	33.96	15.98	26.21	13.62	37.84	7.84	28.52
µg g ⁻¹ ; ± SE	(8.11)	(3.28)	(6.24)	(4.35)	(4.89)	(9.2)	(6.03)	(6.69)	(5.54)	(9.4)	(1.55)	(13.09)
Fall 2011												
Nitrogen	0.75	0.78	0.83	0.73	0.80	0.61	0.79	0.71	0.74	0.64	0.78	0.65
%; ± SE	(0.04)	(0.14)	(0.06)	(0.07)	(0.08)	(0.04)	(0.03)	(0.04)	(0.03)	(0.04)	(0.06)	(0.06)
Phosphorus	7.04	7.35	6.31	7.36	6.67	6.76	6.28	5.74	6.89	5.71	6.3	5.47
µg g ⁻¹ ; ± SE	(0.52)	(0.37)	(0.64)	(1.22)	(1.74)	(1.01)	(0.61)	(0.81)	(1.16)	(0.55)	(0.73)	(0.82)
Potassium	61.66	69.17	60.61	57.98	58.39	58.72	55.93	58.43	57.46	59.4	61.31	60.52
µg g ⁻¹ ; ± SE	(3.04)	(2.45)	(2.95)	(0.76)	(2.27)	(2.38)	(1.3)	(2.95)	(0.53)	(2.24)	(3.05)	(3.15)

Table A5.5 The effect of planting density, fertilizer regime, and humic acid amendment on summer 2010 and fall 2010 *S. patens* tissue nitrogen, phosphorus, and potassium (mean \pm SE, n = 5; Su 2010 nitrogen LSD = 0.24, phosphorus LSD = 5.36, potassium LSD = 45.88; Fa 2010 nitrogen LSD = 0.20, phosphorus LSD = 2.51, potassium LSD = 22.27)

<i>Spartina patens</i>	Low Density						High Density					
	Ambient			Fertilized			Ambient			Fertilized		
Summer 2010												
Metric	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²
Nitrogen	1.39	1.51	1.50	1.42	1.40	1.33	1.44	1.42	1.35	1.63	1.45	1.54
%; \pm SE	(0.07)	(0.14)	(0.09)	(0.10)	(0.07)	(0.04)	(0.11)	(0.08)	(0.11)	(0.10)	(0.08)	(0.11)
Phosphorus	9.4	8	7.39	8.85	9.21	7.48	7.15	6.35	8.23	8.61	9.52	7.44
$\mu\text{g g}^{-1}$; \pm SE	(2.28)	(1.88)	(1.74)	(1.13)	(1.12)	(0.93)	(0.99)	(0.68)	(0.38)	(0.43)	(1.21)	(0.73)
Potassium	45.26	43.95	49.04	67.96	40.9	61.47	52.4	43.79	41.83	46.86	40.26	53.45
$\mu\text{g g}^{-1}$; \pm SE	(2.67)	(5.64)	(1.98)	(13.07)	(5.61)	(4.31)	(8.77)	(6.43)	(4.25)	(6.61)	(4.98)	(5.56)
Fall 2010												
Nitrogen	1.14	1.11	1.17	1.03	1.09	1.00	1.37	1.17	1.37	1.34	1.26	1.21
%; \pm SE	(0.11)	(0.04)	(0.08)	(0.05)	(0.10)	(0.06)	(0.08)	(0.10)	(0.07)	(0.12)	(0.10)	(0.06)
Phosphorus	5.08	6.36	5.23	4.27	4.96	4.88	3	3.35	3.66	3.75	1.81	1.82
$\mu\text{g g}^{-1}$; \pm SE	(1.07)	(1.24)	(1.42)	(1.37)	(1.59)	(1.25)	(0.52)	(0.52)	(1.03)	(1.05)	(0.34)	(0.24)
Potassium	20.39	27.59	20.3	21.34	17.91	18.52	32.76	40.19	44.61	35.23	34.91	39.51
$\mu\text{g g}^{-1}$; \pm SE	(5.21)	(8.72)	(5.5)	(3)	(2.94)	(2.28)	(6.16)	(13.3)	(11.32)	(9.72)	(6.68)	(6.76)

Table A5.6 The effect of planting density, fertilizer regime, and humic acid amendment on spring 2011 and fall 2011 *S. patens* tissue nitrogen, phosphorus, and potassium (mean \pm SE, n = 5; Sp 2011 nitrogen LSD = 0.18, phosphorus LSD = 2.36, potassium LSD = 25.88; Fa 2011 nitrogen LSD = 0.25, phosphorus LSD = 6.32, potassium LSD = 14.14).

<i>Spartina patens</i>	Low Density						High Density					
	Ambient			Fertilized			Ambient			Fertilized		
Spring 2011												
Metric	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²
Nitrogen	1.22	0.91	1.23	1.00	1.17	0.98	1.06	0.88	1.05	0.88	0.95	0.85
%; ± SE	(0.13)	(0.11)	(0.12)	(0.13)	(0.10)	(0.12)	(0.07)	(0.05)	(0.05)	(0.10)	(0.08)	(0.03)
Phosphorus	7.99	11.52	10.81	5.46	5.24	5.82	6.2	6.32	6.27	5.47	5.61	4.87
µg g ⁻¹ ; ± SE	(1.87)	(1.18)	(0.24)	(0.66)	(0.38)	(0.54)	(1.69)	(0.5)	(0.53)	(0.27)	(0.34)	(0.36)
Potassium	35.47	49.58	52.58	56.19	48.56	51.69	46.14	43.7	47.94	55.12	45.69	47.98
µg g ⁻¹ ; ± SE	(8.63)	(2.57)	(3.92)	(2.95)	(2.2)	(3.9)	(10.51)	(5.63)	(3.09)	(4.07)	(3.54)	(2.74)
Fall 2011												
Nitrogen	0.84	0.73	0.96	0.73	0.78	0.69	0.74	0.67	0.72	0.73	0.75	0.63
%; ± SE	(0.14)	(0.03)	(0.09)	(0.02)	(0.14)	(0.05)	(0.04)	(0.04)	(0.02)	(0.09)	(0.08)	(0.06)
Phosphorus	6.78	8.87	6.43	6.36	7.32	9.21	10.49	11.17	6.13	5.07	4.19	6.87
µg g ⁻¹ ; ± SE	(1.57)	(3.97)	(2.81)	(2.08)	(1.7)	(2.86)	(2.21)	(2.17)	(2.64)	(2.37)	(1.51)	(1.32)
Potassium	34.42	24.6	34.05	27.1	21.69	35.64	24.45	29.75	35.62	34.48	23.45	36.3
µg g ⁻¹ ; ± SE	(2.85)	(7.59)	(4.71)	(10.47)	(5.84)	(6.82)	(6.68)	(6.17)	(10.8)	(4.78)	(6.91)	(5.59)

Table A5.7 The effect of planting density, fertilizer regime, and humic acid amendment on Spring 2010 *Uniola paniculata* soil pH, conductivity, moisture, organic matter, ammonium, nitrate-nitrite, and phosphorus (mean +/- SE, n = 5; Sp 2010 pH LSD = 0.73, conductivity LSD = 1277.2, moisture LSD = 4.61, organic matter LSD = 1.21, ammonium LSD = 2.083, nitrate-nitrite LSD = 0.218, phosphorus LSD = 0.333)..

Spring 2010 Metric	Low Density						High Density					
	Ambient			Fertilized			Ambient			Fertilized		
	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²
pH	8.83	9.02	8.88	8.84	9.08	8.55	8.81	8.9	8.89	8.74	8.9	8.61
± SE	(0.14)	(0.08)	(0.09)	(0.16)	(0.08)	(0.22)	(0.07)	(0.08)	(0.12)	(0.11)	(0.14)	(0.14)
Conductivity	141.28	194.40	171.32	136.15	152.20	176.98	119.50	138.92	184.90	146.46	153.98	151.58
µS cm ⁻¹ ; ± SE	(8.34)	(29.55)	(16.93)	(20.71)	(11.30)	(34.07)	(20.96)	(30.83)	(32.80)	(23.38)	(14.01)	(17.70)
Moisture	3.37	5.39	4.90	4.81	5.41	4.11	2.56	3.06	2.18	2.81	2.50	2.56
%; ± SE	(0.89)	(0.51)	(1.52)	(1.08)	(0.42)	(0.56)	(0.83)	(1.37)	(0.56)	(0.87)	(1.11)	(0.86)
Organic Matter	0.74	0.93	0.79	0.72	0.78	0.64	0.71	0.79	0.76	0.66	0.84	0.72
%; ± SE	(0.022)	(0.1)	(0.085)	(0.034)	(0.069)	(0.08)	(0.099)	(0.057)	(0.068)	(0.08)	(0.064)	(0.04)
Ammonium	1.099	1.656	2.602	0.833	2.041	1.14	0.994	1.23	1.36	0.969	1.195	0.951
µg g ⁻¹ ; ± SE	(0.554)	(0.688)	(0.521)	(0.401)	(0.632)	(0.172)	(0.143)	(0.893)	(0.641)	(0.178)	(0.219)	(0.139)
Nitrate-Nitrite	0.197	0.528	0.238	0.138	0.207	0.263	0.194	0.279	0.181	0.404	0.256	0.496
µg g ⁻¹ ; ± SE	(0.025)	(0.248)	(0.043)	(0.063)	(0.036)	(0.039)	(0.038)	(0.116)	(0.02)	(0.169)	(0.027)	(0.143)
Phosphorus	1.404	1.193	1.301	0.836	1.264	1.383	1.454	1.344	1.624	1.654	1.748	1.147
µg g ⁻¹ ; ± SE	(0.311)	(0.136)	(0.183)	(0.328)	(0.319)	(0.256)	(0.200)	(0.339)	(0.131)	(0.123)	(0.060)	(0.179)

Table A5.8 The effect of planting density, fertilizer regime, and humic acid amendment on Summer 2010 *Uniola paniculata* soil pH, conductivity, moisture, organic matter, ammonium, nitrate-nitrite, and phosphorus (mean +/- SE, n = 5; Su 2010 pH LSD = 0.59, conductivity LSD = 1889.3, moisture LSD = 8.04, organic matter LSD = 0.46, ammonium LSD = 5.226, nitrate-nitrite LSD = 0.218, phosphorus LSD = 0.688).

Summer 2010 Metric	Low Density						High Density					
	Ambient			Fertilized			Ambient			Fertilized		
	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²
pH	8.41	8.61	8.45	8.52	8.64	8.4	8.11	8.41	8.35	8.23	8.26	8.27
± SE	(0.32)	(0.21)	(0.16)	(0.24)	(0.21)	(0.14)	(0.46)	(0.25)	(0.26)	(0.27)	(0.24)	(0.29)
Conductivity	118.55	364.26	183.62	202.17	191.66	210.83	147.33	179.62	186.81	145.46	142.52	194.02
µS cm ⁻¹ ; ± SE	(30.61)	(143.15)	(54.11)	(67.91)	(53.42)	(93.27)	(24.72)	(57.43)	(60.45)	(23.67)	(24.87)	(60.95)
Moisture	4.09	5.93	4.15	6.56	5.69	7.83	4.71	7.53	16.96	3.92	5.12	4.68
%; ± SE	(0.68)	(2.51)	(1.37)	(2.18)	(2.17)	(1.52)	(1.69)	(2.89)	(11.95)	(0.86)	(1.40)	(2.38)
Organic Matter	0.59	1.04	0.65	0.79	1.2	0.75	0.57	0.86	0.92	0.66	0.67	0.65
%; ± SE	(0.139)	(0.13)	(0.062)	(0.136)	(0.342)	(0.125)	(0.112)	(0.072)	(0.109)	(0.063)	(0.122)	(0.074)
Ammonium	1.494	1.34	2.237	6.295	9.17	1.413	0.766	1.119	1.506	1.607	1.646	10.401
µg g ⁻¹ ; ± SE	(0.424)	(0.397)	(1.194)	(5.275)	(7.935)	(0.644)	(0.274)	(0.544)	(0.19)	(0.448)	(0.39)	(8.373)
Nitrate-Nitrite	0.186	0.219	0.16	0.259	0.596	0.116	0.236	0.12	0.464	0.291	0.175	0.607
µg g ⁻¹ ; ± SE	(0.062)	(0.021)	(0.026)	(0.139)	(0.325)	(0.048)	(0.142)	(0.06)	(0.313)	(0.095)	(0.025)	(0.443)
Phosphorus	1.000	1.043	0.886	0.892	2.720	0.577	0.588	1.244	0.649	1.213	0.814	0.692
µg g ⁻¹ ; ± SE	(0.286)	(0.308)	(0.283)	(0.347)	(0.385)	(0.346)	(0.339)	(0.190)	(0.305)	(0.226)	(0.323)	(0.293)

Table A5.9 The effect of planting density, fertilizer regime, and humic acid amendment on Fall 2010 *Uniola paniculata* soil pH, conductivity, moisture, organic matter, ammonium, nitrate-nitrite, and phosphorus (mean +/- SE, n = 5; Fa 2010 pH LSD = 0.71, conductivity LSD = 1913.2, moisture LSD = 6.52, organic matter LSD = 0.964, ammonium LSD = 1.50, nitrate-nitrite LSD = 0.197, phosphorus LSD = 0.303).

Fall 2010 Metric	Low Density						High Density					
	Ambient			Fertilized			Ambient			Fertilized		
	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²
pH	7.5	7.67	7.41	7.12	7.34	7.28	7.51	7.53	7.64	7.21	7.26	7.43
± SE	(0.13)	(0.27)	(0.22)	(0.19)	(0.17)	(0.16)	(0.17)	(0.11)	(0.27)	(0.16)	(0.09)	(0.29)
Conductivity	212.10	157.24	217.16	232.56	258.42	166.12	187.56	138.14	204.14	167.60	182.36	189.84
µS cm ⁻¹ ; ± SE	(92.43)	(19.43)	(83.63)	(53.60)	(59.49)	(19.55)	(44.79)	(5.51)	(38.16)	(35.10)	(25.40)	(35.05)
Moisture	4.39	2.28	4.33	5.10	6.50	2.96	8.92	7.10	5.84	5.07	2.53	6.71
%; ± SE	(2.48)	(0.99)	(2.03)	(2.83)	(3.80)	(1.43)	(3.73)	(3.03)	(2.55)	(1.81)	(1.92)	(3.09)
Organic Matter	0.73	0.78	0.73	0.86	0.8	0.72	1.02	0.71	1	0.78	0.72	0.82
%; ± SE	(0.051)	(0.039)	(0.101)	(0.062)	(0.189)	(0.079)	(0.237)	(0.156)	(0.097)	(0.085)	(0.102)	(0.09)
Ammonium	0.615	0.574	0.432	0.632	0.421	0.294	0.862	0.564	0.515	0.6	0.389	0.67
µg g ⁻¹ ; ± SE	(0.244)	(0.101)	(0.25)	(0.199)	(0.261)	(0.114)	(0.565)	(0.27)	(0.129)	(0.167)	(0.152)	(0.315)
Nitrate-Nitrite	0.203	0.252	0.337	0.265	0.12	0.139	0.385	0.107	0.213	0.176	0.505	0.123
µg g ⁻¹ ; ± SE	(0.061)	(0.07)	(0.144)	(0.094)	(0.073)	(0.069)	(0.113)	(0.043)	(0.066)	(0.046)	(0.338)	(0.053)
Phosphorus	2.461	2.387	2.259	2.451	2.191	1.991	2.541	2.528	2.508	2.568	2.616	2.412
µg g ⁻¹ ; ± SE	(0.078)	(0.133)	(0.146)	(0.092)	(0.275)	(0.261)	(0.048)	(0.051)	(0.064)	(0.016)	(0.021)	(0.143)

Table A5.10 The effect of planting density, fertilizer regime, and humic acid amendment on Spring 2011 *Uniola paniculata* soil pH, conductivity, moisture, organic matter, ammonium, nitrate-nitrite, and phosphorus (mean \pm SE, n = 5; Sp 2011 pH LSD = 0.64, conductivity LSD = 578.06, moisture LSD = 4.12, organic matter LSD = .44, ammonium LSD = 1.118, nitrate-nitrite LSD = 0.222, phosphorus LSD = 0.171).

Spring 2011 Metric	Low Density						High Density					
	Ambient			Fertilized			Ambient			Fertilized		
	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²
pH	8.63	8.66	8.57	8.26	8.39	8.52	8.78	8.6	8.97	8.66	8.5	8.62
\pm SE	(0.21)	(0.28)	(0.27)	(0.23)	(0.22)	(0.32)	(0.37)	(0.3)	(0.32)	(0.27)	(0.23)	(0.28)
Conductivity	212.98	204.84	88.98	374.20	476.96	108.44	184.94	89.48	75.32	223.46	198.56	191.90
μ S cm ⁻¹ ; \pm SE	(146.84)	(114.99)	(15.65)	(282.79)	(392.35)	(16.80)	(101.04)	(19.29)	(6.59)	(128.02)	(79.15)	(108.73)
Moisture	0.52	0.70	1.04	0.72	0.76	0.89	0.85	0.75	0.73	0.50	0.52	1.15
%; \pm SE	(0.05)	(0.09)	(0.34)	(0.17)	(0.15)	(0.17)	(0.29)	(0.29)	(0.22)	(0.08)	(0.06)	(0.29)
Organic Matter	0.68	0.76	0.66	0.82	0.93	0.95	0.75	0.77	0.63	0.69	0.67	0.95
%; \pm SE	(0.048)	(0.12)	(0.035)	(0.224)	(0.181)	(0.17)	(0.168)	(0.114)	(0.045)	(0.115)	(0.099)	(0.179)
Ammonium	0.145	0.259	0.403	0.163	0.205	0.37	0.337	0.583	0.636	0.287	0.107	0.412
μ g g ⁻¹ ; \pm SE	(0.039)	(0.159)	(0.261)	(0.051)	(0.028)	(0.236)	(0.189)	(0.429)	(0.34)	(0.144)	(0.007)	(0.207)
Nitrate-Nitrite	0.314	0.47	0.287	0.292	0.373	0.31	0.297	0.333	0.251	0.202	0.315	0.416
μ g g ⁻¹ ; \pm SE	(0.056)	(0.163)	(0.04)	(0.064)	(0.144)	(0.095)	(0.079)	(0.047)	(0.034)	(0.036)	(0.042)	(0.163)
Phosphorus	0.129	0.128	0.103	0.135	0.133	0.084	0.318	0.195	0.351	0.526	0.242	0.238
μ g g ⁻¹ ; \pm SE	(0.129)	(0.128)	(0.103)	(0.135)	(0.133)	(0.084)	(0.214)	(0.123)	(0.145)	(0.085)	(0.148)	(0.133)

Table A5.11 The effect of planting density, fertilizer regime, and humic acid amendment on Fall 2011 *Uniola paniculata* soil pH, conductivity, moisture, organic matter, ammonium, nitrate-nitrite, and phosphorus (mean +/- SE, n = 5; Fa 2011 pH LSD = 0.79, conductivity LSD = 158.31, moisture LSD = 3.72, organic matter LSD = .44, ammonium LSD = 0.482, nitrate-nitrite LSD = 0.223, phosphorus LSD = 0.256).

Fall 2011 Metric	Low Density						High Density					
	Ambient			Fertilized			Ambient			Fertilized		
	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²
pH	7.98	7.52	7.99	7.83	7.75	7.86	7.79	7.89	7.83	7.92	7.77	7.82
± SE	(0.2)	(0.75)	(0.24)	(0.19)	(0.05)	(0.13)	(0.17)	(0.15)	(0.18)	(0.16)	(0.1)	(0.15)
Conductivity	36.58	126.00	29.07	33.84	34.82	51.32	31.18	32.68	26.36	42.38	47.16	39.40
µS cm ⁻¹ ; ± SE	(9.01)	(81.27)	(8.90)	(7.36)	(6.69)	(7.52)	(8.28)	(11.37)	(7.06)	(11.78)	(19.75)	(5.01)
Moisture	4.30	4.59	3.36	5.07	4.83	5.32	3.46	3.93	3.21	3.90	4.47	4.10
%; ± SE	(0.71)	(0.79)	(0.33)	(1.09)	(0.86)	(1.01)	(0.25)	(0.84)	(0.46)	(0.55)	(0.76)	(0.70)
Organic Matter	0.52	0.55	0.42	0.62	0.69	0.64	0.48	0.6	0.47	0.53	0.99	0.67
%; ± SE	(0.109)	(0.114)	(0.097)	(0.167)	(0.176)	(0.133)	(0.07)	(0.212)	(0.096)	(0.124)	(0.375)	(0.104)
Ammonium	0.238	0.265	0.437	0.429	0.996	0.833	0.18	0.222	0.199	0.553	0.577	0.405
µg g ⁻¹ ; ± SE	(0.084)	(0.078)	(0.26)	(0.153)	(0.434)	(0.266)	(0.052)	(0.09)	(0.047)	(0.424)	(0.11)	(0.074)
Nitrate-Nitrite	0.353	0.284	0.641	0.413	0.329	0.649	0.429	0.269	0.405	0.636	0.328	0.726
µg g ⁻¹ ; ± SE	(0.067)	(0.047)	(0.369)	(0.088)	(0.033)	(0.452)	(0.17)	(0.025)	(0.129)	(0.383)	(0.106)	(0.26)
Phosphorus	0.236	0.322	0.210	0.283	0.575	0.486	0.197	0.300	0.241	0.279	0.536	0.460
µg g ⁻¹ ; ± SE	(0.039)	(0.072)	(0.066)	(0.039)	(0.128)	(0.080)	(0.029)	(0.074)	(0.025)	(0.038)	(0.151)	(0.121)

Table A5.12 The effect of planting density, fertilizer regime, and humic acid amendment on Spring 2010 *Panicum amarum* soil pH, conductivity, moisture, organic matter, ammonium, nitrate-nitrite, and phosphorus (mean \pm SE, n = 5; Sp 2010 pH LSD = 0.73, conductivity LSD = 1277.2, moisture LSD = 4.61, organic matter LSD = 1.21, ammonium LSD = 2.083, nitrate-nitrite LSD = 0.218, phosphorus LSD = 0.333).

Spring 2010 Metric	Low Density						High Density					
	Ambient			Fertilized			Ambient			Fertilized		
	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²
pH	8.96	8.89	8.82	9.06	8.88	8.88	8.91	8.90	8.97	8.86	8.66	9.10
\pm SE	(0.12)	(0.27)	(0.06)	(0.15)	(0.18)	(0.05)	(0.08)	(0.08)	(0.16)	(0.10)	(0.23)	(0.15)
Conductivity	77.90	169.44	282.98	131.16	220.52	135.92	213.20	184.40	163.62	122.56	1440.30	165.46
$\mu\text{S cm}^{-1}$; \pm SE	(19.58)	(76.09)	(181.50)	(28.33)	(124.41)	(36.80)	(43.76)	(78.51)	(52.02)	(14.31)	(1315.07)	(34.87)
Moisture	1.86	3.09	3.02	2.44	2.43	3.07	4.07	3.50	2.59	2.84	4.20	2.98
%; \pm SE	(0.37)	(0.77)	(0.71)	(0.41)	(0.88)	(0.99)	(0.71)	(0.71)	(0.74)	(0.44)	(1.02)	(0.46)
Organic Matter	0.640	0.770	0.890	0.720	0.790	0.720	0.820	0.790	0.840	0.750	0.870	0.830
%; \pm SE	(0.065)	(0.087)	(0.109)	(0.080)	(0.202)	(0.115)	(0.077)	(0.086)	(0.090)	(0.059)	(0.113)	(0.151)
Ammonium	0.914	1.719	1.367	0.927	0.886	1.386	1.296	1.242	1.426	0.877	1.372	0.934
$\mu\text{g g}^{-1}$; \pm SE	(0.393)	(0.621)	(0.336)	(0.275)	(0.198)	(0.379)	(0.376)	(0.192)	(0.268)	(0.133)	(0.215)	(0.123)
Nitrate-Nitrite	0.198	0.219	0.526	0.277	0.237	0.200	0.173	0.246	0.163	0.292	0.189	0.578
$\mu\text{g g}^{-1}$; \pm SE	(0.066)	(0.054)	(0.275)	(0.173)	(0.047)	(0.025)	(0.025)	(0.044)	(0.028)	(0.114)	(0.040)	(0.389)
Phosphorus	1.015	0.792	0.831	1.479	0.994	0.729	0.983	0.805	0.659	0.641	1.062	1.170
$\mu\text{g g}^{-1}$; \pm SE	(0.310)	(0.244)	(0.378)	(0.203)	(0.307)	(0.266)	(0.289)	(0.348)	(0.283)	(0.278)	(0.306)	(0.262)

Table A5.13 The effect of planting density, fertilizer regime, and humic acid amendment on Summer 2010 *Panicum amarum* soil pH, conductivity, moisture, organic matter, ammonium, nitrate-nitrite, and phosphorus (mean \pm SE, n = 5; Su 2010 pH LSD = 0.59, conductivity LSD = 1889.3, moisture LSD = 8.04, organic matter LSD = 0.46, ammonium LSD = 5.226, nitrate-nitrite LSD = 0.218, phosphorus LSD = 0.688).

Metric	Low Density						High Density					
	Ambient			Fertilized			Ambient			Fertilized		
Summer 2010	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²
pH	8.96	8.89	8.82	9.06	8.88	8.88	8.91	8.9	8.97	8.86	8.66	9.1
\pm SE	(0.09)	(0.06)	(0.15)	(0.36)	(0.18)	(0.13)	(0.17)	(0.16)	(0.33)	(0.11)	(0.1)	(0.21)
Conductivity	175.72	208.94	186.36	147.37	130.35	201.27	134.18	173.78	483.08	110.46	108.69	169.84
μ S cm ⁻¹ ; \pm SE	(29.41)	(96.57)	(61.14)	(38.11)	(19.94)	(42.77)	(27.50)	(59.44)	(355.77)	(8.24)	(33.50)	(44.14)
Moisture	7.65	11.58	6.46	5.26	6.37	7.39	7.17	7.46	7.58	5.49	7.95	6.46
%; \pm SE	(3.12)	(5.37)	(1.22)	(1.42)	(1.74)	(2.19)	(1.41)	(1.91)	(2.96)	(0.80)	(3.21)	(0.96)
Organic Matter	0.63	0.88	0.85	0.74	0.81	0.84	0.76	0.8	0.89	0.87	1.15	0.99
%; \pm SE	(0.151)	(0.116)	(0.125)	(0.112)	(0.07)	(0.081)	(0.06)	(0.091)	(0.159)	(0.034)	(0.27)	(0.159)
Ammonium	2.136	2.029	1.497	1.909	1.989	1.447	3.035	2.456	1.914	8.557	3.195	1.798
μ g g ⁻¹ ; \pm SE	(0.487)	(0.842)	(0.417)	(0.868)	(0.546)	(0.436)	(1.200)	(0.815)	(0.747)	(5.849)	(1.308)	(0.430)
Nitrate-Nitrite	0.725	0.423	0.397	0.281	0.150	0.204	0.290	0.307	0.501	0.303	0.264	0.704
μ g g ⁻¹ ; \pm SE	(0.330)	(0.147)	(0.048)	(0.092)	(0.056)	(0.067)	(0.025)	(0.048)	(0.190)	(0.028)	(0.045)	(0.338)
Phosphorus	0.417	1.112	0.856	0.788	0.693	0.514	0.999	0.460	0.907	1.286	1.061	1.524
μ g g ⁻¹ ; \pm SE	(0.187)	(0.330)	(0.229)	(0.297)	(0.219)	(0.245)	(0.319)	(0.202)	(0.205)	(0.284)	(0.389)	(0.321)

Table A5.14 The effect of planting density, fertilizer regime, and humic acid amendment on Fall 2010 *Panicum amarum* soil pH, conductivity, moisture, organic matter, ammonium, nitrate-nitrite, and phosphorus (mean \pm SE, n = 5; Fa 2010 pH LSD = 0.71, conductivity LSD = 1913.2, moisture LSD = 6.52, organic matter LSD = 0.964, ammonium LSD = 1.50, nitrate-nitrite LSD = 0.197, phosphorus LSD = 0.303)

Fall 2010 Metric	Low Density						High Density					
	Ambient			Fertilized			Ambient			Fertilized		
	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²
pH	6.6	6.58	6.9	7.11	6.47	6.99	6.5	6.78	6.5	6.56	6.58	6.71
\pm SE	(0.08)	(0.24)	(0.05)	(0.18)	(0.32)	(0.13)	(0.1)	(0.16)	(0.25)	(0.17)	(0.17)	(0.29)
Conductivity	130.88	477.32	947.70	145.74	201.58	1207.40	466.04	282.32	318.84	119.22	3656.92	183.08
μ S cm ⁻¹ ; \pm SE	(16.24)	(300.09)	(806.79)	(12.76)	(53.13)	(1070.66)	(329.69)	(168.71)	(163.59)	(5.93)	(3530.83)	(64.79)
Moisture	9.60	10.79	13.97	9.85	9.59	12.04	21.91	12.28	11.60	10.93	14.10	10.75
%; \pm SE	(1.01)	(2.24)	(2.89)	(0.27)	(0.75)	(3.11)	(6.86)	(2.53)	(1.36)	(1.21)	(4.30)	(1.44)
Organic Matter	0.63	0.76	0.76	0.62	0.7	0.91	0.75	0.72	0.67	0.62	1.02	0.55
%; \pm SE	(0.085)	(0.171)	(0.15)	(0.035)	(0.052)	(0.202)	(0.071)	(0.115)	(0.047)	(0.017)	(0.218)	(0.171)
Ammonium	0.555	0.139	0.205	0.562	0.539	2.429	0.332	0.421	0.434	0.412	0.814	0.126
μ g g ⁻¹ ; \pm SE	(0.228)	(0.054)	(0.050)	(0.191)	(0.309)	(1.453)	(0.142)	(0.097)	(0.194)	(0.127)	(0.550)	(0.024)
Nitrate-Nitrite	0.531	0.146	0.194	0.243	0.496	0.390	0.272	0.407	0.257	0.662	0.216	0.212
μ g g ⁻¹ ; \pm SE	(0.326)	(0.065)	(0.050)	(0.034)	(0.193)	(0.119)	(0.110)	(0.118)	(0.042)	(0.420)	(0.033)	(0.050)
Phosphorus	2.411	2.037	2.524	2.300	1.785	2.396	2.533	2.606	2.479	2.445	2.460	2.670
μ g g ⁻¹ ; \pm SE	(0.152)	(0.514)	(0.115)	(0.163)	(0.330)	(0.183)	(0.038)	(0.032)	(0.106)	(0.166)	(0.085)	(0.041)

Table A5.15 The effect of planting density, fertilizer regime, and humic acid amendment on Spring 2011 *Panicum amarum* soil pH, conductivity, moisture, organic matter, ammonium, nitrate-nitrite, and phosphorus (mean \pm SE, n = 5; Sp 2011 pH LSD = 0.64, conductivity LSD = 578.06, moisture LSD = 4.12, organic matter LSD = .44, ammonium LSD = 1.118, nitrate-nitrite LSD = 0.222, phosphorus LSD = 0.171).

Spring 2011 Metric	Low Density						High Density					
	Ambient			Fertilized			Ambient			Fertilized		
	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²
pH	8.39	8.4	8.39	8.51	8.63	8.41	8.36	8.77	8.45	8.22	8.53	8.49
\pm SE	(0.11)	(0.25)	(0.17)	(0.11)	(0.3)	(0.12)	(0.22)	(0.21)	(0.17)	(0.2)	(0.15)	(0.14)
Conductivity	65.56	214.06	225.42	60.28	127.82	75.54	141.74	103.00	95.98	88.42	180.04	65.32
μ S cm ⁻¹ ; \pm SE	(15.43)	(162.80)	(180.30)	(21.68)	(50.31)	(23.35)	(64.94)	(19.43)	(30.25)	(14.07)	(111.70)	(12.67)
Moisture	1.59	2.78	3.57	0.72	3.61	2.37	1.44	2.19	1.04	0.73	1.89	1.59
%; \pm SE	(0.36)	(1.44)	(1.89)	(0.09)	(2.03)	(1.54)	(0.54)	(1.05)	(0.26)	(0.04)	(0.86)	(0.72)
Organic Matter	0.8	0.81	0.87	0.82	1.1	1.03	0.85	0.86	0.92	0.95	0.89	1.01
%; \pm SE	(0.077)	(0.152)	(0.138)	(0.061)	(0.28)	(0.176)	(0.105)	(0.067)	(0.121)	(0.098)	(0.047)	(0.103)
Ammonium	1.021	1.107	1.485	0.621	1.191	0.852	0.825	1.313	1.419	0.784	1.358	1.877
μ g g ⁻¹ ; \pm SE	(0.510)	(0.606)	(0.370)	(0.201)	(0.521)	(0.374)	(0.181)	(0.712)	(0.539)	(0.157)	(0.392)	(0.501)
Nitrate-Nitrite	0.469	0.297	0.231	0.563	0.394	0.277	0.303	0.448	0.311	0.292	0.559	0.699
μ g g ⁻¹ ; \pm SE	(0.061)	(0.081)	(0.034)	(0.332)	(0.148)	(0.068)	(0.014)	(0.179)	(0.057)	(0.072)	(0.199)	(0.414)
Phosphorus	0.255	0.330	0.290	0.290	0.522	0.385	0.452	0.233	0.298	0.312	0.471	0.538
μ g g ⁻¹ ; \pm SE	(0.082)	(0.057)	(0.060)	(0.087)	(0.145)	(0.145)	(0.079)	(0.098)	(0.082)	(0.096)	(0.193)	(0.307)

Table A5.16 The effect of planting density, fertilizer regime, and humic acid amendment on Fall 2011 *Panicum amarum* soil pH, conductivity, moisture, organic matter, ammonium, nitrate-nitrite, and phosphorus (mean +/- SE, n = 5; Fa 2011 pH LSD = 0.79, conductivity LSD = 158.31, moisture LSD = 3.72, organic matter LSD = .44, ammonium LSD = 0.482, nitrate-nitrite LSD = 0.223, phosphorus LSD = 0.256).

Fall 2011 Metric	Low Density						High Density					
	Ambient			Fertilized			Ambient			Fertilized		
	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²
pH	6.6	6.58	6.9	7.11	6.47	6.99	6.5	6.78	6.5	6.56	6.58	6.71
± SE	(0.17)	(0.24)	(0.32)	(0.4)	(0.18)	(0.36)	(0.12)	(0.33)	(0.18)	(0.16)	(0.22)	(0.17)
Conductivity	18.92	17.54	113.26	9.96	9.44	19.22	18.06	11.94	47.12	10.36	32.34	17.66
µS cm ⁻¹ ; ± SE	(5.93)	(7.44)	(105.93)	(1.85)	(1.60)	(9.13)	(7.05)	(4.14)	(33.16)	(1.69)	(23.58)	(7.29)
Moisture	6.35	6.42	8.13	9.19	7.05	8.20	6.98	7.79	7.41	7.42	8.89	9.93
%; ± SE	(0.91)	(0.86)	(2.01)	(1.60)	(2.45)	(2.30)	(1.07)	(1.85)	(1.23)	(1.15)	(2.16)	(1.07)
Organic Matter	0.76	1.03	0.62	0.93	0.87	0.87	0.62	0.68	0.94	0.76	0.85	1.18
%; ± SE	(0.077)	(0.253)	(0.17)	(0.074)	(0.127)	(0.115)	(0.012)	(0.067)	(0.195)	(0.053)	(0.146)	(0.288)
Ammonium	0.395	0.723	0.224	1.128	1.001	0.827	0.279	0.233	0.393	1.433	1.302	1.257
µg g ⁻¹ ; ± SE	(0.122)	(0.263)	(0.073)	(0.302)	(0.156)	(0.167)	(0.05)	(0.066)	(0.131)	(0.493)	(0.281)	(0.315)
Nitrate-Nitrite	0.44	0.495	0.311	0.655	0.434	0.454	0.411	0.351	0.423	0.726	0.72	0.938
µg g ⁻¹ ; ± SE	(0.069)	(0.114)	(0.025)	(0.121)	(0.091)	(0.102)	(0.021)	(0.029)	(0.059)	(0.185)	(0.102)	(0.199)
Phosphorus	1.32	1.37	1.43	1.27	1.07	1.12	1.28	1.53	1.51	1.98	1.20	2.05
µg g ⁻¹ ; ± SE	(0.184)	(0.181)	(0.094)	(0.120)	(0.123)	(0.042)	(0.264)	(0.025)	(0.059)	(0.548)	(0.166)	(0.656)

Table A5.17. The effect of planting density, fertilizer regime, and humic acid amendment on Spring 2010 *Spartina patens* soil pH, conductivity, moisture, organic matter, ammonium, nitrate-nitrite, and phosphorus (mean \pm SE, n = 5; Sp 2010 pH LSD = 0.73, conductivity LSD = 1277.2, moisture LSD = 4.61, organic matter LSD = 1.21, ammonium LSD = 2.083, nitrate-nitrite LSD = 0.218, phosphorus LSD = 0.333).

Spring 2010 Metric	Low Density						High Density					
	Ambient			Fertilized			Ambient			Fertilized		
	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²
pH	9.02	8.69	9.18	9.14	8.68	8.99	9.16	9.11	8.63	9.12	8.86	9.42
\pm SE	(0.2)	(1.26)	(0.2)	(0.26)	(0.29)	(0.2)	(0.09)	(0.06)	(0.13)	(0.13)	(0.16)	(0.12)
Conductivity	745.10	2401.24	737.22	1559.40	1984.58	2212.06	685.52	176.91	2210.26	282.34	165.44	475.76
μ S cm ⁻¹ ; \pm SE	(503.85)	(2157.26)	(456.12)	(1256.63)	(1786.54)	(1917.93)	(465.63)	(50.22)	(1592.09)	(108.16)	(60.65)	(149.26)
Moisture	11.74	6.33	4.07	5.29	3.76	6.58	3.36	3.18	4.21	1.70	4.11	2.48
%; \pm SE	(7.67)	(1.96)	(1.27)	(1.47)	(0.55)	(3.06)	(0.49)	(0.67)	(0.37)	(0.52)	(1.85)	(0.52)
Organic Matter	1.07	1.29	1.07	1.22	1.17	1.48	0.96	0.93	1.15	0.92	1.13	1.13
%; \pm SE	(0.086)	(0.276)	(0.123)	(0.284)	(0.124)	(0.391)	(0.025)	(0.025)	(0.25)	(0.025)	(0.195)	(0.191)
Ammonium	1.754	2.491	1.744	1.78	1.846	2.49	1.391	1.526	1.842	1.002	1.907	1.427
μ g g ⁻¹ ; \pm SE	(0.373)	(0.432)	(0.486)	(0.532)	(0.353)	(0.757)	(0.284)	(0.334)	(0.37)	(0.218)	(0.575)	(0.551)
Nitrate-Nitrite	0.631	0.228	0.183	0.261	0.275	0.229	0.572	0.222	0.567	0.424	0.169	0.329
μ g g ⁻¹ ; \pm SE	(0.351)	(0.009)	(0.034)	(0.065)	(0.047)	(0.05)	(0.355)	(0.052)	(0.311)	(0.188)	(0.021)	(0.155)
Phosphorus	0.539	1.180	0.760	0.474	0.299	0.541	0.549	0.605	0.545	0.175	0.907	1.087
μ g g ⁻¹ ; \pm SE	(0.298)	(0.372)	(0.355)	(0.229)	(0.271)	(0.381)	(0.356)	(0.297)	(0.334)	(0.164)	(0.370)	(0.354)

Table A5.18 The effect of planting density, fertilizer regime, and humic acid amendment on Summer 2010 *Spartina patens* soil pH, conductivity, moisture, organic matter, ammonium, nitrate-nitrite, and phosphorus (mean +/- SE, n = 5; Summer 2010 pH LSD = 0.59, conductivity LSD = 1889.3, moisture LSD = 8.04, organic matter LSD = 0.46, ammonium LSD = 5.226, nitrate-nitrite LSD = 0.218, phosphorus LSD = 0.688)

Summer 2010 Metric	Low Density						High Density					
	Ambient			Fertilized			Ambient			Fertilized		
	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²
pH	8.68	0.32	8.16	8.23	8.47	7.99	8.36	7.96	7.91	8.32	7.68	7.89
± SE	(0.09)	(0.16)	(0.22)	(0.13)	(0.16)	(0.29)	(0.13)	(0.31)	(0.24)	(0.22)	(0.3)	(0.18)
Conductivity	276.68	1346.88	546.20	236.46	228.94	855.40	214.24	246.64	1311.24	341.82	2349.98	216.28
µS cm ⁻¹ ; ± SE	(68.32)	(695.34)	(386.52)	(46.95)	(38.16)	(510.92)	(55.07)	(120.55)	(1179.83)	(205.54)	(2225.07)	(62.52)
Moisture	7.87	9.43	6.13	6.56	6.89	6.01	5.07	5.63	5.71	4.26	6.54	4.50
%; ± SE	(2.05)	(2.72)	(0.89)	(1.32)	(0.47)	(1.75)	(1.09)	(1.42)	(2.07)	(0.73)	(2.86)	(0.64)
Organic Matter	1.17	1.34	1.12	1.14	1.03	1.24	1.13	1.65	1.38	1.19	1.47	1.28
%; ± SE	(0.163)	(0.097)	(0.103)	(0.122)	(0.124)	(0.19)	(0.084)	(0.391)	(0.325)	(0.136)	(0.368)	(0.177)
Ammonium	2.539	1.769	1.823	1.892	1.894	3.157	3.804	2.859	1.371	2.121	1.395	2.124
µg g ⁻¹ ; ± SE	(0.382)	(0.398)	(0.526)	(0.519)	(0.559)	(1.354)	(1.045)	(0.233)	(0.307)	(0.838)	(0.372)	(0.65)
Nitrate-Nitrite	0.353	0.224	0.306	0.261	0.153	0.415	0.586	0.29	0.286	0.376	0.197	0.211
µg g ⁻¹ ; ± SE	(0.093)	(0.03)	(0.072)	(0.04)	(0.01)	(0.133)	(0.31)	(0.043)	(0.033)	(0.155)	(0.011)	(0.031)
Phosphorus	1.833	2.328	2.481	2.307	2.601	2.548	1.881	1.599	1.732	2.270	2.065	2.258
µg g ⁻¹ ; ± SE	(0.119)	(0.206)	(0.080)	(0.124)	(0.082)	(0.085)	(0.437)	(0.499)	(0.349)	(0.147)	(0.309)	(0.192)

Table A5.19 The effect of planting density, fertilizer regime, and humic acid amendment on Fall 2010 *Spartina patens* soil pH, conductivity, moisture, organic matter, ammonium, nitrate-nitrite, and phosphorus (mean \pm SE, n = 5; Fa 2010 pH LSD = 0.71, conductivity LSD = 1913.2, moisture LSD = 6.52, organic matter LSD = 0.964, ammonium LSD = 1.50, nitrate-nitrite LSD = 0.197, phosphorus LSD = 0.303)

Fall 2010 Metric	Low Density						High Density					
	Ambient			Fertilized			Ambient			Fertilized		
	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²
pH	8.43	7.87	7.54	8.02	8.33	7.29	8.21	8.26	8.06	7.84	7.57	7.93
\pm SE	(0.39)	(0.22)	(0.21)	(0.38)	(0.47)	(0.27)	(0.46)	(0.52)	(0.26)	(0.15)	(0.22)	(0.25)
Conductivity	320.74	700.05	488.12	1371.38	1005.06	277.35	893.98	294.94	527.62	213.52	151.20	267.28
μ S cm ⁻¹ ; \pm SE	(138.62)	(259.70)	(288.96)	(1099.11)	(483.52)	(150.92)	(767.66)	(73.01)	(324.24)	(57.02)	(52.68)	(62.49)
Moisture	2.64	3.94	3.28	2.94	3.48	4.16	1.20	1.53	4.47	0.52	2.61	0.97
%; \pm SE	(0.73)	(1.35)	(1.99)	(1.30)	(2.81)	(2.88)	(0.50)	(0.57)	(2.00)	(0.44)	(1.55)	(0.18)
Organic Matter	1.32	1.15	0.98	1.66	1.51	1.47	0.98	1.14	1.24	1.08	1.14	1.06
%; \pm SE	(0.17)	(0.155)	(0.126)	(0.44)	(0.29)	(0.146)	(0.053)	(0.284)	(0.258)	(0.173)	(0.114)	(0.104)
Ammonium	0.498	1.003	1.887	1.148	1.048	0.82	0.48	6.336	0.897	2.969	0.838	0.46
μ g g ⁻¹ ; \pm SE	(0.138)	(0.787)	(1.476)	(0.59)	(0.404)	(0.436)	(0.315)	(5.229)	(0.239)	(2.61)	(0.453)	(0.13)
Nitrate-Nitrite	0.018	0.475	0.075	0.078	0.165	0.072	0.2	0.118	0.39	0.105	0.169	0.232
μ g g ⁻¹ ; \pm SE	(0.026)	(0.366)	(0.035)	(0.04)	(0.089)	(0.017)	(0.099)	(0.058)	(0.133)	(0.056)	(0.127)	(0.085)
Phosphorus	1.011	1.470	1.416	1.637	1.339	1.075	0.899	1.547	1.077	0.831	1.536	1.131
μ g g ⁻¹ ; \pm SE	(0.323)	(0.242)	(0.148)	(0.448)	(0.367)	(0.302)	(0.366)	(0.404)	(0.322)	(0.328)	(0.123)	(0.470)

Table A5.20 The effect of planting density, fertilizer regime, and humic acid amendment on Spring 2011 *Spartina patens* soil pH, conductivity, moisture, organic matter, ammonium, nitrate-nitrite, and phosphorus (mean \pm SE, n = 5; Sp 2011 pH LSD = 0.64, conductivity LSD = 578.06, moisture LSD = 4.12, organic matter LSD = .44, ammonium LSD = 1.118, nitrate-nitrite LSD = 0.222, phosphorus LSD = 0.171)

Spring 2011 Metric	Low Density						High Density					
	Ambient			Fertilized			Ambient			Fertilized		
	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²
pH	8.94	8.91	8.72	8.95	8.68	8.9	8.64	8.88	9.09	8.46	8.79	8.57
\pm SE	(0.31)	(0.11)	(0.19)	(0.27)	(0.26)	(0.09)	(0.14)	(0.1)	(0.26)	(0.21)	(0.2)	(0.08)
Conductivity	74.96	307.50	945.60	107.77	65.62	505.02	86.62	89.44	132.62	60.56	288.54	43.34
μ S cm ⁻¹ ; \pm SE	(34.86)	(258.77)	(899.10)	(59.52)	(16.54)	(465.26)	(41.10)	(55.19)	(58.39)	(8.62)	(200.79)	(6.09)
Moisture	3.39	5.81	7.90	1.67	2.03	5.00	1.99	1.54	2.51	1.19	3.71	1.07
%; \pm SE	(1.62)	(4.01)	(5.26)	(0.67)	(0.49)	(3.08)	(0.65)	(0.55)	(0.93)	(0.25)	(1.99)	(0.15)
Organic Matter	0.97	1.08	1.25	1	1.13	1.23	0.82	0.89	0.76	0.91	1.25	0.96
%; \pm SE	(0.072)	(0.252)	(0.437)	(0.106)	(0.08)	(0.269)	(0.062)	(0.129)	(0.021)	(0.066)	(0.279)	(0.055)
Ammonium	0.951	1.444	0.904	0.748	1.198	1.311	0.595	0.694	2.992	0.762	1.712	0.939
μ g g ⁻¹ ; \pm SE	(0.491)	(0.392)	(0.129)	(0.232)	(0.347)	(0.27)	(0.136)	(0.334)	(1.09)	(0.155)	(0.446)	(0.43)
Nitrate+Nitrite	0.348	0.628	0.372	0.477	0.295	0.365	0.305	0.389	0.27	0.667	0.688	0.447
μ g g ⁻¹ ; \pm SE	(0.051)	(0.169)	(0.037)	(0.096)	(0.032)	(0.071)	(0.024)	(0.111)	(0.026)	(0.373)	(0.425)	(0.156)
Phosphorus	0.353	0.490	0.308	0.475	0.411	0.365	0.509	0.783	0.403	0.347	0.333	0.504
μ g g ⁻¹ ; \pm SE	(0.111)	(0.067)	(0.104)	(0.123)	(0.072)	(0.092)	(0.103)	(0.340)	(0.125)	(0.090)	(0.089)	(0.048)

Table A5.21 The effect of planting density, fertilizer regime, and humic acid amendment on Fall 2011 *Spartina patens* soil pH, conductivity, moisture, organic matter, ammonium, nitrate-nitrite, and phosphorus (mean +/- SE, n = 5; Fa 2011 pH LSD = 0.79, conductivity LSD = 158.31, moisture LSD = 4.2, organic matter LSD = .44, ammonium LSD = 0.482, nitrate-nitrite LSD = 0.223, phosphorus LSD = 0.256)

Fall 2011 Metric	Low Density						High Density					
	Ambient			Fertilized			Ambient			Fertilized		
	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	250 ml m ⁻²
pH	7.07	6.84	6.8	6.99	7.12	6.96	7.11	6.95	7.22	6.95	6.64	7.03
± SE	(0.14)	(0.11)	(0.22)	(0.25)	(0.26)	(0.18)	(0.27)	(0.18)	(0.34)	(0.23)	(0.12)	(0.29)
Conductivity	89.76	141.50	147.90	89.88	19.36	149.54	20.46	30.62	18.06	16.74	197.52	16.92
µS cm ⁻¹ ; ± SE	(71.31)	(116.87)	(135.06)	(77.43)	(5.58)	(134.89)	(8.94)	(16.43)	(8.07)	(5.74)	(179.17)	(3.88)
Moisture	8.13	7.92	8.74	10.19	10.25	11.32	8.64	8.59	7.23	8.51	10.23	8.62
%; ± SE	(0.36)	(1.59)	(1.67)	(0.76)	(1.02)	(2.47)	(1.50)	(0.72)	(0.87)	(1.28)	(1.95)	(1.41)
Organic Matter	1.05	1.05	0.96	1.11	0.84	1.32	0.81	0.95	0.86	0.94	1.2	1.06
%; ± SE	(0.11)	(0.114)	(0.229)	(0.118)	(0.052)	(0.3)	(0.07)	(0.093)	(0.052)	(0.085)	(0.22)	(0.107)
Ammonium	0.196	0.678	0.278	1.4	1.229	1.17	0.537	0.449	0.813	0.978	1.132	2.411
µg g ⁻¹ ; ± SE	(0.045)	(0.566)	(0.176)	(0.816)	(0.304)	(0.488)	(0.221)	(0.159)	(0.518)	(0.2)	(0.51)	(1.009)
Nitrate+Nitrite	0.288	0.26	0.615	0.312	0.237	0.278	0.445	0.181	0.39	0.307	0.243	0.242
µg g ⁻¹ ; ± SE	(0.04)	(0.072)	(0.315)	(0.06)	(0.042)	(0.045)	(0.15)	(0.028)	(0.124)	(0.065)	(0.025)	(0.082)
Phosphorus	1.419	1.578	1.572	1.338	1.241	1.147	1.529	1.531	1.456	1.269	1.408	1.170
µg g ⁻¹ ; ± SE	(0.039)	(0.024)	(0.037)	(0.099)	(0.132)	(0.293)	(0.017)	(0.017)	(0.079)	(0.142)	(0.064)	(0.169)

Table A5.22 The effect of season, fertilizer regime, and humic acid amendment on high density–*Panicum amarum* soil pH, conductivity, organic matter, ammonium, nitrate-nitrite, and phosphorus (mean +/- SE, n = 5; Fall 2010: pH LSD = 0.36, conductivity LSD = 1508, organic matter LSD = 1.40, ammonium LSD = 3.43, nitrate-nitrite LSD = 0.59, phosphorus LSD = 0.208; Spring 2011: pH LSD = 0.77, conductivity LSD = 1363, organic matter LSD = 0.72, ammonium LSD = 2.77, nitrate-nitrite LSD = 0.44, phosphorus LSD = 0.493).

Metric	Fall 2010				Spring 2011			
	Ambient		Fertilized		Ambient		Fertilized	
	0 ml m ⁻²	125 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²
pH	8.63	8.71	8.64	8.66	8.53	7.89	8.41	8.24
± SE	(0.10)	(0.08)	(0.13)	(0.04)	(0.11)	(0.28)	(0.06)	(0.27)
Conductivity	14.74	18.70	22.47	20.24	6.89	7.76	10.68	10.94
mS cm ⁻¹ ; ± SE	(3.40)	(3.94)	(4.43)	(5.71)	(3.68)	(5.30)	(5.91)	(4.79)
Organic Matter	2.35	2.09	1.97	2.15	1.43	1.64	1.70	1.74
%; ± SE	(0.60)	(0.23)	(0.34)	(0.31)	(0.28)	(0.18)	(0.31)	(0.15)
Ammonium	0.738	0.191	0.466	0.931	1.307	1.862	2.045	3.096
µg g ⁻¹ ; ± SE	(0.638)	(0.073)	(0.366)	(0.802)	(0.46)	(0.796)	(1.058)	(0.944)
Nitrate-Nitrite	0.577	0.563	0.328	0.245	0.231	0.632	0.723	0.185
µg g ⁻¹ ; ± SE	(0.314)	(0.28)	(0.089)	(0.062)	(0.028)	(0.296)	(0.304)	(0.032)
Phosphorus	0.694	0.501	0.57	0.658	0.646	0.655	0.528	0.277
µg g ⁻¹ ; ± SE	(0.028)	(0.095)	(0.07)	(0.047)	(0.041)	(0.021)	(0.181)	(0.056)

Table A5.23 The effect of season, fertilizer regime, and humic acid amendment on high density–*Distichlis spicata* soil pH, conductivity, organic matter, ammonium, nitrate-nitrite, and phosphorus (mean \pm SE, n = 5; Fall 2010: pH LSD = 0.36, conductivity LSD = 1508, organic matter LSD = 1.40, ammonium LSD = 3.43, nitrate-nitrite LSD = 0.59, phosphorus LSD = 0.208; Spring 2011: pH LSD = 0.77, conductivity LSD = 1363, organic matter LSD = 0.72, ammonium LSD = 2.77, nitrate-nitrite LSD = 0.44, phosphorus LSD = 0.493).

Metric	Fall 2010				Spring 2011			
	Ambient		Fertilized		Ambient		Fertilized	
	0 ml m ⁻²	125 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²
pH	8.60	8.80	8.72	8.60	8.34	7.93	8.39	8.10
\pm SE	(0.13)	(0.20)	(0.09)	(0.15)	(0.33)	(0.45)	(0.17)	(0.37)
Conductivity	28.07	18.60	26.92	30.81	8.26	11.98	15.58	12.85
mS cm ⁻¹ ; \pm SE	(10.45)	(6.31)	(6.39)	(6.34)	(3.04)	(6.57)	(4.77)	(4.92)
Organic Matter	1.82	2.41	4.39	2.55	1.47	2.01	1.50	1.80
%; \pm SE	(0.43)	(0.56)	(1.59)	(0.58)	(0.14)	(0.45)	(0.28)	(0.40)
Ammonium	0.176	1.032	0.671	1.695	1.326	1.785	1.897	2.31
μ g g ⁻¹ ; \pm SE	(0.055)	(0.865)	(0.512)	(0.959)	(0.544)	(0.692)	(0.544)	(1.037)
Nitrate-Nitrite	0.38	0.245	0.224	0.22	0.273	0.181	0.245	0.238
μ g g ⁻¹ ; \pm SE	(0.141)	(0.08)	(0.047)	(0.039)	(0.053)	(0.035)	(0.054)	(0.03)
Phosphorus	0.648	0.642	0.608	0.546	0.622	0.633	0.592	0.529
μ g g ⁻¹ ; \pm SE	(0.047)	(0.057)	(0.043)	(0.092)	(0.033)	(0.032)	(0.291)	(0.192)

Table A5.24 The effect of season, fertilizer regime, and humic acid amendment on low density–*Spartina patens* soil pH, conductivity, organic matter, ammonium, nitrate-nitrite, and phosphorus (mean +/- SE, n = 5; Fall 2010: pH LSD = 0.36, conductivity LSD = 1508, organic matter LSD = 1.40, ammonium LSD = 3.43, nitrate-nitrite LSD = 0.59, phosphorus LSD = 0.208; Spring 2011: pH LSD = 0.77, conductivity LSD = 1363, organic matter LSD = 0.72, ammonium LSD = 2.77, nitrate-nitrite LSD = 0.44, phosphorus LSD = 0.493).

Metric	Fall 2010				Spring 2011			
	Ambient		Fertilized		Ambient		Fertilized	
	0 ml m ⁻²	125 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²
pH	8.47	8.75	8.85	8.71	8.48	8.41	8.66	8.18
± SE	(0.12)	(0.15)	(0.14)	(0.06)	(0.10)	(0.12)	(0.02)	(0.28)
Conductivity	26.94	29.69	16.54	18.19	14.95	15.49	17.10	12.20
µS cm ⁻¹ ; ± SE	(5.91)	(4.56)	(1.90)	(4.57)	(5.87)	(3.27)	(3.38)	(6.19)
Organic Matter	2.71	2.50	2.20	2.45	1.80	1.88	1.77	1.76
%; ± SE	(0.52)	(0.39)	(0.26)	(0.67)	(0.12)	(0.25)	(0.30)	(0.27)
Ammonium	1.262	0.493	0.195	0.692	2.604	1.363	2.109	1.689
µg g ⁻¹ ; ± SE	(0.638)	(0.364)	(0.082)	(0.498)	(0.869)	(0.866)	(1.25)	(0.836)
Nitrate-Nitrite	0.877	0.364	0.227	0.302	0.425	0.337	0.367	0.356
µg g ⁻¹ ; ± SE	(0.373)	(0.102)	(0.062)	(0.074)	(0.089)	(0.073)	(0.113)	(0.209)
Phosphorus	0.505	0.62	0.671	0.543	0.557	0.61	0.568	0.349
µg g ⁻¹ ; ± SE	(0.059)	(0.025)	(0.03)	(0.069)	(0.087)	(0.017)	(0.067)	(0.1)

Table A5.25 The effect of season, fertilizer regime, and humic acid amendment on high density–*Spartina patens* soil pH, conductivity, organic matter, ammonium, nitrate-nitrite, and phosphorus (mean +/- SE, n = 5; ; Fall 2010: pH LSD = 0.36, conductivity LSD = 1508, organic matter LSD = 1.40, ammonium LSD = 3.43, nitrate-nitrite LSD = 0.59, phosphorus LSD = 0.208; Spring 2011: pH LSD = 0.77, conductivity LSD = 1363, organic matter LSD = 0.72, ammonium LSD = 2.77, nitrate-nitrite LSD = 0.44, phosphorus LSD = 0.493).

Metric	Fall 2010				Spring 2011			
	Ambient		Fertilized		Ambient		Fertilized	
	0 ml m ⁻²	125 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²
pH	8.69	8.76	8.90	8.79	8.31	8.06	8.32	8.08
± SE	(0.18)	(0.09)	(0.14)	(0.08)	(0.35)	(0.31)	(0.28)	(0.30)
Conductivity	20.58	16.97	17.54	19.91	8.30	6.54	8.86	9.59
µS cm ⁻¹ ; ± SE	(2.80)	(6.04)	(2.96)	(5.87)	(2.86)	(3.66)	(4.16)	(4.64)
Organic Matter	1.76	1.98	1.74	2.14	1.61	1.58	1.81	1.73
%; ± SE	(0.17)	(0.43)	(0.17)	(0.44)	(0.24)	(0.20)	(0.35)	(0.20)
Ammonium	1.298	7.176	1.227	1.375	3.086	1.592	2.902	2.267
µg g ⁻¹ ; ± SE	(0.736)	(4.831)	(0.759)	(0.688)	(1.969)	(0.275)	(1.412)	(0.538)
Nitrate-Nitrite	0.408	0.561	0.293	0.422	0.26	0.444	0.235	0.362
µg g ⁻¹ ; ± SE	(0.155)	(0.408)	(0.072)	(0.248)	(0.058)	(0.197)	(0.032)	(0.161)
Phosphorus	0.647	0.433	0.572	0.539	0.58	0.63	0.416	0.586
µg g ⁻¹ ; ± SE	(0.007)	(0.106)	(0.105)	(0.089)	(0.046)	(0.035)	(0.092)	(0.249)

Table A5.26 The effect of season, fertilizer regime, and humic acid amendment on the high-density *Panicum amarum*, *Distichlis spicata* and *Spartina patens* mixture soil pH, conductivity, organic matter, ammonium, nitrate-nitrite, and phosphorus (mean +/- SE, n = 5; ; Fall 2010: pH LSD = 0.36, conductivity LSD = 1508, organic matter LSD = 1.40, ammonium LSD = 3.43, nitrate-nitrite LSD = 0.59, phosphorus LSD = 0.208; Spring 2011: pH LSD = 0.77, conductivity LSD = 1363, organic matter LSD = 0.72, ammonium LSD = 2.77, nitrate-nitrite LSD = 0.44, phosphorus LSD = 0.493).

Metric	Fall 2010				Spring 2011			
	Ambient		Fertilized		Ambient		Fertilized	
	0 ml m ⁻²	125 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²	0 ml m ⁻²	125 ml m ⁻²
pH	8.86	8.96	8.84	8.67	8.37	8.73	8.16	8.88
± SE	(0.13)	(0.10)	(0.13)	(0.11)	(0.16)	(0.09)	(0.41)	(0.19)
Conductivity	15.33	21.46	18.88	21.88	9.67	14.17	8.49	10.75
µS cm ⁻¹ ; ± SE	(2.79)	(1.61)	(4.44)	(3.09)	(4.61)	(7.44)	(4.94)	(5.18)
Organic Matter	1.76	1.55	1.91	1.49	1.80	1.33	1.61	1.46
%; ± SE	(0.22)	(0.15)	(0.36)	(0.22)	(0.23)	(0.21)	(0.24)	(0.12)
Ammonium	0.1	0.558	0.763	0.1	2.238	1.797	1.967	2.56
µg g ⁻¹ ; ± SE	(0)	(0.458)	(0.425)	(0)	(1.012)	(0.48)	(1.016)	(1.404)
Nitrate-Nitrite	0.553	0.454	0.398	0.36	0.187	0.294	0.516	0.266
µg g ⁻¹ ; ± SE	(0.307)	(0.15)	(0.101)	(0.112)	(0.028)	(0.058)	(0.266)	(0.018)
Phosphorus	0.565	0.525	0.586	0.616	0.662	0.622	0.686	0.576
µg g ⁻¹ ; ± SE	(0.064)	(0.13)	(0.049)	(0.056)	(0.025)	(0.025)	(0.412)	(0.422)

Table A7.1 Total live cover, total dead cover, mean canopy height, and soil redox potential at 1 cm and 15 cm for reference plots in fall 2010, spring 2011, and fall 2011

Measurement	Reference Plot Sampling Season		
	Fall 2010	Spring 2011	Fall 2011
Total Live Cover (%)	65	60	53
± SE	(7)	(11)	(19)
Total Dead Cover (%)	2	13	16
± SE	(1)	(5)	(8)
Mean Canopy Height (cm)	57	50	52
± SE	(4)	(8)	(12)
Soil Redox 1 cm (mV)	72.8	73.8	68.7
± SE	(31.0)	(51.1)	(13.4)
Soil Redox 15 cm (mV)	11.6	-72.0	66.3
± SE	(27.7)	(55.0)	(24.5)

Table A7.2 Effect of propagule establishment technique, elevation, and humic acid on edaphic measurements in fall 2010.

Establishment Technique	Treatment		pH		Conductivity (mS cm ⁻¹)		Salinity (ppt)		Soil Moisture (%)		Bulk Density (g cm ⁻³)		Organic Matter (%)	
	Elevation	Humic Acid	Mean	± SE	Mean	± SE	Mean	± SE	Mean	± SE	Mean	± SE	Mean	± SE
Bare	Low	0 ml m ⁻²	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a
		125 ml m ⁻²	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a
		250 ml m ⁻²	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a
	High	0 ml m ⁻²	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a
		125 ml m ⁻²	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a
		250 ml m ⁻²	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a
Fences	Low	0 ml m ⁻²	8.08	0.03	7.8	1.2	4.4	0.7	37.27	1.58	1.89	0.16	5.78	0.48
		125 ml m ⁻²	8.08	0.05	7.9	1.3	4.4	0.8	36.96	2.59	1.66	0.10	11.48	5.45
		250 ml m ⁻²	8.13	0.05	8.4	0.9	4.7	0.5	35.58	3.56	1.66	0.12	7.00	1.07
	High	0 ml m ⁻²	8.04	0.11	8.0	1.0	4.5	0.6	33.45	2.31	1.69	0.25	7.49	1.44
		125 ml m ⁻²	8.03	0.09	8.3	0.7	4.7	0.4	34.89	2.77	1.83	0.16	6.55	1.50
		250 ml m ⁻²	8.02	0.07	10.2	1.6	5.8	1.0	35.26	1.69	1.91	0.11	5.68	0.61
Propagules Added	Low	0 ml m ⁻²	8.04	0.13	6.6	0.9	3.7	0.6	38.23	1.47	1.47	0.16	6.08	0.67
		125 ml m ⁻²	8.08	0.07	7.6	0.5	4.5	0.2	33.97	0.96	1.88	0.16	5.80	0.82
		250 ml m ⁻²	8.06	0.10	8.1	1.1	4.5	0.6	34.83	2.18	1.62	0.18	7.16	1.40
	High	0 ml m ⁻²	8.04	0.12	8.2	1.2	4.6	0.7	37.05	1.30	1.82	0.13	5.99	0.28
		125 ml m ⁻²	8.05	0.06	9.5	1.8	5.2	1.2	34.61	1.24	1.92	0.08	5.76	0.25
		250 ml m ⁻²	8.07	0.05	9.5	1.8	5.4	1.1	37.26	2.99	1.94	0.12	7.31	1.66
Reference			7.77	0.03	6.7	0.9	11.7	1.5	59.28	4.77	1.51	0.18	4.78	1.27
LSD			0.24		3.5		2.1		6.23		0.43		5.31	

Table A7.3 Effect of propagule establishment technique, elevation, and humic acid on edaphic measurements in spring 2011.

Establishment Technique	Treatment		pH		Conductivity (mS cm ⁻¹)		Salinity (ppt)		Soil Moisture (%)		Bulk Density (g cm ⁻³)		Organic Matter (%)	
	Elevation	Humic Acid	Mean	± SE	Mean	± SE	Mean	± SE	Mean	± SE	Mean	± SE	Mean	± SE
Bare	Low	0 ml m ⁻²	8.30	0.11	11.8	1.4	6.7	0.9	33.27	1.32	2.00	0.14	7.57	0.80
		125 ml m ⁻²	8.46	0.02	11.8	1.7	6.8	1.0	33.12	1.99	1.99	0.18	7.48	0.62
		250 ml m ⁻²	8.36	0.12	11.1	0.6	6.3	0.4	30.43	2.32	1.93	0.18	6.60	0.18
	High	0 ml m ⁻²	8.34	0.05	11.0	0.6	6.3	0.4	33.66	1.96	2.12	0.16	7.40	0.89
		125 ml m ⁻²	8.42	0.03	10.5	0.6	5.9	0.4	33.45	2.36	2.32	0.11	7.16	0.52
		250 ml m ⁻²	8.37	0.05	10.7	1.3	6.1	0.8	31.73	0.83	2.33	0.13	5.93	0.26
Fences	Low	0 ml m ⁻²	8.40	0.04	10.1	0.4	5.7	0.2	37.04	2.18	2.07	0.19	7.71	0.65
		125 ml m ⁻²	8.40	0.02	9.8	1.0	5.5	0.6	33.67	1.11	1.94	0.16	6.76	0.45
		250 ml m ⁻²	8.37	0.03	10.4	1.3	5.9	0.8	36.47	2.76	2.14	0.14	6.78	0.66
	High	0 ml m ⁻²	8.44	0.05	10.5	0.9	5.9	0.6	30.52	1.15	2.21	0.05	6.49	0.21
		125 ml m ⁻²	8.41	0.04	11.0	1.7	6.3	1.0	34.27	1.21	2.07	0.11	6.89	0.24
		250 ml m ⁻²	8.32	0.06	10.7	1.3	6.0	0.8	31.98	1.69	2.19	0.13	6.75	0.63
Propagules Added	Low	0 ml m ⁻²	8.36	0.09	10.3	0.9	5.8	0.5	36.93	2.15	1.89	0.12	7.40	0.47
		125 ml m ⁻²	8.40	0.05	10.6	0.8	6.0	0.5	32.86	1.75	2.25	0.09	6.74	0.45
		250 ml m ⁻²	8.33	0.06	11.1	0.9	6.3	0.6	33.82	1.78	2.15	0.11	6.49	0.32
	High	0 ml m ⁻²	8.48	0.10	11.0	1.8	6.2	1.1	36.82	1.00	1.99	0.06	7.14	0.58
		125 ml m ⁻²	8.36	0.04	11.8	0.8	6.7	0.5	29.90	0.84	2.13	0.12	6.88	0.86
		250 ml m ⁻²	8.22	0.05	11.3	0.8	6.4	0.5	33.41	0.92	2.06	0.13	6.13	0.26
Reference			8.25	0.08	12.0	2.0	6.9	1.2	53.05	4.67	1.67	0.14	7.38	1.09
LSD			0.18		3.2		1.9		4.88		0.38		1.54	

Table A7.4 Effect of propagule establishment technique, elevation, and humic acid on edaphic measurements in fall 2011.

Establishment Technique	Treatment		pH		Conductivity (mS cm ⁻¹)		Salinity (ppt)		Soil Moisture (%)		Bulk Density (g cm ⁻³)		Organic Matter (%)	
	Elevation	Humic Acid	Mean	± SE	Mean	± SE	Mean	± SE	Mean	± SE	Mean	± SE	Mean	± SE
Bare	Low	0 ml m ⁻²	8.46	0.18	5.0	1.0	2.7	0.6	39.75	3.17	2.06	0.20	7.76	1.14
		125 ml m ⁻²	8.43	0.29	4.0	1.0	2.1	0.6	39.75	3.55	2.03	0.21	6.38	0.70
		250 ml m ⁻²	8.48	0.29	5.0	1.6	2.7	0.9	38.51	1.15	1.97	0.07	6.40	1.10
	High	0 ml m ⁻²	8.58	0.12	6.4	1.4	3.5	0.8	39.52	2.08	2.08	0.20	6.14	0.25
		125 ml m ⁻²	8.15	0.24	4.5	0.9	2.4	0.5	42.54	2.16	1.90	0.13	5.36	0.25
		250 ml m ⁻²	8.43	0.28	6.6	2.1	3.7	1.2	37.64	1.37	2.26	0.08	5.80	0.60
Fences	Low	0 ml m ⁻²	8.46	0.15	5.5	1.3	3.0	0.7	38.25	1.90	2.04	0.21	5.39	0.47
		125 ml m ⁻²	8.56	0.12	8.9	4.4	2.2	0.5	38.50	3.17	2.12	0.10	6.67	0.41
		250 ml m ⁻²	7.93	0.35	6.3	0.6	3.4	0.4	41.98	3.22	2.10	0.15	5.73	0.20
	High	0 ml m ⁻²	8.56	0.06	5.2	0.9	2.8	0.5	37.79	2.18	2.16	0.15	5.98	0.52
		125 ml m ⁻²	8.41	0.18	5.2	1.3	1.9	0.6	41.67	1.86	2.03	0.16	6.93	0.82
		250 ml m ⁻²	8.17	0.30	6.8	2.0	3.8	1.2	35.85	2.52	2.18	0.19	6.08	0.69
Propagules Added	Low	0 ml m ⁻²	8.54	0.25	4.9	0.9	2.2	0.7	40.96	2.82	2.02	0.19	7.21	1.25
		125 ml m ⁻²	8.19	0.29	4.5	1.1	2.4	0.6	39.40	2.58	2.26	0.17	6.49	0.64
		250 ml m ⁻²	8.20	0.22	4.8	1.1	2.6	0.6	36.40	4.40	2.21	0.09	6.11	0.81
	High	0 ml m ⁻²	8.52	0.25	4.9	1.2	2.6	0.7	35.48	1.65	2.18	0.16	6.91	0.77
		125 ml m ⁻²	8.29	0.17	7.5	0.8	4.2	0.5	38.43	2.71	2.32	0.17	5.20	0.36
		250 ml m ⁻²	8.24	0.32	5.9	1.0	3.2	0.6	39.62	2.68	2.22	0.07	5.26	0.40
Reference			8.39	0.04	4.9	1.0	2.9	0.6	64.86	7.03	1.73	0.16	6.28	0.72
LSD			0.67		4.5		2.0		7.31		0.44		2.16	

Table A7.5 Effect of propagule establishment technique, elevation, and humic acid on ammonium, nitrate -nitrite, phosphorus, and potassium concentrations of extracted sediments in fall 2010.

Treatment		Humic Acid	Ammonium ($\mu\text{g g}^{-1}$)		Nitrate-Nitrite($\mu\text{g g}^{-1}$)		Phosphorus ($\mu\text{g g}^{-1}$)		Potassium ($\mu\text{g g}^{-1}$)	
Establishment Technique	Elevation		Mean	$\pm SE$	Mean	$\pm SE$	Mean	$\pm SE$	Mean	$\pm SE$
Bare	Low	0 ml m ⁻²	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a
		125 ml m ⁻²	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a
		250 ml m ⁻²	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a
	High	0 ml m ⁻²	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a
		125 ml m ⁻²	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a
		250 ml m ⁻²	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a
Fences	Low	0 ml m ⁻²	2.28	0.55	0.14	0.02	0.342	0.008	133.1	14.7
		125 ml m ⁻²	2.19	0.20	0.15	0.02	0.327	0.010	138.3	9.1
		250 ml m ⁻²	3.28	1.62	0.20	0.05	0.278	0.044	121.8	11.0
	High	0 ml m ⁻²	1.62	0.43	0.16	0.02	0.331	0.008	134.9	14.6
		125 ml m ⁻²	1.62	0.33	0.22	0.07	0.296	0.020	144.5	18.8
		250 ml m ⁻²	2.29	0.39	0.16	0.01	0.324	0.008	160.0	10.9
Propagules Added	Low	0 ml m ⁻²	2.62	0.49	0.15	0.02	0.329	0.005	119.8	7.4
		125 ml m ⁻²	2.20	0.52	0.42	0.17	0.277	0.061	137.1	13.1
		250 ml m ⁻²	2.27	0.57	0.19	0.04	0.331	0.003	133.6	12.6
	High	0 ml m ⁻²	1.15	0.50	0.13	0.02	0.330	0.005	129.0	11.1
		125 ml m ⁻²	2.79	0.65	0.14	0.02	0.342	0.006	161.5	24.7
		250 ml m ⁻²	2.20	0.53	0.21	0.04	0.340	0.006	151.2	16.6
Reference			1.60	0.43	0.24	0.11	0.309	0.010	156.0	20.8
LSD			1.87		0.17		0.066		41.0	

Table A7.6 Effect of propagule establishment technique, elevation, and humic acid on ammonium, nitrate-nitrite, phosphorus, and potassium concentrations of extracted sediments in spring 2011.

Treatment		Humic Acid	Ammonium ($\mu\text{g g}^{-1}$)		Nitrate-Nitrite($\mu\text{g g}^{-1}$)		Phosphorus ($\mu\text{g g}^{-1}$)		Potassium ($\mu\text{g g}^{-1}$)	
Establishment Technique	Elevation		Mean	$\pm SE$	Mean	$\pm SE$	Mean	$\pm SE$	Mean	$\pm SE$
Bare	Low	0 ml m ⁻²	0.07	0.03	0.05	0.03	0.347	0.012	171.9	14.9
		125 ml m ⁻²	0.43	0.17	0.05	0.02	0.339	0.014	166.5	17.1
		250 ml m ⁻²	0.21	0.13	0.04	0.01	0.329	0.019	153.9	10.2
	High	0 ml m ⁻²	0.07	0.02	0.08	0.05	0.346	0.016	149.7	6.2
		125 ml m ⁻²	0.13	0.08	0.08	0.02	0.347	0.015	159.0	11.6
		250 ml m ⁻²	0.10	0.04	0.03	0.01	0.347	0.016	162.7	13.3
Fences	Low	0 ml m ⁻²	0.25	0.17	0.05	0.02	0.338	0.009	160.1	9.6
		125 ml m ⁻²	0.47	0.27	0.06	0.03	0.348	0.016	150.4	10.0
		250 ml m ⁻²	n/a	n/a	0.03	0.01	0.314	0.019	144.5	15.0
	High	0 ml m ⁻²	0.07	0.06	0.05	0.02	0.347	0.010	155.0	7.0
		125 ml m ⁻²	0.12	0.03	0.05	0.02	0.349	0.012	160.5	8.9
		250 ml m ⁻²	0.02	n/a	0.03	0.01	0.349	0.016	162.6	17.1
Propagules Added	Low	0 ml m ⁻²	0.07	0.05	0.02	0.01	0.353	0.010	162.4	13.3
		125 ml m ⁻²	0.02	0.00	0.04	0.01	0.342	0.020	155.6	13.1
		250 ml m ⁻²	0.09	0.04	0.03	0.02	0.356	0.013	152.4	14.7
	High	0 ml m ⁻²	0.07	0.02	0.03	0.01	0.337	0.012	161.8	17.7
		125 ml m ⁻²	0.05	0.01	0.05	0.02	0.350	0.017	176.2	16.5
		250 ml m ⁻²	0.06	0.04	0.03	0.01	0.359	0.014	168.2	12.0
Reference			0.45	0.19	0.01	n/a	0.376	0.008	158.1	26.5
LSD			0.49		0.07		0.042		4.9	

Table A7.7 Effect of propagule establishment technique, elevation, and humic acid on ammonium, nitrate-nitrite, phosphorus, and potassium concentrations of extracted sediments in fall 2011.

Treatment		Humic Acid	Ammonium ($\mu\text{g g}^{-1}$)		Nitrate-Nitrite($\mu\text{g g}^{-1}$)		Phosphorus ($\mu\text{g g}^{-1}$)		Potassium ($\mu\text{g g}^{-1}$)	
Establishment Technique	Elevation		Mean	$\pm SE$	Mean	$\pm SE$	Mean	$\pm SE$	Mean	$\pm SE$
Bare	Low	0 ml m ⁻²	1.05	0.41	0.13	0.01	0.069	0.007	91.5	14.4
		125 ml m ⁻²	0.87	0.43	0.11	0.02	0.068	0.008	91.4	14.8
		250 ml m ⁻²	1.53	0.49	0.17	0.04	0.071	0.011	97.7	7.8
	High	0 ml m ⁻²	1.23	0.55	0.19	0.05	0.121	0.052	106.1	4.9
		125 ml m ⁻²	0.85	0.47	0.16	0.06	0.088	0.010	101.2	14.8
		250 ml m ⁻²	1.80	0.47	0.14	0.01	0.076	0.006	111.2	8.0
Fences	Low	0 ml m ⁻²	1.20	0.56	0.13	0.02	0.060	0.004	90.9	6.4
		125 ml m ⁻²	0.97	0.57	0.25	0.16	0.079	0.018	92.7	8.3
		250 ml m ⁻²	1.55	0.60	0.12	0.02	0.111	0.035	90.6	10.1
	High	0 ml m ⁻²	1.14	0.44	0.23	0.12	0.068	0.007	76.2	8.3
		125 ml m ⁻²	1.01	0.48	0.14	0.02	0.070	0.010	80.9	6.5
		250 ml m ⁻²	1.70	0.54	0.24	0.07	0.086	0.016	96.1	9.8
Propagules Added	Low	0 ml m ⁻²	1.22	0.51	0.27	0.08	0.072	0.013	89.6	10.9
		125 ml m ⁻²	1.31	0.48	0.19	0.07	0.074	0.014	84.6	9.2
		250 ml m ⁻²	1.55	0.55	0.25	0.11	0.072	0.011	92.6	9.1
	High	0 ml m ⁻²	1.65	0.38	0.15	0.03	0.088	0.020	88.9	3.2
		125 ml m ⁻²	1.01	0.46	0.12	0.03	0.069	0.004	109.5	6.6
		250 ml m ⁻²	1.15	0.45	0.15	0.02	0.078	0.009	98.6	12.8
Reference			1.33	0.24	0.17	0.06	0.032	0.006	80.4	15.7
LSD			1.38		0.20		0.052		27.5	

Table A7.8 Average water level and elevations of back-barrier salt marsh restoration site during the study period.

Time Period	Average water level (m NAVD88)	Maximum water level (m NAVD88)	Elevation of low plots (m NAVD88)	Elevation of high plots (m NAVD88)
April 2010 - Oct 2010	0.590	1.070	N/A	N/A
Oct 2010 - Nov 2010	0.505	0.843	0.788	0.808
Nov 2010 - April 2011	0.451	0.909	0.680	0.700
April 2011 - Oct 2011	0.447	1.250	0.649	0.666
Total Decrease	0.143		0.139	0.142