SALT MARSH DIEBACK IN GEORGIA: FIELD SURVEY AND TRANSPLANT EXPERIMENTS

by

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(Under the Direction of Merryl Alber)

ABSTRACT

In winter 2002, portions of the salt marshes of coastal Georgia began experiencing dieback, affecting both *Spartina alterniflora* and *Juncus roemerianus*. During the summer of 2003, a field survey of 18 widely distributed sites along the coast was conducted to document the characteristics of and obvious patterns in dieback areas. Most dieback areas were small (<1 acre), did not show spatial patterns and occurred along the edges of tidal creeks. There were no consistent differences in soil salinity, pH or redox potential between dieback and healthy areas. A transplant study was carried out to determine if healthy plants can survive in dieback areas. Transplant survival was 100% from May to October 2003, and growth was observed in both dieback and healthy (control) areas. The results of this study suggest that drought, along with various contributing factors, was the ultimate cause of salt marsh dieback in coastal Georgia.

INDEX WORDS: Salt marsh dieback, *Spartina alterniflora*, *Juncus roemerianus*, Soil conditions, Transplant experiment, Sapelo Island

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TABLE OF CONTENTS

CHAPTER 1

INTRODUCTION

Preface

In 2001, portions of the salt marshes of coastal Georgia began experiencing dieback. Dieback areas are characterized by loss of vegetation, resulting in large expanses of bare mud that are susceptible to erosion. The two dominant plants in Georgia salt marshes, *Spartina alterniflora* and *Juncus roemerianus*, were both affected. Salt marsh dieback was first reported to the Coastal Resources Division of the Georgia Department of Natural Resources (DNR) in March 2002. Later that month, aerial surveys showed that extensive dieback had already occurred along the Jerico River in Liberty County. Local residents reported that they had first noticed dieback at the Jerico River site during 2001. As more reports of dieback came in, it became clear that the problem was widespread, occurring in all six Georgia coastal counties, and extensive, with the largest site covering more than 600 acres (Jan Mackinnon, personal communication). Chlorotic or standing dead vegetation was rarely, if ever, observed. Salt marsh dieback was also reported in South Carolina in 2002 (South Carolina Department of Health and Environmental Control). Below I describe the Georgia dieback in more detail and compare it with reports of dieback from other locations, but first I briefly review the adaptations of *S. alterniflora* and *J. roemerianus* to the physicochemical environment of marshes.

Physical Stresses and Plant Adaptations in Salt Marshes

The distribution of salt marsh plants is largely dependent on the physical characteristics of the environment. Salinity, in particular, is often considered an important factor regulating the growth and distribution of salt marsh plants (for reviews, see Pielou and Routledge, 1976; Ungar, 1978; Rozema et al., 1985). In a Georgia salt marsh, Nestler (1977) reported an inverse relationship between productivity of *S. alterniflora* and salinity. Pore-water salinities are largely driven by the frequency of tidal flushing in an area. In the low marsh, where flooding is frequent, salinity is relatively constant and similar to that of flooding water (Adam, 1990). At higher elevations, where soils are not as well-flushed by tidal action, interactions between tides and seasonal climate patterns can cause predictable periods of high salinity (Jefferies, 1977; Jefferies and Perkins, 1977). During extremely dry periods, evapotranspiration may result in the formation of a salt crust on the soil surface even in areas that are usually regularly flooded (Adam, 1990). The differences in salinity, which are associated with concomitant differences in pore-water chemistry, result in variability in the growth of individual species. *S. alterniflora*, for example, shows a range of growth forms from tall plants (2-3 m in height) that grow along creek banks where flooding is frequent, soils are well-drained and nutrient availability is high to short plants (10-40 cm in height) that are found in areas further away from creeks where nutrient limitation tends to occur.

Although the growth rate of *S. alterniflora* is increased in fresh or brackish water as compared to salt water, it is well-adapted to handle the stresses associated with high salinity (Smart and Barko, 1980). *S. alterniflora* is able to regulate its salt content by secretion from salt glands on leaves (Waisel, 1972), exclusion from roots (Smart and Barko, 1980) and metabolism of proline and glycinebetaine as osmoregulatory solutes (Cavalieri and Huang, 1979; Cavalieri

and Huang, 1981), and can grow at salinities of up to 45 PSU (Linthurst and Blum, 1981). Hester et al. (2001) demonstrated variability in responses of different strains of *S. alterniflora* to salinity stress, indicating that some populations may be more resistant than others.

J. roemerianus, the other plant affected by dieback, is also adapted to handle salt stress, although its distribution may be limited by high salinities. *J. roemerianus* is capable of growing in a wide range of salinities, from 0 – 25 PSU (Eleuterius, 1984; Woerner and Hackney, 1997). Continuously higher salinities (30 PSU) can cause mortality, although brief periods of hypersalinity on salt flats (up to 360 PSU) may be compensated for with deeply penetrating roots that reach less saline groundwater (Eleuterius, 1984). Higinbotham et al. (in press) found that the transition from *S. alterniflora*-dominated marsh to *J. roemerianus*-dominated marsh in Georgia riverine estuaries corresponded with an average high tide salinity of 21 PSU, even though this halocline occurred at different distances from the mouth of the two estuaries studied (3 to 4 km from the mouth of the Altamaha River, 13 km from the mouth of the Satilla River).

Another physical factor that affects plant growth is the oxidation state of the soils. Salt marsh sediments have a characteristically flat topography and low hydraulic conductivity, which often result in soil water-logging and anoxia (Clarke and Hannon, 1967; Clarke and Hannon, 1969). The physicochemical environment of anoxic soils is very different from that of well-drained soils (for reviews, see Ponnamperuma, 1972; Gambrell and Patrick, 1978; and Armstrong, 1982), with reduced conditions and the potential for accumulation of toxic substances such as sulfides (Mendelssohn et al., 1981; Mendelssohn and McKee, 1988; Koch et al., 1990; Wilsey et al., 1992). Areas of reduced soil conditions tend to occur away from creek banks where flushing rates are low and coincide with areas of reduced plant productivity (Howes et al., 1981).

S. alterniflora has several adaptations that allow it to grow in water-logged, anoxic sediments. In moderately reduced sediments, aerenchyma tissue facilitates transport of oxygen to the roots and rhizomes, allowing *S. alterniflora* to oxygenate the soil (Teal and Kanwisher, 1966). The sediment underneath marsh grasses is therefore more oxidized than that found in bare areas or below the root zone, and larger plants have a greater influence on sediment oxygenation (Howes et al., 1981). In continuously flooded, highly reduced sediments, however, *S. alterniflora* is unable to sufficiently oxygenate the sediments. It survives under these conditions by switching to anaerobic respiration, although growth rates are lower (Mendelssohn et al., 1981). Toxic compounds such as sulfides can also accumulate in reduced soils and may be a major factor associated with reduced growth of *S. alterniflora* in water-logged sediments (Mendelssohn and McKee, 1988).

Although *J. roemerianus* routinely grows in saturated marsh soils, it may be less tolerant of inundation than *S. alterniflora*. In Georgia riverine estuaries, *S. alterniflora* and *S. cynosuroides* are found closest to the creek bank, with *J. roemerianus* occurring behind the creek bank extending all the way back to the upland following the typical pattern of zonation found in the southeastern United States (Higinbotham et al., in press; Wiegert and Freeman, 1990). Whereas *S. alterniflora* reaches its tallest heights along creek banks where inundation is greatest, plant height of *J. roemerianus* is inversely related to inundation (Woerner and Hackney, 1997). This result is supported by observations in a Mississippi marsh, where patches of *J. roemerianus* were flooded significantly less often than *S. alterniflora* (Eleuterius and Eleuterius, 1979). There is little experimental work that examines the tolerance of *J. roemerianus* to water-logging or sulfide accumulation.

Overview of Salt Marsh Dieback

The current salt marsh dieback phenomenon is the largest such event recorded in Georgia and is different from dieback events reported previously in this state. Edwards and Frey (1977) described the occurrence of barren areas on either low marsh mud or high marsh sand on Sapelo Island, GA. These small dieback areas were associated with causeway construction (Edwards and Frey, 1977), the accumulation of *Spartina* wrack (Basan and Frey, 1977), and grazing by herbivores (Basan and Frey, 1977). Small disturbances such as wrack accumulation and grazing are well-documented in salt marshes (Fischer et al., 2000) and occur in different patterns than the current acute dieback. It is interesting to note, however, that an additional type of barren that appears similar to the current dieback was observed by Edwards and Frey (1977), although the authors were unable to determine what had caused it (see Plate 9 of their article).

The salt marsh dieback that Georgia is currently experiencing manifests itself in several different ways (Figure 1-1). The most distinct and puzzling pattern of dieback occurs in a 1-3 m wide strip parallel to the banks of both large and small tidal creeks. It is typically located on the top of the levee as well as the side that faces the creek. This zone along tidal creeks is usually the most productive part of the marsh, supporting growth of tall-form *S. alterniflora*. Interestingly, healthy vegetation is sometimes found in the creek at the base of the levee below the dieback area. This pattern of dieback is common in both *S. alterniflora* and *J. roemerianus* marshes, with the exception that live *J. roemerianus* is never found in the creek below creek bank dieback areas because it does not grow at such low elevations.

Another distinct pattern of dieback in Georgia is the formation of scallop-shaped dieback areas on or behind the creek bank levee (Figure 1-1). In these situations, *S. alterniflora* or *J. roemerianus* located on the levee appears healthy and the dieback has occurred in areas

commonly dominated by short-form *S. alterniflora*. These scallop-shaped areas are actually raised berms located at a higher elevation than the rest of the marsh and are likely areas with high rates of sediment deposition. The surface of the berms is often dry and cracked or covered with precipitated salt "nodules" and is almost always riddled with fiddler crab burrows. Although berm dieback areas are found in many areas along the coast, they seem to be most common on or near the barrier islands.

Other dieback areas do not fit these distinctive patterns (Figure 1-1). Dieback sometimes occurrs in low-lying, waterlogged areas of the marsh. These interior, "midmarsh" dieback areas are usually small, are relatively uncommon and are reminiscent of 'panne' dieback described previously (Goodman et al., 1959; Linthurst and Seneca, 1980; Mendelssohn and McKee, 1988, de Souza and Yoch, 1997). Marsh vegetation along the upland border also dies back in some locations. Typically, this type of dieback affects *S. alterniflora* and may be related to accumulation of wrack. However, wrack was not always present at these sites and regrowth has not occurred in at least one large upland dieback that has been regularly observed. Dieback of high marsh species immediately adjacent to small marsh hammocks has also been observed (Meredith Devendorf, personal communication), but none of these areas were included in this study.

Three areas along the Georgia coast experienced extensive dieback. The largest of these sites is located along the Jerico River on the border between Liberty and Bryan Counties (Figure 1-2). More than 600 acres of *S. alterniflora* and *J. roemerianus* marsh has been reduced to mudflat at this site, from the tidal creeks to the upland edge of the marsh. In addition, *Scirpus* dieback was recently observed at this site (Fred Hay, personal communication). A second large dieback site encompasses the upper reaches of the North and South Newport Rivers in Liberty

County. In this site, vegetative dieback is less extensive than in the Jerico River and is often confined to the edge of the creek bank. The third large dieback area is located near Harriet's Bluff and Burrell's Creek in Camden County. Like the Newport River site, this site is not completely denuded, but is characterized by a large area of creek bank dieback. Each of these large dieback areas is located at the boundary between *S. alterniflora* and *J. roemerianus* dominated marshes and encompasses extensive dieback of both species.

Smaller dieback areas take on various forms and are widely distributed along the coast (Figure 1-2). These sites are usually isolated patches of creek bank or berm dieback in *S. alterniflora* marshes, but sometimes occur in the midmarsh or along the upland border of the marsh as well. Reports of small dieback areas have been more common in Chatham County, although this trend is likely related to higher population densities in this area (so there are more people to notice problems). Aerial surveys conducted in 2002 and 2003 revealed that small dieback areas are common throughout the coastal region (personal observation). However, dieback areas seem to be concentrated along the barrier islands and at the inland reaches of estuaries, with fewer dieback areas in the vast expanses of marsh in between or in riverine estuaries (Figure 1-2). This observation suggests that proximity to upland habitat or freshwater inflow could be important factors determining the location of dieback areas.

Preliminary Studies

A team of researchers from the Georgia Coastal Ecosystems Long Term Ecological Research program (LTER) and the Georgia DNR carried out an initial field survey at the Jerico River dieback site in Liberty County, GA in October 2002 (Appendix I). Vegetation type, stem density, stem height, salinity and faunal densities were recorded along three transects through the

dieback area and one through a nearby healthy reference marsh. All transects were located in areas of marsh dominated by *S. alterniflora*. In the dieback area, live vegetation was sparse and standing dead vegetation was almost entirely absent. Only short dead stubs remained above the surface of the marsh and below-ground tissue appeared to be dead as well. Pore-water salinities ranged from 24-36 PSU and faunal densities were within normal ranges for Georgia salt marshes (for additional observations and results, see Appendix I).

Other efforts to document and describe the dieback event are ongoing. A subset of reported dieback sites are being photographed by the Georgia DNR during monthly aerial surveys in order to document changes over time and identify new sites. Dr. Karen Payne (University of Georgia Marine Extension Service) is working to develop methods to identify dieback areas using GIS to analyze aerial photographs and satellite imagery from before and during the dieback event. On the ground, a monitoring program coordinated by the Georgia Coastal Research Council is documenting changes in the vegetation, fauna and soil pore-water chemistry at 7 dieback sites along the coast on a quarterly basis (GCRC website). As of this writing (Spring 2004), only very limited regrowth of salt marsh vegetation has been observed. However, no new dieback areas have been reported since summer 2003.

Salt Marsh Dieback in Other Areas

Although salt marsh dieback is unprecedented in Georgia, a variety of dieback events have been documented on the Gulf Coast, in other areas of the South Atlantic and in various parts of Europe. The purpose of this section is to review the characteristics and potential causes of these dieback events as an aid in interpreting the observations made in Georgia.

Louisiana has historically experienced high rates of coastal wetland loss associated with both natural and anthropogenic changes, as well as relative sea level rise in the Mississippi River delta (hereafter referred to as historical dieback). Wetland loss rates (of fresh, brackish and salt marshes) have been estimated at 65.6 $km^2 year^{-1}$ (Dunbar et al., 1992) and are controlled in part by salt water intrusion due to relative sea level rise (Burdick et al., 1989). Occurring primarily in the interior portions of *S. alterniflora* salt marshes, this dieback is likely caused by increased depth and duration of submergence leading to reduced conditions, high sulfide concentrations, and plant death (Mendelssohn et al., 1981; Mendelssohn and McKee, 1988; Koch et al., 1990; Wilsey et al., 1992). Investigations of this phenomenon demonstrated that when the marsh surface was raised 20 cm, soils became less reduced, sulfide concentrations decreased and growth of *S. alterniflora*, *Spartina patens*, *Distichlis spicata* and *J. roemerianus* increased (Webb et al., 1995). However, soil pore-water salinity was low in all treatments and controls. These results suggest that submergence, not salinity, was the primary factor controlling historical dieback in Louisiana salt marshes.

Similar salt marsh dieback areas were observed in *S. alterniflora* marshes along the Florida Panhandle between 1990 and 1995, although the cause of these dieback areas has not been identified. Carlson et al. (2001) reported that patches as large as 1 ha in area became chlorotic, wilted and died within a period of one month. Dead vegetation was quickly broken down by snail grazers, although this effect was secondary to the cause of dieback. These dieback areas occurred in low-lying areas of marsh where flooding was more frequent, as was the case for historical dieback in Louisiana. In contrast to Louisiana, however, no primary cause was identified, as salinity, sulfide concentrations, anthropogenic stresses, tide levels and climatic factors were not different between dieback and healthy areas. It may be that this dieback event

was different from historical dieback in Louisiana, but differences in the methods or timing of sampling may also have made it difficult to identify the cause.

In 2000, Louisiana experienced an acute dieback event (also referred to as "brown marsh") that differed from historical dieback because of its large extent and the rapid degradation of salt marsh vegetation. This dieback event affected over 100,000 acres of *S. alterniflora*-dominated salt marsh throughout the Mississippi River deltaic plain, but did not affect *J. roemerianus* or *Avicennia germinans* (McKee et al., 2004). Between May and October 2000, affected areas showed a progression from yellow to brown leaves to bare mud as *S. alterniflora* died and decomposed, usually in interior portions of the marsh. The acute dieback event coincided with Louisiana's worst drought in 100 years (J. Grymes, Louisiana State Climatologist, as cited in McKee et al., 2004). An extensive research effort evaluated potential causes of the brown marsh, including increased interstitial salinity, climatic factors, pathogens, herbivores and toxic metals (Stewart et al., eds., 2001). Although the mechanism of dieback has not been clearly identified, McKee et al. (2004) suggest that severe drought and low tide levels were the ultimate causes of this acute dieback event. Evidence from field samples suggests that soil desiccation occurred and that acidification and subsequent increased bioavailability of the toxic metals Fe and Al were likely the proximate mechanism that caused the plants to die (McKee et al., 2004). These observations were supported by high levels of Fe and Al in some plant tissue samples as well as studies on the relative susceptibility of different plants to changes in salinity and pH (Irv Mendelssohn, personal communication).

Sudden salt marsh dieback has also occurred during a severe drought in 2002-2003 on the Herring and Red Rivers in Cape Cod, Massachusetts. *S. alterniflora* was affected along creek banks and high marsh vegetation (*Distichlis spicata*) was sometimes affected adjacent to marsh

hammocks (Ron Rosza, http://alpha.marsci.uga.edu/coastalcouncil/Presentations/1). Dieback areas occurred in the mid-estuary of each river, and were not observed either upriver in brackish marshes or near the mouth. Analyses of soil pore-water chemistry, pathogens or other potential causes have not been carried out. (It should be noted that *Littoraria irrorata* does not occur as far north as Cape Cod.)

In the Lower Cape Fear estuary of North Carolina, salt marsh dieback areas appeared in 1975 associated with dredging activity. Although both *S. alterniflora* and *J. roemerianus* dominate the marsh in this area, only *S. alterniflora* was affected (Linthurst and Seneca, 1980). Dieback areas were located near frequently dredged channels and were relatively low in elevation. Despite being covered with approximately 15 cm of water most of the time, two of the sites occasionally dried out during periods of lower-than-average tides and precipitation, leaving a crust on the surface of the marsh. At these two sites, soil pH varied from 6.3 – 7.1 during flooding but was very acidic during dry periods when pH values of 4.2 and 4.9 were observed. Eh was negative at all three study sites, indicating reduced soil conditions existed in the dieback areas. Unfortunately, the authors did not compare dieback areas to nearby healthy areas.

Although Linthurst and Seneca (1980) did not determine the specific mechanism causing the Lower Cape Fear dieback event, their restoration efforts provide us with important information for revegation of dieback areas. Plant vigor was an important factor determining the success of transplants, as sprigs with larger culms tended to grow more. They suggest that vigorous culms may have more stored nutrients and larger aerenchyma systems than other plants, making them more suitable for transplanting into reduced sediments. Sprigs from sites similar in elevation to the dieback sites also tended to perform better than those from areas with different

hydrology. Most importantly, however, natural recolonization occurred from seed, which was much more important than sprigging attempts. Natural reseeding achieved near 100 percent revegetation within four years of the onset of dieback. Linthurst and Seneca (1980) concluded that dieback events in the Lower Cape Fear estuary were short-term phenomena and that without knowing the causes of dieback, little could be done to speed recovery.

Dieback has also been seen in response to changes in freshwater flow. In the Cooper River estuary in South Carolina, dieback of *S. alterniflora* marshes was reported in 1992 during a time when water was diverted from the Cooper River resulting in decreased freshwater input into the estuary (Jim Morris, personal communication). Dieback areas were similar to historical dieback in Louisiana, occurring in waterlogged, reduced sediments (de Souza and Yoch, 1997). De Souza and Yoch (1997) observed consistently higher pore-water salinities in dieback areas than in comparable healthy areas, but no differences in soil pore-water sulfide or dissolved ammonium concentrations, microbial biomass or organic matter. Bacterial acetylene reduction activity was reduced in dieback areas compared to nearby healthy areas, but this difference was attributed to the lack of factors associated with healthy *Spartina* plants (deSouza and Yoch, 1997).

Salt marsh dieback events have also affected other species of *Spartina*. In Lymington Estuary, Great Britain, 500 acres of *Spartina townsendii* marshes were affected by a well-documented dieback event and 90 acres of the dieback area were completely denuded (Goodman et al., 1959). Dieback areas typically occurred along channels or in low-lying pan areas where the soil was soft, saturated and had high organic content. Healthy plants transplanted into dieback areas died, whereas controls that remained in the healthy area were unaffected, and moribund plants from dieback areas recovered when grown in sand culture. Sulfide or some

other toxic reduced inorganic ion in the substrate was thought to have caused the dieback, although tests could not confirm this hypothesis (Goodman and Williams, 1961).

Other marsh plants have been affected as well. Decline of the reed *Phragmites australis* has been studied in brackish marshes along the Adriatic coast of Northern Italy. Symptoms of reed dieback include a clumped habit, stunting and death of roots and shoots, weakened stems, impeded aeration of underground tissue due to callus development, blockages within the vascular systems, lignification and suberisation of the lateral and apical regions of adventitious roots and lower levels of starch in rhizomes (Armstrong and Armstrong, 2001). Although freshwater *P. australis* marshes in central Europe often show signs of decline associated with eutrophication, Fogli et al. (2002) did not find eutrophication to be a major cause of dieback in a brackish marsh. The authors concluded that high sulfide levels in permanently waterlogged soils accounted for blockages in aerenchyma channels and low rates of net CO_2 exchange and reduced energy storage, and may have been responsible for dieback of *P. australis* in Mediterranean wetlands.

Although these studies describe dieback of a variety of species in many locations, there are important common characteristics among them. In most studies, marsh dieback occurred in low-lying areas of the marsh where soils tended to be waterlogged. Sometimes, but not always, a reduced soil environment with high sulfide concentrations was observed. Dieback also usually occurred in the interior of the marsh, with the exception of channel dieback in Lymington estuary, Great Britain, where increased inundation was a probable cause of dieback (Goodman et al., 1959). Finally, when transplant studies were conducted, survival was low to moderate in dieback areas. Acute dieback in Louisiana was unique in that dieback may have been caused by desiccation of the interior marsh, as opposed to waterlogging stress as observed in historical

dieback (McKee et al., 2004). These studies of salt marsh dieback at other locations yield important insights for studying marsh dieback in Georgia.

Potential Causes of Dieback in Georgia

A variety of hypotheses have been proposed to explain the cause of salt marsh dieback in Georgia. These hypotheses include point or non-point source pollution, fungal pathogens, snail herbivory and drought related effects. It is important to remember that any potential cause for dieback must account for the patterns of dieback observed throughout the coastal zone and that multiple causes could have interacted to cause dieback.

Point source pollution was one of the original hypotheses put forth to explain marsh dieback in Georgia. The first (and largest) dieback site reported is located in the Jerico River near two potential sources of pollution: a bridge construction site on Interstate 95 and railroad tracks a short distance upstream where herbicides are sometimes applied. However, as additional dieback sites were reported, it became apparent that the dieback affected vegetation throughout the coastal zone in both heavily developed areas and relatively pristine areas such as Sapelo Island National Estuarine Research Reserve. The widely distributed pattern of dieback areas does not support point-source pollution as the cause of marsh dieback, unless there is a source associated with each individual site. In addition, analyses of soil samples revealed that heavy metal concentrations at dieback sites were in normal ranges (Mac Rawson and Gerard Krewer, personal communication). Although other pollutants (such as herbicides) were not tested for, it seems highly unlikely that point-source pollution caused the current salt marsh dieback event in Georgia.

Non-point source pollution has not been directly investigated as a potential cause of dieback, but the limited evidence available suggests that it was not an important cause. Non-point source pollutants (such as fertilizers) are often found in run-off and are carried to estuaries via rivers. If non-point source pollutants were the cause of salt marsh dieback, affected areas should be located near major rivers. However, the exact opposite pattern occurred in Georgia, in which the most extensive areas of dieback are located in estuaries with little freshwater input. Non-point source pollutants could have originated closer to the dieback areas, but the severe drought conditions experienced in Georgia prior to and during the dieback event would have prevented the transport of pollutants into estuaries via run-off. On the other hand, drought could also serve to concentrate pollutants by reducing the flushing rates and increasing the residence time of pollutants. Such concentration of pollutants seems unlikely, however, considering that tests for heavy metals were negative.

Plant pathogens, particularly fungi, also have the potential for causing salt marsh dieback. Numerous fungi make up the decomposer communities of *S. alterniflora* and *J. roemerianus* (Newell, 2001; Kohlmeyer et al., 1999). One possible mechanism for fungal-induced dieback could be the introduction of a new fungal pathogen to east coast salt marshes or migration of an existing salt-tolerant pathogen into estuaries during the drought. Samples of decaying above- and below-ground *S. alterniflora* collected in October 2002 at the Jericho River dieback site did not contain any unusual components in the fungal community when analyzed by both microscopy and molecular methods (Steve Newell and Mary Ann Moran, personal communication) although it is possible that a pathogen that was present in low abundances or that was not targeted by these methods would have gone undetected. These analyses were focused on ascomycetes and it is possible that a pathogenic member of another fungal group,

such as oomycetes, was present. A study of the oomycete community along a transect from a dieback area into a nearby healthy area is ongoing (David Porter, personal communication). It should be noted that a single fungal pathogen would need to be a generalist that infects both *S. alterniflora* and *J. roemerianus* or else multiple fungi would have to be involved. In Louisiana and Florida, species of *Fusarium* that commonly occur in salt marshes were isolated from dieback areas, but pathogenicity has not been demonstrated (Carlson et al., 2001; Schneider et al., GCRC website). Another potential mechanism by which a microbial pathogen could be a problem would be a case of increased susceptibility to a common species when marsh plants are stressed by other factors such as drought or increased pathogenicity in saltier conditions. Not enough is currently known to evaluate the role of microbial pathogens in salt marsh dieback in Georgia.

Another potential cause of salt marsh dieback in Georgia is grazing by the common periwinkle snail, *Littoraria irrorata*. One of the dominant grazers of *S. alterniflora* in Georgia salt marshes, *L. irrorata* occurs at densities that range anywhere from 0 to as high as 2,112 m^{-2} (Dale Bishop, personal communication). Although traditionally considered a strict detritivore, *L. irrorata* can also graze live plant tissue (Silliman and Zieman, 2001). Snails can also cause reductions in plant growth by damaging tissue and facilitating the growth of invasive fungi (Silliman and Newell, 2003). When they occur in high enough densities (600 m^{-2} or more) they can reduce a salt marsh to bare mud in a period of 8 months (Silliman and Bertness, 2002). Although *L. irrorata* densities are generally high on the barrier islands, they are usually much lower at inland sites such as the Jerico River, the most extensive dieback site (Dale Bishop, personal communication). At this site, snail densities of 0 to 48 m^{-2} were observed by LTER scientists in October 2002, far below the moderate density used by Silliman and Bertness (2002)

to demonstrate the impacts of *L. irrorata* grazing on *S. alterniflora* (600 m^{-2}). Although the effects of snail grazing on *S. alterniflora* have not been determined for lower densities, *L. irrorata* commonly occurs on *S. alterniflora* without causing mortality.

It has been suggested that a decline in blue crab populations may have released snails from predation pressure, allowing populations to explode and graze down the marsh (Silliman and Bertness, 2002; Bertness et al., 2004), but there is limited evidence to support this hypothesis. In 2001, at the onset of marsh dieback, the catch of hard shell blue crabs in Georgia reached a record low of 2.70 million pounds, well below the 20 year average of 6.85 million pounds (CRD, 2004). To relate the decline in blue crab catch to snail grazing rates, it is essential to know whether blue crabs are a major predator of *L. irrorata*. Tethering experiments have shown that predation on *L. irrorata* in Georgia salt marshes is high when marine predators are abundant (tidal creeks) and lower when they are excluded by dense vegetation (stands of high marsh *S. alterniflora*) (Silliman and Bertness, 2002). However, the species of predator was not determined and there are no data available to determine the quantitative importance of blue crabs as predators of *L. irrorata*. It is also necessary to show that the decline correlated with an increase in *L. irrorata* densities. In Georgia salt marshes, *L. irrorata* densities may have increased from fall 2000 to fall 2002 (Ogburn et al., 2002), but densities have subsequently decreased (Dale Bishop, personal communication) even though crab catch continues to decline (CRD, 2004), suggesting that *L. irrorata* densities may not be controlled by blue crab predation. Even if data supporting the considerations outlined above were available, other potential factors affecting snail populations (such as other predators, climatic factors and recruitment patterns) would need to be evaluated before determining whether the decline in blue crab catch is related to marsh dieback.

Georgia experienced a severe drought concurrent with the onset of salt marsh dieback. The 3-year period leading up to the dieback, from 1999 – 2001, was the driest 3-year period in 108 years of record-keeping in Atlanta, GA (NCDC, 2002). State-wide, average total precipitation was 10.57 inches below normal for the 2002 water year (October 2001 – September 2002) (USGS, 2002). An important consequence of drought in Georgia is that freshwater input to the coastal zone decreases significantly, as nearly all of the surface water originates from rain that falls within the state. Hence, freshwater inflow to the estuaries is not buffered by rainfall in distant regions as is the case for larger systems like the Mississippi River. During the 2002 water year, when much of the dieback occurred, streamflow in all of Georgia's rivers was well below average (USGS, 2002). In the Altamaha River, one of the major rivers that empty into coastal Georgia, streamflow was measured at only 17 percent of the long term average (USGS, 2002). Reduced freshwater input and increased salinities had various effects on coastal ecosystems, including an epidemic of *Hematodinium* (a dinoflagellate that causes bitter crab disease), which is adapted to high salinities. The drought, however, did not affect the average monthly water level at the coast (NOAA, http://co-ops.nos.noaa.gov/sltrends/residual1980.shtml?).

There are several mechanisms by which drought might be related to salt marsh dieback. These include: 1) Pore-water salinity may have increased beyond the tolerance limits of salt marsh plants; 2) Reduced pore-water flushing rates may have enhanced anaerobic conditions and/or resulted in the accumulation of toxic compounds in water-logged sediments; 3) Temporary sediment desiccation may have reduced water availability and/or caused soil acidification; 4) Indirect effects of increased salinity may have enabled the proliferation of salt tolerant pathogens as described above; or 5) Drought stress may have weakened plant defenses to

herbivores or pathogens. Any or all of these mechanisms may have contributed to marsh dieback, but available data are insufficient to rule out any of these hypotheses.

The work described in this thesis was carried out in order to describe the salt marsh dieback phenomenon in Georgia. Chapter 2 reports a field survey conducted to document patterns and conditions in a variety of dieback areas along the coast. Chapter 3 reports the results of transplant experiments conducted to evaluate the viability of soil in dieback areas and to determine how soil conditions affect transplant survival and growth. In the conclusions, I revisit these various hypotheses and suggest areas requiring further evaluation.

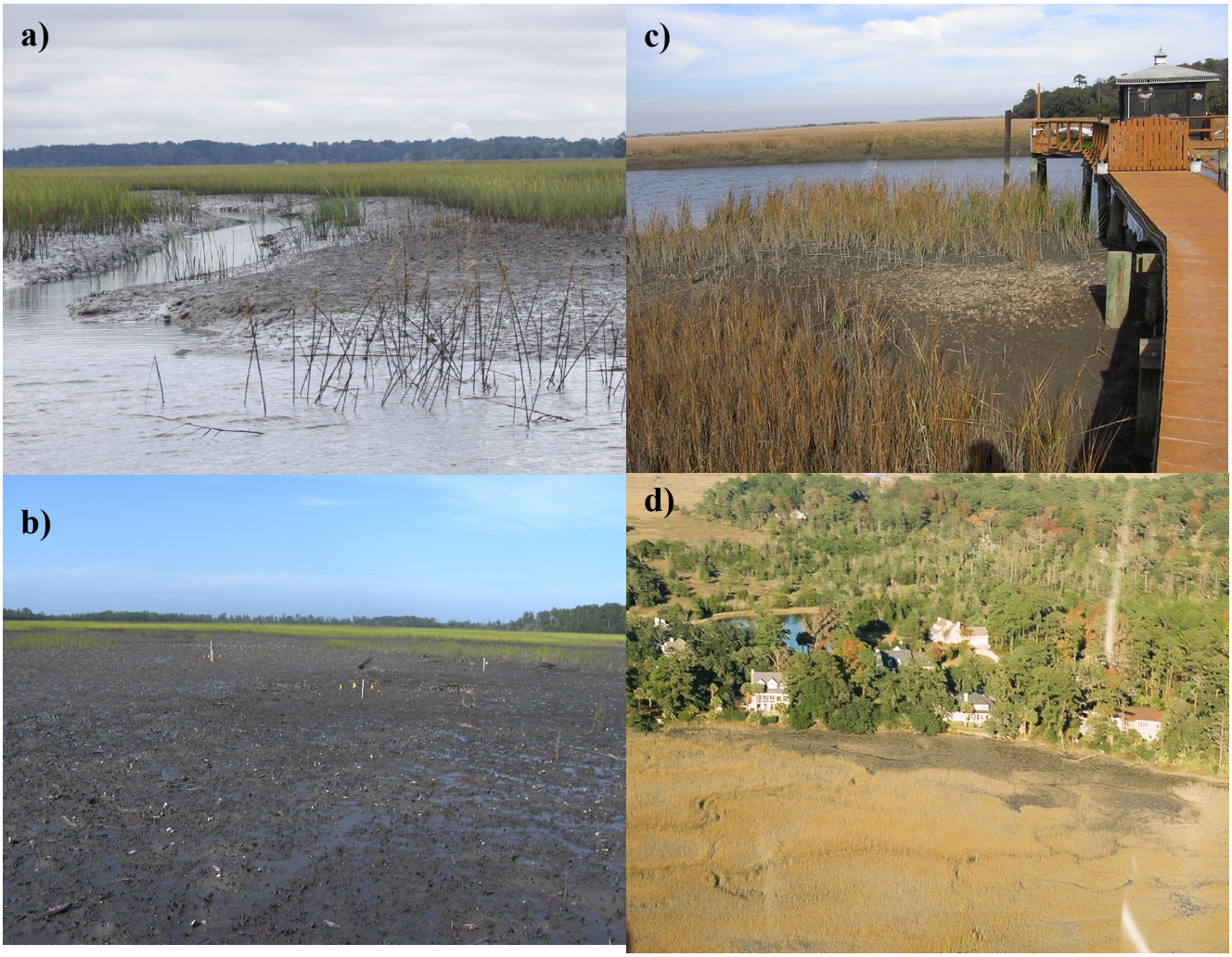

Figure 1-1. Patterns of salt marsh dieback in Georgia (Photos by Matt Ogburn except where noted): a) creek bank dieback; b) midmarsh dieback; c) berm dieback (photo by Nevy Clark); and d) upland dieback.

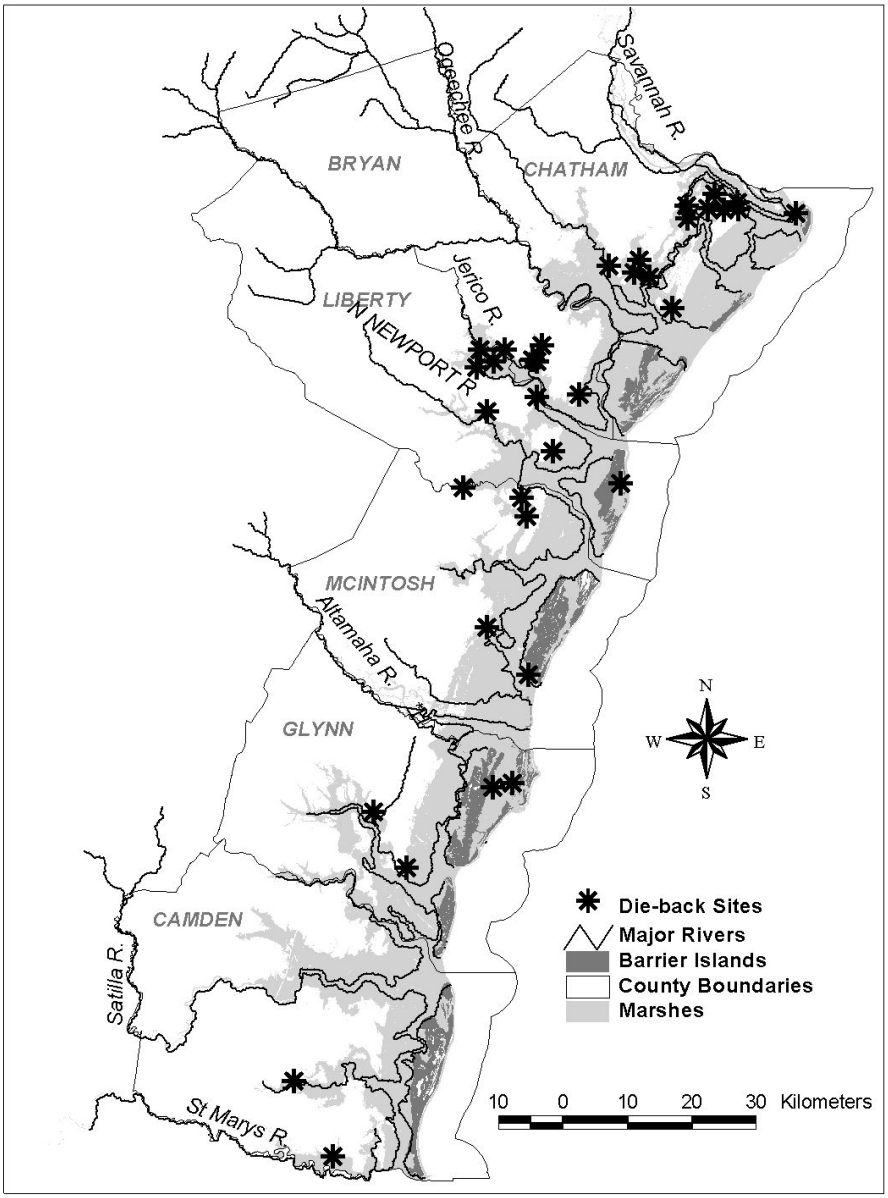

Figure 1-2. Map of salt marsh dieback sites reported to DNR before June 2003.

CHAPTER 2

FIELD SURVEY

Introduction

In 2002, as news of the marsh dieback event spread in coastal Georgia, residents began reporting dieback areas to the Coastal Resources Division of the Georgia Department of Natural Resources (DNR). By June 2003, 37 dieback sites had been reported (Figure 1-2). These sites were widely distributed from north to south, with areas reported in all six coastal counties. Many of the reported dieback sites were concentrated in Chatham County, near Savannah, but this trend may reflect a reporting bias, as population densities are much higher in this area. Dieback sites were also distributed from the westernmost, inland reaches of estuaries to the barrier islands. Early aerial surveys suggested that differences may exist between dieback areas at inland and barrier island sites, with creek bank dieback more common at inland sites and berm dieback more common on barrier islands (personal observations).

When dieback areas were reported, residents often described differences they had noticed as the dieback occurred. Some residents reported seeing differences in elevation, with dieback occurring on mounds that seemed to "rise up" as the dieback proceeded. Others reported that dieback first appeared around the edges of small marsh hammocks, indicating that terrestrial influence may have been important. At some locations, residents reported seeing higher snail densities than usual, although others reported seeing fewer. Many of the dieback areas were located near man-made structures such as docks, dikes, bridges and causeways that could potentially serve as hydrologic barriers. This observation may also be the result of reporting bias, as dieback areas near such structures are probably more likely to be observed and reported.

This chapter describes a survey conducted in order to identify patterns associated with salt marsh dieback in coastal Georgia. I was interested in determining the prevalence and spatial distribution of dieback areas with respect to the plant species, the area affected, the portion of the marsh affected (the dieback pattern), the presence of man-made structures and the importance of terrestrial influence at each site. I was also interested in determining differences in elevation, vegetation, faunal and soil characteristics between dieback areas and nearby healthy areas.

Methods

Eighteen dieback sites were chosen out of a total of 37 areas that had been reported by coastal residents to DNR as of June 2003 (Figure 2-1). Survey sites were distributed from the northern to the southern border of the Georgia coast. Sites were also distributed from east to west, with 5 sites on barrier islands and the remaining 13 further inland. The majority of sites (16) were in marshes dominated by *S. alterniflora*, but 2 sites had *J. roemerianus* die-offs as well. All sites were visited once between June and August, 2003 during low tide.

The sampling protocol used in this survey was adapted from the long term monitoring program developed by the Georgia Coastal Research Council Marsh Dieback Committee (Appendix II). Within each dieback area, a study site was chosen that was both representative of the dieback area as a whole and was accessible. Each dieback site was compared to a nearby control site, which was the closest area of healthy vegetation that resembled the dieback area with respect to plant species, elevation, distance from creeks and soil saturation. The distance between dieback and healthy areas was usually only a few meters and was never more than 20 m. Floral, faunal and physical characteristics were measured in randomly placed 0.5 m x 0.5 m plots. Four plots were sampled in both the dieback and healthy areas at each site.

Data were analyzed using a 2-way Analysis of Variance (ANOVA) using site and status (healthy or dieback) as fixed effects to determine whether there were overall differences in any of the measured variables (stem density, stem height, salinity, etc.) ($\alpha = 0.05$). For variables with a significant interaction, survey sites with significant differences between the healthy and dieback area were identified using Tukey's post hoc test. T-tests were used to examine geographic (inland versus barrier island sites) and species differences (sites with *J. roemerianus* versus *S. alterniflora*). P-values are reported in the text for variables with significant differences ($\alpha = 0.05$) for reference only. Reported values are mean ± 1 standard deviation.

General Characteristics

The area of dieback, the dieback pattern, the relative elevation of each site, the proximity to man-made hydrologic barriers and the degree of terrestrial influence were each classified, as described later. Each dieback site occurred in a well-defined area (i.e. the border between healthy vegetation and bare mud was obvious). Although precise measurements of area were not possible, dieback sites were classified into the size categories based on visual estimates: <1 acre, 1-10 acres, 10-100 acres, or >100 acres.

The patterns of dieback were categorized depending on the portion of the marsh affected. These categories were: 1. "Creek bank dieback" were cases in which dieback occurred in a 1-5 m wide strip along the edges of tidal creeks in the marsh. 2. "Midmarsh dieback" were cases where dieback occurred in low, well-saturated, interior sections of marsh usually surrounded by live vegetation. 3. Dieback was called "berm dieback" when it occurred on elevated mounds above the primary marsh surface. These areas were set back from the creek bank (plants were often healthy on creek edges and between berms), and tended to be covered with a dry, cracked

surface. 4. Areas of dieback along the terrestrial border of the marsh were classified as "upland dieback" (Figure 1-1). In some cases, more than one pattern was present within a dieback area.

The degree of terrestrial influence was estimated using a rank order system that has been used to study the distributions of and interactions between marsh flora and fauna (Steve Pennings, personal communication), where a value of 0 = no terrestrial influence, 1 = adjacent to a thin peninsula (minor terrestrial influence), 2 = upland forms convex border with marsh, 3 = terrestrial border is a straight line (moderate influence), 4 = upland forms concave border with marsh and 5 = nearly surrounded by upland (strong terrestrial influence). Sites were determined to have man-made hydrologic barriers if they were located within 50 m of bridges, causeways, dikes and docks. Relative elevation of dieback and healthy areas was determined visually (i.e. was the dieback area higher or lower than nearby healthy marsh).

Flora and Fauna

Within each 0.5 m^2 quadrat, information was collected on the characteristics of both live and dead *S. alterniflora* and *J. roemerianus*, the two dominant plant species encountered in dieback areas. Live plant stems were counted and classified as > 15 cm (mature stems) or < 15 cm (new shoots), and the density of dead stems was also recorded. These were defined as standing dead stems or remnant stubs that fully protruded above the marsh surface. The heights of the five tallest live stems were also recorded.

The densities of the most abundant marsh epifauna (snails, mussels, and crabs) were recorded in each quadrat. *Littoraria irrorata* were grouped into 2 size classes >10 mm or < 10 mm. This distinction is important because we assumed that snails <10 mm are unable to graze on *S. alterniflora* (Silliman and Bertness, 2002). *Geukensia demissa* were counted and classified

as live or dead. The density of adult *Uca* spp. (*U. pugilator*, *U. pugnax* and *U. minax*) and other crabs (e.g. *Sesarma* spp., *Panopeus* spp.) was estimated by counting the number of crab holes in the surface of the marsh that were greater than 5 mm in diameter. This method is similar to the method used by GCE-LTER scientists to monitor populations of marsh crabs and is an effective way to estimate crab density (Dale Bishop, personal communication).

Soil

Physical characteristics of the soil immediately adjacent to each quadrat were measured at a depth of 10 – 15 cm, which corresponds to the rooting depth of salt marsh plants. To sample pore-water, a shallow well that measured approximately 15 cm deep and 5 cm in diameter was dug into the marsh surface immediately adjacent to each quadrat. Pore-water was allowed to drain into the well for several minutes, and care was taken to prevent surface water from entering. Salinity was measured using a Leica model 10419 temperature-compensated refractometer. pH was determined by inserting a Fisher Scientific accumet® pH probe (13-620-AP50) directly into the well. Temperature was recorded from a soil temperature probe inserted 10 cm into the substrate and allowed to equilibrate for several minutes. Redox measurements were taken at the marsh surface (2 cm depth) and in the rooting zone (15 cm depth) at 13 of the sites using a Mettler-Toledo Combination Redox Electrode (Pt4805-SC-DPAS-K8S/200) attached to a Fisher Scientific accumet® AP62 portable pH/mV meter. A correction factor of +225 mV was added to measured values to account for the potential of the Ag/AgCl reference electrode.

Results

General Characteristics

The majority (16) of the dieback sites surveyed were in *S. alterniflora*-dominated marsh. Only two sites contained *J. roemerianus* (Isle of Hope had both *S. alterniflora* and *J. roemerianus* and Isle of Wight Rd. had only *J. roemerianus* [Table 2-1]). Both of these sites are located in the inland reaches of estuaries (IH and IW in Figure 2-1). Twelve of the 18 dieback sites included in this survey covered less than 1 acre, although sites ranged to as large as 600 acres (Table 2-1). Every size category of dieback area was represented in *S. alterniflora* dominated marshes, whereas only the smallest two size classes were represented in *J. roemerianus* dominated marshes (larger *J. roemerianus* dieback areas have been observed but were not included in this study). Small dieback areas were evenly distributed from north to south and from east to west, whereas large dieback areas were confined to two general areas of the central and southern coastal region and to the western, inland portions of the estuaries. Two of the largest sites were located in Liberty County on the Jerico and North Newport rivers, and a third was further south, near Harriet's Bluff in Camden County (JE, MB and HB in Figure 2-1). These three areas of extensive dieback are located in the two sections of coastal Georgia that are furthest from major rivers. These extensive dieback areas were also located at the upstream end of estuaries near the transition from *S. alterniflora* dominated to *J. roemerianus* dominated marshes.

The creek bank pattern of dieback was most common in the sites surveyed, occurring in 13 sites, whereas midmarsh, berm and upland die-offs were found in 8, 4 and 2 sites, respectively (Table 2-1). Six sites exhibited more than one pattern of dieback, with the largest sites possessing the greatest variety of dieback patterns. The creek bank, midmarsh and berm dieback

patterns were observed in both *S. alterniflora* and *J. roemerianus* dominated marshes, whereas upland dieback only occurred in *S. alterniflora* dominated marshes. There were no trends in the distribution of creek bank or midmarsh sites along either a north-south or east-west axis. The berm and upland dieback types, however, were only observed in the north-central part of the coast and towards the western border of the coastal region.

Elevation differences between paired dieback and reference areas were observed at 10 of the 18 sites, although no consistent trends were detected. Dieback areas were noticeably lower than adjacent live areas at 8 sites, whereas dieback areas were raised at 2 of the sites (Table 2-3). The dieback area in one of the *J. roemerianus* dominated marshes (Isle of Hope) was at a lower elevation than the reference site, whereas at the other *J. roemerianus* area (Isle of Wight Rd.) there was no obvious difference. Dieback areas in *S. alterniflora* dominated marshes had no consistent patterns in terms of elevation. This result is not surprising because the different dieback types were associated with different elevational differences, (berm dieback areas are always higher than nearby healthy marsh, whereas creek bank diebacks tend to be lower). There were no apparent patterns with regard to elevation in terms of the spatial distribution, size or severity of dieback sites.

Man-made hydrologic barriers were observed in close proximity (<50 m) to 11 dieback sites (Table 2-3). This is likely an over-estimate of the prevalence of man-made structures near dieback areas on the coast in general, as dieback sites near bridges, causeways and docks are more visible, and hence more likely to be reported, than those further away from human habitation. The other 7 sites were located in areas of marsh without an apparent alteration. There were no obvious relationships between the presence of man-made structures and any of the other factors included in the survey.

Terrestrial influence at dieback areas was categorized as moderate in the majority of sites, although some sites had no connection to upland areas at all (Table 2-3). Of the study sites, 10 exhibited moderate terrestrial influence (scored as 3), 3 had moderately high terrestrial influence (scored as 4) and 1 had low terrestrial influence (scored as 1). Four sites were separated from upland areas by large tidal creeks and were scored a 0 to indicate that they had no terrestrial influence. There were no obvious relationships between terrestrial influence and any of the other factors included in the survey.

Vegetation

The stem density of live vegetation (either *S. alterniflora* or *J. roemerianus*) was by definition much lower in dieback areas than in adjacent healthy areas, with an overall average of 186 ± 101 stems m^{-2} in healthy areas and 30 ± 55 stems m^{-2} in dieback areas. The effects of site, status and the interaction of site and status were all significant for both stems >15 cm and <15 cm (Table 2-2). The density of both stems >15 cm tall as well as new shoots (<15 cm) was significantly higher in healthy areas as compared to dieback areas when averaged across all sites for both *S. alterniflora* and *J. roemerianus* (Table 2-3). However, *J. roemerianus* sites had significantly fewer ($p = 0.02$) new shoots (9 ± 14 stems) than *S. alterniflora* sites (47 ± 65 stems). There was no significant difference in the density of stems >15 cm between species. There were also no significant differences in live stem density in healthy or dieback areas between inland and barrier island sites, and no apparent trends in relation to size of dieback area, severity of dieback, dieback pattern, relative elevation of dieback and healthy areas, presence of man-made structures or degree of upland influence. For stem height, the effects of site and status were significant, but the interaction between site and status was not (Table 2-2). The overall

height of the five tallest live stems was significantly higher in healthy areas (81 ± 29 cm) than in dieback areas (37 ± 20) (Table 2-4), with no significant differences between species or between inland and barrier island sites. The effects of site, status and site x status were all significant for dead stems (Table 2-2), with 8 sites having significantly more dead stems in the dieback area (Table 2-3). Overall, there were consistently more dead than live stems at all sites in both healthy and dieback areas and significantly more dead stems in dieback than healthy areas (Table 2-3). Comparing species, *J. roemerianus* sites (451 ± 432 stems) had significantly more ($p = 0.004$) dead stems than *S. alterniflora* sites (250 ± 240 stems).

Fauna

L. irrorata were observed at 15 of the 18 sites (although 2 of these had only 2 snails each) (Table 2-5). The effects of site and site x status were significant, whereas the effect of status was not (Table 2-2). The density of snails of all sizes ranged from 0 to 408 m^{-2} (the highest individual measurement was 624 m^{-2}) with no significant differences in average density between healthy and dieback areas or between plant species (Table 2-5). Within sites, there were significantly more snails in the healthy area at Delegal Creek Marina and Sapelo Island, whereas the density of snails was significantly higher in the dieback area at Highway 17 (DE, SA and 17 in Figure 2-1) (Table 2-5). There were no obvious differences in the spatial distribution of large (>10 mm) and small snails (<10 mm) (i.e. where small snails were abundant, large snails were also abundant), indicating that adult densities may reflect recruitment patterns. Snails were seen at most healthy sites (14 out of 18), but only 11 dieback areas (Table 2-5). Heavy snail damage was only observed at one site (DE in Figure 2-1). Densities only exceeded 100 m^{-2} in 3 dieback areas and 6 healthy reference areas. Of these areas with relatively high density, 2 of the 3

dieback areas and 4 of the 6 healthy areas were at barrier island sites. However, when average snail densities were compared between barrier island and inland sites, densities were significantly higher ($p < 0.001$) at barrier island sites (138 ± 181 m^{-2} vs. 34 ± 91 m^{-2}). High snail densities (>100 m^{-2}) were never observed at the largest dieback sites (>10 acres). Snail density may have been related to the portion of marsh affected (dieback pattern), as high densities occurred in 5 of the 8 sites characterized as midmarsh dieback, whereas high densities only occurred in 3 creek bank dieback areas and did not occur in berm or upland dieback areas. This pattern follows observations that snail densities are generally higher in interior portions of the marsh than near tidal creeks (Silliman and Bertness, 2002). There were no patterns relating snail density to north-south distribution, elevation, presence of man-made structures or degree of terrestrial influence.

The effects of site, status and site x status were significant for crab hole density, but for *G. demissa*, only the effect of site was significant (Table 2-2). Crab hole density ranged from 0 to 848 m^{-2} with an average of 148 ± 152. Within sites, crab hole density was significantly higher in the dieback area at 4 sites (TY, MO, SA and SI in Figure 2-1) (Table 2-6). The density of live *G. demissa* ranged from 0 to 88 m^{-2} and averaged 6 ± 13 m^{-2}. There were no obvious relationships between crab hole or *G. demissa* density and plant species, spatial distribution, or any of the other factors included in this study.

Soil

Pore-water salinity ranged from 10 to 48 PSU (the highest individual observation was 50 PSU) and averaged 25 ± 9 PSU across all sites (Table 2-7). The effects of site and status were significant, but the interaction between site and status was not (Table 2-2). Overall, salinity was

significantly higher in healthy areas than dieback areas and was significantly higher ($p < 0.001$) at barrier island sites (34 ± 7 PSU) than at inland sites (21 ± 7 PSU) (Table 2-7).

The effects of site, status and the interaction between site and status were all significant for pH, but only the effect of site was significant for temperature. Pore-water pH varied from 6.30 - 7.36 within sites and averaged 6.72 ± 0.25, and was slightly but significantly higher in the dieback area at 3 individual sites (TA, OS and CA in Figure 2-1) (Table 2-8). Soil temperature varied from 26 to 32 degrees C, with no significant differences between healthy and dieback areas (Table 2-8).

Redox potential (Eh) was measured at only 12 of the 18 survey locations and was highly variable both within and among sites (Table 2-9). Site, status and the interaction of site and status were all significant (Table 2-2), but Eh was not consistently higher or lower in the dieback area at either depth (Table 2-9). At 2 cm depth, Eh ranged from -15 to 356 mV and averaged 168 ± 144 mV. At 15 cm depth, Eh varied from -94 to 225 mV and averaged 34 ± 107 mV. Eh values indicating reduced soil conditions (<0 mV) were recorded at a total of 7 sites in either healthy or dieback areas, but when reduced conditions were observed, Eh was never lower in dieback areas than in healthy areas (Table 2-9). There were no significant differences in redox potential at 2 cm or 15 cm between inland and barrier island sites.

Discussion

This study surveyed 18 salt marsh dieback sites distributed throughout coastal Georgia. The affected area at these sites was generally small (< 1 acre), although some sites were much larger, including the largest site (JE in Figure 2-1) that was estimated at over 600 acres (Jan Mackinnon, personal communication). *S. alterniflora* was the primary species affected at most

of the sites surveyed (14 sites). Four of the largest dieback areas (JE, MB, IW and HB in Figure 2-1), occurred in the inland parts of estuaries near the border of *S. alterniflora* dominated marsh and *J. roemerianus* dominated marsh, but only one of these contained *J. roemerianus* (IW in Figure 2-1). There were no clear differences in soil characteristics, epifaunal densities or spatial distribution between *S. alterniflora* and *J. roemerianus* sites.

Creek bank dieback was the most common pattern of dieback observed in this study. This dieback pattern is characterized by a 1-3 m wide strip of dieback along the bank of both small and large tidal creeks. Creek bank dieback occurred in 13 sites and affected both *S. alterniflora* and *J. roemerianus*. The high prevalence of creek bank dieback observed in Georgia is in stark contrast to previously reported salt marsh dieback events in which dieback occurred almost exclusively in low-lying, interior parts of the marsh (Goodman et al., 1959; Linthurst and Seneca, 1980; Mendelssohn and McKee, 1988 and de Souza and Yoch, 1997). When *S. townsendii* dieback was observed along creek banks in Lymington estuary, Great Britain, slumping of the bank and increased inundation were proposed as possible causes (Goodman et al., 1959). (Creek bank dieback may be the dominant pattern of dieback in the recent dieback in Massachusetts, but this dieback event has not been studied in detail.) Although some of the creek bank dieback areas in this study were lower in elevation than nearby healthy areas, other creek bank dieback areas were at an equal or higher elevation than healthy areas (Table 2-1). When creek bank dieback areas were at a higher elevation than healthy areas, live tall-form *S. alterniflora* occurred closer to the creek. This result suggests that inundation was not the cause of dieback in these areas.

Since the creek bank dieback pattern observed in Georgia appears to be unprecedented, it is important to consider what its causes might have been. Creek bank areas generally have well-

drained, oxidized soils, moderate salinity and pH, and support tall-form *S. alterniflora* (Nestler, 1977; Smart and Barko, 1980; Howes et al., 1981). Even if the dieback and healthy areas at a particular site are of similar elevation, the creek bank is still well-drained, so that the water table is well below the marsh surface during low tide (Howes et al., 1981). Sediments in this zone could potentially have dried out during periods of low tidal amplitude, high temperatures and severe drought (as suggested by a dry, cracked surface observed at some creek bank dieback areas). Soil desiccation alone could also have been responsible for dieback, and/or drying may have been associated with periods of increased salinity and/or low pH. Increased salinity and low pH were not observed in this study, but could have occurred before this study took place. It is also possible that tall-form plants are less stress tolerant than short form plants found in other parts of the marsh (Silliman and Zieman, 2001), which may have increased their susceptibility to fungal pathogens or herbivores. Although there are no data available to determine if fungal activity was higher along creek banks, increased herbivory is an unlikely cause of creek bank dieback, as snail densities were low at the creek bank and leaf tissue damage was not observed on live vegetation immediately surrounding creek bank dieback areas.

Midmarsh, berm and upland dieback patterns were less common in this study, occurring in 8, 4 and 2 sites, respectively. Midmarsh dieback sites were distributed throughout the coastal zone, but berm and upland dieback areas were only observed in the north-west portions of the coast. The low representation and limited spatial distribution of berm dieback areas are surprising, since aerial surveys indicate that berm dieback areas are common throughout the coastal zone and occur frequently on barrier islands (personal observations). Berm dieback areas may have been less common in this survey because they are less visible from the water (since the vegetation in the creek bank is unaffected) and so were reported less often and were therefore not

included in the list from which survey sites were chosen. Preliminary attempts to delineate marsh dieback sites using aerial and satellite photography support the observations made from helicopters that berm dieback areas are common all along the coast (Karen Payne, personal communication).

Similar to creek bank dieback areas, berm dieback areas are likely to have been caused by soil desiccation. Berm dieback areas were located at a higher elevation than surrounding healthy marsh and were often covered with a dry, cracked surface and precipitated salt "nodules", indicating that desiccation was persistent. High densities of fiddler crab holes at these sites may also have contributed to desiccation by increasing the surface area available for evaporation. Although it is unclear whether these areas regularly dried out prior to the dieback, the presence of dead stems indicates that they had supported live vegetation. As with creek bank dieback, soil desiccation alone could have been responsible for dieback, and/or drying may have been associated with periods of increased salinity and/or low pH. An alternative to this hypothesis is that berm dieback is the result of changes in sediment deposition rates during the drought, as it occurs in areas where deposition rates are likely high (creek bank levees) and clearly follows the spatial pattern of deposition (personal observation). Fungal pathogens may also have been important at these sites, but herbivory likely was not (for the same reasons as at creek bank sites: see above).

The potential causes of midmarsh and upland dieback are more difficult to identify. During acute dieback in Louisiana, evidence of sediment desiccation was observed in interior portions of the marsh (McKee et al., 2004). In Georgia, however, dry soil was only observed at 2 of the 8 sites having midmarsh or upland dieback (DE and IW in Table 2-1). Soil water-logging is a potential cause of dieback at 2 midmarsh sites, where negative redox potentials

indicated anoxic soil conditions (TA and SI in Table 2-1). Heavy snail damage was also observed at one midmarsh dieback site (DE in Table 2-1), but it is unlikely that snail herbivory acted alone to cause dieback (see below). A variety of other factors including fungal pathogens and pollution (in runoff from terrestrial sources immediately adjacent to upland dieback areas) could also have interacted with drought to cause dieback at these sites.

Despite anecdotal reports that changes in marsh elevation occurred at the same time as marsh dieback, there were no consistent differences in this study between the elevation of dieback areas and nearby healthy areas. Ten sites showed no clear elevation differences between dieback and healthy areas. Eight of the dieback areas were noticeably lower than adjacent healthy sites, and 2 were higher in elevation. Although elevation was only estimated and not measured (which would have allowed us to detect subtler differences), the results were not in keeping with previously reported dieback events in which dieback occurred in low-lying, waterlogged parts of the marsh (Goodman et al., 1959; Linthurst and Seneca, 1980; Mendelssohn and McKee, 1988; and de Souza and Yoch, 1997). Half of the sites where dieback was considered lower than reference areas were creek bank die-offs where peat collapse or erosion may have reduced elevation in the two years between the onset of dieback and the time this study was carried out. These observations suggest that if elevation contributed to marsh dieback, its effects were probably not related to inundation or soil water-logging.

Several early observations suggested that dieback areas were more common near man-made structures (such as dikes, docks, bridges and causeways) or in estuaries with limited freshwater flow, and this theory is potentially supported by this study. Man-made structures could affect marsh plants by increasing sedimentation or erosion, disrupting flow of surface or groundwater, or causing thick mats of wrack to accumulate. In some locations (Louisiana, North

Carolina and South Carolina), marsh dieback may have been associated with hydrologic alterations (Mendelssohn and McKee, 1988; Linthurst and Seneca, 1980; James Morris, personal communication). Dieback sites caused by wrack accumulation (mats of wrack present) were intentionally left out of this study because they are probably unrelated to the current dieback phenomenon. Eleven out of the 18 dieback sites surveyed here were located within 50 m of man-made structures, indicating that marsh near man-made structures may be more susceptible to dieback. However, this result must be interpreted with caution, as it seems likely that dieback sites near docks, bridges and causeways were more frequently noticed and reported. Even if there is no relationship between dieback areas and man-made structures, however, surficial groundwater could be related to the location of dieback areas either because it may be affected by local topography (i.e. creek bank vs. midmarsh dieback patterns) or because it may be influenced by freshwater input from rivers. The second of these hypotheses is supported by observations that the largest and most severely affected dieback sites were located in estuaries with very little freshwater input from rivers, whereas marshes in estuaries along Georgia's major rivers were rarely affected by dieback (personal observations). GIS analysis of dieback sites is needed to determine the extent to which dieback areas are associated with man-made structures, topography and estuaries with limited freshwater input.

Upland habitat is a potential source of groundwater to the surrounding marsh. Under severe drought conditions, this fresh water flow would be reduced, potentially resulting in dieback. This hypothesis is supported by reports that the first signs of dieback at Melon Bluff Plantation occurred around the edges of marsh hammocks (Laura Devendorf, personal communication), a pattern of dieback also observed in Massachusetts (Ron Rosza, http://alpha.marsci.uga.edu/coastalcouncil/Presentations/1). However, if this were the case

everywhere, one would expect to see dieback originating in upland sections of marsh and spreading toward the creek bank, and this was not observed. Moreover, there were no obvious trends relating dieback areas to the degree of terrestrial influence. Instead, dieback was most prevalent along creek banks and was only rarely observed along the upland edge of marshes. In fact, four of the dieback sites were separated from any terrestrial influence by large tidal creeks.

It has been suggested that populations of marsh snails may have exploded and grazed down the marsh following a release from predation pressure caused by a decline in blue crab populations in recent years (Silliman and Bertness, 2002; Bertness et al., 2004), but this study found little evidence to support that theory. Although heavy snail damage was observed at one site (DE in Figure 2-1), few snails were present at other sites during this survey and snail damage was rarely observed (personal observations). *L irrorata* densities in this study rarely reached the moderate densities (600 per m^2) used by Silliman and Bertness (2002) and never exceeded densities commonly observed in Georgia salt marshes (Dale Bishop, personal communication). Snail densities were also very low (6 ± 9 m^{-2}) at *J. roemerianus* sites, suggesting that snails are an unlikely cause of *J. roemerianus* dieback. It is possible that snail densities peaked and then declined prior to this survey, but observations made in October 2002 suggest this was not the case at the Jerico River dieback site (GCRC, 2002). A more likely hypothesis is that snails may have contributed to dieback by increasing the size of dieback sites in areas where high snail densities do occur (Brian Silliman, personal communication).

Differences in pore-water chemistry between dieback and healthy areas were rarely observed. As would be expected, salinities were significantly higher near the coast than at inland sites, but there were no differences in salinity, pH or redox potential between dieback and healthy areas. These observations demonstrate that if pore-water differences caused the dieback,

the signal was no longer present. It should be noted that this survey was conducted more than a year after the onset of dieback after substantial late fall and spring rains that marked the end of severe drought. If drought resulted in high salinities or spikes of acidity in the soil of dieback areas (as may have occurred in Louisiana [McKee et al., 2004]), subsequent precipitation and tidal flooding may have ameliorated these conditions. Alternatively, by measuring pH and salinity only in cases when pore-water was present, we may have systematically eliminated high salinity or low pH measurements from our analysis. This possibility seems unlikely, however, since dry soils only prevented salinity and pH measurements at one site. The preliminary survey carried out in the Jerico River in October 2002 (before the end of the drought) also failed to detect elevated pore-water salinities (Appendix I).

Although soil conditions were similar between healthy and dieback areas, very little re-colonization of vegetation was observed in dieback areas even though this study was carried out more than a year after the onset of dieback. There were significantly fewer new stems (<15 cm) in dieback areas, and all of these were associated with surviving live vegetation (personal observations). A total of 7 sites (all of which were included in this study) have been monitored quarterly since June 2003 and, as of December 2003, had shown only very limited re-colonization (GCRC website). However, the size of the areas being tracked during the quarterly monitoring effort have not increased in size either, indicating that dieback is no longer occurring at these locations. Two reasons why re-colonization may not have occurred prior to December 2003 are: 1. The cause of dieback, be it pollution, pathogens, herbivores or drought, is still present in the soil of dieback areas; or 2. Natural re-colonization by *S. alterniflora* and *J. roemerianus* is a slow process and has not had time to operate. Transplant studies, such as those

described in the next chapter, can help shed some light on which of these hypotheses is occurring in salt marsh dieback areas in Georgia.

In summary, this study did not find obvious patterns in soil conditions or epifauna, but did provide evidence against elevated salinities, increased water-logging and snail grazing as causes of dieback in Georgia. The Georgia dieback event is most similar to acute dieback in Louisiana, because both dieback events were widespread and occurred during severe drought. The portions of marsh that were most often affected were different at the two locations (creek bank in Georgia vs. interior marsh in Louisiana) and *J. roemerianus* was not affected in Louisiana, but these differences may be due to differences in hydrology between the two sites. If this comparison is accurate, soil desiccation resulting in periods of low pH and increased bioavailability of Fe and Al may be the cause of salt marsh dieback in Georgia, but there are currently no data available to evaluate this hypothesis.

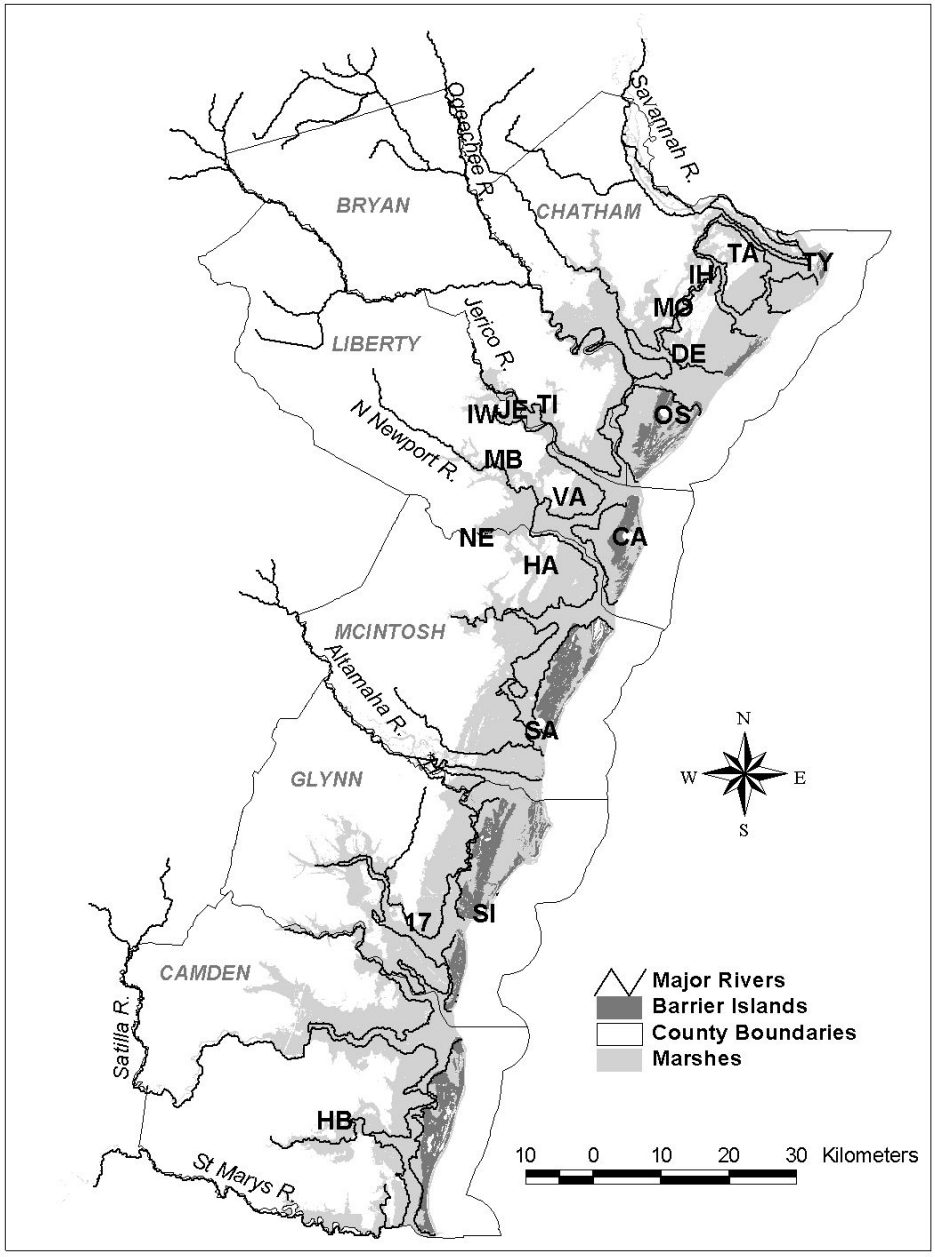

Figure 2-1. Map of study area in Coastal Georgia. Survey sites are designated by 2-letter site codes (see Table 2-1). Major rivers, barrier islands, county boundaries and salt and brackish marshes are also indicated (fresh marshes are not included in the marsh coverage).

Table 2-1. General characteristics of dieback sites included in this study. Dieback sites (and 2-letter site codes) are listed from north to south (see figure 2-1). The species of vegetation is either *S. alterniflora* (S.a.) or *J. roemerianus* (J.r.). Dieback sites were located either on barrier islands (Barrier Is.) or in marshes further inland (Inland). The area covered by dieback sites was estimated, in acres, as <1, 1-10, 10-100 or >100. The pattern or patterns of dieback observed at each site were classified as creek bank (C), midmarsh (M), berm (B) or upland (U) dieback. The elevation of dieback areas was higher than (+), equal to (=) or lower than (-) the elevation of nearby healthy areas (in two sites, part of the dieback area was at a lower elevation than the healthy area, but other parts of the dieback area were equal to [-/=] or higher [-/+] in elevation). Dieback sites were associated with man-made structures when they were located within 50 m of the listed structure. The influence of terrestrial environments was estimated by assigning ranks from 0 = no influence to 5 = nearly surrounded by upland (see text for details).

SITE	CODE	SPECIES	LOCATION	AREA	PATTERN	ELEVATION	STRUCTURES	INTERFACE
Talahi Island	TA	S.a.	Inland	1-10	M	-	dike/dock	0
Tybee Is.	TY	S.a.	Barrier Is.	<1	C	=	causeway	4
Isle of Hope	IH	S.a., J.r.	Inland	<1	C	-	causeway	3
Moon River	MO	S.a.	Inland	<1	C	-	bridge	1
Delegal Ck. Marina	DE	S.a.	Inland	<1	B, M	=	dock	3
Ossabaw Is.	OS	S.a.	Barrier Is.	<1	C	=	none	3
Tivoli Trail	TI	S.a.	Inland	<1	C, B	-/+	none	4
Jerico River	JE	S.a.	Inland	>100	C, M, B, U	=	bridge	3
Isle of Wight Rd.	IW	J.r.	Inland	1-10	B, M	=	bridge	0
Melon Bluff	MB	S.a.	Inland	10-100	C, M, U	=	none	3
Van Dyke Ck.	VA	S.a.	Inland	<1	C	-	none	3
St. Catherine's Is.	CA	S.a.	Barrier Is.	<1	M	-	none	0
South Newport	NE	S.a.	Inland	1-10	C	=	bridge	3
Harris Neck	HA	S.a.	Inland	<1	C	=	none	4
Sapelo Is.	SA	S.a.	Barrier Is.	<1	C	+	dike	3
St. Simons Is.	SI	S.a.	Barrier Is.	<1	C, M	-	causeway	0
Hwy 17	17	S.a.	Inland	<1	M	=	causeway	3
Harriet's Bluff	HB	S.a.	Inland	10-100	C	-/=	none	3

Table 2-2. The p-values for site (survey site), status (healthy or dieback area) and site x status (2-way ANOVA) for each survey variable. Significant results ($\alpha = 0.05$) are indicated by bold type.

RESULTS OF 2-WAY ANOVA			
VARIABLE	**SITE**	**STATUS**	**SITE x STATUS**
Stem density - >15 cm	**<0.001**	**<0.001**	**<0.001**
Stem density - <15 cm	**<0.001**	**<0.001**	**<0.001**
Stem density - dead stems	**<0.001**	**<0.001**	**<0.001**
Stem height	**0.004**	**<0.001**	0.302
L. irrorata density - > 10 cm	**<0.001**	0.062	**<0.001**
L. irrorata density - all sizes	**<0.001**	0.28	**0.025**
Crab hole density	**<0.001**	**<0.001**	**<0.001**
G. demissa density	**<0.001**	0.146	0.052
Salinity	**<0.001**	**0.022**	0.490
pH	**<0.001**	**0.015**	**<0.001**
Temperature	**<0.001**	0.233	0.182
Eh - 2 cm	**<0.001**	**<0.001**	**<0.001**
Eh - 15 cm	**<0.001**	**0.034**	**<0.001**

Table 2-3. The average density (count m^{-2} ± S.D.) of live stems >15 cm and <15 cm and dead stems in quadrats sampled in healthy and dieback areas. All values are for *S. alterniflora* except where indicated (* = both *S. alterniflora* and *J. roemerianus*; ** *J. roemerianus* only). Dead stems included both standing dead stems and low stubs that protruded above the marsh surface. Bold type indicates significantly higher values.

	DIEBACK AREA (no. m^{-2})			**HEALTHY AREA (no. m^{-2})**		
	LIVE STEMS		**DEAD STEMS**	**LIVE STEMS**		**DEAD STEMS**
SITE	**> 15 cm**	**< 15 cm**		**> 15 cm**	**< 15 cm**	
Talahi	4 ± 8	6 ± 12	407 ± 200	88 ± 52	22 ± 15	195 ± 76
Tybee Is.	0	0	211 ± 85	**59 ± 11**	**92 ± 3**	222 ± 37
Isle of Hope *	0	0	205 ± 202	**80 ± 33**	31 ± 6	101 ± 107
Moon River	0	0	199 ± 69	**79 ± 26**	**125 ± 41**	273 ± 38
Delegal Ck. Marina	11 ± 19	1 ± 2	**831 ± 94**	**141 ± 24**	**111 ± 21**	341 ± 81
Ossabaw Is.	39 ± 18	41 ± 19	13 ± 13	**124 ± 38**	30 ± 23	134 ± 88
Tivoli Trail	24 ± 32	29 ± 39	233 ± 99	No Data	No Data	No Data
Jerico River	84 ± 90	68 ± 55	**293 ± 485**	105 ± 30	74 ± 31	34 ± 10
Isle of Wight Rd. **	10 ± 12	1 ± 2	**907 ± 515**	**210 ± 80**	4 ± 6	591 ± 265
Melon Bluff	1 ± 2	0	303 ± 134	**79 ± 10**	**69 ± 29**	162 ± 20
Van Dyke Cr.	0	0	122 ± 59	**68 ± 8**	**54 ± 22**	105 ± 91
St. Catherine's Is.	22 ± 22	19 ± 19	**433 ± 140**	**84 ± 20**	**141 ± 176**	205 ± 43
South Newport	5 ± 6	13 ± 19	**460 ± 84**	**120 ± 15**	**139 ± 7**	134 ± 66
Harris Neck	24 ± 19	3 ± 4	96 ± 31	**74 ± 21**	34 ± 17	80 ± 29
Sapelo Is.	18 ± 36	14 ± 25	**330 ± 183**	**114 ± 25**	13 ± 10	77 ± 22
St. Simons Is.	8 ± 7	8 ± 6	**550 ± 144**	**212 ± 26**	**264 ± 42**	58 ± 34
Highway 17	68 ± 22	40 ± 12	63 ± 22	95 ± 14	39 ± 13	104 ± 74
Harriet's Bluff	0	0	**931 ± 227**	**91 ± 28**	**57 ± 10**	450 ± 128
AVERAGE	13 ± 18	9 ± 14	**379 ± 295**	**107 ± 46**	**76 ± 67**	202 ± 148

Table 2-4. Average height ($\bar{x}$ ± S.D.) of the 5 tallest live stems in quadrats sampled in dieback and healthy areas. All values are for *S. alterniflora* except when indicated (* = both *S. alterniflora* and *J. roemerianus*; ** *J. roemerianus* only). Only one live stem was present when no standard deviation is given. Bold type indicates significantly higher values.

	HEIGHT (cm)	
SITE	**Dieback**	**Healthy**
Talahi	37	64 ± 16
Tybee Is.	0	80 ± 11
Isle of Hope *	0	102 ± 13
Moon River	0	109 ± 9
Delegal Ck. Marina	62 ± 50	55 ± 3
Ossabaw Is.	45 ± 14	55 ± 11
Tivoli Trail	65 ± 46	No Data
Jerico River	51 ± 55	81 ± 26
Isle of Wight Rd. **	51 ± 8	91 ± 7
Melon Bluff	7 ± 14	66 ± 9
Van Dyke Cr.	0	159 ± 23
St. Catherine's Is.	36 ± 4	57 ± 5
South Newport	20	87 ± 16
Harris Neck	55 ± 9	113 ± 43
Sapelo Is.	12 ± 24	59 ± 7
St. Simons Is.	20 ± 6	51 ± 6
Highway 17	66 ± 31	89 ± 10
Harriet's Bluff	0	61 ± 7
AVERAGE	37 ± 20	**81 ± 39**

Table 2-5. Average *L. irrorata* density (count m^{-2} ± S.D.) in quadrats sampled in dieback and healthy areas. Densities of all size classes and of snails >10 mm are shown ("**" denotes barrier island sites). Bold type indicates significantly higher values.

	DENSITY (all sizes)		DENSITY (>10 cm only)	
SITE	**DIEBACK**	**HEALTHY**	**DIEBACK**	**HEALTHY**
Talahi	4 ± 6	113 ± 96	4 ± 6	59 ± 40
Tybee Is. **	0	0	0	0
Isle of Hope	0	17 ± 11	0	16 ± 10
Moon River	0	3 ± 4	0	1 ± 2
Delegal Ck. Marina	91 ± 97	**408 ± 146**	51 ± 58	**260 ± 116**
Ossabaw Is. **	277 ± 339	241 ± 141	222 ± 292	146 ± 138
Tivoli Trail	4 ± 8	No Data	4 ± 8	No Data
Jerico River	12 ± 8	4 ± 8	12 ± 8	4 ± 8
Isle of Wight Rd.	1 ± 2	5 ± 2	1 ± 2	4 ± 0
Melon Bluff	1 ± 2	14 ± 10	1 ± 2	13 ± 8
Van Dyke Cr.	0	0	0	0
St. Catherine's Is. **	352 ± 201	257 ± 83	208 ± 99	172 ± 51
South Newport	0	0	0	0
Harris Neck	0	1 ± 2	0	1 ± 2
Sapelo Is. **	3 ± 6	**136 ± 149**	0	72 ± 77
St. Simons Is. **	2 ± 2	113 ± 43	2 ± 2	**103 ± 42**
Highway 17	**136 ± 87**	9 ± 7	36 ± 27	5 ± 6
Harriet's Bluff	0	1 ± 2	0	1 ± 2
AVERAGE	48 ± 133	84 ± 132	12 ± 91	50 ± 85

Table 2-6. Densities (count m^{-2} ± S.D.) of crab holes and *G. demissa* in dieback and healthy areas ("**" denotes barrier island sites). Bold type indicates significantly higher values.

	CRAB HOLES (no. m^{-2})		*G. DEMISSA* (no. m^{-2})	
SITE	**DIEBACK**	**HEALTHY**	**DIEBACK**	**HEALTHY**
Talahi	11 ± 12	29 ± 20	37 ± 39	8 ± 11
Tybee Is. **	**316 ± 196**	61 ± 71	0	0
Isle of Hope	89 ± 46	77 ± 78	42 ± 30	12 ± 14
Moon River	**444 ± 136**	176 ± 54	5 ± 6	5 ± 6
Delegal Ck. Marina	145 ± 36	155 ± 95	1 ± 2	2 ± 2
Ossabaw Is. **	60 ± 44	66 ± 57	3 ± 4	2 ± 4
Tivoli Trail	109 ± 27	No Data	12 ± 6	No Data
Jerico River	64 ± 116	64 ± 85	1 ± 2	1 ± 2
Isle of Wight Rd.	137 ± 87	60 ± 22	4 ± 5	3 ± 4
Melon Bluff	55 ± 28	24 ± 7	14 ± 17	23 ± 26
Van Dyke Cr.	378 ± 161	316 ± 93	3 ± 6	0
St. Catherine's Is. **	105 ± 43	94 ± 10	0	4 ± 6
South Newport	81 ± 68	43 ± 64	0	3 ± 6
Harris Neck	364 ± 58	372 ± 79	1 ± 2	0
Sapelo Is. **	**332 ± 196**	140 ± 138	8 ± 10	12 ± 16
St. Simons Is. **	**500 ± 247**	50 ± 23	11 ± 8	3 ± 6
Highway 17	120 ± 42	29 ± 27	6 ± 12	4 ± 5
Harriet's Bluff	154 ± 16	67 ± 44	0	1 ± 2
AVERAGE	194 ± 70	107 ± 100	8 ± 16	5 ± 10

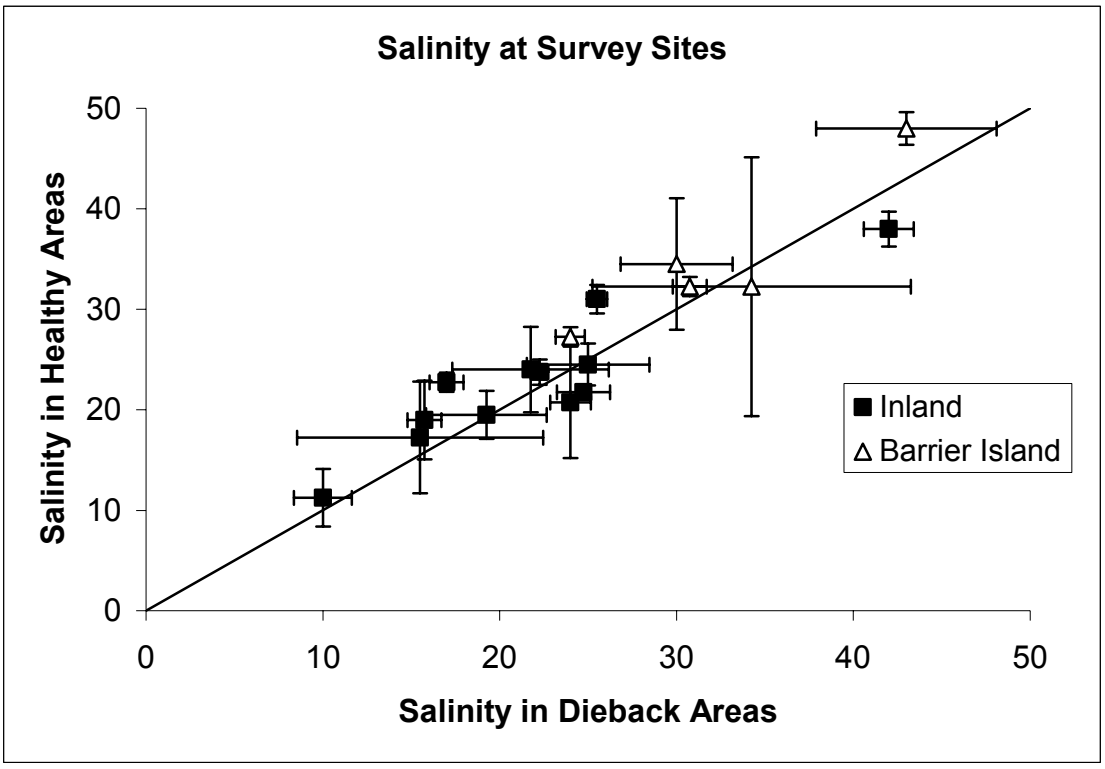

Figure 2-2. Average pore-water salinity at healthy vs. dieback areas at both inland and barrier island sites (error bars are 1 S.D.). Solid line is a 1:1 line, representing the condition where salinity is equal in healthy and dieback areas.

Table 2-7. Salinity ($\bar{x}$ ± S.D.)of pore-water in dieback and healthy areas ("**" denotes barrier island sites). The interaction (2-way ANOVA) between site and dieback vs. healthy area was not significant. Bold type indicates significantly higher values.

	Salinity (PSU)	
SITE	**DIEBACK**	**HEALTHY**
Talahi	24 ± 1	20 ± 6
Tybee Is. **	30 ± 1	32 ± 1
Isle of Hope	16 ± 1	19 ± 4
Moon River	17 ± 1	23 ± 1
Delegal Ck. Marina	42 ± 1	38 ± 2
Ossabaw Is. **	34 ± 9	32 ± 13
Tivoli Trail	22 ± 1	No Data
Jerico River	16 ± 7	17 ± 6
Isle of Wight Rd.	25 ± 3	25 ± 2
Melon Bluff	19 ± 3	20 ± 2
Van Dyke Cr.	25 ± 2	22 ± 1
St. Catherine's Is. **	43 ± 5	48 ± 2
South Newport	10 ± 2	11 ± 3
Harris Neck	22 ± 1	24 ± 1
Sapelo Is. **	35 ± 3	30 ± 7
St. Simons Is. **	24 ± 1	27 ± 1
Highway 17	22 ± 4	24 ± 4
Harriet's Bluff	26 ± 1	31 ± 1
AVERAGE	25 ± 9	**27 ± 9**

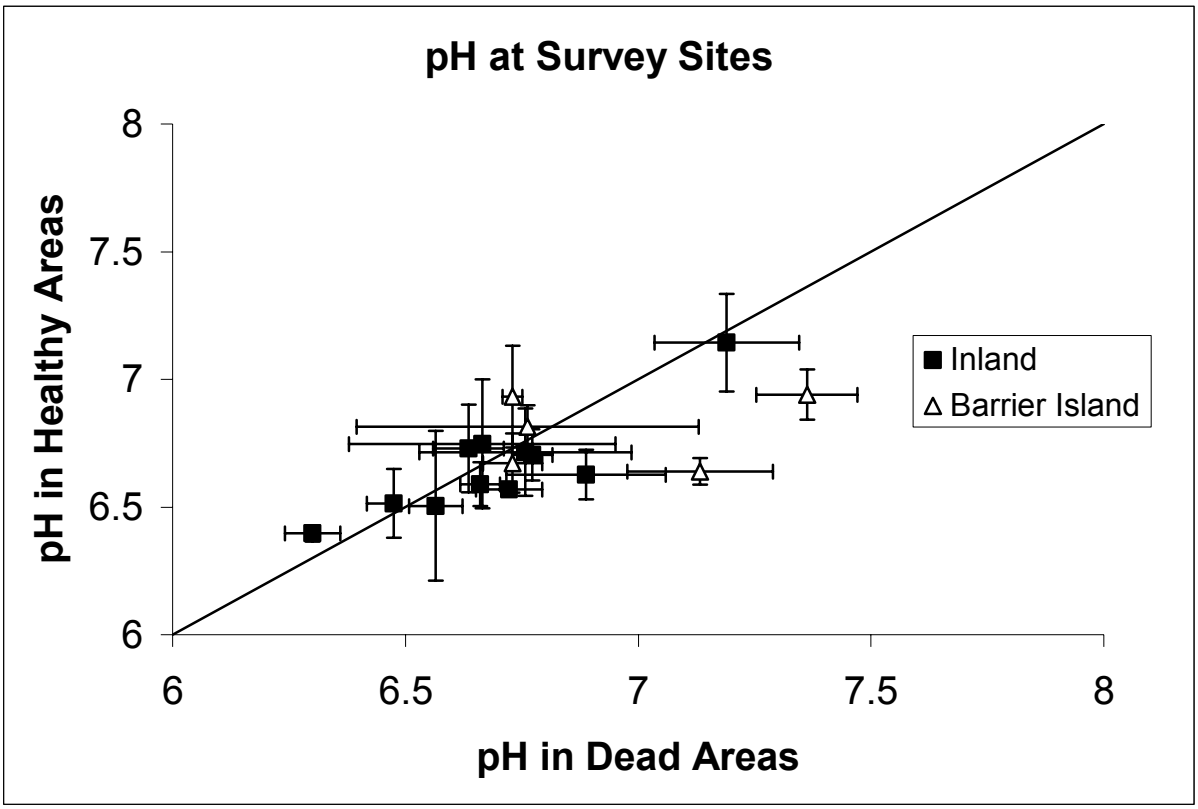

Figure 2-3. pH of pore-water in healthy areas vs. dieback areas at both inland and barrier island sites (error bars are 1 S.D.). Solid line is a 1:1 line, representing equal pH values in the healthy and dieback area within each site.

Table 2-8. Soil pH and temperature ($\bar{x}$ ± S.D.) at dieback and healthy areas ("**" indicates barrier island sites). Bold type indicates significantly higher values.

	pH		Temperature (ºC)	
SITE	**DIEBACK**	**HEALTHY**	**DIEBACK**	**HEALTHY**
Talahi	**6.89 ± 0.17**	6.63 ± 0.10	27 ± 0	27 ± 0
Tybee Is. **	6.73 ± 0.06	6.67 ± 0.12	28 ± 0	28 ± 0
Isle of Hope	6.66 ± 0.04	6.59 ± 0.09	27 ± 0	27 ± 1
Moon River	6.77 ± 0.04	6.71 ± 0.10	28 ± 0	28 ± 0
Delegal Ck. Marina	7.19 ± 0.16	7.14 ± 0.19	28 ± 0	28 ± 0
Ossabaw Is. **	**7.36 ± 0.11**	6.94 ± 0.10	29 ± 0	28 ± 1
Tivoli Trail	6.64 ± 0.07	No Data	30 ± 1	No Data
Jerico River	6.71 ± .26	6.74 ± 0.12	28 ± 3	30 ± 2
Isle of Wight Rd.	6.76 ± 0.23	6.72 ± 0.17	26 ± 1	26 ± 1
Melon Bluff	6.64 ± 0.08	6.73 ± 0.17	26 ± 0	27 ± 1
Van Dyke Cr.	6.48 ± 0.06	6.52 ± 0.13	26 ± 1	26 ± 0
St. Catherine's Is. **	**7.13 ± 0.16**	6.64 ± 0.05	27 ± 1	28 ± 1
South Newport	6.57 ± 0.06	6.51 ± 0.29	28 ± 1	28 ± 2
Harris Neck	6.30 ± 0.06	6.40 ± 0.03	26 ± 0	26 ± 0
Sapelo Is. **	6.76 ± 0.37	6.82 ± 0.08	No Data	No Data
St. Simons Is. **	6.73 ± 0.02	6.93 ± 0.20	28 ± 1	28 ± 2
Highway 17	6.67 ± 0.29	6.75 ± 0.25	29 ± 4	32 ± 1
Harriet's Bluff	6.72 ± 0.07	6.57 ± 0.01	26 ± 0	26 ± 0
AVERAGE	6.75 ± 0.27	6.70 ± 0.22	27 ± 1	28 ± 1

Table 2-9. Redox potential ($\bar{x}$ ± S.D.) at 2 cm and 15 cm depth in dieback and healthy areas. Bold type indicates significantly lower values. "**" denotes barrier island sites.

	DEPTH = 2 cm (mV)		DEPTH = 15 cm (mV)	
SITE	**DIEBACK**	**HEALTHY**	**DIEBACK**	**HEALTHY**
Talahi	-15 ± 16	-2 ± 90	-50 ± 22	-69 ± 22
Tybee Is. **	116 ± 92	35 ± 92	-32 ± 26	-64 ± 38
Isle of Hope	203 ± 78	**40 ± 99**	42 ± 113	**-53 ± 22**
Moon River	229 ± 16	**0 ± 17**	41 ± 37	**-73 ± 12**
Delegal Ck. Marina	356 ± 25	294 ± 45	113 ± 96	**11 ± 25**
Isle of Wight Rd.	350 ± 66	336 ± 22	86 ± 82	164 ± 57
Van Dyke Cr.	**121 ± 61**	269 ± 47	**74 ± 94**	211 ± 8
St. Catherine's Is. **	282 ± 123	238 ± 100	**42 ± 126**	225 ± 46
South Newport	105 ± 69	28 ± 40	7 ± 35	-36 ± 22
Harris Neck	285 ± 50	259 ± 55	170 ± 57	92 ± 96
St. Simons Is. **	14 ± 11	-29 ± 40	-42 ± 12	-94 ± 7
Harriet's Bluff	165 ± 68	**30 ± 41**	12 ± 27	-37 ± 61
AVERAGE	198 ± 134	138 ± 147	47 ± 93	21 ± 118

CHAPTER 3

TRANSPLANT EXPERIMENTS

Introduction

In spring, 2003, at least a year after the onset of salt marsh dieback in coastal Georgia, very little evidence of regrowth or recolonization of dieback areas had been observed. Heavy rains across much of Georgia during the previous fall had signaled the end of severe drought (USGS 2002), however, and coastal residents reported that salt marsh vegetation in general appeared much healthier than it had during the previous several years. Despite the healthier appearance of the marsh, dieback areas did not show any recovery through summer and fall, 2003 (personal observations). By December 2003, the only observed recolonization occurred as a slow expansion of live vegetation along the edges of dieback areas (GCRC website). In areas with extensive dieback, once the root mat of *S. alterniflora* and *J. roemerianus* decomposes, erosion of salt marsh sediments can be a major concern, and this, in fact, was observed in some areas (Jerico River and Melon Bluff, in particular). If dieback areas are not recolonized by natural processes, restoration may therefore be necessary to prevent erosion of the marsh. Before restoration is undertaken, however, it is important to know whether healthy transplants can survive and grow in dieback areas.

There are several possible outcomes of transplant trials in salt marsh dieback areas: 1) Healthy plants transplanted into dieback areas could show reduced survival and/or growth compared to controls, indicating that the conditions causing dieback are still present; 2) Healthy plants transplanted into dieback areas could survive and grow equally as well as controls, which could indicate either that the conditions causing dieback were transitory and were ameliorated

prior to the experiment, or that the transplanting process itself ameliorates the conditions causing dieback; 3) Healthy plants transplanted into dieback areas could grow better than controls due to reduced competition for resources (space, light or nutrients) or 4) healthy plants transplanted into both dieback and healthy areas could die, indicating a failed experiment in which plant death occurred as a result of the transplanting process.

Transplant experiments carried out during previous dieback events at other locations have had various outcomes. In cases where dieback was probably caused by sediment waterlogging and sulfide accumulation, healthy plants transplanted into dieback areas suffered reduced growth compared to controls. In the Lymington Estuary, Great Britain, *S. townsendii* transplants from vigorous swards died when planted in dieback areas, whereas affected plants recovered when moved out of dieback areas (Goodman et al., 1959). In Louisiana, during the historical dieback, swards transplanted into dieback areas in low-lying parts of the marsh showed reduced growth (Mendelssohn and McKee, 1988). This was accompanied by decreases in redox potential and increases in sulfide and NH_4 concentrations and root alcohol dehydrogenase activity (an indicator of anaerobic root metabolism). In contrast, transplants into a dieback area during the recent acute dieback event in Louisiana salt marshes not only survived, but were overgrown by natural recolonization (Irving Mendelssohn, personal communication). These observations indicate that, in some cases, natural processes may restore dieback areas faster than restoration efforts.

In Georgia, two greenhouse experiments were carried out in October 2002 to determine the viability of plant tissue and suitability of soil for plant growth from the Jericho River dieback site. Plugs of *S. alterniflora* from a completely denuded area (rhizomes still present) and a small area of live vegetation located within the dieback area (rhizomes + green stems) were grown in

pots and watered with fresh water. The live vegetation continued to grow whereas the rhizomes without live above-ground tissue were indeed dead and did not re-sprout. This result was supported when rhizomes from dieback areas were analyzed for viability using a vital stain: those that appeared dead did not take up the stain (Chandra Franklin, personal communication).

In a second experiment, healthy young shoots of *S. alterniflora* (14 ± 6 cm tall) were planted into pots containing soil from the dieback area, a nearby healthy marsh (the source of the shoots) or a sandy greenhouse mix typically used for *S. alterniflora*. Two plants were planted in each pot with 5 pots per treatment (total of 10 plants per treatment). Plant height was measured monthly over a period of 3 months, during which time plants were watered daily with fresh water and pots were allowed to drain freely. Survival was nearly 100%, as only two plants died within the first 2 weeks of the experiment, one in the dieback soil and one in the greenhouse mix. This was likely due to transplant stress. Not surprisingly, growth was slightly higher in the greenhouse mix than in marsh soils (Figure 3-1). However, seedlings had similar growth rates in both the dieback and healthy marsh soil, indicating that soil from dieback areas was viable. Although these results were encouraging, it is important to note that the cause of dieback may have been ameliorated under greenhouse conditions. Additional surveys and transplant experiments were necessary for developing a broader understanding of the patterns and characteristics of salt marsh dieback in Georgia.

This chapter describes transplant studies carried out in the field. The study was designed to investigate similarities and differences in the response of both species affected, *S. alterniflora* and *J. roemerianus*, to transplanting. Study sites were chosen to represent two different patterns of dieback, creek bank and midmarsh. Trials were conducted in both inland and barrier island areas, because differences such as salinity may affect the expression of dieback. The purpose of

these efforts was to determine: 1. Whether healthy *S. alterniflora* or *J. roemerianus* plants could survive and grow in dieback areas under field conditions and 2. Whether differences in soil chemistry between dieback and healthy areas could be related to transplant survival and growth.

Methods

Study Sites

Transplant experiments were carried out using four paired experimental plots at two locations on the Georgia coast. The first site (referred to as the Sapelo creek bank site) was located on the southern end of Sapelo Island in MacIntosh County, GA near the University of Georgia Marine Institute (Figures 3-2, 3-3). This barrier island site is approximately 50 m from Doboy Sound and is submerged during high tide except during neap tides. Only *S. alterniflora* occurs at this site. Dieback affected a 1-3 m wide strip along the upper end of a small tidal creek, which is characteristic of the creek bank dieback pattern. A healthy reference area (and source of transplants) was located about 20 m from the dieback area along the same small creek, but was closer to Doboy Sound. The site is located near an old dike, but has been mostly undisturbed in recent years. Dieback was first reported at this site in the late fall of 2002.

The three other sites were located several miles inland from Sapelo Island on Dickinson Creek at Melon Bluff Plantation in Liberty County, GA (Figures 3-2, 3-3). Dieback at Melon Bluff Plantation is reported to have begun during summer and fall 2001. The Melon Bluff creek bank site is a creek bank dieback of tall form *S. alterniflora* comparable to the site on Sapelo Island. This site is also submerged at high tide except during neap tides. At this site, *S. alterniflora* died on the top and sides of the creek bank in a strip 1 - 3 m wide, although there were some small patches of live *S. alterniflora* at the bottom of the creek. A healthy reference

site was located approximately 5 m from the creek directly behind the dieback area. The second site at Melon Bluff was a patch of dieback in an area of short form *S. alterniflora* (referred to as the high marsh *Spartina* site). This site was located near the upper extent of moderate high tides. Dieback occurred in an area of saturated soils and was characteristic of the midmarsh dieback pattern. The marsh surface at this site is composed of a dense, spongy mat of roots about 10-15 cm thick with very soft, wet mud underneath. The healthy control site was located about 5 m from the edge of the die-off in an area with similar elevation and soil saturation. At the third Melon Bluff site, patches of *J. roemerianus* occurred within an area dominated by short-form *S. alterniflora* (referred to as the *Juncus* site). This site was slightly higher in elevation than the high marsh *Spartina* site, and was only flooded during spring tides. Dieback in this area affected both *S. alterniflora* and *J. roemerianus* (though only *J. roemerianus* dieback areas were used) and occurred in the midmarsh pattern. A healthy reference site was located approximately 1 km south of the dieback area, because live *J. roemerianus* closer to the dieback site only remained in very small clumps or as individual stems at the edges of some former patches. The reference area, which was still located on Dickinson Creek, was the closest available source of healthy transplants. The marsh surface at this site was a thick mat of roots similar to the high marsh *S. alterniflora* site.

Experimental Design

At each site, I set up a series of reciprocal transplants, wherein plants from the healthy area were transplanted into either the healthy or dieback zone, and, when possible, plants from the dieback area were likewise transplanted. In each healthy site, 24 swards (plants and the surrounding soil) of *S. alterniflora* or *J. roemerianus* were dug up and placed into 20 cm

diameter x 20 cm tall plastic pots. *S. alterniflora* swards averaged 7 ± 3 stems per pot, whereas *J. roemerianus* swards averaged 14 ± 5 stems per pot. There were no significant differences in initial stem counts or height of the 5 tallest stems between treatments at any site (Table 3-1). Each pot had numerous 3 cm diameter holes drilled in both the sides and bottom to allow free exchange of interstitial water between the transplant and its external environment. Of these 24 pots, 12 were randomly planted into new holes in the dieback area, and the remaining pots were randomly placed back into holes in the healthy area as controls. A second set of 24 swards was dug up in or at the edge of the dieback area and treated similarly at the Sapelo creek bank and high marsh *Spartina* sites (with a similar number of plants per pot). At the other two sites, however (Melon Bluff creek bank and *Juncus*), there were no remaining live plants in the dieback area with which to carry out these treatments. In these cases, only healthy plants were transplanted.

The transplant study was initiated at Melon Bluff Plantation between May 3-9, 2003. Initial measurements (described in detail below) were taken immediately after transplanting was completed. Measurements were subsequently taken on June 14, July 15 and October 12, 2003. At Sapelo Island, transplanting was carried out and initial measurements were taken on May 28, 2003. Subsequent measurements were taken on July 6 and October 4, 2003.

Vegetation Analysis

On each sampling date, the total number of stems per pot and height of all stems were recorded. Gain or loss of individuals was calculated as the difference between the initial and final number of stems in each pot, normalized to the initial numbers. The average height of all stems in a pot could not be used because increases in the number of new stems caused average

plant height to decrease even though individual plants increased in height. To eliminate this effect, the height of the 5 tallest stems in each pot was used as an indicator of plant height. Change in height was calculated as the difference between the initial and final height of the 5 tallest stems in each pot, normalized to the initial height of the 5 tallest stems.

Plant tissue samples were collected by removing one or two outer green leaves from a randomly chosen plant in each pot on each sampling date. Tissue samples were placed in plastic bags and kept on ice for several hours (no more than 24 hours) until taken into the lab. Samples were then rinsed in DI water to remove mud and salts from the leaf surface, placed in paper bags and dried in an oven at 60 degrees C. After drying, leaf tissue was pulverized using a ball grinder. The Sapelo creek bank and *Juncus* sites were chosen for preliminary analyses to determine if differences in elemental composition could be observed between healthy and dieback areas for either species. Samples from May and October were measured for carbon, nitrogen and sulfur concentrations with a CE Elantech Flash Elemental Analyzer 1112. Atomic C:N ratios were then determined.

Soil

On each sampling date, soil pore-water was sampled at rooting depth (10-15 cm) and analyzed for pH, salinity and NH_4 concentration. To obtain samples, shallow wells (measuring 5 cm in diameter and 15 cm deep) were dug into the marsh surface at five locations within the transplant area in both the dieback and healthy areas at each site. (Wells were dug outside the transplant pots to prevent destruction of transplants.) Interstitial water was allowed to percolate into wells for several minutes prior to sampling. Water samples were extracted using a large pipette and stored in 30 ml plastic bottles on ice.

Salinity and pH were measured in the laboratory within several hours of sample collection. Samples were filtered using a Whatman GF/F 47 μM filter placed on plastic filter towers and each sample was divided into two portions. Approximately 15 ml of sample was stored in ashed 20 ml glass scintillation vials and frozen at 0 degrees C for later analysis of NH_4 concentration, and the remaining sample was used to determine salinity and pH. Salinity was measured using a Leica model 10419 temperature-compensated refractometer. pH was determined using a Fisher Scientific accumet® pH probe (13-620-AP50). NH_4 concentration was analyzed colorimetrically and measured with a Shimadzu UV-1601 spectrophotometer (Koroleff, 1983).

Redox potential was measured in the root zone (15 cm depth). Eh was determined using a Mettler-Toledo Combination Redox Electrode (Pt4805-SC-DPAS-K8S/200) attached to a Fisher Scientific accumet® AP62 portable pH/mV meter. A correction factor of +225 mV was added to measured values to account for the potential of the Ag/AgCl reference electrode.

Statistical Analyses

Pair-wise t-tests were used to compare both soil and plant characteristics in healthy vs. dieback areas. P-values were adjusted using the Dunn-Šidák method to achieve an overall alpha of 0.05 ($\alpha' = 1-[1-\alpha]^{1/k}$), where α is the overall alpha, α' is the adjusted alpha used for each test and k is the total number of t-tests carried out (Sokal and Rohlf, 1981). P-values are reported in the text for reference purposes only. Analysis of variance (ANOVA) was used to compare changes in height and plant abundance by source and destination (healthy vs. dieback) for sites where reciprocal transplants were carried out, using transplant source and transplant destination

as fixed effects (SAS Institute 2000). Proportionate data (change in number of stems per pot and average height of the 5 tallest stems) were square-root transformed prior to analysis.

Results

Vegetation Analysis

Survival of live transplants was high in both dieback and healthy areas at all sites. Although occasional stem death occurred early in the experiment in some pots (personal observations), live stems survived throughout the course of the experiment in every pot (Figures 3-4 and 3-5). By March 2004, roots and/or rhizomes of both *S. alterniflora* and *J. roemerianus* had extended through holes in most pots, and live stems sometimes sprouted up next to the pots.

At the Sapelo creek bank site (tall-form *S. alterniflora*), all plants from either the healthy area or the dieback area survived and grew, with few differences between treatments. Plants from the healthy area moved to the dieback area increased from May to October in both the total number of stems per pot (6 ± 2 to 13 ± 4) and height of the five tallest stems (73 ± 12 to 84 ± 12 cm) (Figures 3-4, 3-6). There were no significant differences in growth between healthy and dieback areas (Table 3-1). Plants from the dieback area moved to the healthy area also increased in the total number of stems per pot and average height of the 5 tallest stems with no differences between treatments (Figures 3-5, 3-7). Analysis of variance revealed that the transplant source (but not destination) had significant effects on the total number of stems per pot and height of the 5 tallest stems (Table 3-2). These results support the findings of greenhouse trials in which there was no difference in growth for healthy young *S. alterniflora* shoots transplanted into soil from healthy and dieback areas.

At the Melon Bluff creek bank site (tall-form *S. alterniflora*), all plants survived, but plants grew significantly more in the dieback area. Plants moved from the healthy area to the dieback area had significantly ($p = 0.004$) larger increases in the number of stems per pot than those that remained in healthy areas (Figure 3-4, Table 3-1). Average height of the 5 tallest stems also increased significantly more ($p < 0.001$) in the dieback area as compared to the healthy site (Figure 3-6, Table 3-1). These differences are surprising since, if anything, it might be expected that dieback areas would be less suitable for growth than healthy areas. These results may indicate a release from competition in dieback areas.

At the high marsh *Spartina* site (short form *S. alterniflora*), all stems survived and grew throughout the experiment, but growth responses were inconsistent when plants from the healthy area were moved to either the dieback or healthy area. There were no differences in the total number of stems per pot, but average height of the 5 tallest stems increased significantly more ($p = 0.004$) in the healthy area as compared to the dieback site (Figure 3-6, Table 3-1). When plants from the dieback area were moved, there were significantly larger ($p < 0.001$) increases in the total number of stems per pot in the dieback area as compared to the healthy area (Figure 3-5, Table 3-1), but there were no differences in average height of the 5 tallest stems (Figure 3-7). These inconsistencies make it difficult to draw any clear conclusion about which environment was more conducive to growth. Analysis of variance indicated that both transplant source and final destination were important (Table 3-2).

At the *Juncus* site, *J. roemerianus* transplants survived, but they did not exhibit as much growth as *S. alterniflora* transplants. There were no significant differences in either number of stems per pot or average height of the 5 tallest stems between plants moved to the dieback area as compared to those moved to the healthy area. However there was a net decrease in average

height of the 5 tallest stems of plants in the dieback area (from 83 ± 9 to 74 ± 9 cm) (Figure 3-6). This decrease was the result of the death of a few tall stems in some pots. This may have occurred because plants moved into the dieback area had to be carried between sites by car, whereas those moved to the healthy area were carried a short distance by hand.

Elemental analyses were carried out on tissue samples collected in May and October at the Sapelo creek bank and *Juncus* sites. Values for *S. alterniflora* that originated in both healthy and dieback areas averaged 44.1 ± 1.2 % carbon, 1.2 ± 0.3 % nitrogen and 0.4 ± 0.2 percent sulfur, with an average C:N ratio (atomic) of 46 ± 10 (Table 3-2). *J. roemerianus* (from healthy areas only) averaged 46.7 ± 0.7 % carbon and 1.6 ± 0.2 % nitrogen (sulfur was below the detection limit of 0.19 %), with an average C:N ratio (atomic) of 35 ± 5. These values are similar to those previously reported for *S. alterniflora* and *J. roemerianus* in southeastern salt marshes (Gallagher, 1975). No significant differences were observed between healthy and dieback areas for any metric at either site.

Soil

In the 3 experimental sites that had *S. alterniflora*, soil pore-water salinities were comparable between dieback and healthy areas and through time. At the Juncus site, however, salinities were usually significantly higher in the dieback area as compared to the healthy area. Salinity at the 3 sites containing *S. alterniflora* varied from 11 to 31 PSU and averaged 24 ± 4 PSU (Figure 3-8, Table 3-4). The only significant difference between healthy and dieback areas occurred at the Melon Bluff creek bank site in May (Table 3-4). In the *Juncus* site, however, salinities in the dieback area average 32 ± 3 PSU as compared to 24 ± 3 PSU in the healthy sites, and were significantly higher during 3 of the 4 sampling times (Figure 3-8, Table 3-4). Since the

healthy area of *J. roemerianus* was located about 1 km from the dieback area, it is possible that this difference is related to hydrologic differences between the two areas.

Soil pH ranged from 5.69 to 7.75 and averaged 6.72 ± 0.40, with no differences between dieback and healthy areas or through time except for a small but significant decrease in pH in the healthy area at the *Juncus* site in July (Figure 3-9, Table 3-4).

Redox potential at 15 cm depth varied within and among sites (Figure 3-10). At the 2 creek bank *Spartina* sites, Eh values were positive in both healthy and dieback areas. At the high marsh *Spartina* site, Eh values were negative in both healthy and dieback areas. The *Juncus* site was the only place where significant differences in redox potential were observed between dieback and healthy areas: Eh was positive in the healthy area and negative in the dieback area. This difference may also be related to hydrologic differences between the healthy and dieback areas at the *Juncus* site.

The concentration of NH_4 in pore-water did not vary over the course of the study, and was often significantly higher in dieback areas than in healthy areas (Figure 3-11). NH_4 concentrations were significantly higher at all 4 sampling times at both the Melon Bluff creek bank and *Juncus* sites and at 1 of the 3 sampling times at the Sapelo creek bank site. However, there were no significant differences between healthy and dieback areas at the high marsh *Spartina* site (Table 3-4). These results are in keeping with observations of high NH_4 concentrations in dieback areas at other locations (Linthurst and Seneca, 1980; Mendelssohn and McKee, 1988; de Souza and Yoch, 1997), and are likely due to decomposition of belowground plant tissue (Hackney and de la Cruz, 1980).

Discussion

This study demonstrated that both *S. alterniflora* and *J. roemerianus* could survive when transplanted to dieback areas along the Georgia coast. For *S. alterniflora*, the total number of stems per pot and average height of the 5 tallest stems increased at each of the three experimental sites over the course of the growing season (May to October 2003). These results are in keeping with greenhouse trials in which young *S. alterniflora* shoots (~15 cm tall) survived and grew when transplanted into soil from a dieback area. Growth was less apparent for *J. roemerianus*, although there were no significant differences between healthy and dieback areas and the dieback site retained live vegetation throughout the experiment. These results support the notion that dieback was no longer occurring in summer 2003.

There were very few differences in plant growth between healthy and dieback sites. No significant differences in growth (based either on the number of stems per pot or average height of the 5 tallest stems) were observed at the Sapelo creek bank or *Juncus* sites. Differences were observed at the high marsh *Spartina* site, but the differences were equivocal. The Melon Bluff creek bank site was the only site where consistent differences were observed, with plants in the dieback area growing better as compared to the healthy area. Overall, these results are in keeping with greenhouse trials in which no significant differences in growth were observed for *S. alterniflora* shoots in soil from healthy and dieback areas.

At the Melon Bluff creek bank site, where consistent differences in growth were observed in the field, growth (both the number of stems per pot and average height of the 5 tallest stems) was significantly higher for plants in the dieback area. This difference is potentially related to the observation that NH_4 concentrations were significantly higher in the dieback are, which would have provided a ready source of nutrients. Soil salinity, pH and redox potential were

similar between healthy and dieback areas at this site, so these variables did not have a differential effect on plant growth. Note that at the corresponding creek bank site on Sapelo Island where there were no significant differences in growth between treatments and controls, NH_4 concentrations were fairly similar between healthy and dieback areas.

At the high marsh *Spartina* site, differences in growth between healthy and dieback areas were inconsistent. Plants moved from the healthy area to the dieback had significantly smaller increases in height than those that remained in the healthy area. On the other hand, plants moved from the dieback area to the healthy area had significantly smaller increases in the number of stems per pot than those that remained in the dieback area. These results are contradictory and cannot be explained based on soil conditions, as there were no significant differences between the dieback and healthy areas with respect to soil salinity, pH, redox potential or NH_4 concentration. Although growth was observed in all transplants at this site, we were unable to determine whether plants performed better in the healthy or dieback area.

Plants from the dieback area were transplanted to a healthy area at two sites (Sapelo creek bank and high marsh *Spartina*) to determine if potentially affected plants could recover, but these plants did not perform better than controls that remained in the dieback area. One explanation for this observation might be that affected plants did not recover even when transplanted into a healthy site, but it was clear based both on growth measurements (which were similar to those of plants from healthy areas) and the visual appearance of plants that *S. alterniflora* in the dieback area was healthy throughout the experiment. Analysis of variance, however, suggested that transplant source did matter, but this result may have been due to initial height differences. This observation suggests that the dieback was not spreading and that plants growing on the edge of dieback areas were not negatively affected and remained viable.

At the *Juncus* site, no differences in growth were observed between healthy and dieback areas despite significant differences in soil conditions. Soil pore-water NH_4 concentration was significantly higher in the dieback area as compared to the healthy area, which should have promoted growth as was observed at the Melon Bluff creek bank site. However, soil salinity was also significantly higher in the dieback area and redox potential was negative (positive values were observed in the healthy area). The latter result indicates that the root zone in the dieback area was waterlogged and anoxic even at low tide (when measurements were carried out), whereas the healthy area was not. However, these conditions may not be representative of *J. roemerianus* dieback sites elsewhere in coastal Georgia, as there were no significant differences in salinity or redox potential between the healthy and dieback areas of the two survey sites containing *J. roemerianus*. Our results are not consistent with other studies of *J. roemerianus* in which water-logged soils and salinities consistently higher than 30 PSU (32 ± 3 PSU in the dieback area in our study) resulted in reduced growth or mortality (Eleuterius, 1984; Woerner and Hackney, 1997). However, a single growing season may not have been long enough to observe differences in growth of *J. roemerianus* between healthy and dieback sites. Shoot production and elongation of *J. roemerianus* is spread throughout the year, whereas shoot production and growth of *S. alterniflora* occurs primarily during spring and summer (Eleuterius and Caldwell, 1981). This difference in growth pattern may have made it more difficult to detect changes in the number of stems per pot and average height of the 5 tallest stems of our *J. roemerianus* transplants.

The results of this study suggest that transplanting is feasible means of salt marsh restoration in Georgia, but that *S. alterniflora* is a much better candidate for transplanting than *J. roemerianus*. *S. alterniflora* transplants survived and grew throughout the experiment with few

differences in growth between healthy and dieback areas, whereas growth of *J. roemerianus* was limited. *S. alterniflora* transplants were successful at various sites within a single marsh (creek bank and high marsh) and across the coast (inland and barrier island sites), suggesting that transplants can work in a variety of settings. These results show that the cause of dieback was transitory rather than a persistent change in the soils.

Before large scale restoration efforts are undertaken, restoration trials should be conducted to identify suitable source populations. Analysis of variance revealed that transplant source was an important indicator of growth and was more important than transplant destination at the Sapelo creek bank site (Table 3-2). This result is supported by studies at other locations that indicate that source location is an important factor determining the success of *S. alterniflora* transplants in dieback areas (Linthurst and Seneca, 1980; Carlson et al., 2001).

Another important factor to consider is whether restoration efforts significantly decrease the time to recovery over natural recolonization processes. At the time of this writing (Spring 2004), some natural recolonization has occurred at the two creek bank dieback sites in this study, such that some transplants will soon be overgrown by shoots from nearby live vegetation. Similar recolonization was been observed in Louisiana after the acute dieback event in 2000 (Irv Mendelssohn, personal communication). Some recolonization has also occurred at the *Juncus* site, but new vegetation at this site is almost entirely *S. alterniflora*. This is one of several locations in Georgia where *S. alterniflora* appears to be invading *J. roemerianus* stands following the dieback event (personal observations).

The results of these transplant experiments do not shed much light on the causes of salt marsh dieback in Georgia. The experiments were designed to determine differences in survival and growth between healthy and dieback areas and to identify soil characteristics that might

explain these differences. Instead, there were few differences in the growth of *S. alterniflora* or *J. roemerianus* between healthy and dieback areas, and in some cases, growth of *S. alterniflora* was actually higher in dieback areas than healthy areas. There were also no differences in pore-water pH, salinity or redox potential at most sites. The fact that all transplants survived throughout the growing season shows that transplanting is possible, and that whatever caused the dieback is no longer operating.

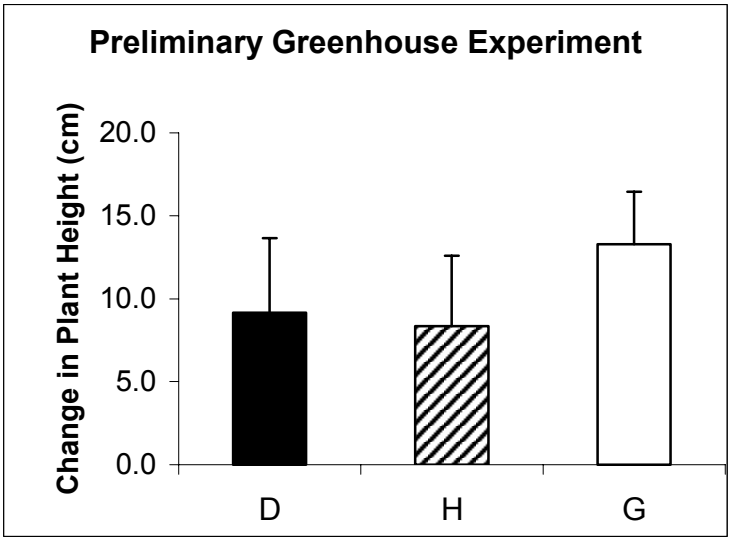

Figure 3-1. The change in plant height for healthy young *S. alterniflora* potted in soil from the Jerico River dieback site (D), soil from a nearby healthy marsh (H) and a sand-based greenhouse mix (G).

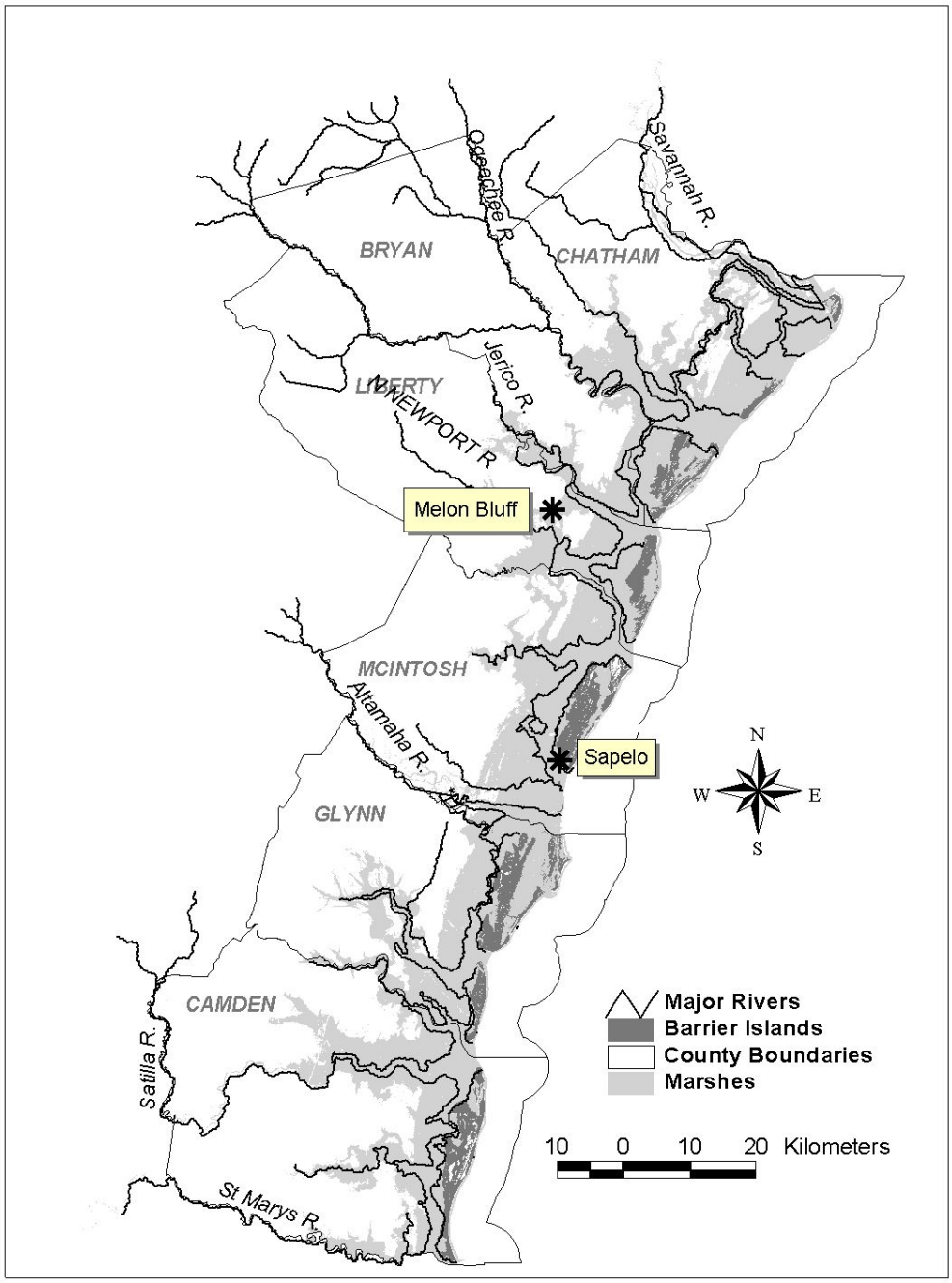

Figure 3-2. Study sites on the Georgia Coast (indicated by *). Melon Bluff Plantation is on the Newport River and Sapelo Island is a barrier island.

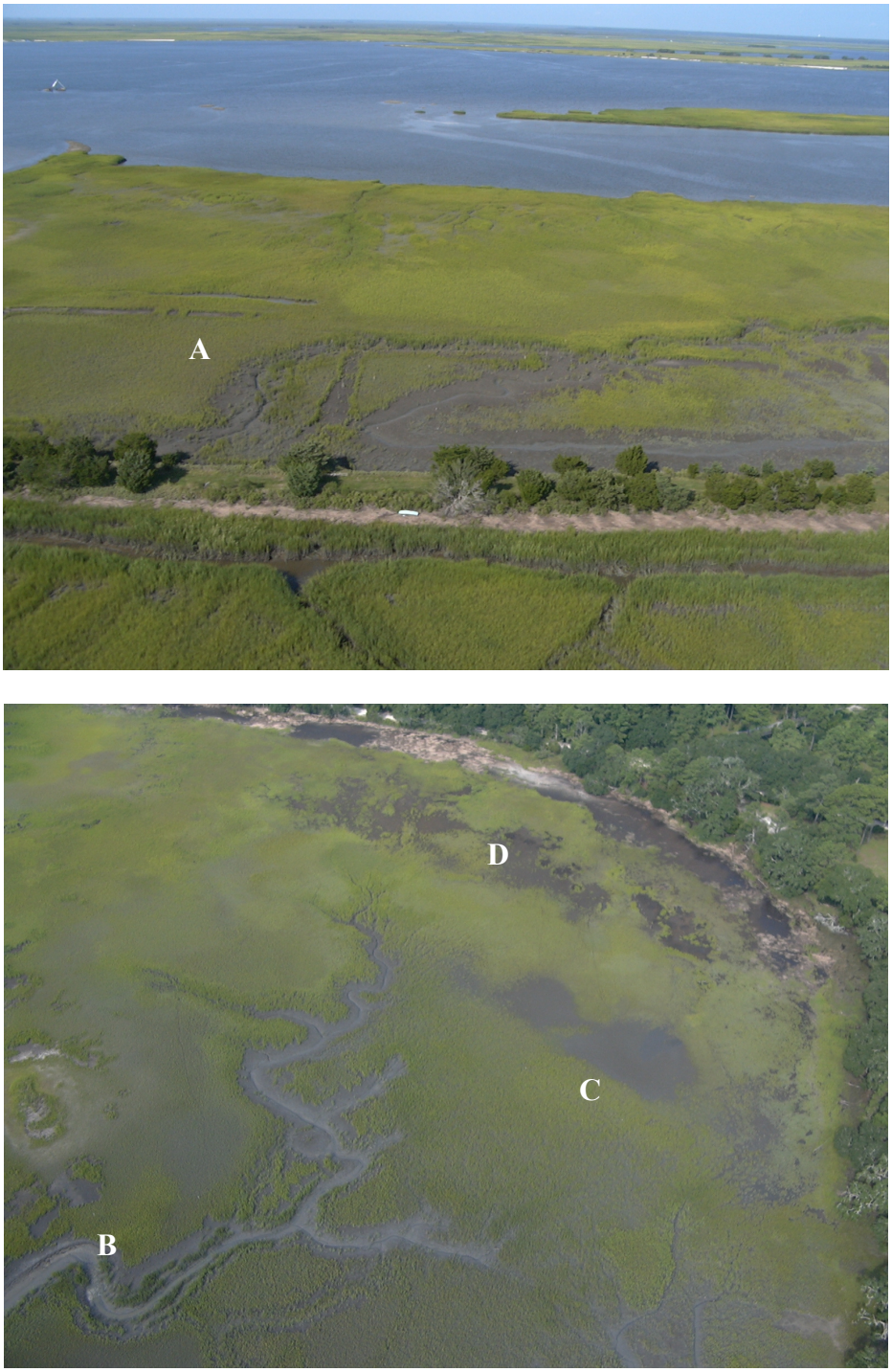

Figure 3-3. Transplant sites located at Sapelo Island (top) and Melon Bluff Plantation (bottom). The location of each experiment is indicated as follows: A) Sapelo creek bank, B) Melon Bluff creek bank, C) High marsh *Spartina* and D) *Juncus*.

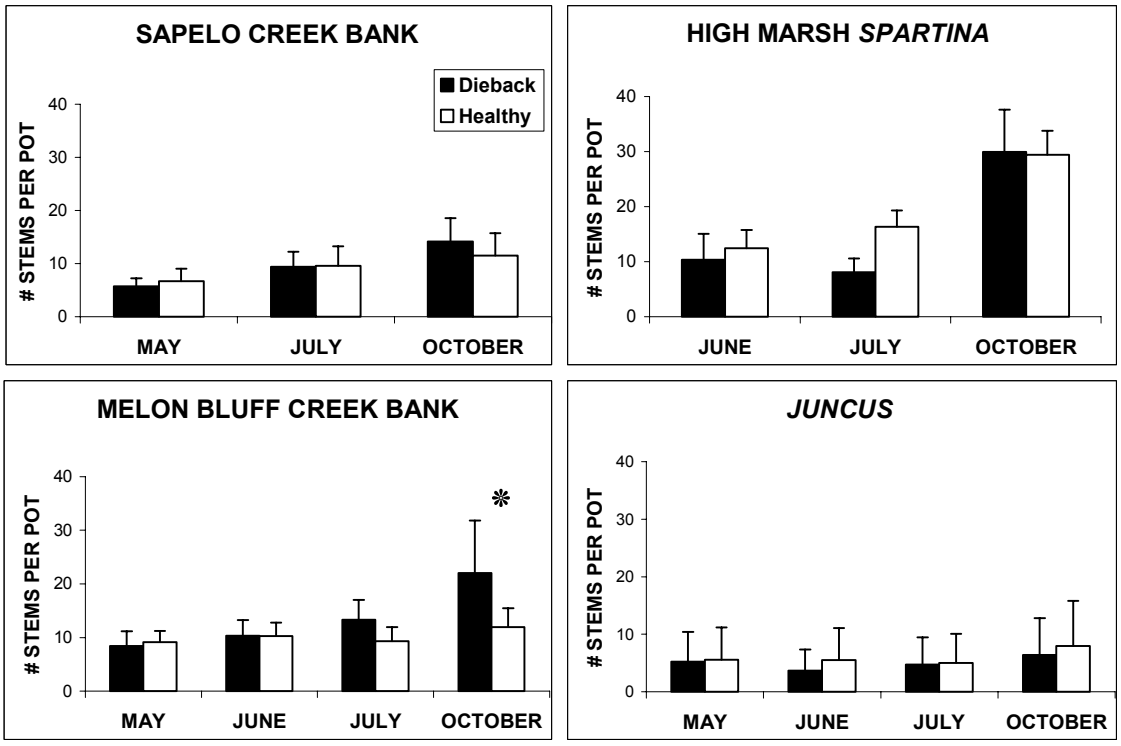

Figure 3-4. The number of stems per pot was determined for healthy *S. alterniflora* and *J. roemerianus* transplanted into dieback areas (black bars) and healthy areas (white bars). Error bars represent standard deviation. Asterisk indicates a significant difference in growth between dieback and healthy areas.

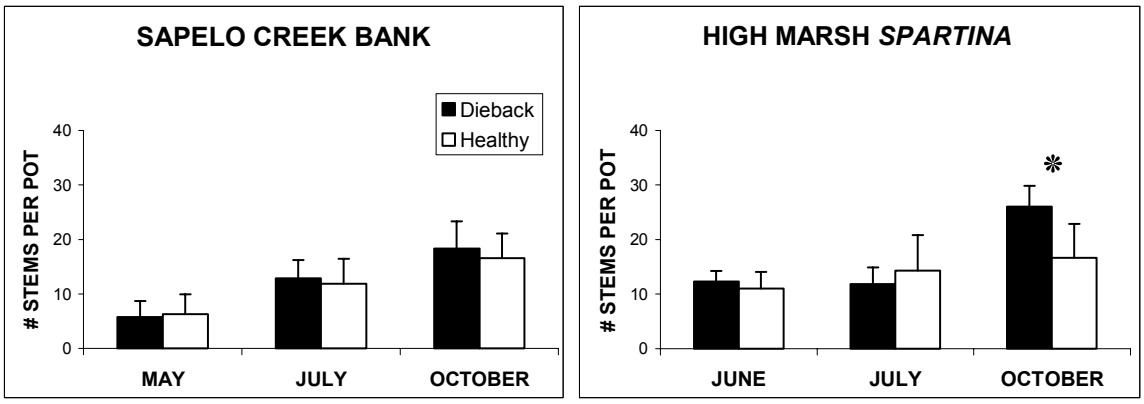

Figure 3-5. The number of stems per pot was determined for *S. alterniflora* at the edge of a dieback area transplanted into dieback areas (black bars) or healthy areas (white bars). Error bars represent standard deviation. Asterisk indicates a significant difference in growth between dieback and healthy areas.

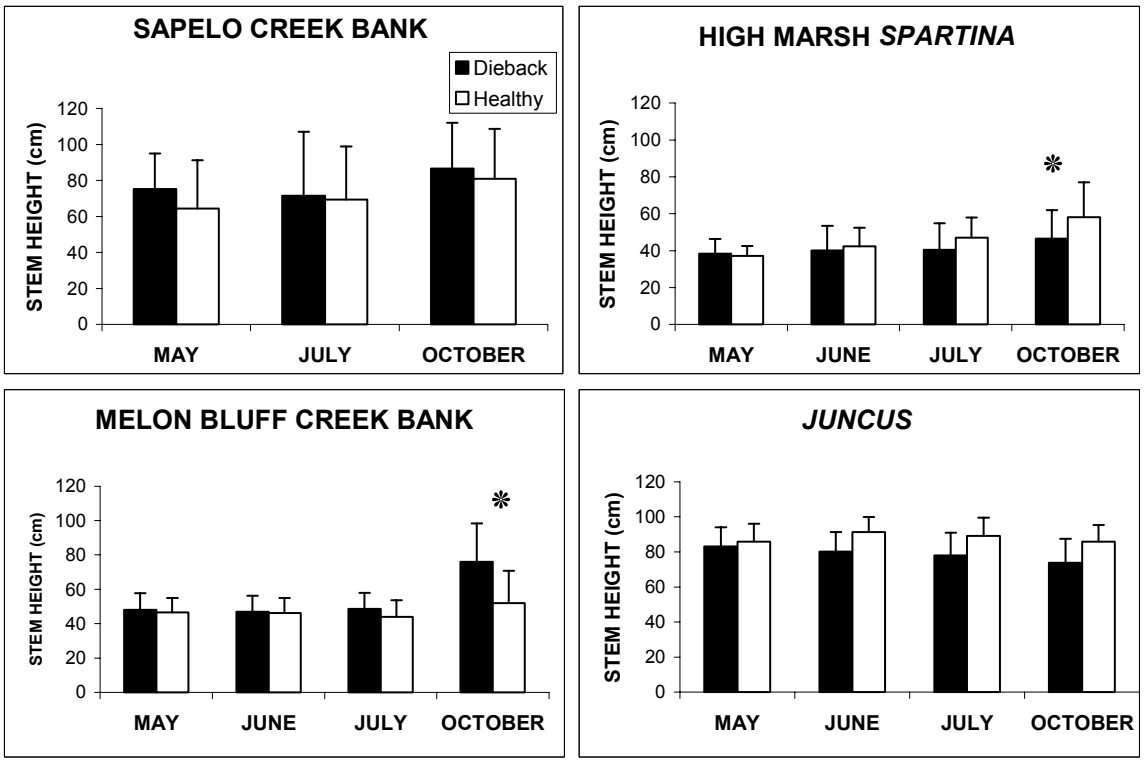

Figure 3-6. Average height of the 5 tallest stems in each pot transplanted from healthy areas into dieback areas (black bars) or healthy areas (white bars). Error bars represent standard deviation. Asterisk indicates a significant difference in growth between dieback and healthy areas.

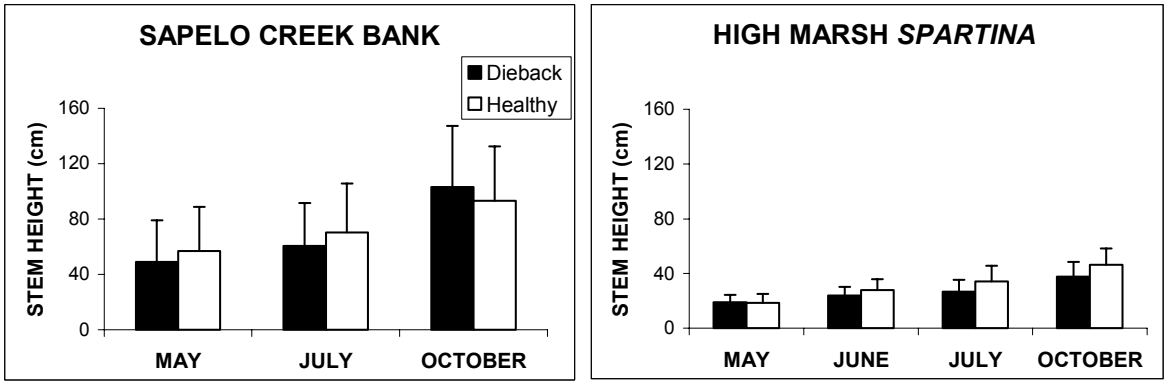

Figure 3-7. Average height of the 5 tallest stems in each pot transplanted from dieback areas into dieback areas (black bars) or healthy areas (white bars). Error bars represent standard deviation. No significant differences in growth were observed.

Table 3-1. Changes in average height of the five tallest stems and number of stems per pot from May (Initial) to October (Final) at each experimental site. Bold type indicates significantly differences ($p < 0.05$) in growth between dieback and healthy areas.

1. Transplants from healthy area to either dieback area (treatment) or healthy area (control).

		Height of 5 Tallest Stems		Number of Stems per Pot	
Site	Treatment	Initial	Final	Initial	Final
Sapelo creek bank	Dieback	78 ± 9	87 ± 15	6 ± 1	14 ± 4
	Healthy (control)	68 ± 13	81 ± 6	7 ± 2	12 ± 4
Melon Bluff creek bank	Dieback	48 ± 5	**76 ± 10**	8 ± 3	**22 ± 10**
	Healthy (control)	46 ± 6	52 ± 2	9 ± 2	12 ± 3
High Marsh *Spartina*	Dieback	38 ± 4	47 ± 10	10 ± 4	30 ± 8
	Healthy (control)	37 ± 4	**58 ± 11**	12 ± 3	29 ± 7
Juncus	Dieback	83 ± 9	74 ± 9	14 ± 5	18 ± 6
	Healthy (control)	86 ± 8	86 ± 7	15 ± 6	22 ± 8

2. Transplants from dieback area to either healthy area (treatment) or dieback area (control).

		Height of 5 Tallest Stems		Number of Stems per Pot	
Site	Treatment	Initial	Final	Initial	Final
Sapelo creek bank	Dieback (control)	53 ± 16	103 ± 31	6 ± 3	18 ± 5
	Healthy	65 ± 22	93 ± 24	6 ± 4	17 ± 5
High Marsh *Spartina*	Dieback (control)	19 ± 4	38 ± 8	12 ± 3	**26 ± 6**
	Healthy	18 ± 6	46 ± 10	11 ± 3	17 ± 6

Table 3-2. Analysis of Variance for the change in height of the 5 tallest stems and number of stems per pot (stem count) for transplant sites where reciprocal transplants were carried out. "Source" is the source of transplants (healthy or dieback area) and "destination" is the location plants were transplanted into (healthy or dieback area). Significance ($p < 0.05$) is indicated by bold type.

High marsh Spartina	**Source**	**Destination**	**Source x Destination**
5 tallest stems	**<0.0001**	**<0.0001**	0.9387
Stem Count	**<0.0001**	**0.0003**	0.7689
Sapelo Creek bank	**Source**	**Destination**	**Source x Destination**
5 tallest stems	**<0.0001**	0.0646	**0.0251**
Stem Count	**0.0019**	0.1180	0.4607

Table 3-3. Elemental composition and C:N ratios of transplants in October, 2003. There were no significant differences ($p < 0.05$) between treatments and controls. "n.d." indicates values were below the detection limit of the instrument.

	Juncus		Sapelo creek bank			
	Healthy to:		Healthy to:		Dieback to:	
Element	Dieback	Healthy	Dieback	Healthy	Dieback	Healthy
%						
Carbon	46.7 ± 0.9	46.5 ± 0.7	44.7 ± 0.6	44.2 ± 1.0	44.8 ± 1.0	44.7 ± 0.6
Nitrogen	1.4 ± 0.2	1.6 ± 0.2	1.2 ± 0.5	1.0 ± 0.3	1.3 ± 0.3	1.2 ± 0.5
Sulfur	n.d.	n.d.	0.6 ± 0.2	0.5 ± 0.1	0.6 ± 0.1	0.4 ± 0.1
C/N	38.2 ± 5.1	35.1 ± 4.6	43.6 ± 14.7	49.5 ± 11.5	41.6 ± 9.3	43.6 ± 7.0

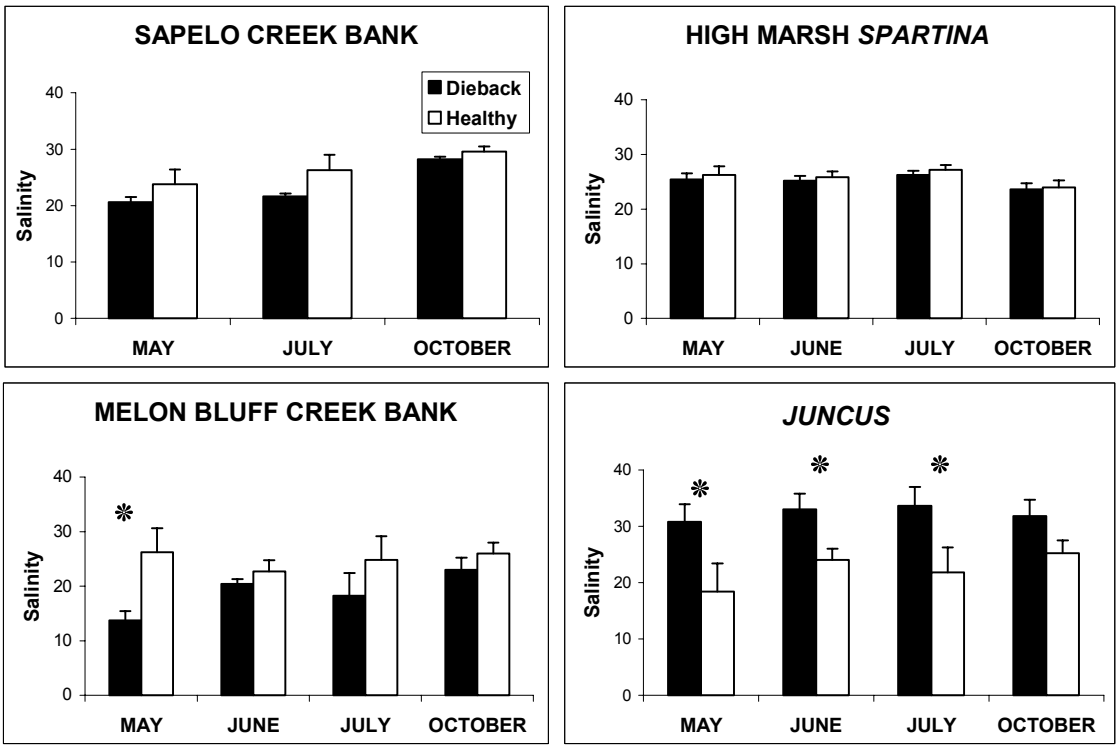

Figure 3-8. Soil pore-water salinity (PSU) measured at five locations within each dieback (black bars) and healthy area (white bars). Error bars represent standard deviation. Asterisks indicate a significant difference ($p < 0.05$) in salinity between dieback and healthy areas.

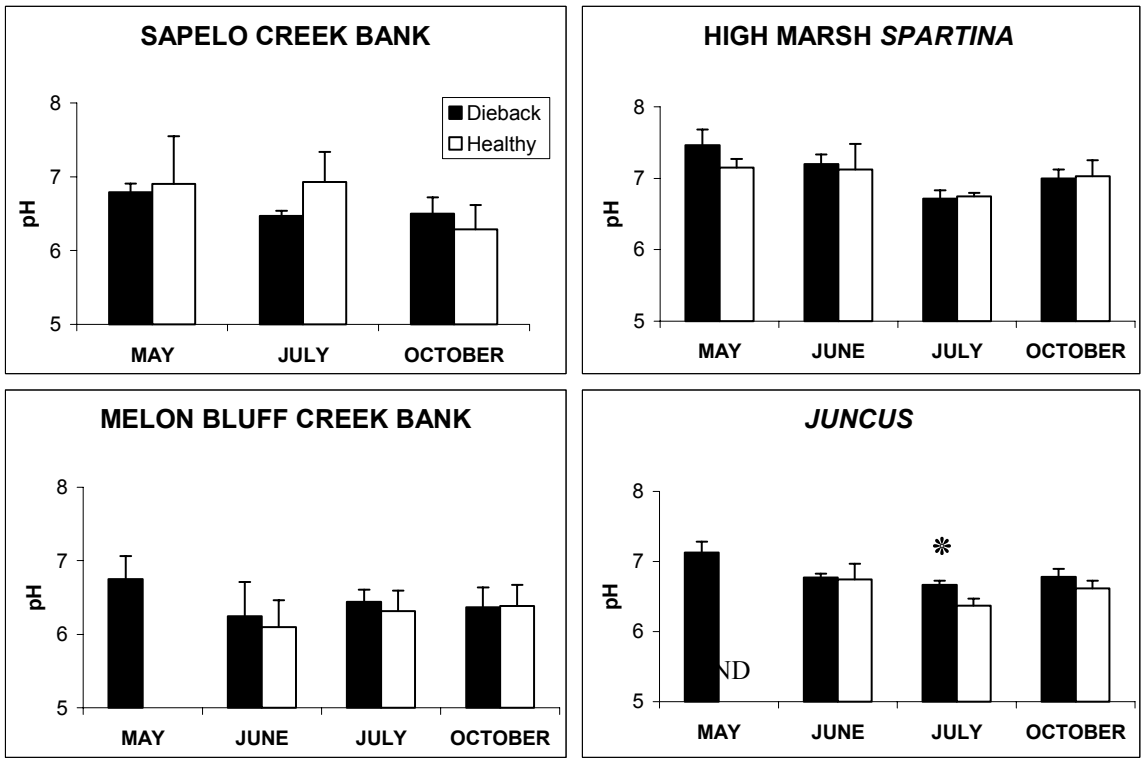

Figure 3-9. Soil pore-water pH measured at five locations in each dieback (black bars) and healthy area (white bars). Error bars represent standard deviation. Asterisks indicate a significant difference ($p < 0.05$) in pH between dieback and healthy areas.

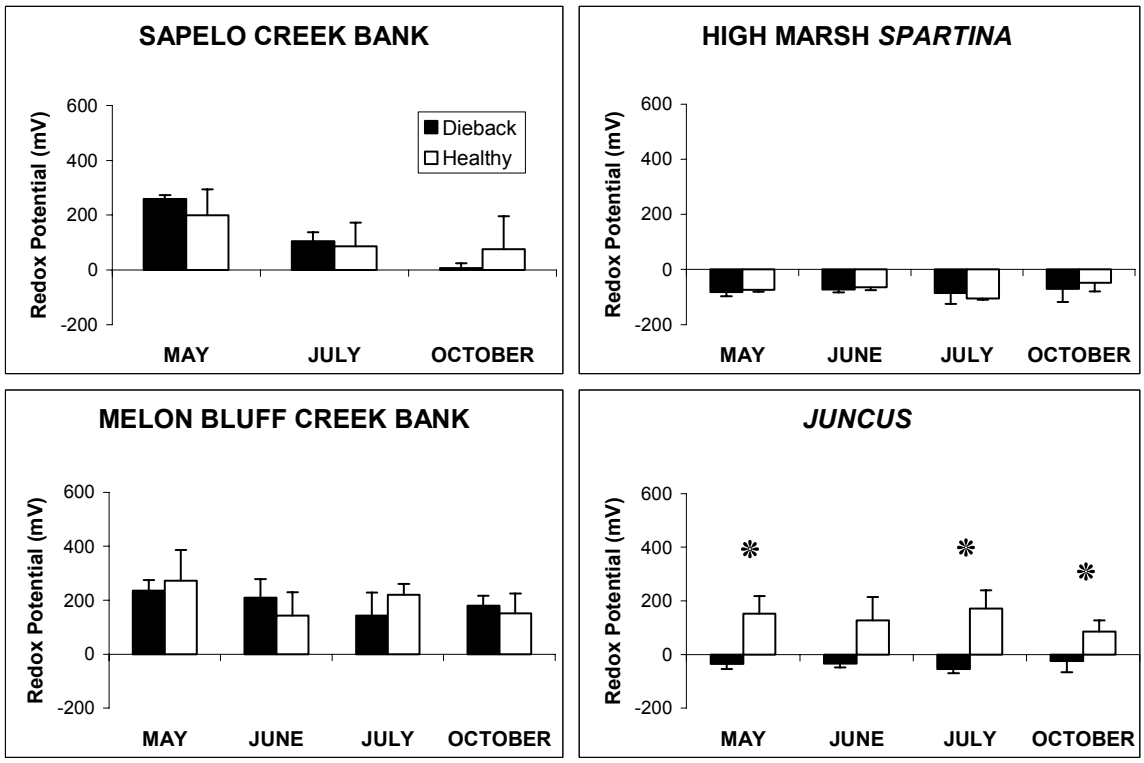

Figure 3-10. Soil redox potential at 15 cm depth measured at five locations in each dieback (black bars) and healthy area (white bars). Error bars represent standard deviation. Asterisks indicate a significant difference ($p < 0.05$) in redox potential between dieback and healthy areas.

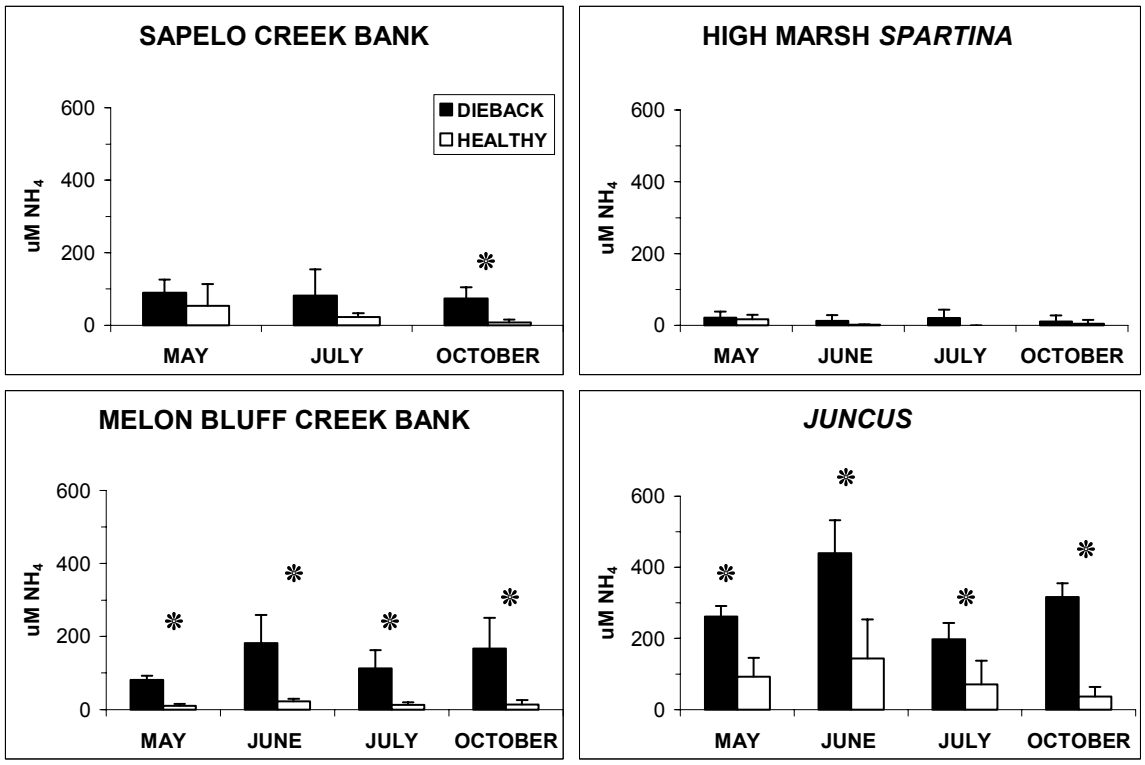

Figure 3-11. Concentration of NH_4 in soil pore-water measured at five locations in each dieback (black bars) and healthy area (white bars). Error bars represent standard deviation. Asterisks indicate a significant difference ($p < 0.05$) in NH_4 concentration between dieback and healthy areas.

Table 3-4. Soil salinity, pH, redox potential (Eh) and NH4 concentration measured in dieback and healthy areas at each experimental site. Values are average ± standard deviation. Bold type indicates significant higher values ($p < 0.05$).

SITE	SALINITY (PSU) DIEBACK	SALINITY (PSU) HEALTHY	pH DIEBACK	pH HEALTHY	Eh (mV) DIEBACK	Eh (mV) HEALTHY	NH_4 (uM) DIEBACK	NH_4 (uM) HEALTHY
CREEK BANK SPARTINA - SAPELO ISLAND								
MAY	21 ± 1	24 ± 3	6.79 ± 0.12	6.90 ± 0.64	259 ± 14	199 ± 93	89 ± 36	54 ± 60
JULY	22 ± 1	26 ± 3	6.47 ± 0.07	6.93 ± 0.41	109 ± 33	86 ± 86	81 ± 72	22 ± 10
OCTOBER	28 ± 0	30 ± 1	6.50 ± 0.22	6.29 ± 0.33	7 ± 16	75 ± 120	**73 ± 31**	8 ± 8
CREEK BANK SPARTINA - MELON BLUFF								
MAY	14 ± 2	**26 ± 4**	6.75 ± 0.31	NO DATA	235 ± 40	273 ± 114	**81 ± 11**	10 ± 6
JUNE	20 ± 1	23 ± 2	6.24 ± 0.47	6.10 ± 0.36	210 ± 68	144 ± 86	**182 ± 78**	23 ± 7
JULY	18 ± 4	25 ± 4	6.44 ± 0.16	6.32 ± 0.28	144 ± 85	220 ± 41	**113 ± 50**	13 ± 7
OCTOBER	23 ± 2	26 ± 2	6.37 ± 0.27	6.38 ± 0.29	181 ± 37	152 ± 73	**167 ± 84**	13 ± 13
HIGH MARSH SPARTINA - MELON BLUFF								
MAY	25 ± 1	26 ± 2	7.47 ± 0.21	7.15 ± 0.12	-82 ± 15	-74 ± 7	21 ± 17	16 ± 12
JUNE	25 ± 1	26 ± 1	7.20 ± 0.13	7.12 ± 0.36	-73 ± 11	-65 ± 10	12 ± 17	1 ± 2
JULY	26 ± 1	27 ± 1	6.71 ± 0.12	6.75 ± 0.05	-85 ± 40	-105 ± 4	20 ± 24	0
OCTOBER	24 ± 1	24 ± 1	7.00 ± 0.13	7.03 ± 0.22	-70 ± 48	-49 ± 31	10 ± 17	4 ± 11
JUNCUS - MELON BLUFF								
MAY	**31 ± 3**	18 ± 5	7.12 ± 0.16	NO DATA	-35 ± 18	**152 ± 66**	**261 ± 29**	92 ± 53
JUNE	**33 ± 3**	24 ± 2	6.77 ± 0.05	6.74 ± 0.22	-34 ± 14	126 ± 88	**440 ± 92**	144 ± 109
JULY	**34 ± 3**	22 ± 4	**6.67 ± 0.05**	6.37 ± 0.10	-54 ± 16	**172 ± 68**	**198 ± 45**	66 ± 12
OCTOBER	32 ± 3	25 ± 2	6.78 ± 0.11	6.61 ± 0.11	-24 ± 42	**85 ± 42**	**316 ± 39**	26 ± 15

CHAPTER 4

CONCLUSIONS

Salt marsh dieback in Georgia is distinctly different from previous reports of dieback events at other locations. In Louisiana (historical dieback), Florida, South Carolina, North Carolina and Great Britain, salt marsh dieback has characteristically occurred in low-lying parts of the marsh where inundation and water-logging contribute to anoxic conditions and high sulfide concentrations (Goodman et al., 1959; Linthurst and Seneca, 1980; Mendelssohn and McKee, 1988; and de Souza and Yoch, 1997). In Georgia, however, dieback areas were most common in well-drained soils along creek banks and on relatively dry, elevated berms on the creek bank levee. Waterlogged, anoxic conditions were only observed in a few low-lying dieback areas. The hydrologic setting of dieback areas in Georgia was not consistent with the hypothesis that salt marsh dieback was caused by waterlogging stress and sulfide toxicity.

Of the dieback events at other locations, acute salt marsh dieback in Louisiana bears the closest resemblance to the Georgia dieback event. McKee et al. (2004) suggested that acute salt marsh dieback was related to drought and may have been caused by periods of soil desiccation resulting in decreased pH and increased bioavailability of the toxic metals Fe and Al. Similarly, salt marsh dieback in Georgia occurred during a period of severe drought. Areas of dry, cracked marsh soil were observed and vegetation died back in areas of marsh (creek banks and berms) where desiccation could have occurred during periods of drought and low tidal amplitude. Unfortunately, pH and plant elemental composition were not measured at the onset of the dieback. Multiple species were affected in both Georgia in Louisiana, suggesting that species

specific pathogens were not the cause at either location. Snail herbivory may also have contributed to dieback at both locations by enlarging dieback areas at some sites where snails naturally occur in high densities, but was not likely a causal agent, as high densities of snails and severe snail damage were not observed at most dieback sites.

Despite these similarities, some aspects of salt marsh dieback in Georgia were different from acute salt marsh dieback in Louisiana. The most striking difference is that *J. roemerianus* was highly affected in Georgia marshes but was unaffected in Louisiana. The pattern of dieback was also different among locations. In Louisiana, acute dieback occurred primarily in the interior parts of the marsh (McKee et al., 2004), but in Georgia, dieback occurred most often along creek banks and on elevated berms. This difference may be explained by differences in sediment deposition patterns or hydrology in marshes at the two locations. Alternatively, different patterns of dieback could be the result of different causes at the two locations.

Although we are confident in suggesting that drought was the ultimate cause of salt marsh dieback in Georgia, the specific mechanism by which dieback occurred cannot be determined from our data. Two drought-related mechanisms seem unlikely given the results of this study. Increased soil pore-water salinity has often been suggested as a potential cause, but salinities were within normal ranges for Georgia salt marshes both during our study (which took place after the dieback occurred) and during initial field sampling efforts in October 2002 (during the dieback). Sulfide toxicity in waterlogged soils also probably did not occur, as the soil in dieback areas tended to be oxidized by the time we measured it. Other possible causes such as temporary sediment desiccation and acidity leading to Fe and Al toxicity, the proliferation of salt-tolerant pathogens, and weakened plant defenses against common pathogens cannot be evaluated based on the results of this study.

Monitoring programs and further experimental manipulations will be helpful for advancing our understanding of this phenomenon. Studying the processes underlying dieback events as they occur is the only definitive way to determine the mechanisms by which salt marsh dieback causes plant death. Monitoring efforts are needed to identify the early stages of future dieback events so that hypotheses about the causes of dieback can be tested. These efforts should be particularly intense during periods of moderate to severe drought, when salt marsh dieback is most likely to occur again. Additionally, monitoring should be carried out at the most extensive dieback areas (such as the Jerico River, Melon Bluff and Harriet's Bluff), as these areas may be the most susceptible to future dieback events.

Continued monitoring efforts are also needed at Georgia Coastal Research Council (GCRC) monitoring sites to be able to document the natural progression and recovery of dieback sites. At the time of this writing, recolonization of dieback areas has only occurred by rhizome growth from nearby healthy areas or from clumps of live vegetation remaining in the dieback area. Seedling germination apparently has not occurred, so areas with no nearby live vegetation have no source for recolonization. If this remains the case, large dieback areas such as the 600 acre site on the Jerico River will not recover for some time and restoration efforts may be needed to revegetate these areas. The survival and growth of transplants in this study indicates that restoration is possible. However, before large scale restoration efforts are undertaken, trials should be carried out to determine if restoration speeds the recovery of dieback areas and to determine optimal sources and transplanting methods to make restoration as cost effective as possible.

Until dieback occurs again, experimental manipulations can be used to investigate the causes of dieback. Greenhouse experiments could be carried out to determine the responses of

marsh plants to various stressors, but such experiments cannot simulate the variety of stressors present in the field. Experimental field simulations of potential mechanisms of salt marsh dieback would be much more effective. Experiments could test for the effects of soil desiccation, spikes in acidity accompanied with increased bioavailability of Fe and Al and the susceptibility of drought-stressed vegetation to pathogens and herbivores. In addition to testing individual stressors, experimental manipulations could be designed to examine the interactions between multiple factors that might combine to cause salt marsh dieback. Understanding the causes of salt marsh dieback would be useful for predicting and possibly preventing future dieback events.

If dieback events begin to occur on a regular basis, it will be essential to understand the larger impacts of salt marsh dieback on coastal ecosystems. The salt marsh is one of the most productive ecosystems on earth and forms the basis of estuarine food webs (Pomeroy and Wiegert, 1981). A wide variety of animals inhabit salt marshes, including many commercially important species such as blue crabs, shrimp and various fish that use the marsh as a nursery habitat. Marshes also serve as filters for sediments and pollutants reaching the coastal zone via rivers or coastal runoff. Salt marsh dieback may interrupt these ecosystem functions by limiting production, reducing cover for juvenile species and increasing erosion. Changes in ecosystem function during dieback events need to be assessed to determine the broader impacts of salt marsh dieback events.

The economic costs of failing to understand the causes, progression and consequences of salt marsh dieback may be substantial. Salt marsh dieback affects the viewshed (aesthetic quality of the landscape) of coastal communities and could reduce both property values and tourism in areas where dieback is commonly occurring. Decreases in the nursery habitat of

fishery species could lead to reduced catches and closed fisheries if dieback becomes widespread. Finally, increased erosion of marsh sediment could fill in coastal waterways and increase the need for dredging. Although these predictions may be extreme, the fact remains that salt marsh dieback is a serious and potentially reoccurring problem in coastal ecosystems for which the causes and consequences are poorly understood.

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APPENDIX I: "DEAD MARSH" BIOBLAST OVERVIEW

Georgia Coastal Ecosystems (GCE) LTER Program

Field Sampling: Jan McKinnon, Brooks Good, Jill Huntington (Georgia Department of Natural Resources Coastal Resources Division); Susan White, Matt Ogburn, Steve Pennings, Dale Bishop (Georgia Coastal Ecosystems Long Term Ecosystem Research Program)

Participating GCE Laboratories: Tim Hollibaugh, Steve Pennings, Mary Ann Moran, Steve Newell, Merryl Alber, Samantha Joye

Summary

On October 16, 2002, a team of scientists and graduate students from the Georgia Coastal Ecosystems LTER accompanied DNR Coastal Resource Division staff on a site visit to the Jericho River in Liberty County, GA, where the salt marsh is exhibiting signs of die-off (the so-called "dead marsh" phenomenon). The group performed transects at 3 sites exhibiting signs of die-back (one in an area that was completely devoid of vegetation and two that were only partially denuded) as well as at a nearby control site. Each transect was a total of 200 feet in length and samples were taken for analysis of both physical (water and soil) and biological (plants, animal, microbes) characteristics. Interstitial water samples are being analyzed for salinity, sulfide and sulfate levels, dissolved inorganic nitrogen concentrations, and potentially for metals. Soil samples are being analyzed for salinity and mineral content. Plants were identified, counted, classified as live (either tall or short shoots) or dead, and samples were obtained for plant tissue content (CNS, potentially metals). Infaunal animals were sampled with cores and field counts were made of crab holes and periwinkle snail density (*Littoraria irrorata*). Samples of both stems and rhizomes obtained from live and dead areas are being analyzed to characterize bacterial and fungal community composition. Samples were also obtained to perform two growth trials in a greenhouse: one to determine whether *Spartina alterniflora* rhizomes from the dead marsh are viable if given fresh water and the second to determine whether transplants from a nearby healthy marsh can survive in soil from the dead marsh site.

General Observations

The most extensive die-offs are upstream of I-95, but there are also abundant smaller die-offs downstream. The places where dead and dying marsh have been observed do not exhibit an obvious pattern. In areas where the main channel was curving, the die-offs appeared somewhat more extensive on the inside (accreting) side of the channel rather than on the outside (eroding) side.

We have made a preliminary classification into the following four categories: marshes where only the grass along the creekbank is dead; marshes where a live creekbank occurs in front of a fairly large patch of dead area; marshes that are completely denuded; and marshes where live and dead areas are interspersed that do not readily fit any of the above patterns (Figure 1). Many of the observations made on the ground along the Jericho River fall into the first category,

where only the area where tall-form Spartina generally occurs along the creekbank has been affected. The second type of marsh die-off pattern, where a live creekbank fronts a dead area, is readily obvious in aerial photographs but this type of pattern was not observed on the ground. In a few cases small patches of live tall-form Spartina on little mud islands (usually sections of creekbank that had slumped into the creek to form small bars) were observed, and there were also some areas where mid-marsh stands of Spartina were observed to be less dense than healthier creek areas. The third type of marsh die-off looks essentially like a mud flat, with areas where the marsh has started to visibly slough off into the water. The fourth type is more difficult to categorize except to say that there is no obvious pattern to the places where live and dead plants occur.

The plants that were affected by the die-off are *S. alterniflora* and *Juncus roemerianus*. Where both species were affected, Juncus was affected equally or more severely than Spartina. The upland border, with Juncus and shrubs, was not usually affected, and there have been observations of *Salicornia* species invading bare mud and also of live *Borrichia frutescens* in an otherwise bare area. Where *S. alterniflora* has been affected there is very little standing dead (or brown marsh), as has been described in Louisiana. However, large patches of standing dead plants were observed in the Juncus marsh.

Macroinvertebrates (*Littoraria irrorata, Geukensia demissa, Uca pugnax*) appeared reasonably abundant in both Spartina- and Juncus-dominated marshes, with few dead shells littering the marsh. At the Juncus marsh site (Figure 2), high numbers of *Melampus bidentatus* were massed in groups in small depressions at the base of plants. Most were large adults, and they were distributed all the way to the creekbank, which is generally not observed at the GCE marsh sites. Although densities appeared higher than normal, it may have been that the snails were aggregated and just easier to see due to the lack of plant cover. Plants at this site also showed signs of grasshopper grazing.

Methods:

Three transects were performed at "dead marsh" sites along the Jericho River (Figure 2). Transect A was in an area that was completely denuded, although evidence of *S. alterniflora* was visible from the dead stubs remaining in the soil. Transects B and C were on the opposite sides of the marsh from transect A in areas where live and dead *S. alterniflora* was interspersed with bare mud. Transect D was a control and was performed in a marsh that contained live, healthy Spartina plants (estimated at > 95%) and was considered unaffected. Note that none of these transects were done in a Juncus marsh.

Each transect was 200 feet long and was permanently marked with PVC poles at the creekbank end and the inland end. Water, soil and plant samples were taken at 5 points (every 50 feet) along each transect. Epifaunal animal observations were recorded at each sample point and cores for infaunal animals were collected at 0, 100, and 200 feet along each transect. Samples were also collected for a comparative analysis of microbial (bacterial and fungal) composition in live and dead areas near Transect A and in the healthy site near Transect D.

Interstitial water samples were collected with PVC sippers at a depth of approximately 15 cm in the rooting zone of *S. alterniflora*. Salinity was measured with a refractometer. Sulfide samples (10 ml) were collected in acid-washed glass vials and immediately fixed with ZnAc and kept cold. The remaining water was returned to the laboratory, where aliquots were filtered through GF/F filters for analysis of dissolved inorganic nitrogen (NH_4, $NO_2 + NO_3$). Additional water was filtered through 0.2-µm acrodisc filters and fixed with HNO_3 for sulfate analysis. It

may also be possible to perform additional analyses on these samples for dissolved minerals and heavy metals.

Soil samples were collected from a depth of approximately 15 cm in the *S. alterniflora* rooting zone. Each sample was separated into two aliquots: one for measurement of interstitial salinity (using the dry/wet weight method routinely used in the GCE) and one for routine analysis of salts, metals, etc.

Plants were identified, counted, and classified as live or dead. Dead plants were quantified by counting vegetation stubs (no standing dead plants were observed). Live plants were categorized as either tall (> 15 cm) or short (< 15 cm) shoots. Tissue samples were obtained for plant tissue CNS content and potentially for metals.

Infaunal animals were sampled with cores (10 cm diameter x 15 cm deep), refrigerated until they could be washed through a 500 μm mesh sieve, and fixed in 10% formalin with Rose Bengal stain. Organisms were sorted from organic material and debris and transferred to 70% ethanol for preservation. Aliquots of the material passing through the 500 μm mesh screen were subsequently washed over a 63 μm mesh sieve to collect meiofaunal organisms. These samples were preserved as above. Animals in each core will be identified to the lowest taxonomic level possible and counted. Field counts were made of crab holes and periwinkle snail density (*L. irrorata*) in a manner similar to that being performed as part of the LTER invertebrate sampling protocol. The quadrat size used for the crab hole counts was 500 cm^2 and that for the snail counts was 2500 cm^2. No *M. bidentatus* were observed along these transects.

Microbial community composition is being analyzed on samples obtained from *S. alterniflora* leaves and rhizomes collected at the "Dead" marsh from patches of live *S. alterniflora* adjacent to Transect A (at a distance of approximately 400 feet from the creekbank) and at the reference site (Transect D). Both bacterial and fungal DNA is being extracted from these samples for determination of community composition via molecular methods. Samples are also being examined microscopically to identify fungi.

Additional field work *S. alterniflora* "scalloped" edges (borders where live Spartina and dead marsh are clearly defined, typically along a creekbank) were flagged adjacent to Transects B and C to follow future changes in border position on the creekbank.

Greenhouse trials Samples were also obtained to perform two growth trials in a greenhouse: one to determine whether *S. alterniflora* rhizomes from the dead marsh are viable if given fresh water and the second to determine whether transplants from a nearby healthy marsh can survive in soil obtained from the dead marsh site. For the rhizome viability trial, five blocks of soil (approximately 25 cm^2) were collected from both denuded areas and live areas near Transect A and transported back to the greenhouse in Athens. These pots are being watered regularly and any new growth will be monitored.

Soil samples for the transplant trial (25 cm^2 blocks) were collected from both the denuded marsh (Transect A) and the control site (Transect D). Healthy seedlings, ranging in size from 5 – 15 cm, were collected from the control site (Transect D). In the greenhouse, *S. alterniflora* was transplanted into 5 replicate pots from each site as well as 5 pots filled with a sand/peat moss (75/25) combination as a control. Two plants (one larger and one smaller) were transplanted into each pot. Survival and plant height are being monitored.

Results to date

Most of the analyses described here are ongoing. Results to date are limited to field observations of salinity, plant characteristics, and epifauna.

Salinity Salinity was measured two ways: on water samples obtained via PVC sippers and by rehydrating dried soil samples with a known amount of deionized water. Salinity in the water samples ranged from 26 to 35. These were approximately the same as salinities obtained in the soil samples, which ranged from 21 to 36 (Table 1).

Table 1. Salinities (PSU) along each transect, obtained by measuring interstitial water or soil samples.

Distance (ft)	0	50	100	150	200
Transect A - water	26	31	NA	30	32
- soil	25	24	25	29	31
Transect B - water	27	30	30	34	NA
- soil	33	23	36	32	30
Transect C - water	NA	28	33	35	34
- soil	21	31	34	28	24
Transect D - water	30	33	NA	28	27
- soil	28	34	26	36	28

Plants *S. alterniflora* was the dominant grass in all transects. (*J. roemerianus* was observed near Transect A but was not part of the transect). *S. alterniflora* characteristics varied among the transects (Figure 3). In Transect A (the denuded marsh), the area contained mostly dead vegetation stubs and no live stems were observed. In Transect B live plants were observed over the first 100 feet of the transect, and dead stubs were observed between 50 and 200 feet. Transect C was patchy, with high densities of dead stubs observed in the middle of the transect. Transect D, the healthy marsh, had a fairly even distribution of live plants (averaging 12.6 ± 3.4 plants per 500 cm^2). No dead stubs were recorded at this site, but they may have been covered by the incoming tide. The proportion of tall (> 15 cm) versus short (< 15 cm) shoots was fairly similar in all locations where live plants were observed (Figure 4).

Epifauna The density of *L. irrorata* showed interesting differences among the four transects (Figure 5). In Transect A, where no live plants were present, no snails were found at all. In transects B and C, snail densities were highest at the 0 and 50 foot sampling sites (where they ranged between 20 and 48 snails per m^2). These locations did not correspond to the highest densities of either live or dead plants. In the reference Transect (D), snail densities were much lower. For comparison, *L. irrorata* densities observed across GCE sites in October 2001 averaged 17 m^{-2} at creekbank sites and 181 m^{-2} at mid-marsh sites.

The number of crab holes was highest at the creekbank in the dead marsh site (Figure 6). Crab hole density was considerably lower in the other two impacted sites, ranging from 0 to 9 holes per 500 cm^2. Crab holes were not counted at Transect D because the flooding tide had submerged the site and holes could not be located. During the concurrent GCE sampling, crab hole density averaged 13 ± 9 per 500 cm^2 and ranged from 0 to 70.

Figure 1. Dead marsh types. Photographs taken along the Georgia coast in spring 2002 show die-off categories discussed in the text: die-off concentrated at the creekbank (top left); die-off behind the creekbank (top right); die-off affects the entire marsh (bottom left); die-off pattern is erratic (bottom right).

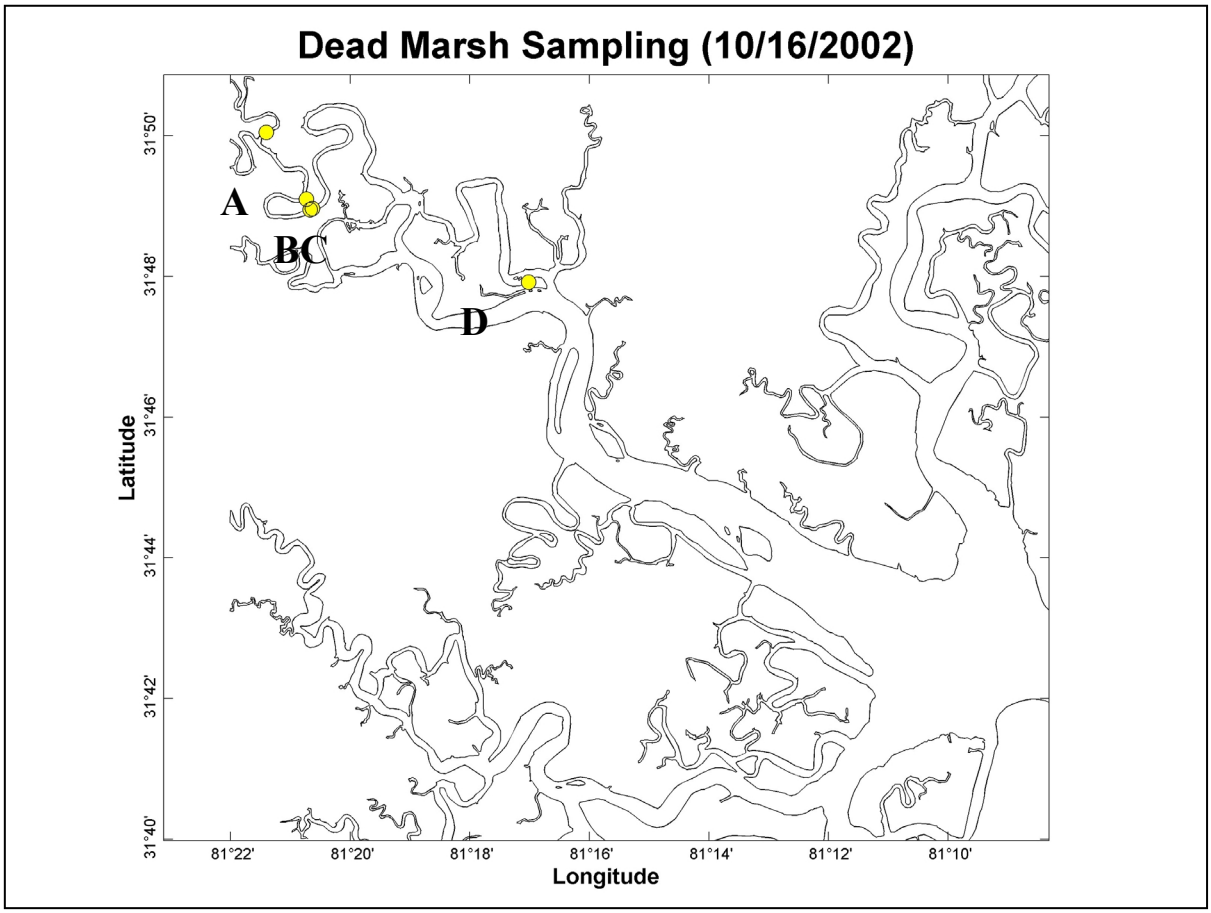

Figure 2. Study site. Circles show the location of the sites along the Jerricho River that are discussed in the text. The site furthest upstream is the site of the Juncus marsh; sites labeled A through D are where transects were sampled.

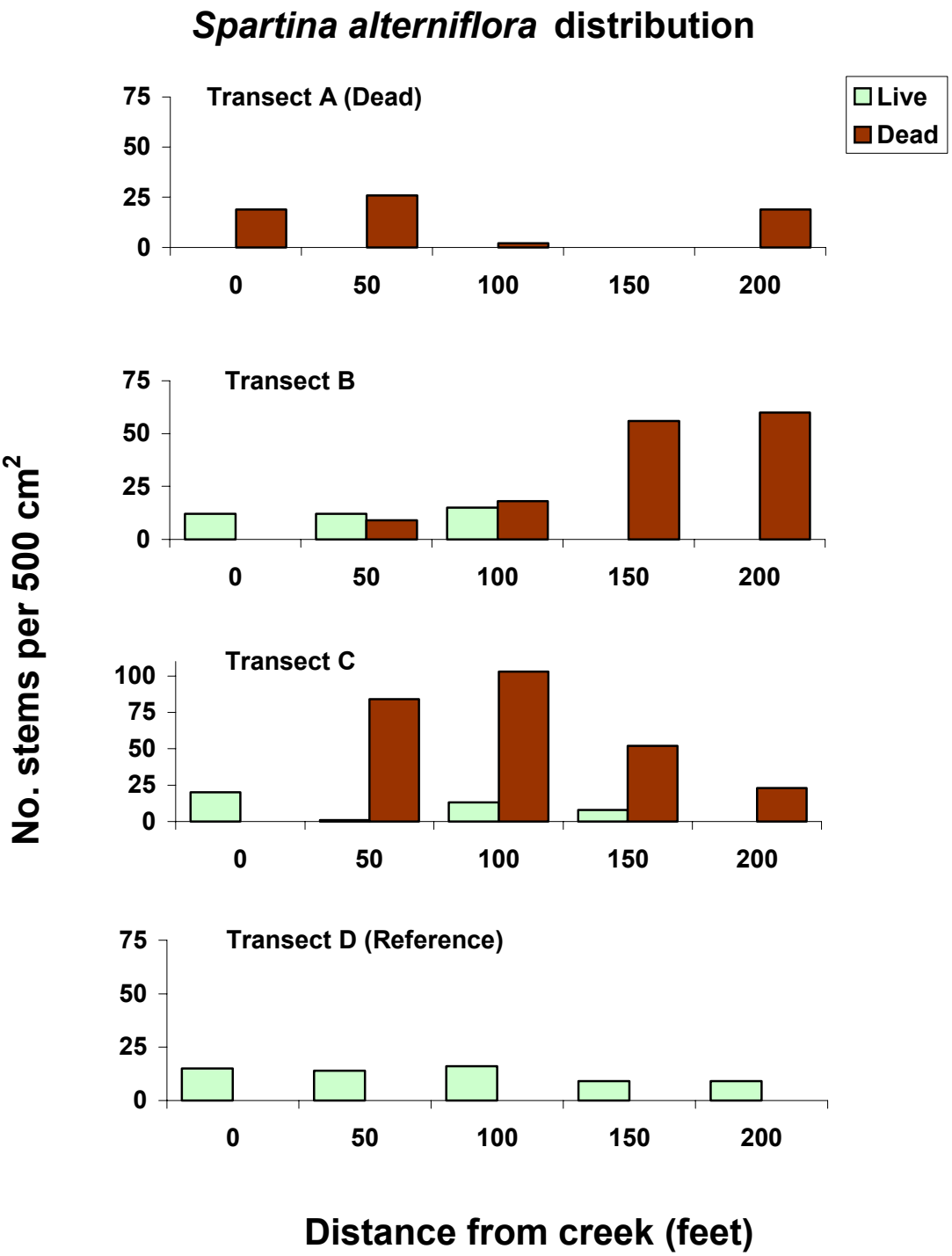

Figure 3. Distribution of *Spartina alterniflora*. The four graphs depict the number of live and dead *S. alterniflora* stems along Transects A through D.

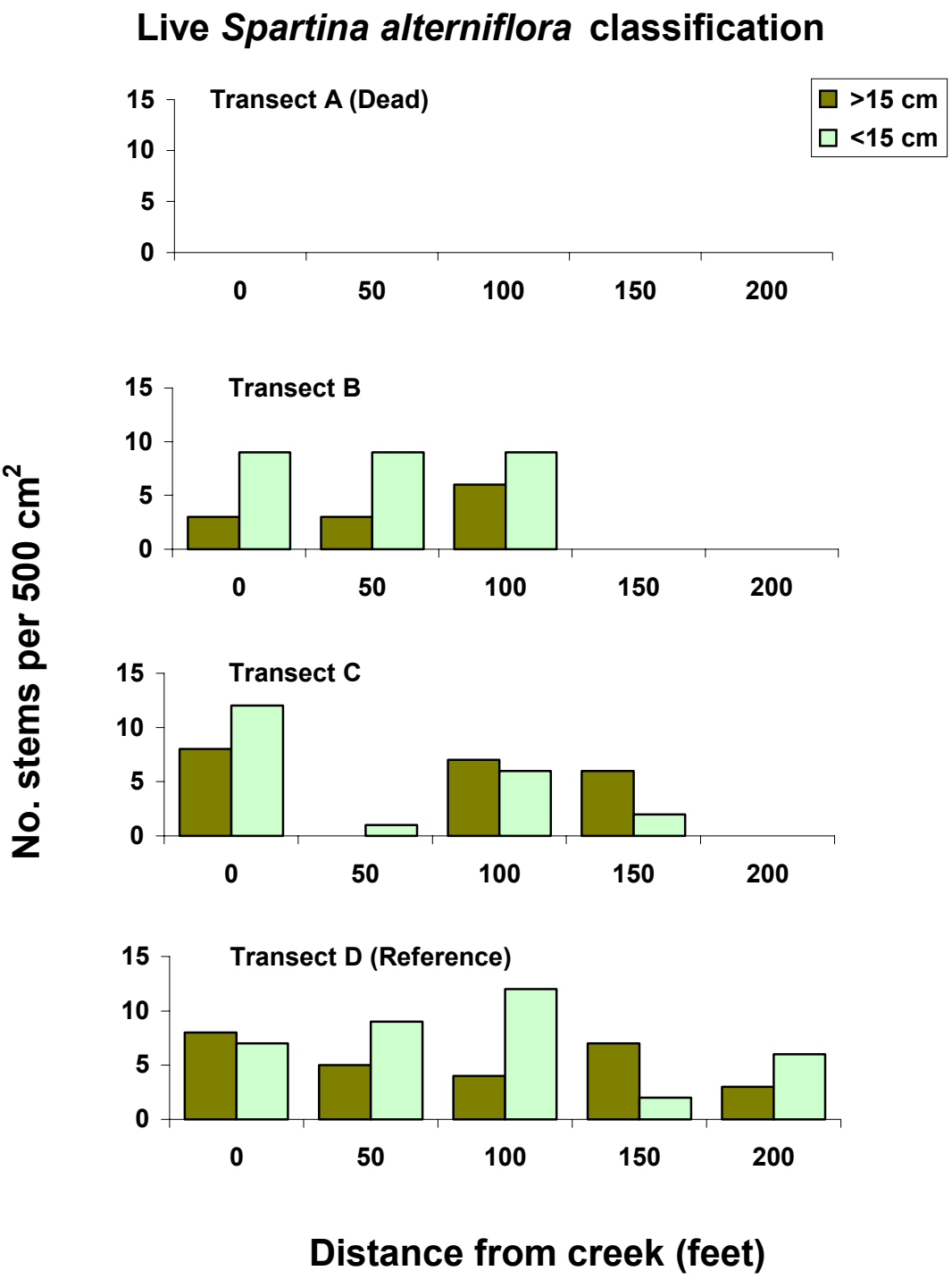

Figure 4. Classification of live *S. alterniflora*. The four graphs depict the number of live shoots classified as tall (> 15 cm) or short (< 15 cm) along each transect.

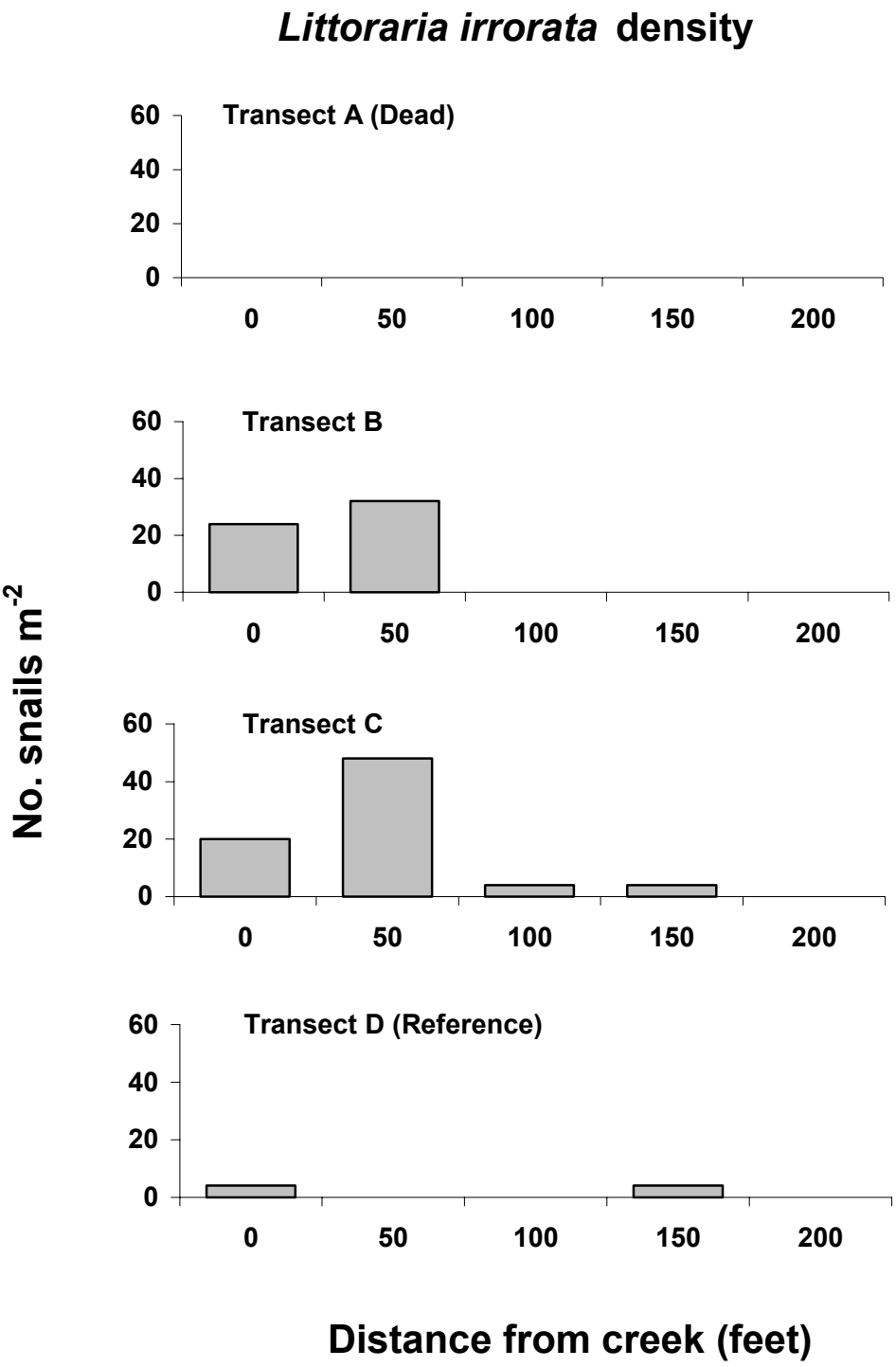

Figure 5. Distribution of *Littoraria irrorata*. The four graphs depict the density of *L. irrorata* along each transect.

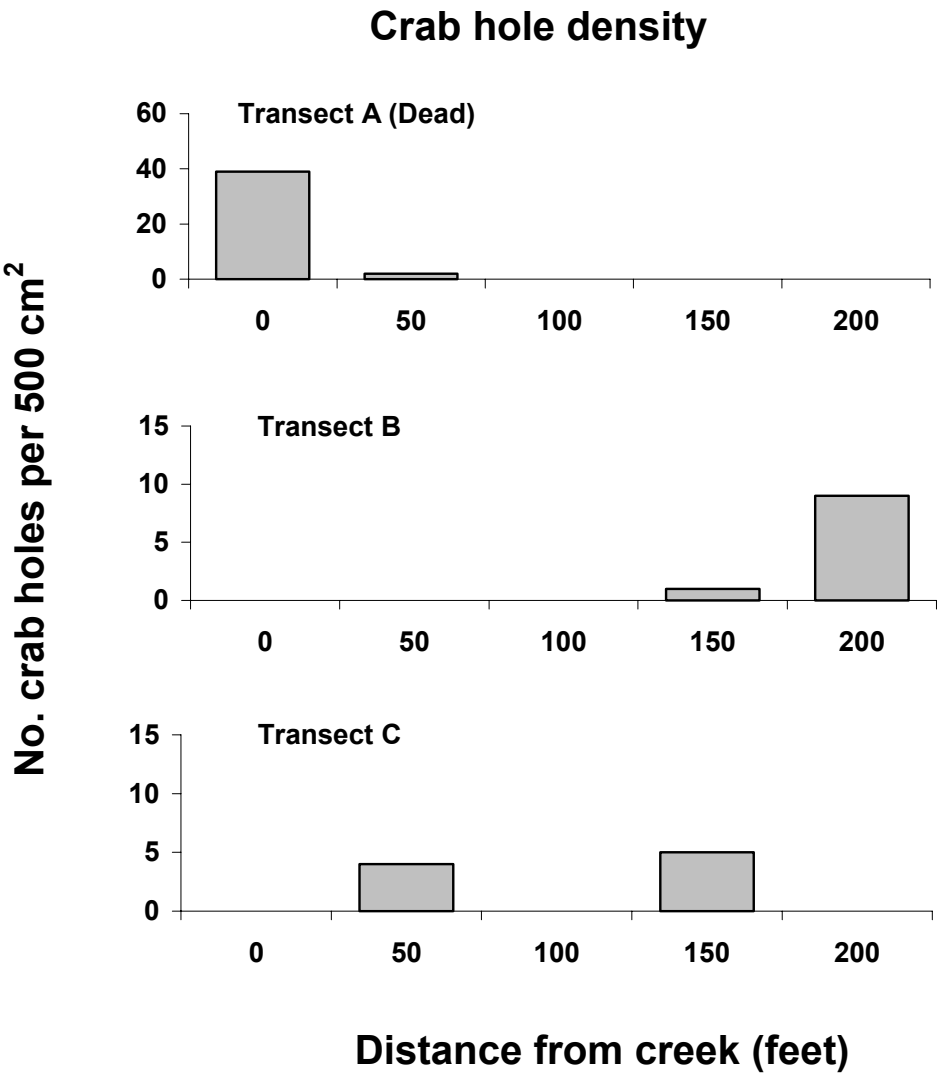

Figure 6. Distribution of crab holes. The four graphs depict the density of crab holes along each transect.

APPENDIX II: MARSH SAMPLING PROTOCOL

Any comments on the protocol should be directed to Matt Ogburn (ogburn@uga.edu). Groups interested in participating in this standardized monitoring effort should contact Joe Richardson (richards@savstate.edu).

Download and print standardized data sheet (PDF)
You'll need one copy for each quadrat (18).

Overview:
This protocol provides a standardized method for monitoring physical, chemical, and biological characteristics of marshes. We recommend that sites be established in both marsh die-off and control areas, and that they be monitored quarterly (March, June, September, and December).

Setup:
Individual sites may require adjustment of the protocol.

Ladders (flat on the marsh surface with a plank across them) are recommended for accessing some areas.

At each sampling site, set up 3 transects 10 m apart (ideally) in a dieback area and an unaffected area. Transects should; 1) run the length of the area, 2) be 2 m wide, and 3) run perpendicular from a creek bank to the marsh interior. Make sure not to walk within the transect area to minimize impact to the marsh. Mark the location of 3 permanent quadrats (0.5 m x 0.5 m) evenly spaced along the length of each transect with a PVC pole. Prior to installation, the PVC should be calibrated as follows: a zero point should be marked with a Sharpie or labeling tape, and then the pole should be marked at 5 cm intervals +/-25 cm above and below the zero point. When the pole is placed in the marsh, the zero point should sit at the marsh surface so changes in marsh surface height can be measured over time.

If a clear transition zone is evident (strong demarkation between live marsh and dieback area), use permanent flags to mark the transition (the border between mud and plants) as a way to document changes in the extent of the dieback area.

Take a photograph (if possible) of each quadrat, the transects, and any usual features. If a GPS unit is available, record the location of each transect.

There should be a total of 9 quadrats in the 'healthy' marsh area and 9 in the dieback area (3 quadrats/transect x 3 transects) for a total of 18 quadrats per site.

Sampling frequency:
Sampling should be carried out quarterly (March, June, September, December) at low tide (begin approximately 2 hrs before low tide). Be sure to record the time of sampling.

Before starting vegetation counts, prepare holes (see below) for collecting porewater.

Vegetation:
Marsh vegetation should be monitored within each 0.5 m x 0.5 m quadrat set up along the transect. In addition, the location of major vegetation boundaries along the transect (dead marsh - live marsh, Spartina - Juncus, etc.) should be recorded.

Stem counts:
Plant stems should be counted in each quadrat. Only stems that are rooted inside the quadrat should be counted. Separate counts should be made for each species present. Three categories of stems should be used: live stems >15 cm (tall shoots), live stems <15 cm (short shoots), and dead stems.
Plant height:
The height of the five tallest plants in each quadrat should be measured to estimate vegetation height. Where more than one species is present, plant heights should be recorded for each species.
Leaf color:
Observations of leaf color should be recorded.

Epifauna:
Note live and dead fauna. All counts are done in the same quadrats used for the vegetation survey.

If snails and crab holes are too numerous to count in a the 0.5 x 0.5 m quadrat, use a designated (southeast, or lower-right) corner of the permanent quadrat, and count in 0.25 m x 0.25 m areas. Be sure to specify quadrat size on the data sheet.

Snails:
Species should be identified and separate counts made for each species. Snails should be counted only if they are on the ground or on plant stems rooted inside the quadrat (snails on overhanging stems should not be counted). For periwinkles, the number greater or less than 10 mm in size (measured from the aperture to the apex of the shell) are recorded separately.
Crabs:
Crab densities should be measured by counting the number of crab holes > 5 mm (= approximately the diameter of a pencil).
Mussels:
Counts are made in 0.5 m x 0.5 m quadrats. Note whether dead mussels are present.

General:
Note the presence or absence of a clear transition zone, and the distance for each quadrat from this area. If possible, sketch the site and label the quadrats.
Other observations:

How does the soil look, feel, smell? Is sulfide obviously present? Is there erosion? Unusual drainage patterns?

Physicochemical Characteristics:
If possible, get a temperature reading (useful if corrections are needed for salinity or pH) on the soil (we will try to obtain some probe-type soil thermometers).

Set this up before vegetation counts.
Porewater collection and analysis
Use a broomstick-sized rod (or PVC) to core a 15 to 20-cm deep hole adjacent to each quadrat to allow it to fill with porewater. Sample the water in this hole (~15 cm below marsh surface) for measurements of salinity (with a refractometer), pH (use a meter, not test strips or dye kit), and Eh (if possible).