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## Physical and microbial responses of dredged sediment to two-soil-stabilizing amendments, xanthan gum and guar gum, for use in coastal wetland restoration

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PHYSICAL AND MICROBIAL RESPONSES OF DREDGED SEDIMENT TO TWO SOIL -  
STABILIZING AMENDMENTS, XANTHAN GUM AND GUAR GUM, FOR USE IN COASTAL  
WETLAND RESTORATION

A Thesis

Submitted to the Graduate Faculty of the  
Louisiana State University and  
Agricultural and Mechanical College  
in partial fulfillment of the  
requirements for the degree of  
Master of Science

in

The Department of Oceanography and Coastal Sciences

by  
Lauren Land  
B.S., University of Maryland – College Park, 2008  
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## **ABSTRACT**

In wetland sediments, organic matter provides a substrate for microbial activity. During metabolism, microbes release extracellular polymeric substances, which accumulate to bind soil particles. A similar concept can be implemented on a large scale to reduce wetland loss in Louisiana. Hypothetically, hydraulically dredged sediment can be amended with polymer and deposited on subsiding marshes as a restoration method where the polymer increases sediment stabilization until plants become established. This lab study focused on investigating the influence of natural polymer additions on particle aggregation to increase sediment stability and the effects on microbial activity.

Sediments from three sites (i.e. freshwater, intermediate, and marine) were used, which varied in moisture content, organic matter content, salinity, and texture. The soil amendments were xanthan gum, a microbially produced polymer, and guar gum, a plant polysaccharide. Following polymer application, sediment-polymer mixtures were incubated for 1, 8, 16, or 26 weeks before analysis. Response variables included moisture content, redox potential, pH, dewatering, consolidation, aggregate size, microbial biomass, and basal respiration.

Polymer addition increased microbial activity in the first week. Lower redox potentials indicate that more carbon substrates were available to serve as electron donors for microbial use. High respiration rates suggest a microbial response to polymer addition with increased activity and growth, followed by rapid turnover of the biomass. At the 0.5% polymer concentration level, microbes assimilated carbon as indicated by respiration similar to control samples. At the 1% polymer concentration level, increased respiration indicates a transition to an increasing biomass pool.

Microbial response to added polymer carbon indicates that microbial communities degraded the polymers within one week of application. No evidence of increased aggregation was found, supported by no polymer effects on dewatering and consolidation.

Natural polymer additions may not achieve the goal of increasing sediment stability, due to their water-solubility and simple structure, which contributed to rapid degradation by microbes. High moisture content of wetland sediments may require the use of synthetic polymers for aggregation. A material that maintains structure in water and resists microbial activity may be more successful in stabilizing wetland sediments.

## **CHAPTER 1: INTRODUCTION, LITERATURE REVIEW, AND PROJECT OBJECTIVES**

### **1.1 The Function of Wetlands**

In 1854, Henry David Thoreau wrote “without the wetland, the world would fall apart. The wetland feeds and holds together the skeleton of the body of nature.” Across the landscape, wetlands comprise the transition between dry upland and open water systems. Wetlands function as sinks, sources, and transformers of nutrients that provide ecosystem services benefitting humans, plants, and animals. Examples of ecosystem services include water filtration, water storage, diverse wildlife habitat, nutrient cycling, and storm protection. Wetlands are some of the most productive and most complex ecosystems on earth (Reddy and DeLaune 2008).

Unique properties of wetland soils determine their role in biogeochemical cycling, which determines the biota that can survive. The three main components of a wetland are hydrology, hydric soils, and hydrophytic vegetation (Reddy and DeLaune 2008). Hydrology defines the length of time and frequency of the water saturation process during which water moves up through the soil depth and remains at or near the sediment surface. Some wetlands are flooded for a few weeks every year whereas others are completely submerged all year round.

Water drives the availability of oxygen, which defines the presence of hydric soils. Soils remaining saturated for a long period of time exhibit anaerobic conditions, which determine the plants that will survive. Hydrophytic plants have adapted to tolerate saturated sediments with no oxygen and loose soil structure (Reddy and DeLaune 2008).

The presence of water and inherent soil saturation determines how wetlands fill the role of source, sink, or transformer. Wetlands can remove pollutants, nutrients, and suspended solids from the water column; these materials are retained on the soil surface by deposition. In addition, wetlands transform chemicals to be exchanged with atmospheric gases or to be made available for use by plants or microbes.

## **1.2 Wetlands in Coastal Louisiana**

In coastal Louisiana, sediment deposition on the deltaic plain began about 5,000 years ago as sea level rose following the most recent ice age. The Mississippi River drains about 41% of the contiguous 48 states; therefore, the River transports a significant sediment load to Louisiana ([www.lacoast.gov](http://www.lacoast.gov)). Throughout history, sediment in an expanding deltaic lobe accumulated until water from the River found a faster route to the Gulf of Mexico. As the river changed course, sediment in the old deltaic lobe compacted and subsided, allowing Gulf waters to encroach and form estuaries. The River then changed course again, abandoning the newly formed delta, and found a shorter path to the sea. Sediment layers from the past 5,000 years provide evidence of the Mississippi River changing course across the south Louisiana coastal plain, resulting in seven different delta systems (Roberts 1997; Reed 2002).

Over the last 1200 years, the River has delivered sediment to the Plaquemines-Balize delta plain, otherwise known as the Bird's Foot delta (Torqvist et al. 1996). This area of land stretches beyond Venice, Louisiana and extends into the Gulf. The Mississippi River Delta Basin covers 210,841 hectares, of which 83% is open water. The remaining area is freshwater and brackish marshes. The average water discharge rate of the River is 13,309 cubic meters per second, and the average suspended sediment load is 395,533 metric tons per day ([www.lacoast.gov](http://www.lacoast.gov)). Unfortunately for wetlands building and maintenance, levee construction of the lower River and much of the Bird's Foot Delta extends the River's discharge to the edge of the continental shelf into the Gulf where the depth abruptly increases to 305 meters. Consequently, most of the sediment currently being carried by the River is deposited into the Gulf and is not available to build land.

Scientists estimate that 70% of total land area in the Mississippi River Delta Basin has been lost since 1932 ([www.lacoast.gov](http://www.lacoast.gov)). Reasons for land loss are both natural and anthropogenic. Compaction of unconsolidated sediments from the River results in a subsidence rate of 1.5 meters per century



([www.lacoast.gov](http://www.lacoast.gov)). Sea level rise, hurricanes, and tidal erosion have also increased land loss. Human activities play a very large role in land loss in coastal Louisiana. Before 1950, the River brought 440 million metric tons of sediment to Louisiana each year; after 1950, the construction of dams along the entire course of the Mississippi reduced sediment load by 50% (Davis 2010). Navigation channel dredging and canal construction for oil and gas exploration have also contributed to land loss.

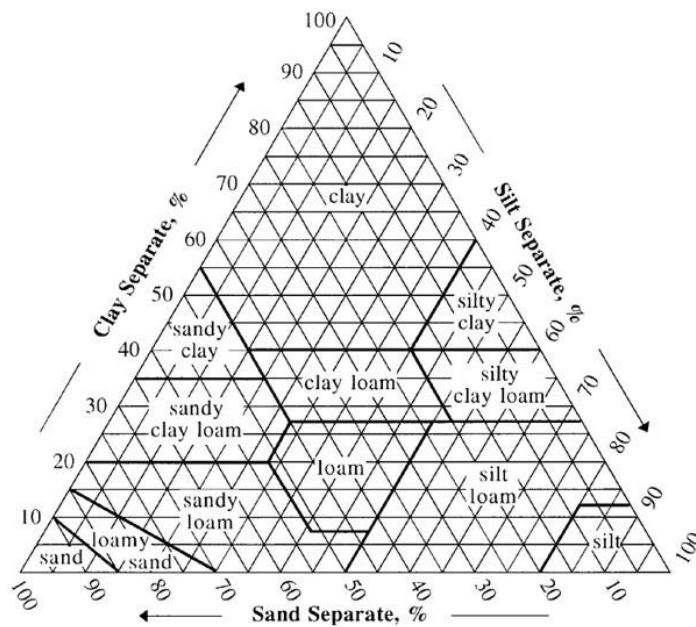
Stabilization of the River through stream channelization and levee construction causes several problems for coastal Louisiana. These activities prevent natural flooding events every spring, which eliminates sediment replenishment in the adjacent marshes and prevents the River from changing course. During hurricanes, forces scour sediment because of failing levees and unstable wetlands. In 2005, Hurricanes Katrina and Rita led to 56,203 hectares of wetland loss (Barras 2006). At a rate loss of 6475 to 7770 hectares of wetland per year, the state of Louisiana needs a restoration solution that will protect coastal communities from increasing encroachment by the waters of the Gulf of Mexico.

### **1.3 Physical Properties of Soils and Sediments**

#### **1.3.1 Grain Size Classes**

In soils and sediments, structure and texture determine the separation of aggregates and pore spaces, which influence the chemical properties that drive biological interactions. In 1940, soil physicist Leonard David Baver defined soil structure as the arrangement of soil particles, including both grains and aggregates (Warkentin 2008).

Soil particles comprise different size classes determined by particle diameter. Clay particles range from 0 to 2  $\mu\text{m}$  in diameter, silt particles range from 2-50  $\mu\text{m}$  in diameter, and sand particles range from 50  $\mu\text{m}$  to 1 mm in diameter (USDA). The silt and sand categories can be further divided into fine and coarse fractions. The USDA uses a texture triangle (Figure 1.1) to categorize soils depending on the relative proportion of clay, silt, and sand in a given sample.



**Figure 1.1 Texture Triangle used by the U.S. Department of Agriculture (USDA NRCS)**

Generally, smaller soil particles have higher surface area available for chemical and biological activity; however, a single soil particle does not function alone. Soil particle associations are called aggregates. By 1955, soil scientist J.P. Martin defined a soil aggregate as “a naturally occurring cluster or group of soil particles in which the forces holding the particles together are much stronger than the forces between adjacent aggregates” (Martin 1955). The forces that cement soil particles are both physical and biological in nature. Aggregation affects physical properties of soil including effective particle size (i.e. size of aggregates), soil density, soil stability, and soil structure, which affect the movement of fluids, solutes, and heat through soil (Warkentin 2008).

### **1.3.2 Physical Mechanisms of Aggregation**

Aggregate formation results from three physical forces: compression, tension, and shear forces. Compression pushes soil particles together and creates aggregates whereas tension pulls soil particles apart and breaks aggregates. Shear forces act in a direction parallel to the plane of the soil particles, which moves aggregates. Surface tension, microbial by-products, and electrostatic interactions hold soil particles together within aggregates (Frey 2005). As aggregates get larger, porosity increases, leading to destabilization as more surface is available for forces to break aggregates apart (Frey 2005).

In saturated sediments, water in the pore spaces weakens the effect of the electrical double layer on clay particles, which decreases flocculation and results in aggregate destabilization (Frey 2005).

#### **1.4 Chemical Properties of Anaerobic Sediments**

In fully saturated organic wetland sediments, as much as 90% of the soil volume can consist of water (Reddy and DeLaune 2008). The value decreases to 50% of soil volume in fully saturated mineral wetland sediments (Reddy and DeLaune 2008). With increasing saturation, redox potential becomes increasingly more reduced. Redox potential ranges from +700 to -300 millivolts but tends to be more negative in wetland sediments, indicating low oxygen status and greater electron availability (Reddy and DeLaune 2008). Without oxygen, alternate compounds serve as electron acceptors during microbial decomposition of organic matter. The reduction of electron acceptors such as nitrate ( $\text{NO}_3^-$ ), ferric iron ( $\text{Fe}^{3+}$ ), manganic manganese ( $\text{Mn}^{4+}$ ), and sulfate ( $\text{SO}_4^{2-}$ ) yields much less energy than the reduction of oxygen. Therefore, anaerobic respiration is much slower than aerobic respiration, resulting in slow organic matter decomposition and eventual organic matter accumulation (DeBusk and Reddy 1998; Reddy and DeLaune 2008). The pH of wetland soils generally approaches neutrality, with typical values from 6.0 – 8.0 (Reddy and DeLaune 2008). In most cases, sediment saturation slightly lowers pH. As organic matter accumulates, more electron donors are present and pH increases. (Reddy and DeLaune 2008).

#### **1.5 Physical and Chemical Factors Influencing Microbial Communities in Soils**

Particle size, hydrological regime, organic matter quality, and nutrient availability influence microbial communities in soils. As particle size changes, changing redox conditions and availability of organic carbon substrates determine microbial activity.

Particle size is one of the most important factors influencing size of the microbial community. Clay soils, compared to sandy soils, have a greater capacity for retaining carbon in the soil organic matter component because the carbon is protected in smaller pore spaces (Van Veen and Kuikman

1990). Clayey soils also have greater surface area for organic matter to bind to clay particles. In addition, soils with higher clay content have enhanced biomass retention after substrate addition due to the following properties: lower turnover rate of microbial products, increased retention of microbial biomass and organic matter, increased nutrient adsorption, and greater protection from microbial predators (Wardle 1992). Microbes are physically protected in clay soil aggregates, which increase efficiency of microbial utilization of substrates.

Pore space is another physical property that influences the prevalence of microbes in soil. Generally, bacteria are found in pore spaces larger than 0.8  $\mu\text{m}$  (Frey 2005). In sandy soils, microbes live on both the exterior and interior of aggregates whereas in clay soils, microbes live mostly on the surface of aggregates because the pores are too small for further penetration (Frey 2005). Saturation of 60% or more of the pore space with water maximizes microbial activity because moisture provides bacteria with easier access to organic substrates (Frey 2005). In the clay fraction of soils, species richness and diversity are high but microbial activity is lower, possibly due to the presence of refractory carbon in smaller pore spaces (Zhang et al. 2007). In silty and sandy soils, microbial diversity is lower but microbial activity is higher because particulate organic matter is more available (Zhang et al. 2007).

Changes in pore size determine soil structure and influence microbial activity. Altering pore geometry affects the exposure of organic substrates and water to soil microbes. As time progresses, biotic responses to habitat and particle movement change pore geometry and stability. As soil structure changes, microbes respond to the changing availability of organic material. Eventually, altered pore structure determines future microbial events (Feeney et al. 2006).

Soil moisture is another physical factor affecting microbial community composition. In soils with higher water content, increased connectivity among pathways results in a balance of microbial activity. Higher moisture content enhances nutrient availability, which leads to faster microbial

growth; however, higher water content limits gaseous diffusion, which decreases microbial activity (Or et al. 2007). Therefore, wetland soils and sediments may experience a somewhat constant level of microbial activity.

Hydrological movement in soils and sediments is another important factor in determining microbial community composition. Plant life cycle, organic matter accumulation, nutrient transformation, and microbial activity are all influenced by the extent of flooding in a soil (Boon et al. 1996; Bossio et al. 2006; and Groffman et al. 1996). Wet environments create opportunities for several methods of bacterial dispersion including active movement using flagella, random movement of particles suspended in a liquid (i.e. Brownian motion), and convective transport by water flow (Or et al. 2007). In addition, bacteria that attach to solid substrates grow more rapidly than drifting planktonic species (Or et al. 2007). Bacterial movement through the liquid environment increases access to organic matter substrates and promotes interspecies competition for materials.

The quality of available organic carbon substrates is a third important factor in determining microbial community composition. For example, microbial growth and activity increase when soils receive readily hydrolysable carbon. When a nutrient amendment is added to soil, microbial activity increases for some amount of time (Wardle 1992). In addition, the slow release of nutrients sustains microbial activity over long periods of time. The active microbial biomass is responsible for litter decomposition, nutrient cycling, and energy flow (Wardle 1992). By acting as a “transformation station” the microbial biomass converts organic materials into bioavailable nutrients that can be utilized by plants and other soil organisms (Van Veen and Kuikman 1990).

Unlike a nutrient amendment, soils with plant cover receive continuous inputs of carbon and nitrogen, which consistently stimulate microbial activity (Tiessen 1988; Nguyen 2000). As roots age or as distance from the roots increases, microbial population size decreases because less carbon substrates

from plant roots are available. More mature marshes with higher organic carbon content exhibit higher microbial activity and greater decomposition than young marshes (Costa et al. 2007).

Within soil aggregates, pore spaces protect organic matter. When forces disrupt soil structure, organic matter becomes more available, which incites a burst of microbial activity. The spaces in between layers of expandable clays act as a sink for microbial waste products. When these soils are physically disrupted and waste products are released, microbial activity increases from the additional availability of substrate (Ransom et al. 1999).

On the microbial level, soil aggregates influence community composition by presenting nutrient gradients from the surface to the interior of aggregates. Soil complexes adsorb more carbon, nutrients and water, which increase microbial biomass and activity. As the process continues, aggregate formation creates larger storage capacities for more carbon, nutrients, and water (Smucker and Hopmans 2007). Between the outside and inside of these micro-ecosystems, microbial communities exponentially increase in size.

In general, soil structure affects the size and connectivity of pores, which affects the movement and growth of soil microbes. Increased soil moisture increases nutrient availability to microbes. As microbes move around and interact with organic carbon substrates, microbial activity increases and promotes soil aggregation.

## **1.6 The Relationship between Microbes, Organic Matter, and Aggregation**

### **1.6.1 Microbial Mechanisms of Aggregation**

On the microbial level, three mechanisms lead to aggregate formation: adhesion, immobilization, and retention.

Microbes and soil particles are drawn together by Van der Waals interactions. The distance between the two constituents remains constant over time due to the balance between attractive and repulsive electrostatic interactions. For particles smaller than 2  $\mu\text{m}$  in diameter, electrostatic bonding

between oxides, polymers, and microbes leads to adhesion and clay flocculation (Gomez-Suarez et al. 2002).

Electrostatic interactions are present due to charges on the microbes, soil particles, and electrolytes in soil solution. When charges on microbes and soil particles are similar, increasing ionic strength of the soil solution decreases the physical distance between microbes and particles because less energy is required for microbial adhesion (Gomez-Suarez et al. 2002). For example, in solutions of high ionic strength, microbes readily bind to soil particles because the cations in solution attract the two negatively charged constituents to one another (Gomez-Suarez et al. 2002).

Through the immobilization mechanism, perpendicular and lateral forces bind microbes to the solid surface. On an ideally homogeneous surface, microbes move about freely on the solid surface. In reality, however, heterogeneous soil structure causes colloidal particles to deposit on the soil surface and extend vertically from the substratum. Surface structure of microbes is also heterogeneous due to biological activity (Geoghegan et al. 2008). As microbial cells grow and respond to the environment, their surface structure changes. Cell surface macromolecules and surface functional groups create bonds with each other that influence microbial adhesion to soil particles. Lateral interactions between attached microbes and rough spots on the substratum result in immobilization. The combination of microbially produced substances and clay particles enhances these interactions on the soil surface, leading to aggregation of silt-sized particles ranging from 2-50  $\mu\text{m}$  (Gomez-Suarez et al. 2002).

Through the retention mechanism, microbial cells and subsequently produced extracellular polymeric substances [EPS] bind clay, silt, and sand particles together into aggregates larger than 50  $\mu\text{m}$ . Retention refers to the capacity of microbes to remain adhered to the surface after being subjected to an external force. Larger aggregates with more pore space will have greater microbial activity to increase retention through formation of a biofilm (Gomez-Suarez et al. 2002).

### **1.6.2 The Role of Organic Matter in Biofilm Formation**

In addition to the production of EPS, microbial activity leads to the production of a biofilm. A biofilm is the accumulation of microbes, EPS, multivalent cations, biogenic particles, and dissolved compounds (Rillig 2005). After organic matter particles initially adhere to soil particles, microbes coaggregate and transport themselves to the soil-organic-matter complexes by diffusion, convection, sedimentation or self motility. Single organisms and microbial aggregates adhere to particles and anchor themselves by producing EPS. As aggregation continues, microbial growth occurs inside and outside pore spaces (Gomez-Suarez et al. 2002).

Water-stability of soil aggregates depends on available organic materials (Tisdall and Oades 1982). Free organic matter provides a substrate for microbial production of EPS, which stabilize clay floccules and aggregates smaller than 50  $\mu\text{m}$  in diameter. The short chain length of polysaccharides inhibits development of any larger aggregates. As organic matter declines, polysaccharide production decreases along with the number of stable aggregates (Tisdall and Oades 1982).

Organic matter provides a “nucleus” for soil aggregation. Organic compounds from plants pass through microbial biomass and enter the soil organic matter pool as small particles. Organic particles adsorb to soil particles, which initiates a tiny aggregate that grows larger as microbial activity continues (Tiessen and Stewart 1988). With a greater amount of fine pores associated with organic matter, increased water retention makes conditions more suitable for microbial activity (Zhang et al. 2005). Higher microbial activity leads to more EPS production, which leads to greater aggregation as polymers trap soil particles.

### **1.6.3 Natural Aggregation in Soils**

Cycles of drying and wetting increase aggregation in soils with organic matter. Rewetting a dry soil increases the release and decomposition of soluble organic matter within aggregates. Since different microbial communities exist in the outside and inside layers of aggregates, the continuous



release of dissolved organic matter leads to microbial activity throughout the soil, increasing soil stability and strength (Park et al. 2007). Sediments with EPS have higher erosion thresholds throughout periods of desiccation (Perkins et al. 2004).

Besides organic matter, other environmental conditions influence microbial adhesion to the soil surface. Changes in solution ionic strength affect clay colloid transport and biofilm stability in the soil. In solutions of high ionic strength, adding a clay colloid and a bacterial biofilm to a sandy soil column results in bacterial mobility because the biofilm binds to the clay colloids and moves through the sand. In solutions of low ionic strength, bacteria do not attach to clay particles; thus, detachment of biofilm cells occurs in the soil (Leon-Morales et al. 2004).

When physical forces bring particles and aggregates together, microbial activity causes particle rearrangement. As bacterial activity increases, organic matter decomposition weakens bonds between soil particles. Simultaneously, as bacteria metabolized organic substrates, they released EPS, which bind soil particles together. Both actions of stabilization and destabilization lead to particle rearrangement (Feeney et al. 2006).

## **1.7 Polymers**

### **1.7.1 Properties of Polymers**

Polymers are hydrocarbon chains with attached organic and inorganic functional groups; they are classified as non-ionic, cationic, or anionic. Each type interacts with soil in a different way. Non-ionic polymers bind to soil particles through hydrogen bonding. Polymers spread throughout the soil and replace adsorbed water molecules around the clays. For cationic polymers, negatively charged soil particles attract positively charged macromolecules and cause adsorption. For anionic polymers, cation bridges form between the polymer and anionic soil constituents. The cation bridges result from dissolved ions in the soil solution; between two negatively charged groups, these bridges cause aggregation (Seybold 1994).

The location of charges on the polymer chain influences the degree of flocculation. The addition of a coagulating salt enhances adsorption of polysaccharides onto clay particles, which results in colloidal flocculation (Labille et al. 2005). Macromolecules with acidic groups on the side chains have greater capacity for attachment to clay particles. Consequently, ionic interactions between side groups and cations in solution increase flocculation between negatively charged clay particles.

Properties of polymers, soil, and soil solution influence adsorption and aggregation. Molecular weight, size, conformation, and type of surface charge are all aspects of the polymer that affect adsorption onto soils (Letey 1994; Seybold 1994). Since polymers interact with the clay fraction of soils, soil characteristics that influence adsorption include particle surface area, type and amount of clay, soil structure, and pore size distribution (Letey 1994; Seybold 1994). Characteristics of the soil solution that affect adsorption of polymers to clays include pH, ionic strength, and electrolyte concentration (Letey 1994; Seybold 1994).

Polymer-induced aggregation occurs through the formation of bridges between the polymer, the soil surface, and the soil solution. Soil stabilization occurs when reactive surface sites on clay particles are saturated with polymer (Letey 1994). Therefore, the optimum polymer concentration level depends on total available surface area of the colloidal suspension.

Since polymeric substances exist naturally in the soil, degradation of the material will occur to some extent. The biodegradation of natural polymers follows steps of decomposition (Ratajska and Boryniec 1998). First, microbes mineralize the biodegradable fragments extending out from the surface of the polymers. The polymeric material starts to unfold, which increases the particle surface area to volume ratio available for microbial activity. Additionally, the material becomes more susceptible to permeation of water. As deeper layers of the polymer become exposed, water transports microbes to those surfaces. Over time, the polymers undergo molecular and morphological changes, increasing degradation by microbes (Ratajska and Boryniec 1998).

### **1.7.2 Natural Polymers**

Microbial biofilms, EPS, and natural polymers can be used interchangeably when describing their functions in soil. A positive correlation exists between soil concentrations of EPS and aggregate stability (Rillig 2005). As EPS content of intertidal sediments increases, critical erosion velocity also increases, suggesting that microbial biofilms increase sediment cohesiveness and sediment stability (Widdows et al. 2006). Even though bacterial polymers degrade 1-2 weeks after application, the soil is most stable 3-4 weeks after application because microbes metabolize and excrete polymeric substances that increase soil aggregation (Martens et al. 1992).

Extracellular polymeric substances also increase soil resistance to desiccation. Polysaccharide structure involves a main backbone surrounded by attached polar groups resulting in a three-dimensional network with a high capacity to absorb solution (Mikkelsen 1994). Therefore, soils with EPS have higher volumetric water content and water-saturated pores (Rillig 2005).

### **1.7.3 Synthetic Polymers**

Synthetic polymers can be manufactured in two ways: through polymerization of vinyl and acrylic monomers or through modification of macromolecules to achieve a polymer with desired properties. Water-soluble synthetic polymers have polar groups, which allow absorption of water into the polymer chain. Some examples are polyvinyl alcohol [PVA] and polyacrylamide [PAM]. PAM is a high molecular weight synthetic organic polymer that becomes more viscous with increasing weight (Seybold 1994).

Water-insoluble polymers can be hydrophobic or cross-linked. Hydrophobic polymers have no polar groups and have linear structure. Cross-linked polymers have physical and chemical cross-links throughout the macromolecule chain, which allow the polymer to swell but not dissolve. Most synthetic polymers are applied with sodium, which dissociates and exposes polyanions on the polymer

chain. The negatively charged groups attract cationic groups, which leads to formation of soil aggregates (Bouranis et al. 1995).

Over the past 60 years, the use of synthetic polymers on agricultural soils has led to improvements in soil structure, plant growth, water retention, and erosion rates. As early as the 1930s, scientists learned more about particle and pore arrangement in the soil and understood that clay and organic matter contents influence the size and stability of aggregates (Warkentin 2008). Scientists also learned that through organic matter addition and decomposition, biological and chemical processes stabilized soils. In 1951, a synthetic polyacrylic molecule known as Krilium was introduced. Krilium stabilized soil aggregates, but unlike organic matter, it persisted in the soil (Warkentin 2008). From the 1950s to the 1970s, farmers emphasized the importance of soil stability to increase crop production. After the addition of synthetic polymers, soil aggregation improved, erosional soil loss decreased, and biological activity in the soil remained constant (Bouranis et al. 1995; Warkentin 2008).

In the early 1990s, the use of PAM led to decreased irrigation demands and decreased soil erosion. Later in the 1990s, scientists studied anionic PAM and found that the anionic form acted as a bridge between cationic particles, which improved mechanical aspects of the soil and increased stable aggregates (Warkentin 2008).

Extensive research has been completed looking at the impact of synthetic polymers on soil stabilization. The charge on the polymer determines the interaction between polymer and soil solution (Letey 1994). For example, adsorption of anionic polymers onto clays increases with higher electrolyte concentration because cations in solution neutralize the negative charges on the polymer. Polymers form a network around soil particles and do not penetrate into aggregates; therefore, soil stability depends on the uninterrupted network of polymeric molecules around the aggregates (Letey 1994). In addition to increasing flocculation, the combination of PAM and an electrolyte source slows the physical disintegration of soil aggregates and reduces the chemical dispersion of clays (Mamedov et al.

2007). Furthermore, high molecular weight PAM is more efficient than low molecular weight PAM at flocculation. Higher molecular weight polymers have a longer “grappling distance”, which assists with interparticle bridging (Mamedov et al. 2007).

## **1.8 Current Field Applications of Polymers**

### **1.8.1 Agricultural Soils**

The use of polymers to stabilize agricultural soils has already been emphasized. Agricultural soils have poor structure, causing increased erosion and runoff. Anionic, heavy molecular weight PAM is the most common type of commercial polymer used on agricultural soils whereas a cationic, low molecular weight guar derivation is the preferred natural polysaccharide (Graber et al. 2006). The success of the polymers depends on how the materials are applied to soils.

When added directly to the soil surface, molecular weight determines the effectiveness of the polymer. For example, to maintain a high infiltration rate in soil, a small amount of high molecular weight polymer is required instead of a large amount of low molecular weight polymer. Adding a source of electrolytes with polymers further reduces runoff and erosion (Graber et al. 2006). Polymers added directly to the soil surface must be dissolved in water and distributed by spraying. In some cases, this method is inefficient because a large volume of water is required to dissolve the polymer (Graber et al. 2006).

When added to irrigation water, polymer conformation and quality of surrounding water determines the effectiveness of the polymer. PAM is more effective than a natural polysaccharide at cementing aggregates because it is longer and has limited capacity to absorb water (Graber et al. 2006).

When applying polymers through furrow irrigation, medium to finely textured soils exhibit less surface sealing and increased infiltration. PAM applied through furrow irrigation results in less runoff, less soil detachment and reduced sediment transport capacity across agricultural lands (Graber et al. 2006).

In some cases, agricultural soils receive synthetic polymers in combination with natural organic matter amendments. In the presence of excess carbon, microbes that degrade PAM may choose another substrate, which could be organic matter (Peterson et al. 2007). In addition, soluble calcium in the soil increases efficiency of PAM due to cation bridge formation between soil particles and the polymer (Petersen et al. 2007).

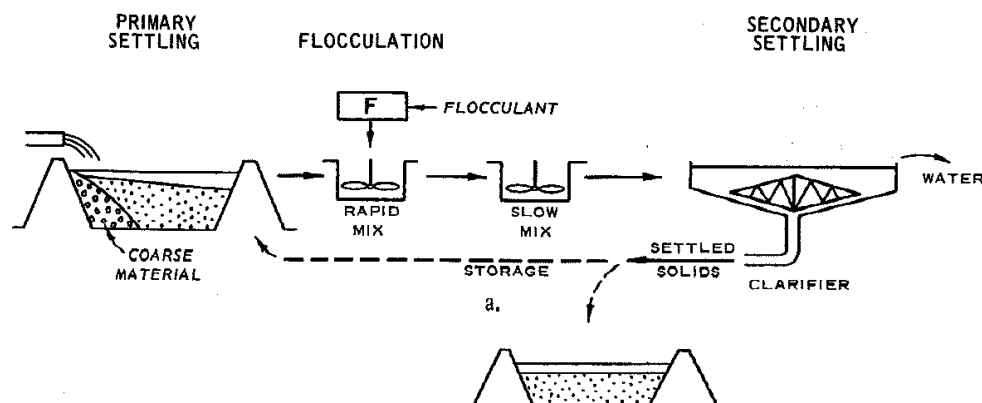
### **1.8.2 Semi-Arid Soils**

Polymers have also been used to stabilize semi-arid soils. In the Biancana Badlands of Tuscany, adding gypsum along with polymer promoted clay flocculation and restored hydraulic conductivity (Agostini et al. 2003). In addition, when soil is dried, polymers become irreversibly bound to the soil (Letey 1994).

In dry soils with low organic matter content, polymers increase stability. In semi-arid soils low in organic matter, high molecular weight anionic polymers most effectively increase infiltration, reduce runoff, and reduce soil loss (Abu-Zreig 2006). As charge density and molecular weight of polymers increase, effectiveness increases.

### **1.8.3 Flocculation of Suspended Clays with Contaminants in Dredged Material**

Polymers have also been used as flocculating agents for suspended solids in dredged slurries. The Army Corps of Engineers has tried various methods to increase sedimentation of fine-grained particles in the water column at a dredged material disposal facility in order to remove chemical toxins associated with clay particles (ACE Technical Report DS-78-14). For one method, polymers are directly injected into a pipeline where the slurry mixes before being discharged. The optimum setup to reduce suspended solids includes a polymer feed system, a rapid-mix facility, a slow-mix facility, and a settling basin with a long enough detention time that newly formed flocs can settle out of the water column (Barnard and Hand 1978, Figure 1.2).



**Figure 1.2 Army Corps of Engineers design for removing contaminants from the water column in a containment area (Barnard and Hand 1978).**

In a treatment situation, electrochemical properties of the dredged material slurry can be altered to enhance particle bridging with long polymer chains. For freshwater dredged material, cationic polymers are most effective. For saltwater dredged material, anionic and nonionic polymers are most effective. To achieve optimum flocculation, a rapid mixing time of 5 to 15 minutes allows full dispersal of clay particles and polymer molecules (Barnard and Hand 1978). Following that with a slow mixing time of 10 to 30 minutes provides time for complete floccule formation before the suspension is allowed to settle. In a pipeline, the injection rate of the polymer depends on the solid concentration and the flow velocity of the slurry (Barnard and Hand 1978).

The intensity and time of agitation for mixing of polymers and soil are factors determining adsorption. Greater agitation intensity decreases the amount of polymer adsorbed, and longer agitation time decreases the degree of aggregation (Harris and Mitchell 1973). One end of a polymer chain attaches to solid surfaces, which leaves the other end suspended in the soil solution to facilitate aggregation (Harris and Mitchell 1973).

#### **1.8.4 Xanthan Gum and Guar Gum**

Xanthan gum and guar gum are two natural polymers that are commercially available and have different molecular properties and charges. Xanthan gum is an extracellular polysaccharide produced

by the bacterium *Xanthomonas campestris* (Kim 2006). The structure of xanthan gum consists of repeating pentasaccharide monomers with varying amounts of acetyl and pyruvate substituents (Jong 2007). The carboxylic acid groups attached to the backbone provide xanthan gum a negative charge.

Xanthan gum increases the viscosity of a polymer solution at relatively low concentrations. Xanthan gum has pseudoplastic properties, which means that the viscosity of the polymer in solution decreases as the applied shear rate increases (Nugent 2009). Materials creating pseudoplastic solutions become oriented in the direction of shear and provide less resistance. When the ionic strength of a solution increases, the macromolecular interactions between salt cations and pyruvate groups increase, inherently increasing viscosity and pseudoplasticity of aqueous solutions of xanthan gum (Smith et al. 1980). At higher gum concentrations in solutions of high ionic strength, interactions in the macromolecular dimension increase viscosity (Smith et al. 1980).

Xanthan gum has previously been applied to soils to improve aggregation. A soil from southeast Scotland experienced increased aggregation and stability due to an increase in tensile strength after addition of xanthan gum (Czarnes et al. 2000). Tensile strength describes the bond energy within a soil aggregate and how quickly energy is released when bonds break during rapid wetting. An increase in tensile strength indicates an increase in the bond energy between particles. In untreated soils, wetting releases energy that disrupts interparticle bonds and destabilizes soil structure. In xanthan-amended soils, the increase in tensile strength enables soil to resist disruption and destabilization when wetted. Additionally, xanthan gum absorbs water; therefore, when an amended soil is wetted, the xanthan gum expands, forming a fibrous network and stabilizing the sediment (Czarnes et al. 2000).

Guar gum, another natural polymer, is extracted from the seed of a leguminous shrub known as *Cyamopsis tetragonoloba* (Kim 2006). Guar gum consists of repeating galactose and mannose units



(Jong 2007). The absence of functional groups on the polymer provides guar gum a neutral charge.

Similar to xanthan gum, guar gum creates viscous and pseudoplastic aqueous solutions (Nugent 2009).

## **1.9 Project Objectives**

In South Louisiana, the Office of Coastal Protection and Restoration has implemented marsh restoration projects using dredged sediment from the Mississippi River. The sediment is hydraulically dredged from the bottom of a river, bayou, or channel and pumped as a slurry to be distributed on the soil surface as a substrate for marsh building. The goal of this research was to investigate a technique to increase sediment stability, thereby improving the efficiency of marsh restoration. By adding a polymer to a pipeline carrying hydraulically dredged material, stability of the deposited sediment might be improved against rainfall and tidal events until plants become established.

The main objectives of this study pose the following questions:

1. Do the polymers increase aggregation of wetland sediments? Is there an optimum concentration level of polymer?
2. Is there a difference in aggregation between xanthan gum (anionic polymer) and guar gum (nonionic polymer)?
3. Does salinity increase the effectiveness of the polymers?
4. Do the polymers negatively affect microbial communities in wetland sediments?
5. Is there a threshold at which microbial activity decreases the aggregative properties of the polymers?
6. Is there a length of time over which the polymers remain effective?

These questions were addressed in a laboratory study.

## **CHAPTER 2: THE ROLE OF SOIL-STABILIZING AMENDMENTS ON AGGREGATION OF DREDGED SEDIMENTS FOR COASTAL WETLAND RESTORATION**

### **2.1 Introduction**

In the soil and sediment environment, soil structure and texture determine the distribution of aggregates and pore spaces, which influence the chemical factors that drive biological interactions. In 1940, soil structure was defined as the arrangement of soil particles, including both grains and aggregates (Warkentin 2008). By 1955, a soil aggregate was defined as “a naturally occurring cluster or group of soil particles in which the forces holding the particles together are much stronger than the forces between adjacent aggregates” (Martin 1955). The forces that cement soil particles together are both physical and microbial in nature. Aggregation affects physical properties of soil including soil density, soil stability, and soil structure, which affect the movement of fluids, solutes, and heat through soil (Warkentin 2008). All of these factors in combination affect microbial activity.

The three physical forces leading to aggregate formation include compressive, tensile, and shear forces. Compressive forces push soil particles together to create aggregates whereas tensile forces pull soil particles apart to break aggregates. Shear forces act in a direction parallel to the plane of soil particles and consequently move aggregates. Within aggregates, surface tension, microbial by-products, and electrostatic interactions hold soil particles together (Frey 2005). Sediment aggregates have an optimum size yielding maximum stability. When aggregates get too large, greater porosity causes destabilization because physical forces acting on the particle surfaces break the aggregates apart (Frey 2005).

When physical forces bring particles and aggregates together, microbial activity causes rearrangement of soil structure. Cycles of particle stabilization take place with bacteria and roots (Feeney et al. 2006). An increase in bacterial activity leads to organic matter decomposition, which weakens bonds between soil particles. Simultaneously, as bacteria metabolize organic substrates, they

release extracellular polymeric substances [EPS], which bind soil particles together. Both actions of stabilization and destabilization lead to particle rearrangement.

On the microbial level, three mechanisms lead to aggregate formation: adhesion, immobilization, and retention. Through the adhesion mechanism, electrostatic bonding flocculates clay particles. For particles smaller than two micrometers in diameter, electrostatic interactions result from charges on microbes, soil particles, and electrolytes in soil solution. As ionic strength of the soil solution increases, physical distance between microbes and particles decreases because less energy is required for adhesion, which leads to clay flocculation (Gomez-Suarez et al. 2002).

Through the immobilization mechanism, perpendicular and lateral forces bind microbes to the solid surface. On an ideally homogeneous surface, microbes move about freely. In reality, however, heterogeneous soil structure causes colloidal particles to deposit on the soil surface and extend vertically from the substratum. Lateral interactions between attached microbes and rough spots on the substratum result in immobilization. The combination of microbially produced polymers and clay particles enhances interactions on the surface, leading to aggregation of silt-sized particles ranging from 2 to 50  $\mu\text{m}$  (Gomez-Suarez et al. 2002).

Through the retention mechanism, microbial cells and microbial by-products bind clay, silt, and sand particles together into aggregates larger than 50  $\mu\text{m}$ . Larger aggregates have higher porosity than small aggregates, leading to less dense and less compact aggregates with larger microbial populations in the pore spaces (Frey 2005). Retention refers to the capacity of microbes to remain adhered to the surface after being subjected to an external force. Larger aggregates with more pore space will have greater microbial activity to increase retention through formation of a biofilm (Gomez-Suarez et al. 2002).

In the 1940s, scientists knew that organic matter and iron oxides were natural agents that bound soil particles. Additionally, scientists knew that organic matter decomposed more rapidly in aerobic

soils, which includes upland agricultural soils. In 1951, a synthetic polyacrylic molecule known as Krilium was introduced for use on agricultural soils. Krilium stabilized soil aggregates, but persisted in the soil, unlike organic matter (Warkentin 2008). Use of synthetic polymers on agricultural soils improved soil structure, plant growth, water retention, and erosion rates (Seybold 1994). In the early 1990s, use of polyacrylamide [PAM] decreased irrigation demands and soil erodability. Later in the 1990s, scientists studied anionic PAM and found that the anionic form acted as a bridge between cationic particles, which improved mechanical aspects of soil and increased stable aggregates (Warkentin 2008).

Properties of polymers, soil, and soil solution influence adsorption and aggregative success of polymers. Molecular weight, size, conformation, and surface charge are all aspects of the polymer that affect adsorption onto soils (Letey 1994; Seybold 1994). Polymers interact more with the clay fraction of soils; therefore, particle surface area, type and amount of clay, soil structure, and pore size distribution are soil characteristics that influence polymer adsorption (Letey 1994; Seybold 1994). The pH, ionic strength, and electrolyte concentration of the soil solution also influence the adsorption of polymers onto clays (Letey 1994; Seybold 1994).

Polymers are classified as non-ionic, cationic, or anionic. Each type interacts with soil in a different way. Non-ionic polymers move toward soil particles by Van der Waals forces and replace adsorbed water molecules around clays. For cationic polymers, positively charged macromolecules adsorb to negatively charged soil particles. For anionic polymers, cation bridges form between the polymer and anionic soil particles. Cation bridges result from dissolved ions, such as  $\text{Ca}^{2+}$  and  $\text{Mg}^{2+}$  in the soil solution; between two negatively charged groups, cation bridges cause aggregation (Seybold 1994).

Extensive research has been completed looking at the impact of synthetic polymers on soil stabilization. Polymer charge determines interactions between polymer and soil solution. For example,

adsorption of anionic polymers onto clays increases with higher electrolyte concentration because cations in solution neutralize negative charges on the polymer (Letey 1994). Polymers form a network around soil particles and do not penetrate into aggregates; therefore, soil stability depends on the uninterrupted network of polymeric molecules around aggregates. In addition to increasing flocculation, application of PAM combined with an electrolyte source slows the physical disintegration of soil aggregates and reduces the chemical dispersion of clays (Mamedov et al. 2007). Furthermore, high molecular weight PAM is more efficient than low molecular weight PAM at flocculation. Higher molecular weight PAM has a longer “grappling distance”, which assists with interparticle bridging (Mamedov et al. 2007).

In reducing sediments and wetland soils, organic matter decomposes slower than in aerobic soils and may increase aggregation. Water-stability of soil aggregates depends on available organic materials (Tisdall and Oades 1982; Martens et al. 1992). Free organic matter provides a substrate for microbial production and decomposition of extracellular polymeric substances (EPS).

As soil organisms decompose organic materials, microbial secretions bind soil particles and small organic particles together (Tiessen & Stewart 1988; Zhang et al. 2005). An increase in the amount of fine pores associated with organic matter improves water retention and makes conditions more suitable for microbial activity. Higher microbial activity leads to more EPS production, which leads to greater aggregation as polymers trap soil particles (Zhang et al. 2005). Even after microbial growth stops, microbial by-products remain in the soil and continue to aggregate soil particles (Frey 2005; Martens et al. 1992). In general, as the amount of organic matter declines, EPS production and the number of stable macroaggregates also declines.

Microbial secretions eventually produce a biofilm. A biofilm is the accumulation of microbes, EPS, multivalent cations, biogenic particles and dissolved compounds (Rillig 2005). Biofilm formation increases soil aggregation. Microbes transport themselves to soil-organic-matter complexes by

diffusion, convection, sedimentation, or self motility. After single organisms adhere to soil and organic matter particles, microbes aggregate together and anchor themselves by producing EPS. Soils exhibit a positive correlation between concentrations of EPS and aggregate stability (Rillig 2005). As a result, changes in biota can have impacts on sediment erodability. As the EPS content of soil increases, critical erosion velocity also increases, suggesting that microbial biofilms increase sediment cohesiveness and sediment stability (Widdows et al. 2006).

Polymeric substances are carbon compounds that exist naturally in soil; therefore, degradation will occur to some extent. Over time, polymers undergo molecular and morphological changes, increasing degradation by microbes (Ratajska and Boryniec 1998). First, microbes metabolize organic functional groups extending out from the surface of the polymers. The polymeric material unfolds, which increases the particle surface area to volume ratio available for microbial activity. Additionally, the material becomes more susceptible to permeation by water. As deeper layers of the polymer become available, water transports microbes to those surfaces.

Extracellular polymeric substances also increase soil resistance to desiccation. Polysaccharide structure consists of a carbon backbone surrounded by attached polar groups resulting in a three-dimensional network with a high capacity to absorb solution (Mikkelsen 1994). Therefore, soils with EPS have higher volumetric water content and water-saturated pores (Rillig 2005).

Previously, polymers have been used as flocculating agents for suspended solids in dredged slurries and other soft sediment applications. Polymers quicken the consolidation of contaminated dredged sediments. Both anionic and nonionic polymers cause significantly greater consolidation than sediments with no polymer treatment (Reddy et al. 2006).

The difference in moisture content over time between sediments with and without a polymer treatment provides information about dewatering and consolidation. Moisture content is inversely related to the amount of dewatering. As moisture content increases, dewatering decreases because

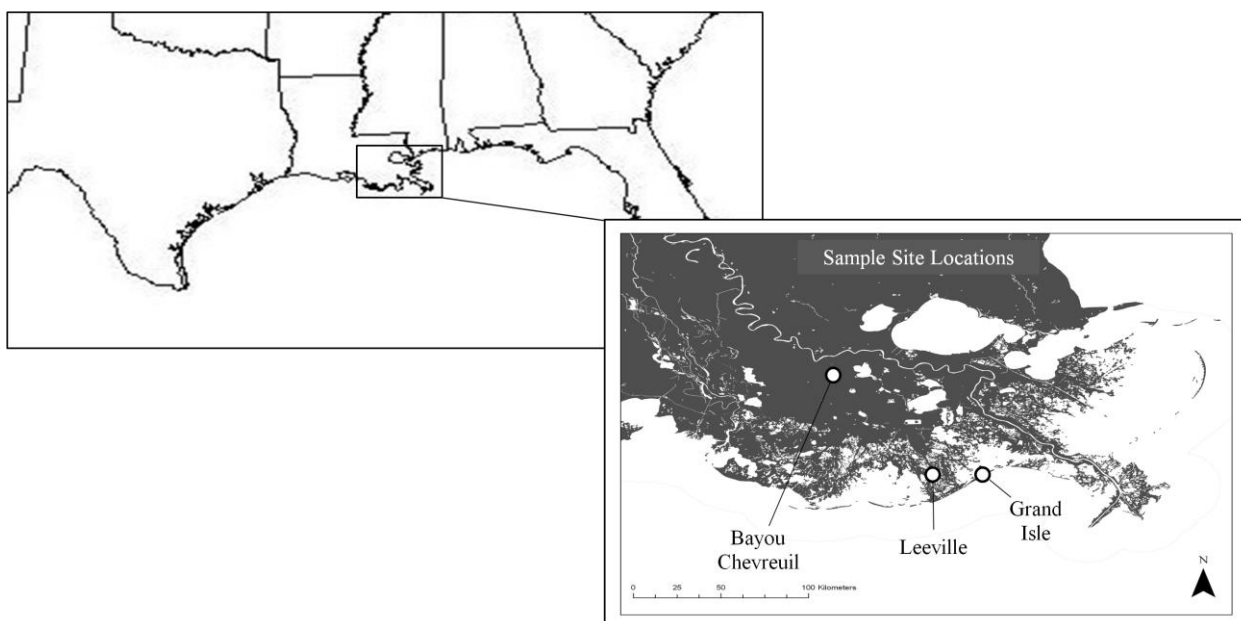
sediment holds more moisture. Similarly, the amount of dewatering is directly related to the amount of sediment consolidation. As dewatering increases, consolidation increases. As water leaves pore spaces, soil particles move closer together and consolidate. Results from moisture content, dewatering, and sediment consolidation should support each other.

This study determined the impact of the addition of two natural polymers on several physical characteristics of lab-simulated, water-saturated hydraulically dredged sediments. Natural polymers are being studied as a soil amendment with the goal of stabilizing hydraulically dredged sediments used for wetland restoration until marsh plants become established. Quantifying the amount of dewatering, sediment consolidation, and aggregation of samples with a polymer treatment will provide information about the potential of natural polymers to increase sediment stability. Therefore, the null hypothesis states that over time, addition of polymer will have no impact on physical characteristics including moisture content, dewatering, sediment consolidation, and aggregation. The alternative hypothesis states that over time, the addition of polymer will decrease moisture content, increase dewatering, increase sediment consolidation, and increase aggregation.

## **2.2 Materials and Methods**

### **2.2.1 Sediment Sample Locations**

Sediment was collected at three sites in southern coastal Louisiana (Figure 2.1). At each site, a Peterson hand-operated dredge was used to collect sediment to be placed in 20-liter plastic buckets. Bayou Chevreuil is located in St. James Parish intersecting LA Route 20 (29.91° N 90.73° W) and represents a freshwater site containing sediment with high clay content. Bayou LaFourche is located in LaFourche Parish alongside LA Route 1 (29.25° N 90.21° W) in Leeville and represents an intermediate salinity site containing sediment with moderate clay content. The third site, Grand Isle, is located in Grand Isle State Park in Jefferson Parish (29.26° N 89.95° W) and represents a marine site containing sediment with low clay content.



Map Courtesy of Andrew Tweel (LSU)

**Figure 2.1 Location of the three sampling sites in coastal Louisiana.**

### **2.2.2 Sediment Characterization**

To ensure sediment homogeneity, the sediment from each 20-liter bucket was pushed through a 0.635 cm sieve to remove large plant debris and sticks. An electric drill attached to a paint mixer was used to homogenize the sediment for ten minutes in each bucket. To ensure that all buckets within each sediment were homogeneous, the following soil properties for each bucket were compared: moisture content, organic matter content, and particle size distribution as determined by the hydrometer method. All three sediments were characterized for the following properties: moisture content, organic matter content, redox potential, pH, soil salinity, cation exchange capacity, exchangeable metals, and particle size distribution.

For moisture content, three sub-samples of each sediment type were dried at 105° C until a constant weight was reached (Gardner 1986). The following formula was used to calculate percent moisture content on a wet weight basis (DeAngelis 2007):

$$[(g \text{ beaker} + g \text{ mud}) - (g \text{ beaker} + g \text{ dry sediment})] / [(g \text{ beaker} + g \text{ mud}) - g \text{ beaker}] * 100$$



To determine organic matter content, the loss-on-ignition method was used (Nelson and Sommers, 1996). In a muffle furnace, dry (105°C) sediment samples were heated to 435°C for 5 hours. The following formula was used to calculate percent organic matter content:

$$[(\text{g beaker} + \text{g sediment})_{105} - (\text{g beaker} + \text{g sediment})_{435}] / [(\text{g beaker} + \text{g sediment})_{105} - \text{g beaker}] * 100$$

For redox potential, platinum-tipped electrodes were cleaned and tested as described by Patrick et al. (1996). Four platinum electrodes were inserted into sediment samples and used in conjunction with a calomel reference electrode to obtain a reading in millivolts ( $E_c$ ); these values were corrected to a standard hydrogen reference electrode for final readings ( $E_h$ ).

For soil pH, a calibrated combination pH electrode with a Ag/AgCl reference was used (Thomas 1996). Soil porewater was collected by centrifuging field moist sediment in a Fisher Scientific accuSpin 3/3R centrifuge at 3500 rpm (3021 g radial centrifugal force) for 15 minutes. The supernatant water was analyzed for salinity with an Accumet AB30 conductivity meter (Rhoades 1996).

Cation Exchange Capacity (CEC) was determined by the Unbuffered Salt Extraction Method according to Sumner and Miller (1996). All three sediments were saturated with 0.2 M  $\text{NH}_4\text{Cl}$ , washed with deionized water, and saturated with 0.2 M  $\text{KNO}_3$  to displace the  $\text{NH}_4^+$ . The extracted supernatant was analyzed for exchangeable  $\text{NH}_4^+$  (US EPA-103-A Rev. 4) using a SEAL AQ2 automated discrete analyzer. The following equation was used to calculate CEC (Sumner and Miller 1996):

$$(\text{mg } \text{NH}_4^+ \text{-N/ L}) * (\text{mL extractant}) * (\text{valence of } \text{NH}_4^+) / (\text{g dry sediment}) * (\text{atomic weight of } \text{NH}_4^+)$$

The results have units of centimoles of cation charge per kilogram of sediment, which is equal to milliequivalents per 100 grams of sediment.

Particle size distribution and textural class were determined by the hydrometer procedure according to Gee and Bauder (1986). Sediments were pre-treated to remove carbonates and soluble salts using sodium acetate, organic matter using hydrogen peroxide, and free iron oxides using citrate-

bicarbonate, sodium dithionite, and sodium chloride. Values from hydrometer readings were used in calculations according to Patrick (1958) to determine the percentages of sand, silt and clay in all three sediments.

### 2.2.3 Experimental Setup

The polymer treatments included two natural polymers (xanthan gum and guar gum) that are commercially available and have different molecular properties and charges (Table 2.1). Xanthan gum is an extracellular polysaccharide produced by the bacterium *Xanthomonas campestris* (Kim 2006). The structure of xanthan gum consists of repeating pentasaccharide monomers with varying amounts of acetyl and pyruvate substituents (Jong 2007). The carboxylic acid groups attached to the backbone provide xanthan gum a net negative charge.

**Table 2.1 Properties of xanthan gum and guar gum.**

Polymer	Source	Molecular Formula	Molecular Weight	Charge	Charge Density	% Total Carbon
Xanthan Gum	microbial extracellular polymer	$(C_{35}H_{49}O_{29})_n$	$0.9 - 1.6 \times 10^6$ Da	anionic	0.25 <sup>a</sup>	40.84
Guar Gum	plant polysaccharide	$(C_{18-20}H_{30}O_{15})_n$	$1.0 - 2.0 \times 10^6$ Da	non-ionic	0	43.25

<sup>a</sup>Charge Density in mol/mol monosaccharide

Guar gum is extracted from the seed of a guar gum plant, a leguminous shrub known as *Cyamopsis tetragonoloba* (Kim 2006). Guar gum consists of repeating galactose and mannose units (Jong 2007). The absence of carboxylic acid groups on the polymer results in an overall neutral charge. Both xanthan gum and guar gum polymer solutions were made to concentration levels of 1% and 2% by weight. To create a 1% polymer solution, 2 g of polymer powder were added to 198 g of water made up to the appropriate salinity. To create a 2% polymer solution, 4 g of polymer powder were added to 196 g of water made up to the appropriate salinity. Experimental units with polymer

treatments received a polymer solution of either 1% or 2% concentration: the 2:1 sediment-to-polymer ratio resulted in final concentrations of polymer at 0.5% and 1%, respectively.

The experimental units were 16-oz. polyethylene cups containing 350 grams of wet sediment mixed with 175 grams of 1% or 2% polymer solution made up with the appropriate salinity solution. Two different salinity solutions were applied to each sediment-polymer combination in order to simulate *in situ* salinity ranges. Salinity treatments were 1 and 5 ppt for Bayou Chevreuil sediments, 5 and 10 ppt for Leeville sediments, and 15 and 25 ppt for Grand Isle sediments. Control experimental units received 175 mL of water of the appropriate salinity.

Due to different moisture contents for the three sediments, different masses of polymer carbon were added to the sediments for each concentration of polymer solution (Table 2.2). All powder-water mixtures were blended in a kitchen blender for 30 seconds to obtain a well-mixed polymer solution.

**Table 2.2 Polymer added carbon to each experimental sample. Units are g C kg<sup>-1</sup> dry sediment.**

Sediment	Concentration	Added C from Xanthan Gum	Added C from Guar Gum
Chevreuil	0.5%	9.22	9.76
	1.0%	18.4	19.6
Leeville	0.5%	7.08	7.48
	1.0%	14.2	15.0
Grand Isle	0.5%	3.65	3.86
	1.0%	7.29	7.73

A randomized block design was implemented to evaluate how several dependent variables over 26 weeks were affected by sediment type (i.e. sampling location), salinity, polymer and polymer concentration. Response variables that were measured included pH, moisture content, sediment consolidation, grain size analysis, and aggregate size analysis. Each treatment was prepared in triplicate in 16-ounce opaque plastic cups.

There were 432 experimental units (3 sediment types x 2 salinities x 2 polymers x 3 concentrations x 4 time periods x 3 replicates). A Barnstead Max-Q 2508 reciprocating shaker was used to mix each sediment and polymer combination. With a fixed circular orbit of 1.2 cm, each

sediment-polymer mixture shook at the maximum setting (400 rpm) for fifteen minutes on the dual action setting (circular and reciprocating movements). Then, each mixture was poured into the cups and set on the lab bench for the appropriate time period before being analyzed for dependent variables.

Destructive sampling was employed. Consequently, at the end of each designated time period (i.e. weeks 1, 8, 16, and 26), pH and moisture content were measured. Then, the samples were stored at 4°C until analysis for aggregate size.

#### **2.2.4 Decanting and Flooding Events**

To maintain saturation of the sediment surface, each sample was re-flooded every 8 to 10 days for the duration of the time periods for 8 weeks, 16 weeks, and 26 weeks. At each decanting and flooding event, supernatant fluid was removed; the volume of the removed fluid was recorded in mLs. A line was drawn on each cup at the beginning of each time period to represent the initial height of the mixture. To monitor any sediment consolidation at each flooding event, the distance from the sediment surface to the initial height line was recorded in centimeters. After noting observations of the sediment surface, each sample was re-flooded to the initial height line with water of the appropriate salinity. The volume of water added was recorded in mLs.

#### **2.2.5 pH**

At the beginning and end of each time period, pH was measured in the same manner as described earlier for sediment characterization.

#### **2.2.6 Moisture Content**

At the beginning and end of each time period, moisture content was measured in the same manner as described earlier for sediment characterization. Initial moisture content was defined by the moisture content determined 24 hours after sediments and polymers were mixed. Final moisture content was defined by the moisture content determined at the end of each time period. The difference

between initial and final moisture contents provided information about the dewatering efficiency of each sediment type (Appendix A).

### **2.2.7 Sediment Consolidation**

Sediment consolidation from flooding event to flooding event was accounted for using the following formula:

$$\text{height}_{\text{current event}} - \text{height}_{\text{previous event}}$$

By subtracting the consolidated distance at one flooding event from the consolidated distance at the previous flooding event, height change over 10-day periods was calculated. The overall change in height over 8 weeks, 16 weeks, and 26 weeks was found by summing the cumulative height changes of all flooding events from each respective time period.

### **2.2.8 Aggregate Size Analysis**

Sediment samples were analyzed on a Beckman Coulter LS 13 320 Series Multi-Wavelength Laser Diffraction Particle Size Analyzer with an aqueous liquid module and sonicator system made available through Dr. Alex Kolker, LUMCON. With 116 size channels and 132 detectors, the LS 13 320 can measure particles from 0.017  $\mu\text{m}$  – 2000  $\mu\text{m}$ . The LS 13 320 uses Mie scattering, Fraunhofer diffraction, and PIDS (Polarization Intensity Differential Scattering) technology.

To find differences in aggregation from the polymer, aggregate size analysis was completed on samples of wet sediment from each experimental unit. Samples for aggregate size analysis did not receive any pre-treatment in an effort to maintain natural aggregates (Matthews 1991). These samples did not receive sonication and were analyzed by the sands standard operating protocol on the LS 13 320 (developed in the Kolker lab at LUMCON).

For all analyses, the LS 13 320 reported the percent volume of the sample falling into several size fractions: less than 2  $\mu\text{m}$ , greater than 2  $\mu\text{m}$ , less than 63  $\mu\text{m}$ , greater than 63  $\mu\text{m}$ , and greater than 1000  $\mu\text{m}$ . The LS 13 320 also reported mean aggregate size diameter for each sample. Size classes

were assigned according to the following parameters: 0-2  $\mu\text{m}$  for the clay fraction, 2-63  $\mu\text{m}$  for the silt fraction, and greater than 63  $\mu\text{m}$  for the sand fraction.

### **2.2.9 Statistical Analysis**

SAS 9.1 software (2009) was used to analyze the data. SigmaPlot 11.0 software (2008) was used to graph the data. For pH, moisture content, and aggregate analysis, an ANOVA along with stepwise variable selection reduced each model to the most significant effects. To enter the model, factors had to be significant at an alpha value of 0.10; to stay in the model, factors had to be significant at an alpha value of 0.05.

For total sediment consolidation, an ANOVA with a Tukey adjustment and least squares means analysis identified significant effects. To look for patterns in consolidation over time, an ANOVA with repeated measures was run on the consolidation data from each flooding event during the 26-week time period. Stepwise variable selection, as previously described, reduced the model to the most significant effects. After the test of Type III fixed effects with a Tukey adjustment, least squares means analysis identified differences between significant effects for all dependent variables. An alpha value of 0.05 was used for all analyses.

## **2.3 Results**

### **2.3.1 Sediment Characteristics**

General sediment characteristics differed from one another (Table 2.3). The Bayou Chevreuil sediment was classified as clay with particle size distribution values of approximately 70% clay, 21% silt, and 9% sand. Cation exchange capacity for Bayou Chevreuil was the highest of all sediments at 125 centimoles of charge per kilogram of soil. Moisture content (wet weight basis) was the highest for the Bayou Chevreuil sediment at 75%; organic matter was also the highest at 14%. The Bayou Chevreuil sediment represented a freshwater site with a porewater salinity of 0.5 parts per thousand (ppt) and hereby will be referred to as the freshwater site.

The Leeville sediment was classified as silty clay with particle size distribution values of approximately 43% clay, 38% silt and 19% sand. Cation exchange capacity for Leeville was lower than that of the freshwater sediment at 85 centimoles of charge per kilogram of soil. Moisture content (wet weight basis) was also slightly lower than that of the freshwater sediment at 67%; organic matter follows the same pattern at 8%. The Leeville sediment represented an intermediate salinity site with a porewater salinity of 4.6 ppt and hereby will be referred to as the intermediate site.

The Grand Isle sediment was classified as a sandy loam with particle size distribution values of approximately 16% clay, 13% silt and 71% sand. Cation exchange capacity for Grand Isle was the lowest of all three sediments at 28 centimoles of charge per kilogram of soil. Moisture content (wet weight basis) was also the lowest of all three sediments at 36%; organic matter follows the same pattern at 1.5%. The Grand Isle sediment represented a marine site with a porewater salinity of 15.5 ppt and hereby will be referred to as the marine site.

**Table 2.3 Characteristic properties of the Freshwater, Intermediate, and Marine sediments.**

Characterization of Soil			
Soil Properties	Freshwater	Intermediate	Marine
Moisture Content % (wet weight)	74.8	67.1	36.3
OM Content %	13.8%	8.34%	1.56%
Redox Potential (mV)	-26	-18	-207
Soil pH	6.50	6.90	6.90
CEC (cmol charge kg <sup>-1</sup> dry sediment)	125.0	84.6	27.7
Porewater Salinity (ppt)	0.50	4.60	15.5
% Sand	9.17	19.2	70.8
% Silt	20.8	38.3	13.3
% Clay	70.0	42.5	15.8
Textural Class	Clay	Silty Clay	Sandy Loam

The redox potential of the marine sediment represented strongly reducing conditions compared to the moderately reducing conditions shown by the redox potential of the freshwater and intermediate sediments. Lower redox potential of the marine sediment suggests the presence of organic matter that was more microbially available. Due to the low organic matter content of this sediment, a more

strongly reducing redox potential may result from recently deposited organic material from natural events. The sediment was collected three months after Hurricanes Gustav and Ike.

The proximity of the marine sediment to the coastline increases the possibility that fresh organic matter was recently deposited, perhaps lowering redox potential. In addition, the presence of seawater at the marine sediment suggests the presence of sulfate reducing bacteria, which are prevalent in strongly reducing sediments containing sediment.

### 2.3.2 pH

The presence of a polymer generally had no significant effect on pH compared to control samples, with a few exceptions for the freshwater and marine sediments (Table 2.4, Appendix B).

**Table 2.4. Mean pH values for the a) Freshwater, b) Intermediate, and c) Marine sediments at weeks 1, 8, 16, and 26. Letters indicate significant differences across time periods and between polymers for each sediment type. Crosses indicate significant differences between the control and a polymer treatment within a time period.**

a. Freshwater	Week 1	Week 8	Week 16	Week 26
0%	6.75 a	7.07 ab	7.04 ab	7.26 b
Xanthan Gum 0.5%	6.09 a	7.13 b	7.16 bc	7.62 c
Xanthan Gum 1%	5.66 a <sup>+</sup>	7.24 b	7.25 b	7.62 b
Guar Gum 0.5%	6.55 a	6.89 b	7.09 bc	7.30 c
Guar Gum 1%	6.04 a	7.05 b	7.20 b	7.47 b
b. Intermediate	Week 1	Week 8	Week 16	Week 26
0%	7.12 a	7.40 a	7.76 ab	7.92 b
Xanthan Gum 0.5%	7.23 a	8.08 b	7.97 b	7.93 b
Xanthan Gum 1%	7.34 a	7.75 b	7.90 b	7.84 b
Guar Gum 0.5%	6.52 c	7.27 d	7.76 d	7.98 d
Guar Gum 1%	6.07 c	7.35 d	7.64 d	7.72 d
c. Marine	Week 1	Week 8	Week 16	Week 26
0%	7.26 a	7.02 a	7.47 a	7.41 a
Xanthan Gum 0.5%	6.98 a	7.80 b <sup>+</sup>	7.37 b	7.78 b
Xanthan Gum 1%	7.27 a	7.60 b	7.70 b	7.54 b
Guar Gum 0.5%	5.95 c <sup>+</sup>	7.30 d	7.49 d	7.17 d
Guar Gum 1%	5.53 c <sup>+</sup>	7.25 d	7.14 d	7.02 d



Since salinity was significant for only one sediment type, the effect was ignored. Therefore, for all pH results presented below, values for different salinity levels have been averaged for each sediment type.

Upon closer analysis, the pH for the freshwater sediment was significantly lower than the pH for the intermediate sediment at all weeks ( $P<0.0001$ , Table 2.5).

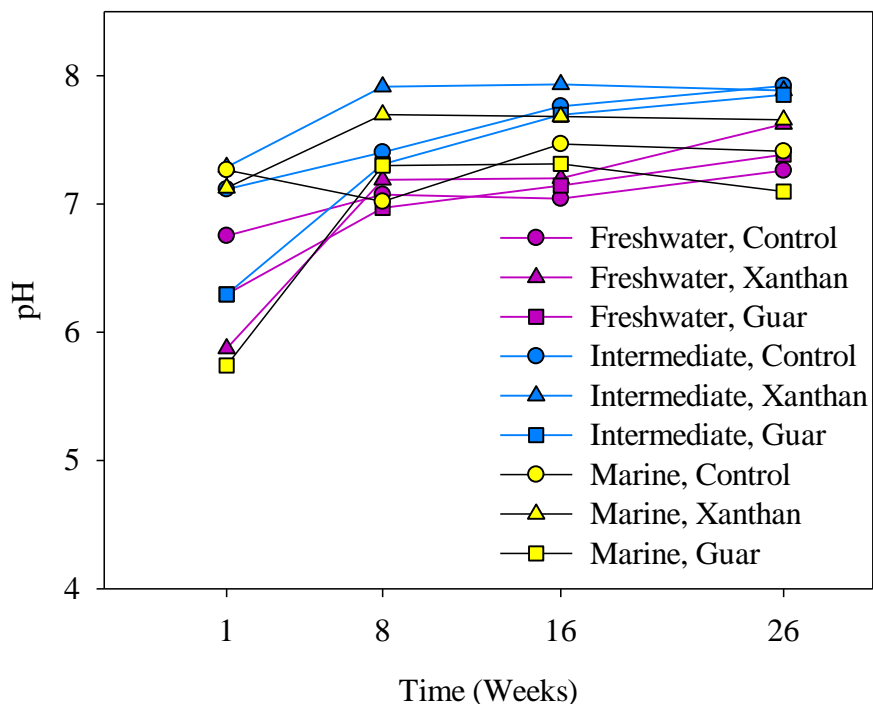
**Table 2.5 Mean pH values for the Freshwater, Intermediate, and Marine sediments at weeks 1, 8, 16, and 26. Values for the polymer types, concentration levels, and salinities have been averaged due to no significant differences. Letters indicate significant differences between sediments at each week.**

Week	Freshwater	Intermediate	Marine
1	6.22 a	6.86 b	6.60 b
8	7.08 a	7.57 b	7.40 b
16	7.15 a	7.80 b	7.49 c
26	7.46 a	7.88 b	7.38 a

The pH for the freshwater sediment was also significantly lower than the pH for the marine sediment at weeks 1, 8, and 16 ( $P<0.0001$ ). By week 26, pH was not significantly different. The pH for the intermediate sediment was not significantly different from that of the marine sediment at week 1 or week 8; however, the pH for the intermediate sediment was significantly higher than the pH for the marine sediment at week 16 ( $P=0.0017$ ) and week 26 ( $P<0.0001$ ).

For the freshwater sediment, only one polymer treatment was significantly different. At week one, the sediment with the 1% xanthan gum treatment had a pH of 5.85, which was significantly lower ( $P<0.0001$ ) than any other sample, possibly due to dissociation of hydrogen ions from the carboxylic acid functional groups of xanthan gum. In addition, the freshwater sediment has less buffering capacity for changes in pH due to the low inorganic carbon content of freshwater aquatic systems. For all other freshwater sediment samples, the control, 0.5%, and 1% concentration levels showed no significant differences for any time period. The pH of the samples with and without polymer at week 26 was significantly higher than at week 1. With all concentration levels combined, the average pH of the

freshwater sediment increased over time (Table 2.5). The pH of the control samples and the samples with polymer increased over time; however, the pHs of the control samples and samples with polymer were not significantly different (Figure 2.2).



**Figure 2.2. Mean final pH values for the Freshwater, Intermediate, and Marine sediments at weeks 1, 8, 16, and 26 for the control, xanthan gum, and guar gum treatments. Values for the 0.5% and 1% concentration levels and salinities have been averaged due to no significant differences.**

For the intermediate sediment, the control, 0.5%, and 1% concentration levels showed no significant differences for any time period. The pH of the samples with and without polymer at week 26 was significantly higher than at week 1. With all concentration levels combined, the average pH of the intermediate sediment increased over time (Table 2.5). The pH of the control samples and the samples with polymer increased over time; however, the pHs of the control samples and samples with polymer were not significantly different.

For the marine sediment, any differences between the control samples and samples with polymer occurred in the first 8 weeks. After week 1, the pH of samples with polymer was significantly lower than the control samples. After week 8, the pH of samples with polymer was significantly higher

than the control samples. Beyond week 8, the pH of the control, 0.5%, and 1% concentration levels showed no significant differences. The pH of the control samples did not change significantly from week to week. With all concentration levels combined, the average pH of the marine sediment increased over time (Table 2.5). In general, for each sediment type, the pH increased from week 1 to week 8 and then stabilized over the rest of the time period (Figure 2.2). The presence of a polymer had no significant effect on the pH compared to the control samples, with a few exceptions for the marine sediment.

### **2.3.3 Moisture Content**

The presence of a polymer did not have a significant effect on final moisture content values, with one exception for the marine sediment (Appendix B). After the sediment-polymer mixtures equilibrated for 24 hours, moisture content for all three sediments was significantly different ( $P < 0.0001$ ). The initial moisture content of the sediments increased from the marine to the intermediate to the freshwater sites with average values of approximately 54%, 73%, and 80%, respectively. With the passage of time, moisture content values for all three sediments were significantly different from each other at weeks 1, 8, 16, and 26 ( $P < 0.0001$ , Table 2.6).

The freshwater sediments with 1% polymer had initial moisture contents lower than any other control or treated sample. The combination of polymer and low ionic strength could have caused the polymer to penetrate the expanding interlayers of clays, which would have pushed water molecules out of interstitial spaces. As time progressed, however, moisture content of samples with and without polymer was not significantly different, suggesting that changes in moisture content were from consolidation and dewatering. Moisture contents for all three sediments were always significantly different from one another (Figure 2.3).

For the freshwater sediment, moisture content values for the 0%, 0.5%, and 1% polymer concentration levels were not significantly different from each other for any time period; however, as

time progressed, moisture content values decreased. Overall, moisture content for the freshwater sediment decreased about 7% over 26 weeks with no significant differences between polymer concentration levels.

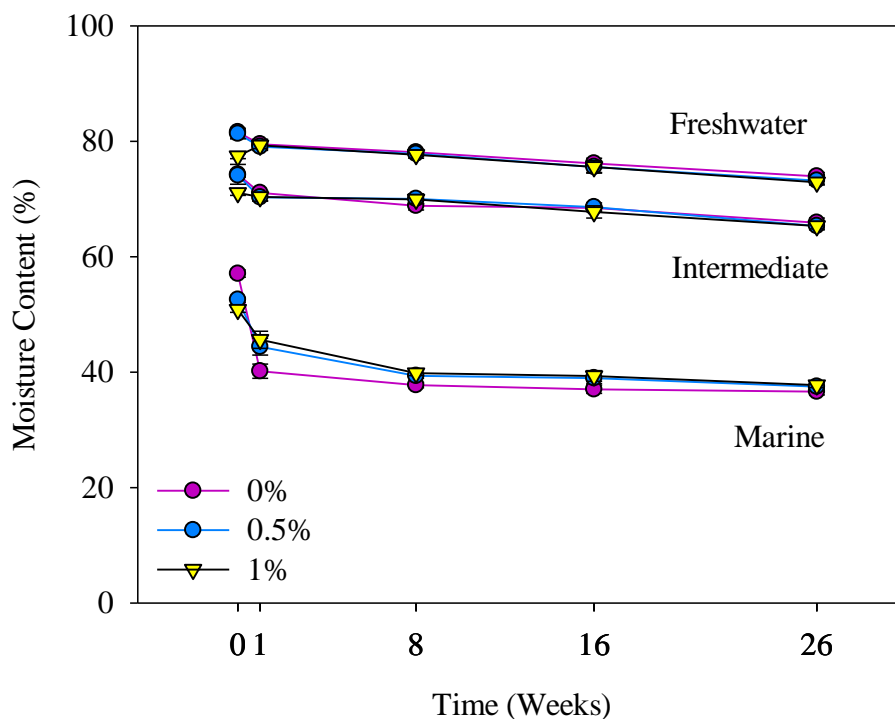
**Table 2.6 Mean and Standard Error for Moisture Content values (%) for the Freshwater, Intermediate, and Marine sediments at weeks 0, 1, 8, 16, and 26 for the 0%, 0.5%, and 1% concentration levels. Values for polymer types and salinities have been averaged due to no significant differences. Letters indicate significant differences between time periods within a concentration level for each sediment. Crosses indicate significant differences between concentration levels within a week for each sediment. (Note: Week 0 N=48, Weeks 1-26 N=12)**

Sediment	Week	0%	0.5%	1%
Freshwater	0	81.6 $\pm$ 0.333 a	81.3 $\pm$ 0.926 a	77.4 $\pm$ 0.428 a <sup>+</sup>
	1	79.5 $\pm$ 0.834 b	79.0 $\pm$ 0.626 b	79.3 $\pm$ 0.579 ab
	8	78.3 $\pm$ 0.593 b	77.7 $\pm$ 0.531 ab	77.6 $\pm$ 0.598 ab
	16	76.2 $\pm$ 0.875 c	75.6 $\pm$ 0.959 c	75.6 $\pm$ 1.07 ac
	26	73.9 $\pm$ 0.670 d	73.2 $\pm$ 0.588 d	72.9 $\pm$ 0.461 d
Intermediate	0	75.9 $\pm$ 0.407 a	74.1 $\pm$ 0.315 a	71.2 $\pm$ 0.428 a
	1	71.1 $\pm$ 0.392 b	70.3 $\pm$ 0.586 b	70.3 $\pm$ 0.725 ab
	8	68.6 $\pm$ 0.776 ab	70.0 $\pm$ 0.849 b	70.3 $\pm$ 0.753 ab
	16	68.5 $\pm$ 0.475 c	68.6 $\pm$ 0.748 c	67.7 $\pm$ 1.07 c
	26	65.9 $\pm$ 0.777 d	65.3 $\pm$ 0.696 d	65.3 $\pm$ 0.504 d
Marine	0	57.1 $\pm$ 0.624 a <sup>+</sup>	52.5 $\pm$ 0.418 a	50.8 $\pm$ 0.458 a
	1	40.2 $\pm$ 1.22 b <sup>+</sup>	44.4 $\pm$ 1.45 b	45.6 $\pm$ 1.48 b
	8	37.7 $\pm$ 0.471 c	39.3 $\pm$ 0.415 c	39.8 $\pm$ 0.435 c
	16	37.0 $\pm$ 0.713 c	38.9 $\pm$ 0.925 c	39.3 $\pm$ 0.919 c
	26	36.6 $\pm$ 0.565 c	37.5 $\pm$ 0.699 c	37.8 $\pm$ 0.438 c

Similarly, for the intermediate sediment, moisture content values for the 0%, 0.5%, and 1% polymer concentration levels were not significantly different from each other for any time period; however, as time progressed, moisture content values decreased. Overall, moisture content for the intermediate sediment decreased about 7% over 26 weeks with no significant differences between concentration levels.

For the marine sediment, moisture content values for the control samples were significantly different from the samples with 0.5% polymer ( $P=0.0037$ ) and 1% polymer for the 1 week and 8 week time periods ( $P<0.0001$ ). For the control samples, moisture content decreased over time. By week one,

moisture content significantly decreased ( $P<0.0001$ ) from 57% to 40%. Moisture content continued to decrease over time with an overall change of 20%. For the marine sediments with polymer treatment, moisture content values also decreased over time. By week one, moisture content significantly decreased ( $P<0.0001$ ) from 52% to 45%. Moisture content continued to decrease over time with an overall change of 14%.



**Figure 2.3 Mean Moisture Content (%) for the Freshwater, Intermediate, and Marine sediments at the 0%, 0.5%, and 1% concentration level at weeks 0, 1, 8, 16, and 26. Values for polymer types and salinities have been averaged due to no significant differences.**

In general, the presence of a polymer only had a significant effect on moisture content values for the marine sediment in the first week (Table 2.6). For those samples with polymer, moisture content was significantly higher than the control samples, possibly due to the amount of pore space in the marine sediment. High sand content led to greater and larger pore space, which increased the polymer's adsorption of water. For the freshwater and intermediate sediments, the presence of a polymer was not significant even though moisture content values significantly decreased over 26 weeks. After the polymer degraded, sediments consolidated, thereby decreasing moisture content.

### 2.3.4 Sediment Consolidation

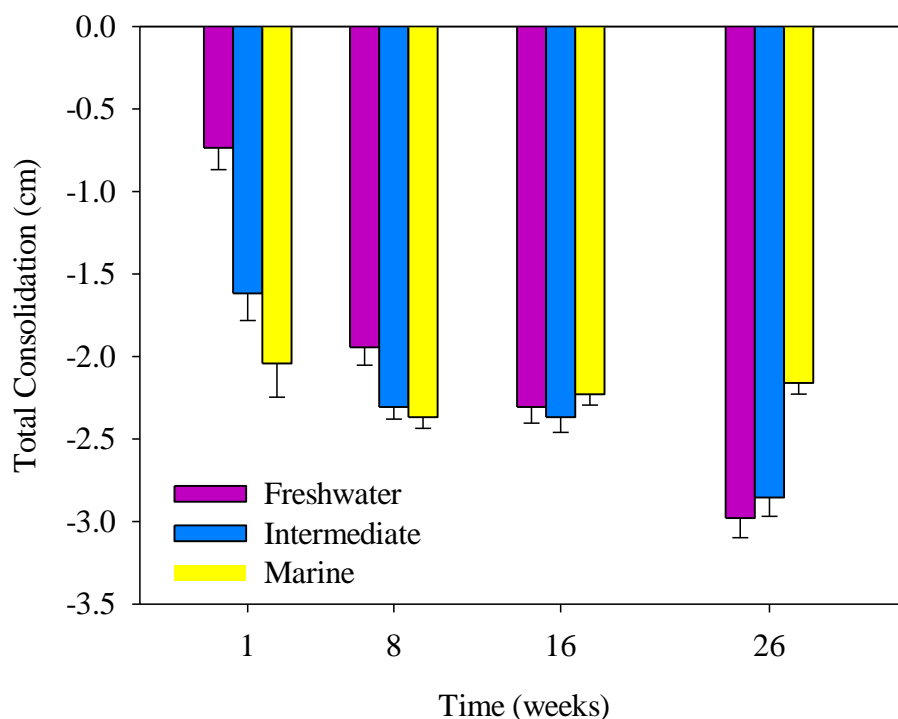
Monitoring the amount of consolidation provides insight into whether or not polymers prevent movement of sediment particles, thereby creating a more stable material. The presence of a polymer did not significantly affect sediment consolidation (Table 2.7, Appendix B). For the freshwater sediment, total consolidation significantly increased over time ( $P<0.0001$ , Table 2.7). With all concentration levels averaged, the freshwater sediment consolidated an average of 0.7 cm, 1.9 cm, 2.3 cm, and 3.0 cm after 1 week, 8 weeks, 16 weeks, and 26 weeks, respectively. The total consolidation of sediment with a polymer treatment was not significantly different from the total consolidation of sediment with no polymer treatment.

**Table 2.7 Total consolidation in centimeters for the Freshwater, Intermediate, and Marine sediments at weeks 1, 8, 16, and 26. Values for polymer types and salinities have been averaged due to no significant differences. Letters indicate significant differences between time periods within a sediment and concentration combination.**

Sediment	Week	0%	0.5%	1%
Freshwater	1	$-1.17 \pm 0.0258$ a	$-0.542 \pm 0.206$ a	$-0.500 \pm 0.174$ a
	8	$-2.06 \pm 0.116$ ab	$-1.88 \pm 0.212$ ab	$-1.90 \pm 0.229$ ab
	16	$-2.13 \pm 0.113$ b	$-2.44 \pm 0.207$ b	$-2.35 \pm 0.178$ b
	26	$-2.65 \pm 0.143$ c	$-3.23 \pm 0.231$ c	$-3.06 \pm 0.211$ c
Intermediate	1	$-1.37 \pm 0.302$ a	$-1.98 \pm 0.299$ a	$-1.50 \pm 0.238$ a
	8	$-2.35 \pm 0.084$ b	$-2.31 \pm 0.178$ b	$-2.25 \pm 0.111$ b
	16	$-2.38 \pm 0.104$ b	$-2.60 \pm 0.161$ b	$-2.13 \pm 0.183$ b
	26	$-2.88 \pm 0.084$ b	$-3.0 \pm 0.204$ b	$-2.69 \pm 0.267$ b
Marine	1	$-2.02 \pm 0.434$ a	$-2.23 \pm 0.347$ a	$-1.88 \pm 0.296$ a
	8	$-2.46 \pm 0.091$ a	$-2.46 \pm 0.130$ a	$-2.19 \pm 0.120$ a
	16	$-2.42 \pm 0.117$ a	$-2.25 \pm 0.102$ a	$-2.02 \pm 0.095$ a
	26	$-2.40 \pm 0.100$ a	$-2.19 \pm 0.098$ a	$-1.90 \pm 0.117$ a

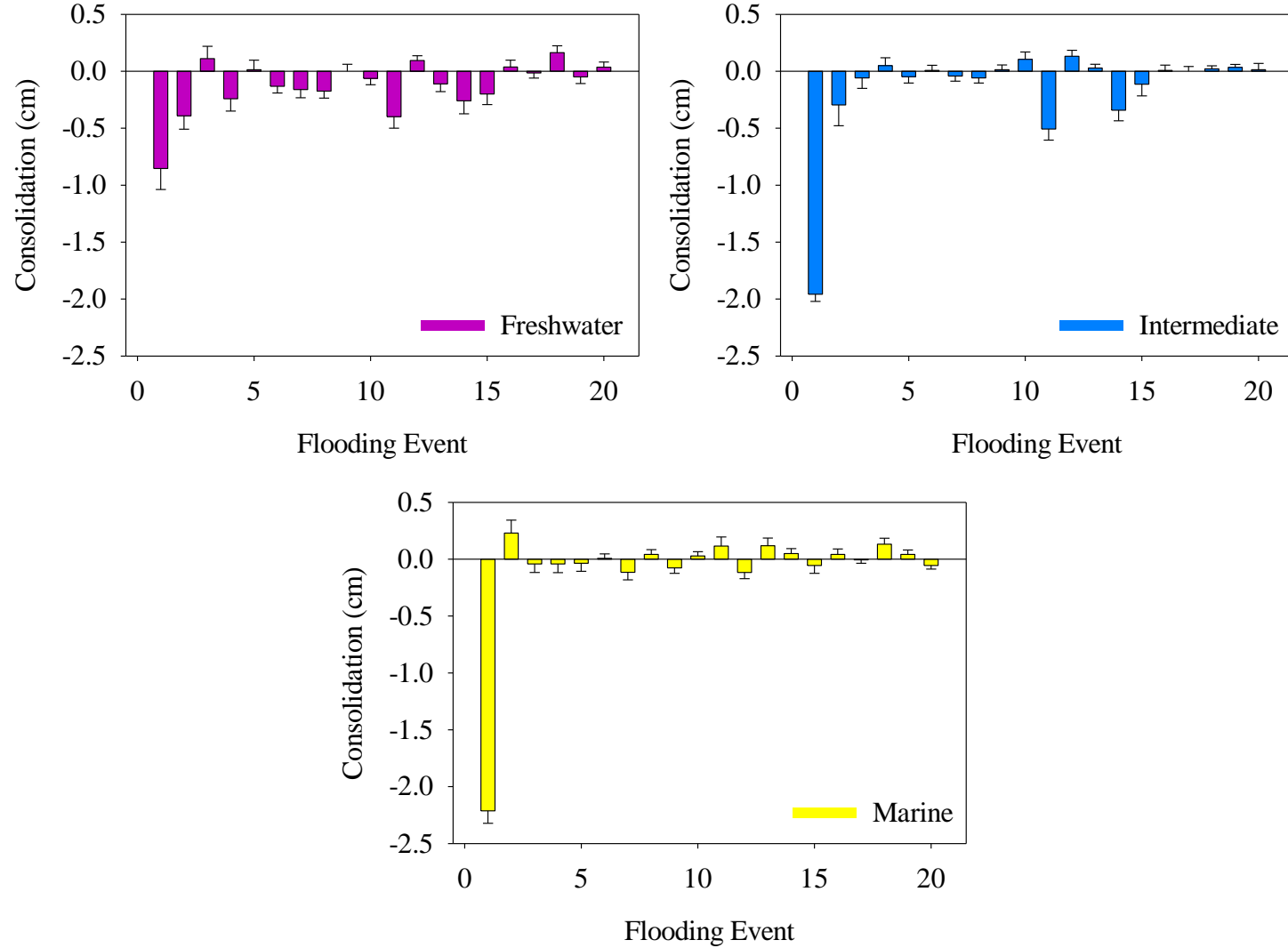
For the intermediate sediment, the total consolidation after 26 weeks was significantly more than the total consolidation after 1 week ( $P=0.0043$ , Table 2.7). With all concentration levels averaged, the intermediate sediment consolidated 1.6 cm, 2.3 cm, 2.4 cm, and 2.9 cm after 1 week, 8 weeks, 16 weeks, and 26 weeks respectively. For the marine sediment, the total consolidation was not significantly different for any time period. With all concentration levels averaged, the marine sediment

consolidated 2.0 cm, 2.4 cm, 2.2 cm, and 2.2 cm after 1 week, 8 weeks, 16 weeks, and 26 weeks, respectively. Overall, the total consolidation over time for all three sediments was not significantly different between samples with a polymer treatment and samples without a polymer treatment (Figure 2.4).



**Figure 2.4 Total consolidation (cm) for the Freshwater, Intermediate, and Marine sediments at weeks 1, 8, 16, and 26. Values for the polymer type, concentration levels, and salinities have been averaged due to no significant differences.**

The presence of a polymer had no significant effect on sediment consolidation at flooding events throughout the 26 week period (Figure 2.5, Appendix B). The greatest amount of consolidation occurred by the first flooding event, which took place 8 to 10 days after the sediments and polymers were mixed. The only other significant amount of consolidation took place at event 11, which happened 22 days after event 10 due to the decanting and flooding schedule. Trends in total consolidation for each time period (Figure 2.4) could be interpreted as consolidation happening in the first week.



**Figure 2.5 Mean consolidation (cm) over 26 weeks for the Freshwater, Intermediate, and Marine sediments at the re-flooding events. Values for the polymer types, concentration levels, and salinities have been averaged due to no significant differences.**



### **2.3.5 Aggregate Size Analysis**

Analysis of wet sediment samples on the LS 13 320 was completed to reveal any differences in aggregation from the polymer treatments. The presence of a polymer had no significant effect on aggregate formation. Clay, silt, and sand size classes were significantly different from each other for the three sediments at all time periods ( $P < 0.0126$ , Table 2.8, Appendix B).

After averaging the percent volume values for all polymer concentrations and time periods within each size class, the typical aggregate distribution of the three sediments is the following: the freshwater sediment had approximately 7% of aggregates in the clay fraction, 48% in the silt fraction, and 45% in the sand fraction. The intermediate sediment had approximately 5% in the clay fraction, 60% in the silt fraction, and 35% in the sand fraction; the marine sediment had approximately 4% in the clay fraction, 32% in the silt fraction, and 64% in the sand fraction.

In general, the marine sediment had the highest percentage of aggregates in the sand fraction. The freshwater sediment and the intermediate sediment had the highest percentage of aggregates in the silt fraction; the intermediate sediment had a higher percentage in the silt fraction than the freshwater sediment. These patterns are similar to initial sediment characterization trends.

In the freshwater sediment, the percentage of aggregates in the sand fraction increased with time, which was reflected in a decrease in the silt fraction over time (Figure 2.6). The percentage of clay did not significantly change over time. For both the intermediate and marine sediments, the percentage of aggregates in the silt fraction increased as time progressed, reflected by a decrease in the sand fraction over time. Additionally, the percentage of aggregates in the clay fraction of the intermediate and marine sediments increased over time.

### **2.3.6 Mean Aggregate Diameter Size**

Over time, the mean aggregate diameter size was significantly different among all three sediments (Table 2.9, Appendix B), but the presence of a polymer had no significant effect on mean

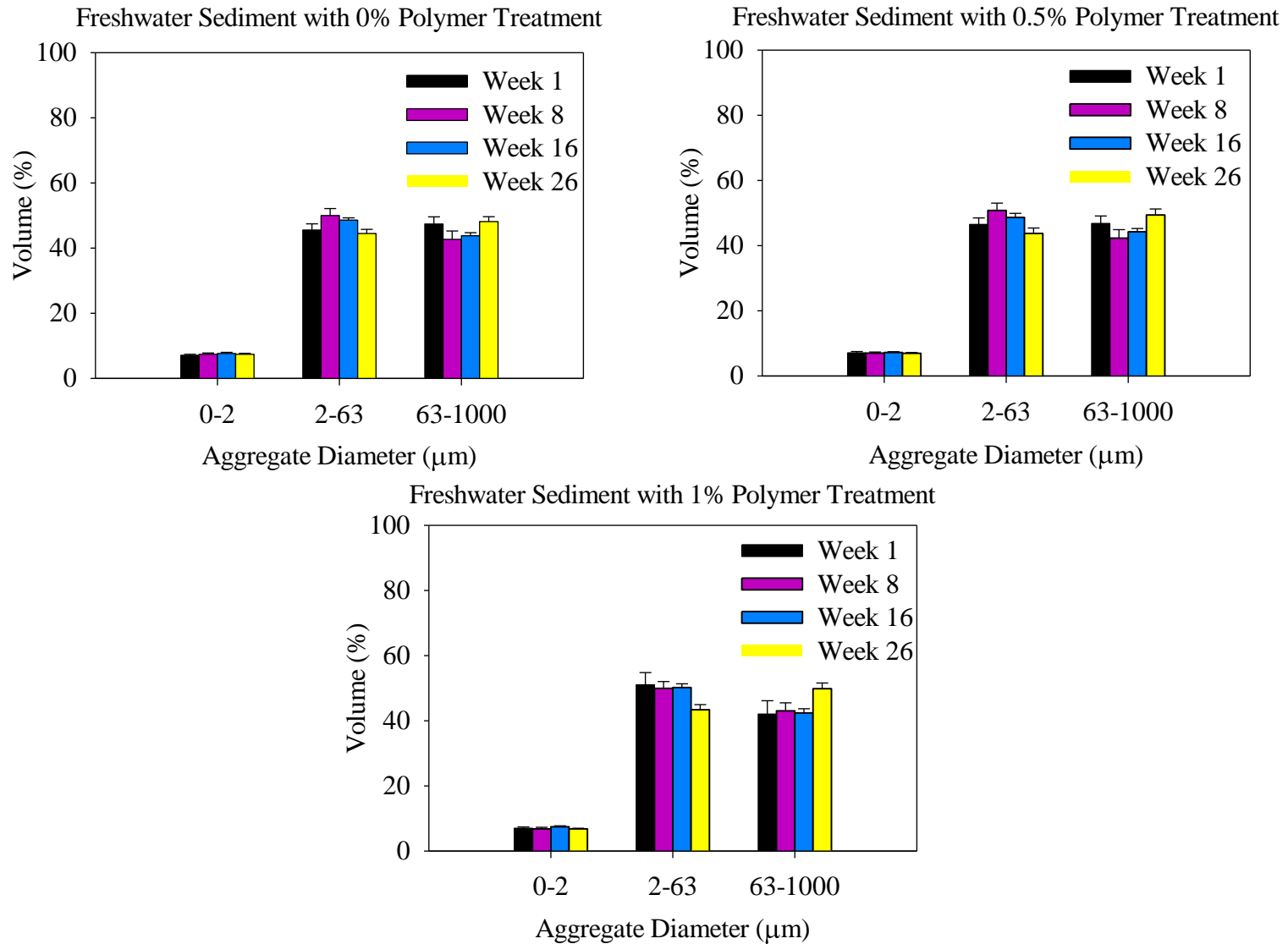
**Table 2.8 Mean percentage of aggregates for the freshwater, intermediate, and marine sediments in the A) clay, B) silt, and C) sand size classes at the 0%, 0.5%, and 1% concentration levels at weeks 1, 8, 16, and 26. Values for polymer types and salinities have been averaged due to no significant differences. Letters indicate significant differences between time periods within each concentration level of each size fraction of each sediment type.**

A	Freshwater			Intermediate			Marine		
	0%	0.5%	1%	0%	0.5%	1%	0%	0.5%	1%
1	7.09 $\pm$ 0.29 a	7.06 $\pm$ 0.41 a	6.97 $\pm$ 0.38 a	5.01 $\pm$ 0.09 a	5.01 $\pm$ 0.08 a	4.88 $\pm$ 0.13 a	3.75 $\pm$ 0.23 a	3.79 $\pm$ 0.27 a	3.40 $\pm$ 0.16 a
8	7.37 $\pm$ 0.38 a	6.95 $\pm$ 0.37 a	6.76 $\pm$ 0.50 a	5.42 $\pm$ 0.05 ab	5.11 $\pm$ 0.07 ab	5.09 $\pm$ 0.06 ab	4.18 $\pm$ 0.22 ab	3.98 $\pm$ 0.17 ab	4.03 $\pm$ 0.20 ab
16	7.65 $\pm$ 0.35 a	7.17 $\pm$ 0.27 a	7.41 $\pm$ 0.32 a	5.85 $\pm$ 0.07 b	5.50 $\pm$ 0.05 b	5.64 $\pm$ 0.07 b	4.99 $\pm$ 0.23 c	4.58 $\pm$ 0.20 c	4.58 $\pm$ 0.19 c
26	7.38 $\pm$ 0.27 a	6.90 $\pm$ 0.28 a	6.76 $\pm$ 0.24 a	5.90 $\pm$ 0.06 b	5.70 $\pm$ 0.06 b	5.75 $\pm$ 0.07 b	4.56 $\pm$ 0.23 bc	4.30 $\pm$ 0.22 bc	4.34 $\pm$ 0.21 bc

B	Freshwater			Intermediate			Marine		
	0%	0.5%	1%	0%	0.5%	1%	0%	0.5%	1%
1	45.5 $\pm$ 1.92 ab	46.5 $\pm$ 2.01 ab	51.1 $\pm$ 3.76 ab	54.2 $\pm$ 0.82 a	55.9 $\pm$ 0.97 a	54.7 $\pm$ 1.35 a	26.2 $\pm$ 1.79 a	28.6 $\pm$ 2.03 a	26.3 $\pm$ 1.35 a
8	50.0 $\pm$ 2.17 a	50.8 $\pm$ 2.23 a	49.9 $\pm$ 2.09 a	60.9 $\pm$ 0.32 b	61.4 $\pm$ 0.66 b	60.6 $\pm$ 0.66 b	31.1 $\pm$ 1.66 b	33.0 $\pm$ 1.50 b	32.4 $\pm$ 1.54 b
16	48.6 $\pm$ 0.68 a	48.6 $\pm$ 1.24 a	50.2 $\pm$ 1.11 a	61.4 $\pm$ 0.44 b	62.1 $\pm$ 0.50 b	62.4 $\pm$ 0.55 b	33.4 $\pm$ 1.69 b	36.0 $\pm$ 1.67 b	35.9 $\pm$ 1.67 b
26	44.5 $\pm$ 1.27 b	43.7 $\pm$ 1.62 b	43.4 $\pm$ 1.55 b	61.1 $\pm$ 0.34 b	61.6 $\pm$ 0.40 b	61.9 $\pm$ 0.31 b	32.6 $\pm$ 1.42 b	35.8 $\pm$ 1.50 b	35.5 $\pm$ 1.29 b

C	Freshwater			Intermediate			Marine		
	0%	0.5%	1%	0%	0.5%	1%	0%	0.5%	1%
1	47.4 $\pm$ 2.16 a	46.8 $\pm$ 2.28 a	42.0 $\pm$ 4.09 a	40.8 $\pm$ 0.91 a	39.1 $\pm$ 1.04 a	40.4 $\pm$ 1.47 a	70.1 $\pm$ 2.02 a	67.7 $\pm$ 2.29 a	70.3 $\pm$ 1.48 a
8	42.7 $\pm$ 2.55 a	42.3 $\pm$ 2.60 a	43.0 $\pm$ 2.45 a	33.6 $\pm$ 0.37 b	33.5 $\pm$ 0.70 b	34.2 $\pm$ 0.72 b	64.8 $\pm$ 1.88 b	62.7 $\pm$ 1.62 b	63.5 $\pm$ 1.73 b
16	43.8 $\pm$ 0.96 ab	44.2 $\pm$ 1.40 ab	42.4 $\pm$ 1.26 ab	32.8 $\pm$ 0.49 b	32.4 $\pm$ 0.48 b	32.0 $\pm$ 0.52 b	61.6 $\pm$ 1.91 b	59.4 $\pm$ 1.86 b	59.6 $\pm$ 1.86 b
26	48.1 $\pm$ 1.49 c	49.4 $\pm$ 1.86 c	49.8 $\pm$ 1.76 c	33.0 $\pm$ 0.35 b	32.7 $\pm$ 0.42 b	32.3 $\pm$ 0.35 b	62.8 $\pm$ 1.64 b	59.9 $\pm$ 1.71 b	60.1 $\pm$ 1.50 b

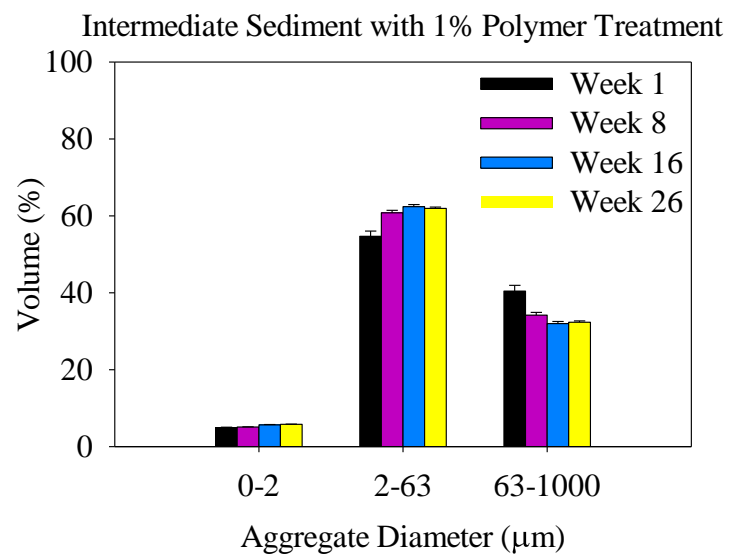
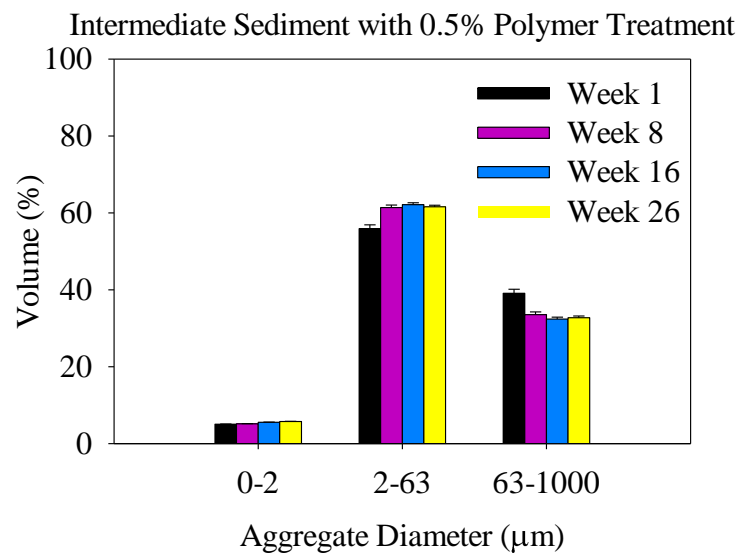
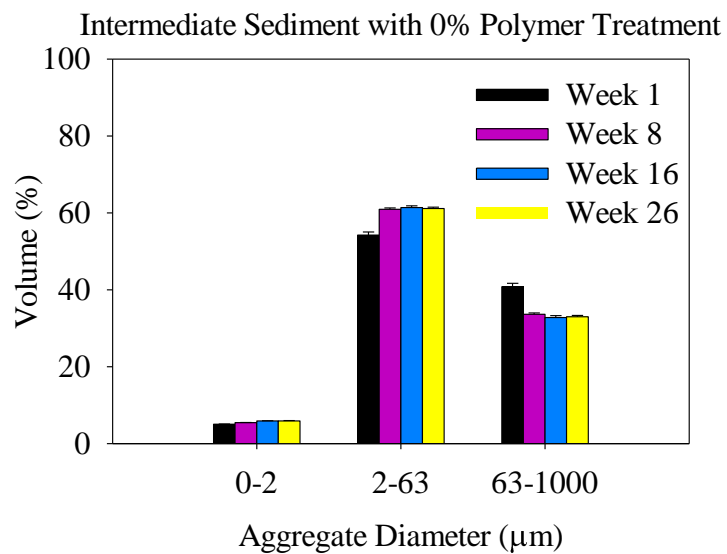
A)



**Figure 2.6 Mean percent volume of aggregates in the clay, silt, and sand fractions for the A) Freshwater, B) Intermediate, and C) Marine sediments at weeks 1, 8, 16, and 26 for the 0%, 0.5%, and 1% concentration levels. Values for polymer types and salinities have been averaged due to no significant differences.**

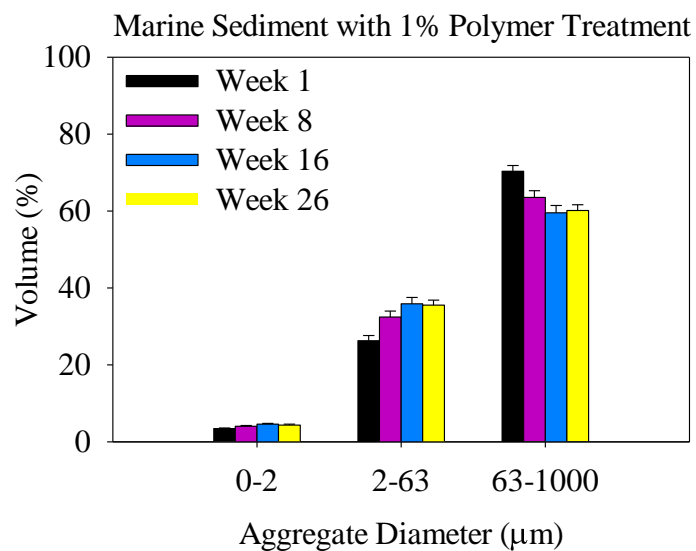
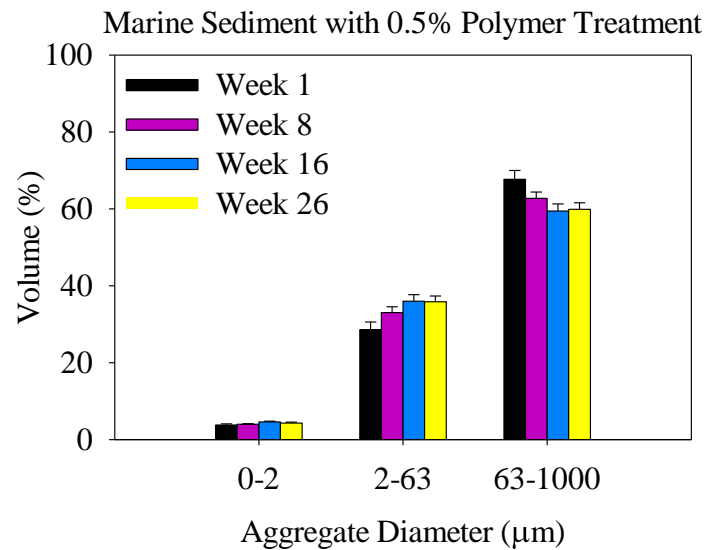
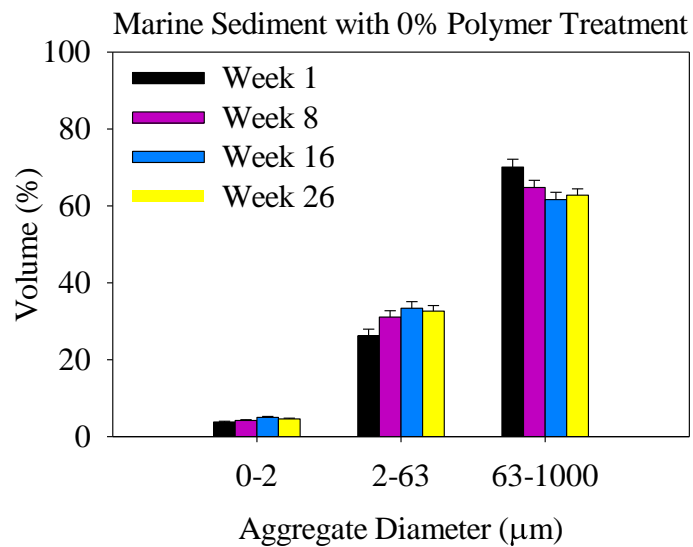
B)

fig. cont'd.



C)

fig. cont'd.



**Table 2.9 Mean and Standard Error Aggregate Diameter Size (units of  $\mu\text{m}$ ) for the Freshwater, Intermediate, and Marine sediments. For the last column, all polymer types, concentration levels, and salinities were averaged due to no significant differences. Letters indicate significant differences between weeks within each sediment type. Crosses indicate significant differences between sediments.**

Sediment	Week	0%	Xanthan 0.5%	Xanthan 1%	Guar 0.5%	Guar 1%	Avg
Freshwater	1	$95.9 \pm 10.8$	$93.3 \pm 14.6$	$86.5 \pm 18.4$	$80.5 \pm 13.9$	$86.8 \pm 15.8$	$89.8 \pm 6.07 \text{ a}^+$
	8	$101 \pm 10.3$	$100 \pm 15.5$	$110 \pm 15.0$	$102 \pm 14.9$	$106 \pm 13.8$	$104 \pm 5.68 \text{ a}$
	16	$122 \pm 5.55$	$124 \pm 12.0$	$120 \pm 8.46$	$133 \pm 20.5$	$104 \pm 12.5$	$121 \pm 4.45 \text{ b}^+$
	26	$172 \pm 10.3$	$173 \pm 3.70$	$141 \pm 10.6$	$188 \pm 25.1$	$186 \pm 6.18$	$173 \pm 6.70 \text{ c}^+$
Intermediate	1	$72.9 \pm 3.02$	$74.4 \pm 5.40$	$71.0 \pm 4.43$	$64.5 \pm 1.49$	$80.3 \pm 10.5$	$72.7 \pm 2.35 \text{ a}^+$
	8	$60.7 \pm 0.474$	$60.4 \pm 2.28$	$60.8 \pm 1.66$	$59.0 \pm 0.631$	$61.7 \pm 1.14$	$60.6 \pm 0.521 \text{ a}^+$
	16	$63.4 \pm 2.27$	$63.5 \pm 3.30$	$67.8 \pm 2.96$	$64.8 \pm 3.80$	$62.4 \pm 4.84$	$64.2 \pm 1.41 \text{ a}^+$
	26	$66.2 \pm 2.38$	$65.6 \pm 3.21$	$66.1 \pm 3.03$	$69.4 \pm 3.48$	$66.6 \pm 3.43$	$66.7 \pm 1.29 \text{ a}^+$
Marine	1	$121 \pm 5.55$	$111 \pm 10.4$	$123 \pm 5.64$	$115 \pm 8.33$	$123 \pm 4.66$	$119 \pm 3.04 \text{ a}^+$
	8	$102 \pm 3.81$	$98.7 \pm 4.26$	$98.9 \pm 4.24$	$97.6 \pm 3.84$	$102 \pm 5.33$	$100 \pm 1.88 \text{ b}$
	16	$101 \pm 5.13$	$99.2 \pm 7.47$	$98.9 \pm 6.57$	$98.4 \pm 7.24$	$96.2 \pm 6.15$	$99.2 \pm 2.71 \text{ b}^+$
	26	$104 \pm 4.71$	$105 \pm 9.75$	$97.9 \pm 5.72$	$96.5 \pm 5.46$	$101 \pm 6.32$	$101 \pm 2.64 \text{ b}^+$

aggregate diameter size. For the freshwater sediment, the average aggregate diameter size increased over time with the following values: 90  $\mu\text{m}$ , 104  $\mu\text{m}$ , 121  $\mu\text{m}$ , and 173  $\mu\text{m}$ . By week 26, the increase in aggregate diameter was significantly different from week 1, possibly due to clay particle aggregation from consolidation and natural organic materials. For the intermediate sediment, the average aggregate diameter was not significantly different at any week. The following values represent the average aggregate diameter over weeks 1, 8, 16, and 26: 73  $\mu\text{m}$ , 61  $\mu\text{m}$ , 64  $\mu\text{m}$ , and 67  $\mu\text{m}$ . For the marine sediment, the average aggregate diameter decreased over time with the following values: 119  $\mu\text{m}$ , 100  $\mu\text{m}$ , 99  $\mu\text{m}$ , and 101  $\mu\text{m}$ . By week 26, the decrease in aggregate diameter was significantly different from week 1, possibly due to less pore space from consolidation.

## **2.4 Discussion**

### **2.4.1 pH**

The presence of a polymer generally had no significant effect on pH compared to control samples, with a few exceptions for the marine sediment. The pH of the sediments generally remained in the 6.0 to 8.0 range, which includes the expected pH of marsh soils and flooded soils (Reddy and DeLaune 2008). Over time, pH of the three sediments increased, which is consistent with the generally observed relationship that an increase in the amount of organic matter raises pH due to an increase in electron donors (Reddy and DeLaune 2008).

The initial decrease in pH can be explained by a flush in microbial activity resulting from agitation of sediment and polymer. The control samples were also agitated to maintain consistency across experimental units. For all samples, the agitation increased microbial activity, which increased carbon dioxide production, lowering pH due to carbonic acid formation. As time progressed, microbial activity and carbon dioxide production decreased, thereby reducing carbonic acid formation and raising pH. At any time during the experiment, pH did not enter the strongly acidic range, which may be one factor limiting adsorption of polymers onto clays. Several studies have shown that low pH

increases polymer adsorption due to the formation of cation bridges from excess hydrogen ions in soil solution (Theng 1982; Wallace et al. 1986b; Gu and Doner 1993). In this experiment, pH remained in the neutral range, which may be one reason why aggregation from the polymers did not occur.

#### **2.4.2 Moisture Content and Sediment Consolidation**

The relationship between moisture content and sediment consolidation over time relates to particle size distribution of the three sediments. The freshwater sediment had relatively high clay content, which explains why the freshwater sediment had the highest moisture content, least amount of consolidation in the first week, and significantly increasing consolidation over time. Clay particles have the smallest diameter of soil particles and take the longest time to settle. Additionally, clay particles have the highest amount of surface area, which contributes to their ability to hold more water than a soil with larger particles.

On the other end of the spectrum for particle size, the marine sediment was mostly sand. High sand content explains why the marine sediment had the lowest moisture content and total consolidation in the first week. Sand particles have a relatively large diameter and settle out of the water column very rapidly. As sand particles settle, relatively large amounts of water expel from the pore spaces and collect on the surface.

The intermediate sediment had less clay than the freshwater sediment but more clay than the marine sediment. Moderate clay content explains why the intermediate sediment had intermediate results for moisture content and sediment consolidation.

When considering the cumulative amount of consolidation over 26 weeks, the significant amount of consolidation happened between the beginning of the experiment and the first flooding event, which took place 8 to 10 days later. Any other significant consolidation occurred after an extended period of no flooding, which supports general observations that drying cycles will cause sediment to compact.



Relationships between moisture content, sediment consolidation, and particle size corroborate the indication that polymers did not increase sediment stability because they were not present in the sediment long enough due to microbial degradation (supporting data in chapter 3). The processes discussed above agree with expected behaviors of the sediments given their particle size distributions.

#### **2.4.3 Aggregation in Wet Sediments**

In general, the presence of a polymer did not have any significant effect on increasing aggregation in the clay, silt, or sand fractions of any of the three sediments. The polymer addition was anticipated to increase particle aggregation as a process to enhance stability of newly deposited dredged sediment. Differences in size classes between initially characterized sediments and sediments for aggregate analysis result from natural processes and naturally occurring soil organic matter.

In both the freshwater and intermediate sediments for aggregate analysis, the increase in the sand fraction compared to initially characterized sediments suggests aggregation of silt and clay particles from organic materials. In the marine sediments for aggregate analysis, the increase in the silt fraction compared to initially characterized sediments suggests that clay and silt particles filled the pore spaces between sand grains and formed aggregates. Once again, differences between size classes of initially characterized sediments and wet sediment samples reinforce the importance of naturally occurring soil organic matter in natural aggregation processes. Possibly, the polymer had no effect on aggregation because the particles in the natural sediments were already fully aggregated (Appendix C).

Environmental conditions in the soil influence electrostatic interactions that affect adsorption of different types of polymers to clay particles (Ruehrwein and Ward 1952; Martin 1972; Theng 1982; Wallace 1986; Gu and Doner 1993; Seybold 1994;). In a lab study conducted by Rick Nugent, both xanthan gum and guar gum increased the liquid limits of kaolinite clay (Nugent et al. 2009). For the current study with field sediments, however, several environmental factors explain why polymers did not increase aggregation.

In this study, guar gum was the chosen neutral polymer. Nonionic or neutral polymers typically maintain a randomly coiled structure, which renders them less effective at flocculating clay particles (Theng 1982). Nonionic polymers adsorb to clays through Van der Waals forces by replacing water molecules around the expanding double layer of clays (Seybold 1994). Addition of electrolytes to the soil environment reduces the amount of expansion between layers of clay particles, which makes it difficult for nonionic polymers to penetrate any clay aggregates (Theng 1982). As a result, nonionic polymers spread out and coat the soil surface. Previous flocculation tests indicate that any stabilizing effect of guar gum is enhanced when mixed with an acidic solution (Wallace 1986). In addition, plant polysaccharides (i.e. guar gum) are unstable in solution against microbial decomposition (Martin 1971; Wallace et al. 1986a).

In this study, xanthan gum was the chosen anionic polymer. Adsorption of anionic polymers onto clay particles depends on pH, ionic strength of the soil solution, and the type of cations present in soil solution. Naturally, an anionic polymer and negatively charged clay particles will repel each other. Soils with low pH or high ionic strength experience greater adsorption of anionic polymers to clay particles (Theng 1982; Wallace et al. 1986b; Gu and Doner 1993). By lowering pH or increasing ionic strength of the soil solution, more cations are available to form bridges between the polymer and clay particles. In addition, hydrogen ions in low pH soils may neutralize the anionic charge on the polymer, allowing polymer penetration into the spaces in between layers of expanding clays. Helical conformation allows anionic polymers a large “grappling” distance, which enables extension into the soil solution, promoting interparticle bridging (Reuhrwein and Ward 1952; Theng 1982; Gu and Doner 1993). Adding a flocculating agent to the solution before adding an anionic polymer may enhance binding properties because the polymer will interact with aggregates instead of particles (Ruehrwein and Ward 1952).

Xanthan gum does not respond to environmental conditions in a manner similar to other anionic polymers. For example, the performance of xanthan gum is unaffected by a wide range of ionic strength and pH (Becker 1998). In addition, the viscosity of xanthan gum does not increase with increasing ionic strength beyond a salt concentration of 1% weight per volume (Smith et al. 1981). For the current study, all salt concentrations were equivalent to or higher than 1% weight per volume. Results contradict previous conclusions that high ionic strength increases the effectiveness of anionic polymers. At higher salt concentrations, cations may be inundating the polymer and inhibiting any conformation changes, which reduces the polymer's ability to adsorb to clay particles.

In some cases, addition of anionic polysaccharides and humic acids promotes clay dispersion due to electrostatic repulsion between the soil amendment and clay particles (Gu and Doner 1993). Subsequently, dispersed particles of organic matter and clay lower the hydraulic conductivity (i.e. the rate of water movement through a porous medium) of soils, which reduces stability. When applied in combination with aluminum, soil amendments increase aggregation through the formation of cation bridges (Gu and Doner 1993).

## **2.5 Conclusion**

Experimental results show that natural polymers and applied environmental conditions did not increase aggregation of wet sediments. Strong evidence of microbial degradation of the polymer within the first week suggests that microbes rapidly metabolized the added polymers. The sediments then naturally settled, indicated by consolidation over time.

The high capacity of polymers to absorb water might allow electrolytes to fill the spaces between polymer molecules and clay particles, thereby inhibiting the ability of the polymer to expand and form bridges. Microbial decomposition of added polymers causes additional release of microbial polysaccharides, which may account for differences in aggregation over time; however, dispersion of clay particles might limit the effectiveness of microbial polysaccharides for aggregation. Furthermore,

additional microbial by-products and microbial biomass turnover from polymer addition may provide additional substrates for decomposition of soil organic carbon, thus removing components for natural aggregation.

A solution that may work in the future is to combine the xanthan gum and guar gum polymer treatments. Some studies have shown that mixing natural anionic polymers and polysaccharides leads to cross-linking, which can increase binding abilities of soil amendments (Wallace et al. 1986b).

Another option is to mix polysaccharides with synthetic anionic polymers. After cross-linking occurs, the polysaccharide is more resistant to microbial breakdown because of protection from the synthetic polymer (Wallace et al. 1986c).

High moisture content of wetland sediments may require the use of synthetic polymers for aggregation. A material that does not lose structure in water (i.e. insoluble polymer) and that resists microbial activity may be more successful in stabilizing wetland sediments. Another option is to have a period of drying to allow the polymers to bind tightly to clay particles. Further research is needed to explore these options for wetland sediment stabilization.

## **CHAPTER 3: MICROBIAL RESPONSE TO CARBON INPUT FROM SOIL-STABILIZING AMENDMENTS USED WITH DREDGED SEDIMENT FOR COASTAL WETLAND RESTORATION**

### **3.1 Introduction**

In soils and sediments, measures of microbial biomass and respiration rates provide insight about the microbial community. Microbial biomass is defined as the component of soil that contains both active and dormant microbial life stages (Sparling 1985). Respiration rates represent the physiologically active component of the biomass since only microbes in the active life stage respond to substrate addition or nutrient input (Sawada et al. 2008). The active microbial biomass is responsible for litter decomposition, nutrient cycling, and energy flow (Wardle 1992). By acting as a “transformation station” microbial biomass converts organic materials into bioavailable nutrients that can be utilized by plants and other soil organisms (Van Veen and Kuikman 1990).

Physical properties such as structure and texture of the soil environment influence size of the microbial community. Clay soils, compared to sandy soils, have a greater capacity for retaining carbon in the soil organic matter component because the carbon is protected in smaller pore spaces (Van Veen and Kuikman 1990). Clayey soils also have greater surface area for organic matter to bind to clay particles. In addition, soils with higher clay content have enhanced biomass retention after substrate addition for the following reasons: lower turnover rate of microbial products, increased retention of microbial biomass and organic matter, increased nutrient adsorption, and greater protection from microbial predators (Wardle 1992). Microbes are protected in clay soil aggregates, which increase efficiency of microbial utilization of substrates.

Hydrological regime and quality of organic carbon in the soil are the two most important factors influencing microbial community composition and activity. Since hydrology primarily controls oxygen availability and redox status, the extent of flooding in a soil influences the rate of organic matter accumulation and decomposition, nutrient transformations, and availability of organic substrates

to microbes (Boon et al. 1996; Bossio et al. 2006; Groffman 1996). As redox potential decreases below the point of anoxia, sediments become more reducing with greater organic matter accumulation and reduced organic matter decomposition.

The amount and activity of microbial biomass in soils respond to available organic matter substrates. Several studies have used glucose as a model to look at microbial response to substrate addition. Higher amounts of glucose addition yield a long-term increase of microbial biomass whereas lower amounts of glucose addition yield an initial large increase in biomass followed by a sharp decline (Tsai et al. 1997). The breakdown of old biomass and the creation of new biomass may account for long-lasting impacts in cases of high amounts of glucose addition.

Carbon substrates serve as material for production of new cells, indicated by assimilation of carbon into storage compounds. Assimilation of carbon represents the passive component of the microbial biomass pool (Sawada et al. 2008). Microbes store and conserve carbon to be used for cell maintenance and survival. Carbon also serves as a source of energy, indicated by respiration of carbon dioxide. Increased respiration rates following substrate addition indicate use of carbon for structural compounds as the active biomass grows in size. Glucose addition increases the active component of microbial biomass (Wardle and Parkinson 1991). At a threshold concentration level of glucose addition, the microbial community switches from assimilating carbon into storage compounds to incorporating carbon into structural compounds (Sawada et al. 2008). Generally, respiration of 50% of carbon input indicates complete degradation of the material; the rest of the carbon is assimilated into biomass (Shen and Bartha 1996).

Microbial community species composition also responds to substrate input. In heterogeneous microbial communities, low substrate addition produces a response in K-strategist microbes whereas high substrate addition produces a response in r-strategist microbes, as indicated by growth rates (Shen and Bartha 1996). With K-strategist species, microbes metabolize substrate at a slow rate over a long

period of time, and the microbial community may not increase in size. With r-strategist species, microbes metabolize substrate much faster, but they collectively may not be able to decompose the substrate at the current population capacity; thus, the microbial biomass undergoes rapid turnover. As the community changes, respiration continues to produce carbon dioxide (Shen and Bartha 1996).

Microbial interactions change after substrate addition. For example, high amounts of organic substrate addition induce the production of secondary metabolites and microbial interactions like antagonism and competition (Griffiths et al. 1999). The addition of readily decomposable carbon causes an increase in r-strategist species whereas the presence of non-labile carbon in the form of recalcitrant organic matter causes an increase in K-strategist species (Dilly 2006). Uncertainties exist as to whether the change in the ratio of r-strategists to K-strategists indicates a change in community composition or a shift in species transition states (Maly et al. 2009).

Several measures of microbial activity exist to decipher community responses to fluctuations in environmental conditions. For example, physiological measurements such as respiration may reflect the activity of a group of microbial species as representing the entire microbial community. By looking at total respiration per unit of biomass, otherwise known as  $qCO_2$  or the metabolic quotient, some insight into community composition can be obtained (Anderson 2003). If the metabolic quotient is lower, then the active component of microbial communities may be increasing; microbial species efficiently use energy from carbon inputs and adjust to environmental fluctuations (Anderson and Domsch 1993). In addition, a lower metabolic quotient indicates that microbial communities are responding to greater availability of carbon for microbial use, which can be verified by a high ratio of microbial carbon to soil organic carbon, or  $C_{mic}:C_{org}$  (Anderson 2003).

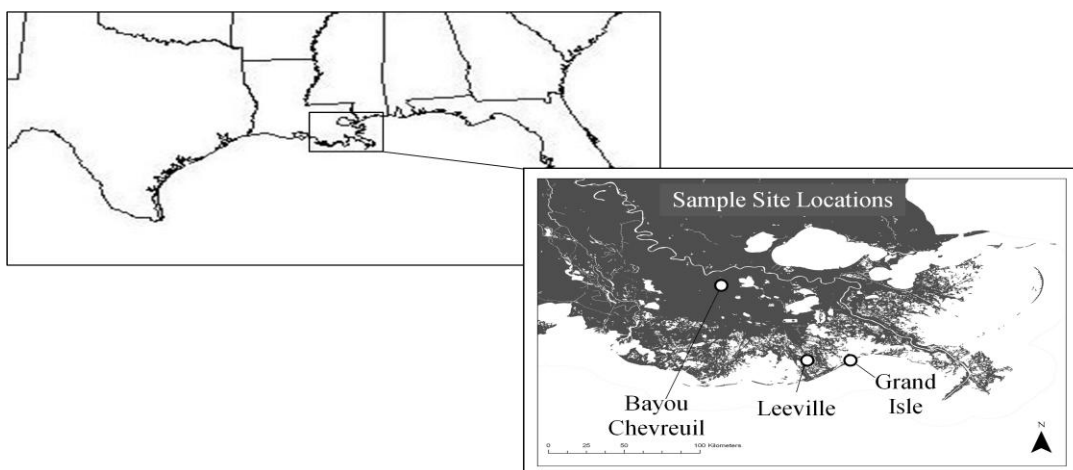
This study determined the impact of the addition of two natural polymers on microbial properties of lab-simulated, water-saturated hydraulically dredged sediments. Natural polymers are being studied as a soil amendment with the goal of stabilizing hydraulically dredged sediments used

for wetland restoration until marsh plants become established. Quantifying the microbial biomass carbon pool and microbial respiration rates will provide information about any effect the polymer additions have on microbial activity. The two polymers are potential sources of hydrolysable carbon substrate that may be available for microbial degradation. Therefore, the null hypothesis states that over time, the addition of polymer will have no impact on microbial biomass carbon or microbial respiration. The alternative hypothesis states that over time, the addition of polymer will either increase or decrease microbial biomass carbon, microbial respiration of carbon dioxide, or both in hydraulically dredged sediments.

### 3.2 Materials and Methods

#### 3.2.1 Sediment Sample Locations

Sediment was collected at three sites in southern coastal Louisiana (Figure 3.1). At each site, a Peterson hand-operated dredge was used to collect sediment to be placed in 20-liter plastic buckets.



Map Courtesy of Andrew Tweel (LSU)

**Figure 3.1 Location of the three sampling sites in coastal Louisiana.**

Bayou Chevreuil is located in St. James Parish intersecting LA Route 20 (29.91° N 90.73° W) and represents a freshwater site containing sediment with high clay content. Bayou LaFourche is located in LaFourche Parish alongside LA Route 1 (29.25° N 90.21° W) in Leeville and represents an intermediate salinity site containing sediment with moderate clay content. The third site, Grand Isle, is



located in Grand Isle State Park in Jefferson Parish (29.26° N 89.95° W) and represents a marine site containing sediment with low clay content.

### 3.2.2 Sediment Characterization

To ensure sediment homogeneity, sediment from each 20-liter bucket was passed through a 0.635 cm sieve to remove large plant debris and sticks. An electric drill attached to a paint mixer was used to homogenize the sediment for ten minutes in each bucket. To ensure that all buckets for each sediment type were homogeneous, the following soil properties for each bucket were compared: moisture content, organic matter content, and particle size distribution as determined by the hydrometer method.

All three sediments were characterized for the following properties: moisture content, organic matter content, redox potential, pH, soil salinity, cation exchange capacity, exchangeable metals, and particle size distribution.

For moisture content, three sub-samples of each sediment type were dried at 105° C until a constant weight was reached (Gardner 1986). The following formula was used to calculate percent moisture content on a wet weight basis (DeAngelis 2007):

$$[(\text{g beaker} + \text{g mud}) - (\text{g beaker} + \text{g dry sediment})] / [(\text{g beaker} + \text{g mud}) - \text{g beaker}] * 100$$

To determine organic matter content, the loss-on-ignition method was used (Nelson and Sommers, 1996). In a muffle furnace, dry (105°C) sediment samples were heated to 435°C for 5 hours. The following formula was used to calculate percent organic matter content:

$$[(\text{g beaker} + \text{g sediment})_{105} - (\text{g beaker} + \text{g sediment})_{435}] / [(\text{g beaker} + \text{g sediment})_{105} - \text{g beaker}] * 100$$

For redox potential, platinum-tipped electrodes were cleaned and tested as described by Patrick et al. (1996). Four platinum electrodes were inserted into sediment samples and used in conjunction with a calomel reference electrode to obtain a reading in millivolts ( $E_c$ ); these values were corrected to a standard hydrogen reference electrode for final readings ( $E_h$ ).

For soil pH, a calibrated combination pH electrode with a Ag/AgCl reference was used (Thomas 1996). Soil porewater was collected by centrifuging field moist sediment in a Fisher Scientific accuSpin 3/3R centrifuge at 3500 rpm (3021 g radial centrifugal force) for 15 minutes. The supernatant water was analyzed for salinity with an Accumet AB30 conductivity meter (Rhoades 1996).

Cation Exchange Capacity (CEC) was determined by the Unbuffered Salt Extraction Method according to Sumner and Miller (1996). All three sediments were saturated with 0.2 M  $\text{NH}_4\text{Cl}$ , washed with deionized water, and saturated with 0.2 M  $\text{KNO}_3$  to displace the  $\text{NH}_4^+$ . The extracted supernatant was analyzed for exchangeable  $\text{NH}_4^+$  (US EPA-103-A Rev. 4) using a SEAL AQ2 automated discrete analyzer. The following equation was used to calculate CEC (Sumner and Miller 1996):

$$(\text{mg } \text{NH}_4^+ - \text{N} / \text{L}) * (\text{mL extractant}) * (\text{valence of } \text{NH}_4^+) / (\text{g dry sediment}) * (\text{atomic weight of } \text{NH}_4^+)$$

The results have units of centimoles of cation charge per kilogram of sediment, which is equal to milliequivalents per 100 grams of sediment.

Particle size distribution and textural class were determined by the hydrometer procedure according to Gee and Bauder (1986). Sediments were pre-treated to remove carbonates and soluble salts using sodium acetate, organic matter using hydrogen peroxide, and free iron oxides using citrate-bicarbonate, sodium dithionite, and sodium chloride. Values from hydrometer readings were used in calculations according to Patrick (1958) to determine the proportion of sand, silt, and clay in all three sediments.

### 3.2.3 Experimental Setup

The polymer treatments included two natural polymers (xanthan gum and guar gum) that are commercially available and have different molecular properties and charges (Table 3.1). Xanthan gum is an extracellular polysaccharide produced by the bacterium *Xanthomonas campestris* (Kim 2006). The structure of xanthan gum consists of repeating pentasaccharide monomers with varying amounts

of acetyl and pyruvate substituents (Jong 2007). The carboxylic acid groups attached to the backbone provide xanthan gum a net negative charge.

**Table 3.1 Properties of xanthan gum and guar gum.**

Polymer	Source	Molecular Formula	Molecular Weight	Charge	Charge Density	% Total Carbon
Xanthan Gum	microbial extracellular polymer	$(C_{35}H_{49}O_{29})_n$	$0.9 - 1.6 \times 10^6$ Da	anionic	0.25 <sup>a</sup>	40.84
Guar Gum	plant polysaccharide	$(C_{18-20}H_{30}O_{15})_n$	$1.0 - 2.0 \times 10^6$ Da	non-ionic	0	43.25

<sup>a</sup>Charge Density in mol/mol monosaccharide

Guar gum is extracted from the seed of a guar gum plant, a leguminous shrub known as *Cyamopsis tetragonoloba* (Kim 2006). Guar gum consists of repeating galactose and mannose units (Jong 2007). The absence of carboxylic acid groups on the polymer results in an overall neutral charge. Both xanthan gum and guar gum polymer solutions were made to concentration levels of 1% and 2% by weight. To create a 1% polymer solution, 2 g of polymer powder were added to 198 g of water made up to the appropriate salinity. To create a 2% polymer solution, 4 g of polymer powder were added to 196 g of water made up to the appropriate salinity. Experimental units with polymer treatments received a polymer solution of either 1% or 2% concentration: the 2:1 sediment-to-polymer ratio, resulted in final concentrations of polymer at 0.5% and 1%.

The experimental units were 16-oz. polyethylene cups containing 350 grams of wet sediment mixed with 175 grams of 1% or 2% polymer solution made up with the appropriate salinity solution. Two different salinity solutions were applied to each sediment-polymer combination in order to simulate *in situ* salinity ranges. Salinity treatments were 1 and 5 ppt for Bayou Chevreuil sediments, 5 and 10 ppt for Leeville sediments, and 15 and 25 ppt for Grand Isle sediments. Control experimental units received 175 mL of water of the appropriate salinity. Due to different moisture contents for the three sediments, different masses of polymer carbon were added to the sediments for each

concentration of polymer solution (Table 3.2). All powder-water mixtures were blended in a kitchen blender for 30 seconds to obtain a well-mixed polymer solution.

**Table 3.2 Polymer added carbon to each experimental sample. Units are g C kg<sup>-1</sup> dry sediment.**

<b>Sediment</b>	<b>Concentration</b>	<b>Added C from Xanthan Gum</b>	<b>Added C from Guar Gum</b>
Chevreuil	0.5%	9.22	9.76
	1.0%	18.4	19.6
Leeville	0.5%	7.08	7.48
	1.0%	14.2	15.0
Grand Isle	0.5%	3.65	3.86
	1.0%	7.29	7.73

A randomized block design was implemented to evaluate how several dependent variables over 26 weeks were affected by sediment type (i.e. sampling location), salinity, polymer and polymer concentration. Response variables that were measured included redox potential, pH, microbial biomass carbon, and microbial basal respiration rates. Each treatment was prepared in triplicate in 16-ounce opaque plastic cups. There were 432 experimental units (3 sediment types x 2 salinities x 2 polymers x 3 concentrations x 4 time periods x 3 replicates). A Barnstead Max-Q 2508 reciprocating shaker was used to mix each sediment and polymer combination. With a fixed circular orbit of 1.2 cm, each sediment-polymer mixture shook at the maximum setting (400 rpm) for fifteen minutes on the dual action setting (circular and reciprocating movements). Then, each mixture was poured into the cups and set on the lab bench for the appropriate time period before being analyzed for dependent variables.

Destructive sampling was employed. Consequently, at the end of each designated time period (i.e. weeks 1, 8, 16, and 26), redox potential and pH were measured. Then, the samples were stored at 4°C until analysis for microbial biomass and microbial respiration. For the duration of each time period, decanting and re-flooding took place every 8 to 10 days to maintain sediment saturation. Any remaining supernatant fluid was decanted; then, each sample was re-flooded with water of the appropriate salinity.

### **3.2.4 Redox Potential**

Changes in redox potential reflect changes in the microbial community in response to fluctuating oxygen status. Redox potential tends to be more negative in fully saturated sediments, indicating low oxygen status and greater electron availability for microbial activity (Reddy and DeLaune 2008). Therefore, at the beginning and end of each time period, redox potential was measured in the same manner as described earlier for sediment characterization. One platinum electrode was used for each sample.

### **3.2.5 pH**

Changes in pH also reflect changes in the microbial community. Therefore, at the beginning and end of each time period, pH was measured in the same manner as described earlier for sediment characterization.

### **3.2.6 Microbial Biomass Carbon Determination**

The Chloroform Fumigation Extraction Method (Vance et al. 1987), as modified by White (2006), was followed to determine the pool of microbial biomass carbon in each sample. From each cup, two samples were weighed out into 25-mL centrifuge tubes and designated as fumigate (F) or non-fumigate (NF). Every tenth sample of both F and NF samples were represented in triplicates. All F samples received half a mL of chloroform and were placed in a glass vacuum dessicator along with a glass beaker containing approximately 50 mL chloroform and 5 boiling stones. By connecting a vacuum pump hose to the stopcock opening on the dessicator, pressure decreased to -25 psi, which caused the chloroform in the samples and the beaker to boil; the chloroform was brought to a boil a total of 3 times. Then, the dessicator was sealed to expose the samples to the chloroform fumes for 24 hours. After this time, the dessicator was opened and any excess chloroform fumes were removed with the vacuum pump.

After fumigation, all samples received the same treatment. Twenty-five mLs of  $K_2SO_4$  extractant were added to each tube. Samples were placed on a Barnstead Max-Q 2508 reciprocating shaker set at the maximum reciprocating setting (250 rpm) for 30 minutes. Then, samples were centrifuged in a Fisher Scientific accuSpin 3/3R centrifuge set to run at 4000 rpm (3452 g radial centrifugal force) for 10 minutes. After centrifugation, the supernatant was filtered through a vacuum filtration apparatus using Whatman 42 filter paper. To prepare for carbon analysis, approximately 25 mLs of each sample were acidified with five drops of concentrated HCl and left in the fume hood overnight to purge any inorganic carbon. The following day, the samples were diluted with distilled water and analyzed for total carbon on a Shimadzu TOC-V series carbon analyzer. The output gave the concentration of organic carbon in mg/L for each sample.

To determine mg soluble C  $kg^{-1}$  dry sediment for both fumigate and non-fumigate samples, the following formula was used:

$$\text{Total extractant volume (L)} * \text{TOC mg/L} * \text{dilution factor} / \text{kg dry weight of sediment}$$

The amount of microbial biomass carbon was found from the following formula according to Vance et al. (1987):

$$\text{Biomass C} = k_{EC} \times E_c$$

A value of 2.70 was used for the conversion factor,  $k_{EC}$  (Sparling et al. 1990).  $E_c$  was found by subtracting the soluble carbon of non-fumigate samples from the fumigate samples.

The ratio of microbial carbon to sediment organic carbon ( $C_{mic}:C_{org}$ ) was found by dividing microbial biomass carbon values by the organic matter content for each sediment type (Rinklebe 2006). The  $C_{mic}:C_{org}$  ratio was calculated for the 0%, 0.5%, and 1% concentration levels for the Bayou Chevreuil, Leeville, and Grand Isle sediments at weeks 1, 8, and 16. The ratio was not calculated for week 26 samples due to no significant difference in microbial biomass from week 16 to week 26 for any of the sediments.

### 3.2.7 Microbial Basal Respiration Measurements

At the end of week one, 5 g of sediment sample were weighed into 60 mL glass serum bottles. Ten mLs of DI water were added, and the bottles were sealed with a rubber septa and aluminum cap. Using a vacuum pump and tubing connected to a syringe needle, headspace gas was removed to -68 kPa. Then, each sample was flushed with nitrogen gas for five minutes. Each sample was brought to a pressure in the range of 15 to 20 kPa. The bottles were kept at a temperature of 25 °C for the duration of the study. Once every day, the samples were gently shaken to maintain a homogeneous matrix.

After 35 days, the pressure of each serum bottle was measured using a SPER Scientific Manometer. Then, gas samples were withdrawn (either 50 or 100 µL) and injected into a gas chromatograph for carbon dioxide analysis. For every sample, the gas chromatograph reported the peak area of carbon dioxide that was detected based on a standard curve. All samples were analyzed on a Shimadzu (Kyoto, Japan) GC-2014 (thermal conductivity detector at 160 °C; packed Poropak N column (6 ft; 80/100 mesh) column supplied by Sigma-Aldrich (St. Louis, MO) with an oven temperature of 80 °C).

Gas samples were repeatedly withdrawn and analyzed every ten days from day 35 to day 125, which was determined as a time well past the peak of carbon dioxide evolution. To calculate the moles of CO<sub>2</sub> in each sample, the ideal gas law equation ( $PV=nRT$ ) was used, where  $P$  = pressure (atm),  $V$  = volume CO<sub>2</sub> injected (µL),  $n$  = moles CO<sub>2</sub>,  $R$  = gas constant (82057 µL atm mol<sup>-1</sup> K<sup>-1</sup>), and  $T$  = temperature (Kelvin). Using the molecular weight of carbon and the dry weight of each sediment sample, moles of CO<sub>2</sub> were converted to grams CO<sub>2</sub>-C kg<sup>-1</sup> dry sediment.

The metabolic quotient, or  $q\text{CO}_2$ , was calculated for weeks 1, 8, and 16. Basal respiration was divided by microbial biomass carbon. The following formula was used to find the metabolic quotient in units of µg C g C<sub>mic</sub><sup>-1</sup>, where C<sub>mic</sub> is microbial biomass carbon (Rinklebe and Langer 2006):

$$q\text{CO}_2 = [(g \text{ CO}_2\text{-C kg}^{-1}\text{dry sediment day}^{-1}) / (g \text{ C}_{\text{mic}} \text{ kg}^{-1} \text{ dry sediment})] * 10^6$$

To obtain an appropriate basal respiration rate for week 1, the daily rate of carbon dioxide production from day 0 to day 35 was calculated. For week 8, the daily rate of carbon dioxide production from day 45 to day 55 was calculated. For week 16, the daily rate of carbon dioxide production from day 105 to day 115 was calculated.

### **3.2.8 Statistical Analyses**

SAS 9.1 software (2009) and SigmaPlot 11.0 software (2008) were used to analyze the data. For redox potential, pH, and microbial respiration, an ANOVA along with stepwise variable selection reduced each model to the most significant effects. To enter the model, factors had to be significant at an alpha value of 0.10; to stay in the model, factors had to be significant at an alpha value of 0.05. The model statement for the microbial biomass data did not need to be reduced. After a test of Type III fixed effects with a Tukey adjustment, least squares means analysis was evaluated to look for differences between any significant effects for all dependent variables. An alpha value of 0.05 was used for all analyses.

An ANOVA with a Type III test of fixed effects and a least squares means analysis with a Tukey adjustment was used to distinguish any differences between the  $C_{mic}:C_{org}$  ratios and metabolic quotients for each sediment by concentration combination.

## **3.3 Results**

### **3.3.1 Sediment Characteristics**

General sediment characteristics differed from one another (Table 3.3). The Bayou Chevreuil sediment was classified as clay with particle size distribution values of approximately 70% clay, 21% silt, and 9% sand. Cation exchange capacity for Bayou Chevreuil was the highest of all sediments at 125 centimoles of charge per kilogram of sediment. Moisture content (wet weight basis) was the highest for the Bayou Chevreuil sediment at 75%; organic matter was also the highest at 14%. The



Bayou Chevreuil sediment represented a freshwater site with a porewater salinity of 0.5 parts per thousand (ppt) and hereby will be referred to as the freshwater sediment.

The Leeville sediment was classified as silty clay with particle size distribution values of approximately 43% clay, 38% silt and 19% sand. Cation exchange capacity for Leeville was lower than that of the freshwater sediment at 85 centimoles of charge per kilogram of sediment. Moisture content (wet weight basis) was also slightly lower than that of the freshwater sediment at 67%; organic matter follows the same pattern at 8%. The Leeville sediment represented an intermediate salinity site with a porewater salinity of 4.6 ppt and hereby will be referred to as the intermediate sediment.

**Table 3.3 Characteristic properties of the Freshwater, Intermediate, and Marine sediments.**

Characterization of Sediment			
Soil Properties	Freshwater	Intermediate	Marine
Moisture Content % (wet weight)	74.8	67.1	36.3
OM Content %	13.8%	8.34%	1.56%
Redox Potential (mV)	-26	-18	-207
Soil pH	6.50	6.90	6.90
CEC (cmol charge kg <sup>-1</sup> dry sediment)	125.0	84.6	27.7
Porewater Salinity (ppt)	0.50	4.60	15.5
% Sand	9.17	19.2	70.8
% Silt	20.8	38.3	13.3
% Clay	70.0	42.5	15.8
Textural Class	Clay	Silty Clay	Sandy Loam

The Grand Isle sediment was classified as a sandy loam with particle size distribution values of approximately 16% clay, 13% silt and 71% sand. Cation exchange capacity for Grand Isle was the lowest of all three sediments at 28 centimoles of charge per kilogram of sediment. Moisture content (wet weight basis) was also the lowest of all three sediments at 36%; organic matter follows the same pattern at 1.5%. The Grand Isle sediment represented a marine site with a porewater salinity of 15.5 ppt and hereby will be referred to as the marine sediment.

The redox potential of the marine sediment represented strongly reducing conditions compared to the moderately reducing conditions shown by the redox potential of the freshwater and intermediate

sediments. Lower redox potential of the marine sediment suggests the presence of organic matter that was more microbially available. Due to the low organic matter content of this sediment, a more strongly reducing redox potential may result from recently deposited organic material from natural events. The sediment was collected three months after Hurricanes Gustav and Ike. The proximity of the marine sediment to the coastline increased the possibility that fresh organic matter was recently deposited, perhaps lowering redox potential. In addition, the presence of seawater at the marine site suggests the presence of sulfate-reducing bacteria, which are prevalent in strongly reducing sediments containing sulfate.

### 3.3.2 Redox Potential

The presence of a polymer did have a significant effect on redox potential in the experimental units, specifically for the intermediate and marine sediments (Table 3.4, Appendix B).

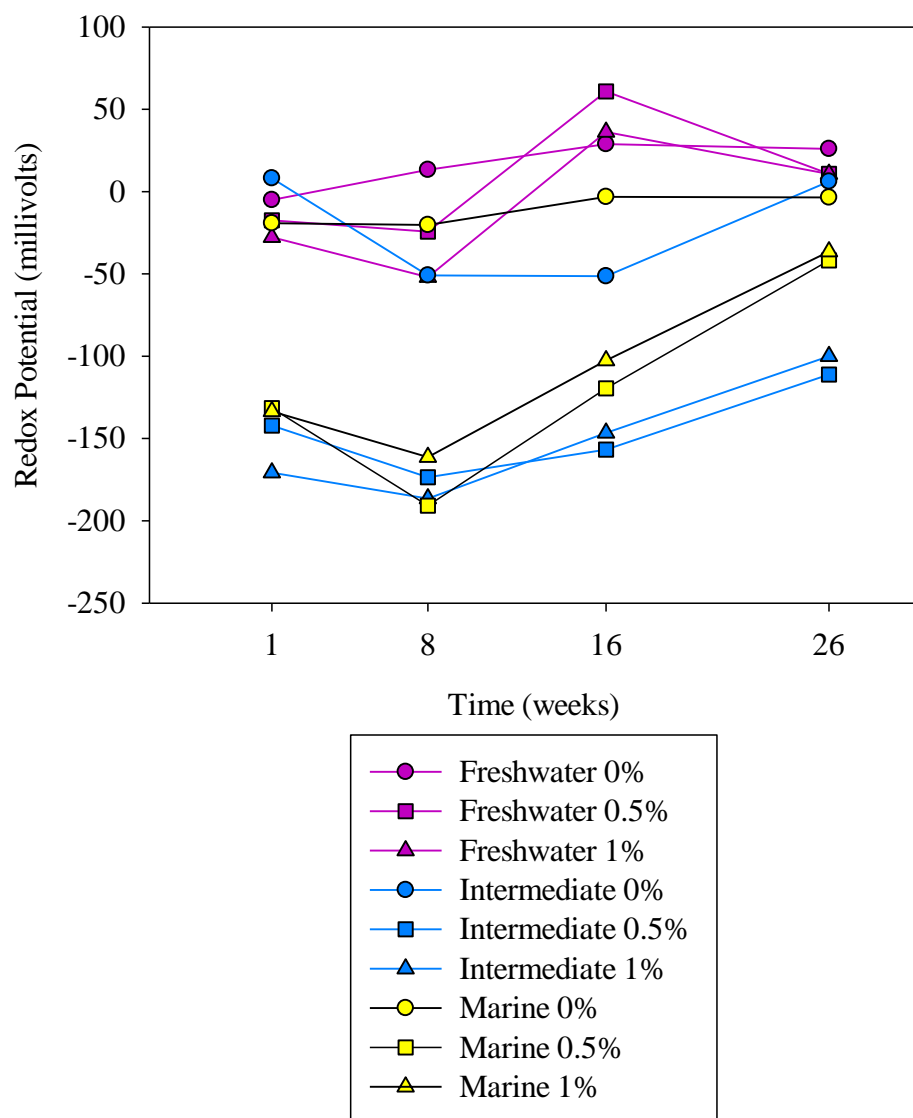
**Table 3.4 Mean redox potential values (millivolts) for the a) Freshwater, b) Intermediate, and c) Marine sediments at weeks 1, 8, 16, and 26. Values for polymer types, the 0.5% and 1% concentration levels, and salinities were averaged due to no significant differences. Letters indicate significant differences between time periods for each sediment. Crosses indicate significant differences between the control samples and samples with polymer.**

a. Freshwater	Week 1	Week 8	Week 16	Week 26
Control	-5 a	13 a	29 a	26 a
Polymer	-23 a	-38 a	49 b	11 ab
b. Intermediate	Week 1	Week 8	Week 16	Week 26
Control	8 a	-51 a	-51 a	6 a
Polymer	-157 ab <sup>+</sup>	-181 b <sup>+</sup>	-152 ab <sup>+</sup>	-106 c <sup>+</sup>
c. Marine	Week 1	Week 8	Week 16	Week 26
Control	-19 a	-20 a	-3 a	-4 a
Polymer	-133 a <sup>+</sup>	-176 a <sup>+</sup>	-112 b <sup>+</sup>	-39 ab

At the 0% concentration level, the redox potential values for all three sediments across time periods were not significantly different. The redox potential values averaged across all time periods for the control samples were as follows: freshwater sediment, 16 mV; intermediate site, -22 mV; and marine sediment, -12 mV. The length of time between sediment collection and experimental setup might

explain the rise in redox potential for the marine sediment compared to the initial sediment characteristic properties.

Redox potential of the freshwater sediment amended with a polymer was not significantly different from redox potential of the control samples (Figure 3.2).



**Figure 3.2 Mean final redox potential values (millivolts) for the Freshwater, Intermediate, and Marine sediments for weeks 1, 8, 16, and 26. Control values for all three sediments were averaged due to no significant differences. Values for polymer types, the 0.5% and 1% concentration levels, and salinities for each sediment have been averaged due to no significant differences. (Control N = 36, Polymer N = 24)**

Averaged across all time periods, redox potential values for the freshwater sediment were approximately 8 mV for the 0.5% level and -8 mV for the 1% level. Polymer did significantly affect

redox potential values for both the intermediate and marine sediments compared to control samples ( $P < 0.0001$ , Table 3.4). The two concentration levels were not significantly different from each other.

For the intermediate sediment, the redox potential values averaged across all time periods were -146 mV for the 0.5% polymer level and -151 mV for the 1% polymer level. These values were significantly lower than the redox potential value of -22 mV for the intermediate control sediment. For the marine sediment, the redox potential values averaged across all time periods were -121 mV for the 0.5% polymer level and -108 mV for the 1% polymer level. These values were significantly lower than the redox potential value of -12 mV for the marine control sediment. In general, the redox potential for the freshwater sediment was significantly higher (i.e. more positive) than the redox potential for the intermediate and marine sites for weeks 1, 8, and 16 ( $P < 0.0001$ , Figure 3.2).

Higher redox potential of the freshwater sediment could be related to the characteristically lower pH of the sediment compared to the other sediments. The redox potential values for the intermediate and marine sediments were not significantly different from each other at any time period. By week 26, the redox potential for the freshwater sediment was significantly higher than the intermediate sediment.

Sediments with and without a polymer treatment tended to have lower redox potential values by week 8 followed by increasing redox potential values for the remaining time (Figure 3.2). For the intermediate and marine sediments, the redox potential values at week 8 were significantly lower than those at week 16 ( $P < 0.0230$ ) and week 26 ( $P < 0.0001$ ). Lower clay content of the intermediate and marine sediments may have allowed polymers to be more available for microbial activity, contributing to increased concentrations of electron donors and lower redox potential.

### **3.3.3 pH**

The presence of a polymer generally had no significant effect on pH compared to control samples, with a few exceptions for the marine sediment (Table 3.5, Appendix B).

**Table 3.5. Mean pH values for the a) Freshwater, b) Intermediate, and c) Marine sediments at weeks 1, 8, 16, and 26. Letters indicate significant differences across time periods and between polymers for each sediment type. Crosses indicate significant differences between the control and a polymer treatment within a time period.**

a. Freshwater	Week 1	Week 8	Week 16	Week 26
0%	6.75 a	7.07 ab	7.04 ab	7.26 b
Xanthan Gum 0.5%	6.09 a	7.13 b	7.16 bc	7.62 c
Xanthan Gum 1%	5.66 a <sup>+</sup>	7.24 b	7.25 b	7.62 b
Guar Gum 0.5%	6.55 a	6.89 b	7.09 bc	7.30 c
Guar Gum 1%	6.04 a	7.05 b	7.20 b	7.47 b
b. Intermediate	Week 1	Week 8	Week 16	Week 26
0%	7.12 a	7.40 a	7.76 ab	7.92 b
Xanthan Gum 0.5%	7.23 a	8.08 b	7.97 b	7.93 b
Xanthan Gum 1%	7.34 a	7.75 b	7.90 b	7.84 b
Guar Gum 0.5%	6.52 c	7.27 d	7.76 d	7.98 d
Guar Gum 1%	6.07 c	7.35 d	7.64 d	7.72 d
c. Marine	Week 1	Week 8	Week 16	Week 26
0%	7.26 a	7.02 a	7.47 a	7.41 a
Xanthan Gum 0.5%	6.98 a	7.80 b <sup>+</sup>	7.37 b	7.78 b
Xanthan Gum 1%	7.27 a	7.60 b	7.70 b	7.54 b
Guar Gum 0.5%	5.95 c <sup>+</sup>	7.30 d	7.49 d	7.17 d
Guar Gum 1%	5.53 c <sup>+</sup>	7.25 d	7.14 d	7.02 d

Since salinity was significant for only one sediment type, the effect was ignored. Therefore, for all pH results presented below, values for different salinity levels have been averaged for each sediment type.

Upon closer analysis, the pH of the freshwater sediment was significantly lower than the pH of the intermediate sediment at all weeks ( $P<0.0001$ , Table 3.6). The pH of the freshwater sediment was also significantly lower than the pH of the marine sediment at weeks 1, 8, and 16 ( $P<0.0001$ , Table 3.5). By week 26, pH was not significantly different. The pH of the intermediate sediment was not significantly different from that of the marine sediment at week 1 or week 8; however, the pH of the intermediate sediment was significantly higher than the pH of the marine sediment at week 16 ( $P=0.0017$ ) and week 26 ( $P<0.0001$ ).

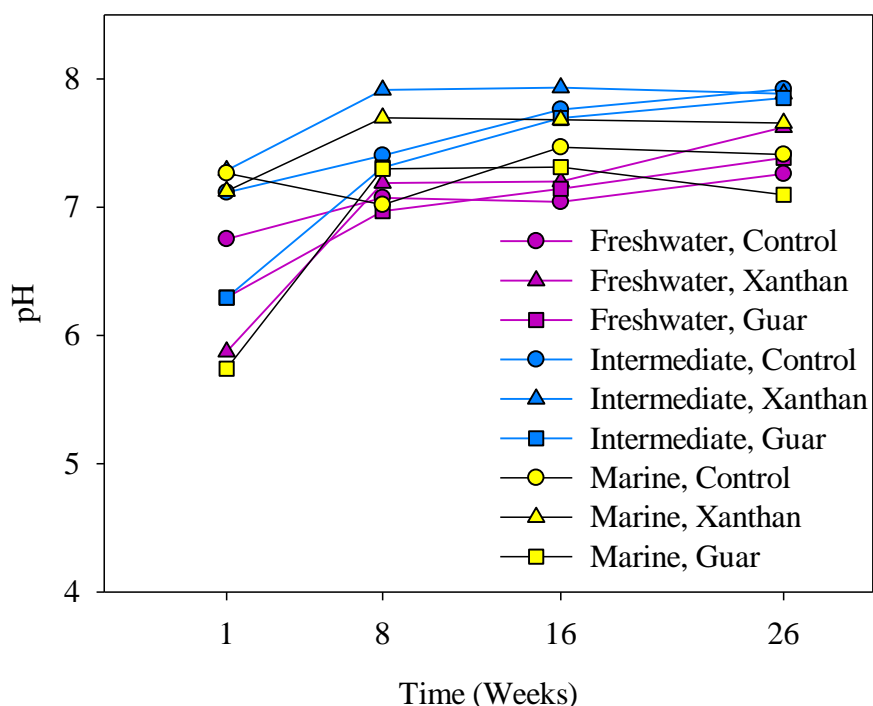
For the freshwater sediment, only one polymer treatment was significantly different. At week one, the sediment with the 1% xanthan gum treatment had a pH of 5.85, which was significantly lower ( $P<0.0001$ ) than any other sample, possibly due to dissociation of hydrogen ions from the carboxylic acid functional groups of xanthan gum. In addition, the freshwater sediment has less buffering capacity for changes in pH due to the low inorganic carbon content of freshwater aquatic systems. For all other freshwater sediment samples, the control, 0.5%, and 1% concentration levels showed no significant differences for any time period.

**Table 3.6 Mean pH values for the Freshwater, Intermediate, and Marine sediments at weeks 1, 8, 16, and 26. Values for polymer types, concentration levels, and salinities have been averaged due to no significant differences. Letters indicate significant differences between sediments at each week.**

Week	Freshwater	Intermediate	Marine
1	6.22 a	6.86 b	6.60 b
8	7.08 a	7.57 b	7.40 b
16	7.15 a	7.80 b	7.49 c
26	7.46 a	7.88 b	7.38 a

The pH of the samples with and without polymer at week 26 was significantly higher than at week 1. With all concentration levels combined, the average pH of the freshwater sediment increased over time (Table 3.6). The pH of the control samples and the samples with polymer increased over time; however, the pHs of the control samples and samples with polymer were not significantly different (Figure 3.3).

For the intermediate sediment, the control, 0.5%, and 1% concentration levels showed no significant differences for any time period. The pH of the samples with and without polymer at week 26 was significantly higher than at week 1. With all concentration levels combined, the average pH of the intermediate sediment increased over time. The pH of the control samples and the samples with polymer increased over time; however, the pHs of the control samples and samples with polymer were not significantly different.



**Figure 3.3. Mean final pH values for the Freshwater, Intermediate, and Marine sediments at weeks 1, 8, 16, and 26 for the control, xanthan gum, and guar gum treatments. Values for the 0.5% and 1% concentration levels and salinities have been averaged due to no significant differences.**

For the marine sediment, any differences between the control samples and samples with polymer occurred in the first 8 weeks. After week 1, the pH of samples with polymer was significantly lower than the pH of the control samples. After week 8, the pH of samples with polymer was significantly higher than the pH of the control samples. Beyond week 8, the pH of the control, 0.5%, and 1% concentration levels showed no significant differences. The pH of the control samples did not change significantly from week to week. With all concentration levels combined, the average pH of the marine sediment increased over time.

In general, for each sediment type, the pH increased from week 1 to week 8 and then stabilized over the rest of the time period. All pH values were lower than typical seawater pH values of 7.8-8.3 (Garrison 2010). The presence of a polymer had no significant difference on the pH compared to the control samples, with a few exceptions for the marine sediment. For the freshwater sediment, the pH values ranged from 5.66 – 7.62. For the intermediate sediment, the pH values ranged from 6.07 – 8.08.

For the marine sediment, the pH values ranged from 5.53 – 7.80. The initial decrease in pH can be explained by a flush in microbial activity resulting from agitation of sediment and polymer, which served as a readily available energy source. As time progressed, microbial activity and carbon dioxide production decreased, thereby reducing carbonic acid formation and raising pH.

### 3.3.4 Microbial Biomass

The presence of a polymer had no significant effect on the pool of microbial biomass carbon (MBC) (Appendix B). The freshwater sediment had the highest amount of MBC with an average value of 12.8 g C kg<sup>-1</sup> dry sediment (Table 3.7).

**Table 3.7 Mean Microbial Biomass Carbon values (g C kg<sup>-1</sup> dry sediment) for the Freshwater, Intermediate, and Marine sediments for the 0%, 0.5%, and 1% concentration levels. Values for the polymer types and salinities have been averaged due to no significant differences. The last column gives the overall average amount of MBC for each sediment at weeks 1, 8, 16, and 26. Letters indicate significant differences between weeks within a sediment type.**

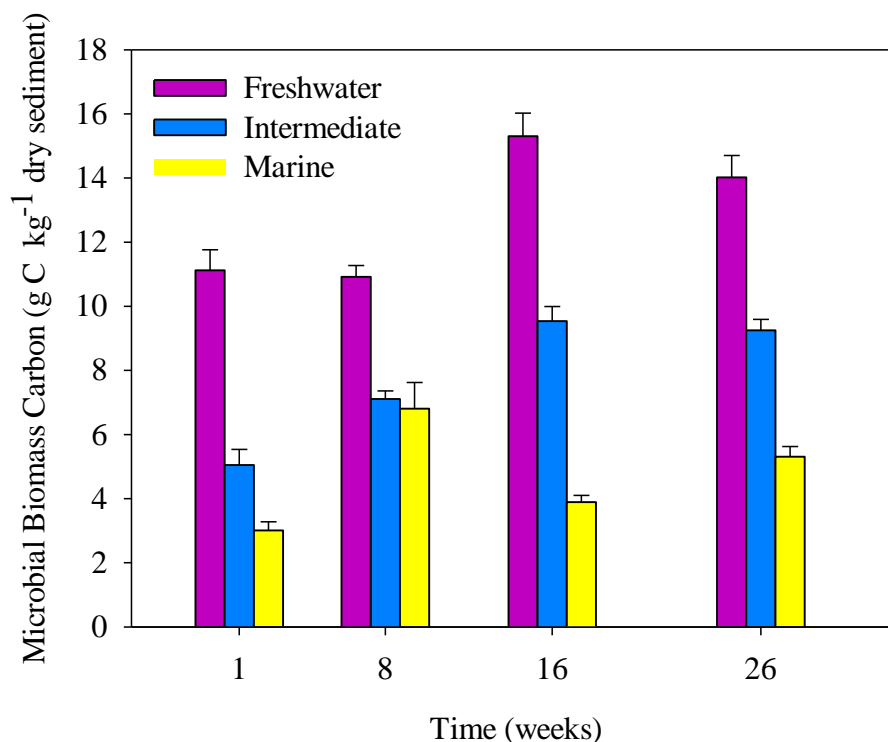
Sediment	Week	Min	Max	0%	0.5%	1%	Overall Mean ± Std Err
Freshwater	1	1.06	18.5	11.3	12.2	9.95	11.1 ± 0.64 a
	8	6.86	16.8	11.4	10.8	10.6	10.9 ± 0.35 a
	16	6.77	22.0	15.1	15.8	15.0	15.3 ± 0.72 b
	26	8.48	26.1	13.5	14.6	13.9	14.0 ± 0.68 b
Intermediate	1	0.00	10.4	6.08	5.41	3.66	5.05 ± 0.49 a
	8	4.99	11.0	6.98	7.16	7.21	7.11 ± 0.25 ab
	16	4.48	14.5	10.1	9.66	8.87	9.54 ± 0.45 b
	26	5.88	14.0	9.05	9.10	9.59	9.25 ± 0.34 b
Marine	1	0.00	6.74	2.97	2.87	3.17	3.01 ± 0.27 a
	8	2.23	16.3	6.70	6.75	6.99	6.81 ± 0.81 b
	16	1.72	6.97	3.57	3.98	4.14	3.89 ± 0.21 a
	26	1.84	9.36	5.34	5.57	5.03	5.31 ± 0.31 ab

The intermediate sediment had an intermediate amount of MBC with an average value of 7.74 g C kg<sup>-1</sup> dry sediment. The marine sediment had the lowest amount of MBC with an average value of 4.76 g C kg<sup>-1</sup> dry sediment.

For the freshwater sediment, MBC increased over time. From week 1 to week 8, MBC was not significantly different. By week 16, the MBC significantly increased ( $P < 0.0001$ ). The MBC at week 26



was not significantly different from week 16; however, it was significantly higher than the MBC at week 1 ( $P=0.0143$ ) and week 8 ( $P=0.0057$ ). For the intermediate sediment, MBC also increased over time. From week 1 to week 8, MBC was not significantly different. As in the freshwater sediment, the MBC significantly increased by week 16 ( $P<0.0001$ ). The MBC at week 26 was not significantly different from week 16; however, it was significantly higher than the MBC at week 1 ( $P<0.0001$ ). For the marine sediment, MBC peaked in week 8, decreased by week 16, and increased again by week 26. The MBC at week 8 was significantly higher than the MBC at week 1 ( $P=0.0001$ ) and week 16 ( $P=0.0133$ ). By week 26, the MBC was not significantly different than the MBC at week 1, 8, or 16 (Table 3.7, Figure 3.4).



**Figure 3.4 Microbial biomass carbon (g C kg<sup>-1</sup> dry sediment) for the Freshwater, Intermediate, and Marine sediments at weeks 1, 8, 16, and 26. Values for polymer type, concentration level, and salinity have been averaged due to no significant differences. (N=36)**

Even though the polymer did not significantly affect MBC, the size of the MBC pool increased over time. The amount of MBC correlated with organic matter content for each sediment type. For example, the freshwater sediment had the highest MBC and the highest organic matter content whereas

the marine sediment had the lowest MBC and the lowest organic matter content. The microbial community metabolized additional carbon and assimilated it into the biomass pool (Figure 3.4).

Overall, the presence of a polymer had no significant effect on the ratio of MBC to organic carbon for each sediment type (Table 3.8, Appendix B). The ratio for the marine sediment was significantly higher than the ratios for the freshwater and intermediate sediments at weeks 1, 8, and 16. The ratios for the freshwater and intermediate sediments were not significantly different from each other. Higher ratios for the marine sediment indicate that more carbon was available for microbial processing (Anderson 2003). The microbial community responded by assimilating carbon and increasing biomass. Possibly, higher ratios for the marine sediment may be further evidence of freshly deposited organic matter from hurricanes.

**Table 3.8 Mean  $C_{mic}:C_{org}$  ratio (%) for the Freshwater, Intermediate, and Marine sediments at the 0%, 0.5%, and 1% levels at weeks 1, 8, and 16. Values for polymer types and salinities have been averaged due to no significant differences. Letters indicate significant differences between weeks.**

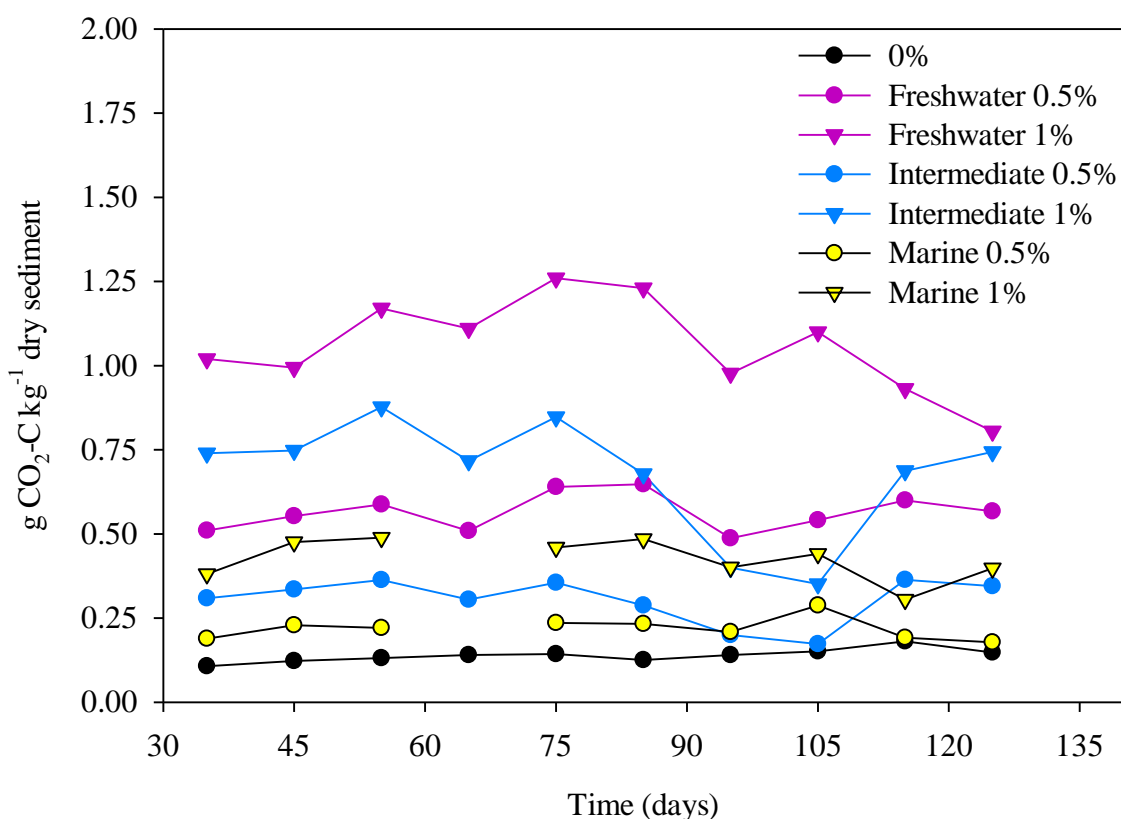
Sediment	Polymer Concentration	Week 1	Week 8	Week 16
Freshwater	0%	0.816 a	0.823 a	1.09 a
	0.5%	0.880 a	0.782 a	1.15 a
	1%	0.721 a	0.769 a	1.09 a
Intermediate	0%	0.729 a	0.837 a	1.21 a
	0.5%	0.649 a	0.858 a	1.16 a
	1%	0.439 a	0.864 a	1.06 a
Marine	0%	1.91 b	4.30 c	2.29 b
	0.5%	1.84 b	4.33 c	2.55 b
	1%	2.04 b	4.48 c	2.65 b

### 3.3.5 Microbial Basal Respiration

The 1% polymer treatment did have a significant effect on basal respiration rates (Appendix B). In general, the freshwater sediment had the highest respiration rates, followed by the intermediate and the marine sediments. For all control samples, respiration was not significantly different among the three sediments. The freshwater sediment respiration rates for the control samples were highest with a

total cumulative respiration of 25.8 g CO<sub>2</sub>-C kg<sup>-1</sup> dry sediment. The control samples for the intermediate sediment had a total cumulative respiration of 12.0 g CO<sub>2</sub>-C kg<sup>-1</sup> dry sediment. The control samples for the marine sediment had the lowest total cumulative respiration of 4.33 g CO<sub>2</sub>-C kg<sup>-1</sup> dry sediment. Respiration for control samples resulted from organic carbon already present in the sediment and was not associated with the polymers.

For all three sediments, the microbial community in samples with 1% polymer respired significantly more than the control samples ( $P<0.0001$ , Figure 3.5).



**Figure 3.5** Cumulative respiration curves for the Freshwater, Intermediate, and Marine sediments at the 0%, 0.5%, and 1% concentration levels. The 0% concentration values were averaged for all sediments due to no significant differences. Values for polymer types and salinities have been averaged due to no significant differences. (Control N=36, Polymer N=12)

For the freshwater sediment, the amount of respiration for control samples was also significantly less than the samples with 0.5% polymer only on days 75 ( $P=0.0408$ ) and 85 ( $P=0.0402$ ). For the

intermediate and marine sediments, the amount of respiration from the 0.5% polymer was not significantly different than the control samples.

For the freshwater and intermediate sediments, the samples with 1% polymer had significantly more respiration than the samples with 0.5% polymer ( $P<0.0001$ ). For the marine sediment, the 1% polymer treatment induced respiration nearly significant from the 0.5% polymer treatment ( $P=0.0550$ ). At each concentration level, the microbes in the freshwater sediment respired the most, followed by the intermediate sediment, and then by the marine sediment.

At the 0.5% concentration level, the microbes in the freshwater sediment respired significantly more than microbes in the intermediate ( $P=0.0037$ ) and marine sediments ( $P<0.0001$ ). At the 1% concentration level, the respiration for all three sediments was significantly different from each other ( $P<0.0045$ ). Due to differences in soil moisture content, the freshwater sediment received the most carbon from the polymer addition, followed by the intermediate sediment and the marine sediment (Table 3.9).

**Table 3.9 Amount of carbon added from the polymer and the cumulative amount of carbon respired after 125 days for the Freshwater, Intermediate, and Marine sediments at the 0%, 0.5%, and 1% concentration levels. Values for polymer types and salinities have been averaged due to no significant differences.**

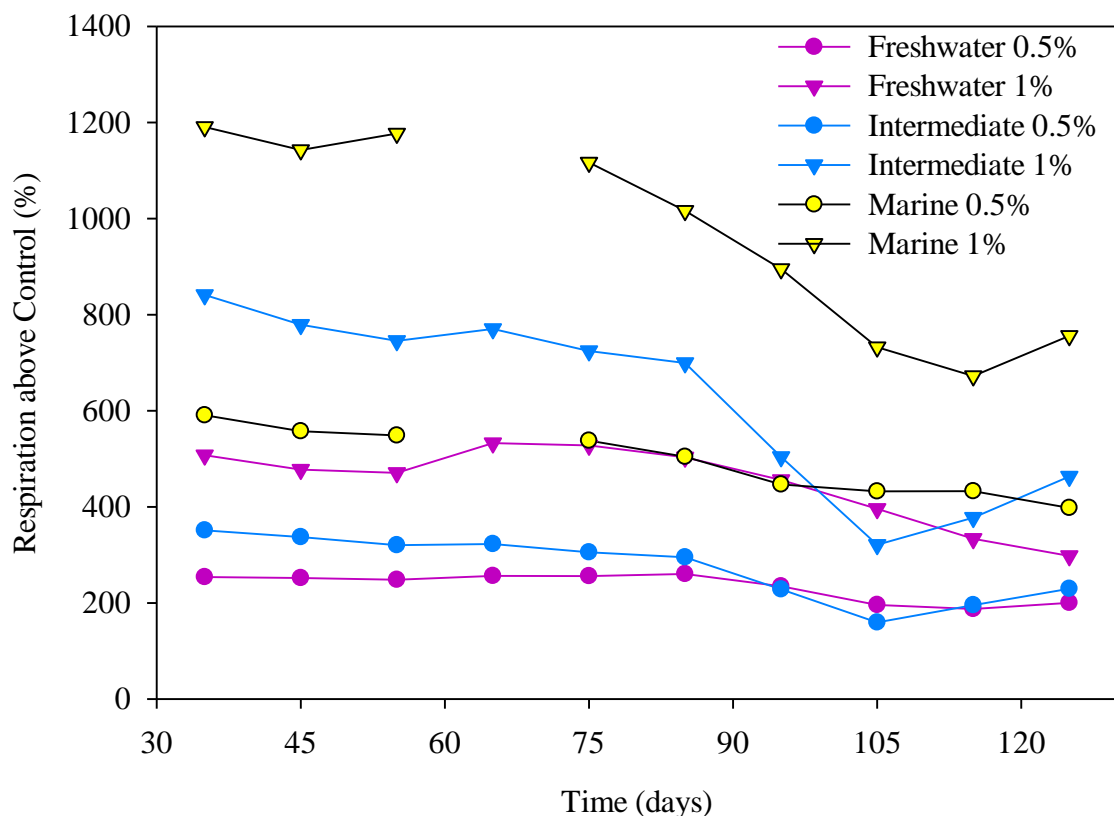
Sediment	Concentration of Polymer (%)	Added C from Polymer (g C kg <sup>-1</sup> dry sediment)	50% Respiration Rule (g C kg <sup>-1</sup> dry sediment)	Cumulative Area Under the Curve (g C kg <sup>-1</sup> dry sediment )	Respiration above Control (g C kg <sup>-1</sup> dry sediment)	Respiration above Control (%)
Freshwater	0	-	-	25.8	-	-
	0.5	9.49	4.75	60	34.2	233
	1	19	9.5	115	89.2	446
Intermediate	0	-	-	12	-	-
	0.5	7.28	3.64	32.5	20.5	271
	1	14.6	7.30	73.4	61.4	612
Marine	0	-	-	4.33	-	-
	0.5	3.76	1.88	21.2	16.9	490
	1	7.51	3.76	41.1	36.8	949

A commonly accepted rule for microbial respiration states that nearly 50% mineralization of a substrate indicates complete degradation of that substrate (Shen and Bartha 1996). At the 0.5% and 1% polymer concentration levels, the microbial consortia in all three sediments completely degraded the polymer (Table 3.9). At the 0.5% polymer concentration level, the freshwater sediment had a cumulative respiration of  $34.2 \text{ g C kg}^{-1}$  dry sediment above the control, which was approximately 7 times more carbon than for complete degradation of the added polymer. The intermediate sediment had a cumulative respiration of  $20.5 \text{ g C kg}^{-1}$  dry sediment above the control, which was about 2.8 times the amount needed for complete degradation. The marine sediment had a cumulative respiration of  $16.9 \text{ g C kg}^{-1}$  dry sediment above the control, which is 4.5 times the amount required for complete degradation of the polymer. At the 1% polymer concentration level, microbes in all three sediments respired more carbon than needed for polymer degradation in the amounts of 9 times, 8.4 times, and 10 times more carbon for the freshwater, intermediate, and marine sediments, respectively. Results suggest a priming effect in which the polymer stimulates the microbes to metabolize carbon from microbial biomass turnover and soil organic carbon.

By subtracting the amount of respiration for the control samples from the amount of respiration for the samples with polymer, the respiration of materials in addition to soil organic carbon can be determined. The respiration for the samples with polymer was much higher than the control samples (Figure 3.6). For each sediment type, the cumulative amount of respiration for the samples at the 1% level was about 2 times higher than the amount of respiration for the samples at the 0.5% level. Relative to the control samples, the microbes in the marine sediment respired the most, followed by the intermediate sediment, then by the freshwater sediment.

The marine sediment had the greatest respiration above the control, which could be related to its low clay content and reducing redox potential, possibly due to fresher, more available organic matter. The polymer provided a source of carbon that was highly available to microbes. The

characteristics of the marine sediment stimulate microbial processing of the substrate, which corresponds to the greatest respiration above the control.



**Figure 3.6 Percent respiration over the control samples for the Freshwater, Intermediate, and Marine sediments at the 0.5% and 1% concentration levels. Values for polymer types and salinities have been averaged due to no significant differences.**

The 1% polymer treatment resulted in a metabolic quotient (i.e ratio of basal respiration to MBC) significantly higher than that of control samples and samples with 0.5% polymer (Table 3.10).

For the control and 0.5% polymer treatment samples, the metabolic quotient was not significantly different for any of the three sediments at weeks 1, 8, or 16 (Appendix B). The metabolic quotient at the 1% level at week 1 increased: marine sediment < intermediate sediment < freshwater sediment. By week 8, the metabolic quotients for all sediment and concentration combinations were not significantly different. High metabolic quotients for the samples with the 1% polymer treatment at week 1 indicate that the microbial community responded quickly to the added carbon input by an increase in microbial biomass.

**Table 3.10 Mean metabolic quotient ( $\mu\text{g CO}_2\text{-C g}^{-1} \text{C}_{\text{mic}}$ ) for the Freshwater, Intermediate, and Marine sediments at the 0%, 0.5%, and 1% levels for weeks 1, 8, and 16. Values for polymer types and salinities have been averaged due to no significant differences. Letters indicate significant differences between weeks.**

Sediment	Polymer Concentration	Week 1	Week 8	Week 16
Freshwater	0%	0.550 a	0.148 a	0.401 a
	0.5%	1.32 a	0.289 a	0.619 a
	1%	4.20 bc	1.64 a	1.10 a
Intermediate	0%	0.447 a	0.166 a	0.230 a
	0.5%	1.67 a	0.420 a	0.425 a
	1%	6.47 bc	1.98 a	0.881 a
Marine	0%	0.400 a	0.125 a	0.122 a
	0.5%	3.14 ab	0.237 a	0.239 a
	1%	3.65 b	0.382 a	0.507 a

### 3.4 Discussion

For all sediments, the polymer induced an environmental change that elicited varied responses from the microbial communities. Polymer addition immediately increased microbial activity as evidenced by lower redox potential, lower pH, and higher metabolic quotients after one week.

Changes in redox potential and pH affect microbial community composition and provide insight into changes in the microbial community. After polymer addition and flooding, a more reducing redox potential indicates an increased availability of organic matter to serve as electron donors for microbial use (Reddy and DeLaune 2008). Microbes convert some of the organic material to carbon dioxide, which combines with soil moisture and forms carbonic acid, thus lowering the pH (Reddy and DeLaune 2008). In this study, as microbial activity stabilized over time, less respiration suggests less carbon dioxide production, which would raise pH.

Immediately high respiration rates suggest a microbial response from r-strategist species, which typically respond to substrate inundation with increased activity and growth, followed by rapid turnover of the biomass. Microbial biomass turnover may contribute to increasing biomass pools over time. Maintenance of reduced redox potentials indicates microbial metabolism, which suggests that

even though the polymer was metabolized, soil microbes were still actively mineralizing soil organic carbon, biomass turnover, and microbial by-products.

The comparison between the amount of carbon added from a substrate and the amount of carbon recovered in the microbial biomass provides information about the microbial community. At the 0.5% polymer level, added carbon from the polymer was fully recovered in the microbial biomass but not reflected in respiration differences from control samples. Such a relationship represents a passive microbial community that increased its carbon storage. The 1% polymer level induced a response visible in respiration rates, which represents a transition to an actively growing microbial biomass.

The cumulative amount of respiration and percent respiration over controls indicate that more carbon was metabolized than was added from the polymer. In the first week, metabolic quotients for the samples with 1% polymer suggest that microbial communities initially respired at a very high rate given the amount of microbial biomass. The physiologically active component of the microbial biomass in the sediments responded to the polymer with increased respiration, which again suggests r-strategist species. Even after the polymer was metabolized, microbial communities still respired at a very high rate; however, the metabolic quotients decreased with time, which indicates an increase in the size of the microbial biomass.

After the polymer was metabolized, the microbial communities metabolized carbon from other sources, possibly including carbon in the soil organic matter fraction. For both the freshwater and intermediate sediments, microbial biomass increase was sustained over 16 weeks, possibly because those sediments have higher soil organic matter content. In addition, the cumulative amount of respiration was highest for the freshwater sediment and intermediate for the intermediate sediment, which corresponds with patterns in organic matter content.



In the marine sediment, the biomass reached a peak by week 8 but then decreased for the rest of the study, possibly due to low organic matter content of the sediment. Beyond 8 weeks, little available organic carbon remained in the soil for microbes to metabolize. Furthermore, the cumulative amount of respiration was lowest for the marine sediment, which relates to having the lowest organic matter content.

Over time, microbial communities in the sediments switched from metabolizing added carbon to assimilating carbon into the biomass. For week 8 and week 16, metabolic quotients for the control samples and samples with polymer were not statistically different. In addition, microbial biomass for the control samples and samples with polymer was not statistically different at these times. The data provide evidence that the polymer was completely metabolized after one week, and for the rest of the study, microbial communities mineralized soil organic carbon and microbial turnover products, which both contributed to the increase in biomass.

### **3.5 Conclusion**

At the microscopic level in the soil, relatively high levels of polymer addition cause an increase in the active component of the biomass. At low levels of substrate addition, microbes in the passive component of the biomass assimilate carbon from the polymer into storage compounds. When respiration greatly increases in relation to biomass, microbes increasingly incorporate the substrate into structural compounds, which means that the physiologically active component of the biomass is responding and growing.

In all three sediments, microbial respiration in sediments with a polymer treatment is above that of the control samples in addition to being above the amount of respiration required to indicate complete mineralization of the polymer. Two possible explanations to justify the increase in respiration include mineralization of soil organic carbon or mineralization of additionally secreted microbial by-products and biomass turnover. In either case, metabolism of these materials removes organic material

from the sediment that might otherwise lead to aggregation of soil particles. The use of natural polymers to stabilize hydraulically dredged sediments may, in fact, be stimulating the growth of the microbial community, which inherently leads to increased mineralization of organic matter and release of carbon dioxide. While the addition of natural polymers does not directly harm microbial communities in sediments, they may be decomposed quickly and thus preventing any natural processes of aggregation.

## **CHAPTER 4: OVERALL PROJECT CONCLUSION**

### **4.1 Relationship between Physical and Microbial Properties of Sediments**

The rationale for this study was to investigate the influence of two natural polymers on physical properties and microbial properties of hydraulically dredged sediments. The goal was to increase particle aggregation and stabilize dredged material used for marsh restoration. In addition, any adverse impacts on microbial activity from the polymer amendments needed to be investigated.

In wetland soils and sediments, soil moisture increases nutrient availability to microbes. Microbes also become more motile and disperse easier in wet environments. As microbial motility increases, interactions with organic carbon substrates and metabolic activity increase, which results in the release of extracellular polymeric substances (EPS). Over time, these substances combine with other soil particles, nearby organic compounds, and microbial complexes to create a three-dimensional matrix that accumulates and increases sediment stability. Biotic processes that disrupt bonds between particles and organic matter cause destabilization; however, disruption also alters the exposure of organic substrates to microbes. Once again, metabolism increases and leads to the production and release of EPS. Continuous cycles of stabilization and destabilization alter soil structure and produce biofilms that increase soil aggregates. This study looked at taking these relationships on the microbial scale and applying them on a large scale to increase wetland sediment stability on a marsh restoration site with dredged material for a period of time until plants become established.

To further understand the influence of biopolymers on sediment stability, an engineering study completed by another student and faculty member investigated the interactions between biopolymer solutions (i.e. xanthan gum and guar gum) and research-grade kaolinitic clay particles (Nugent et al. 2009).

Liquid limit describes the amount of water required for a clay material to flow as a liquid. At higher biopolymer concentrations in pore fluid, the liquid limit of kaolinite increased as solution

viscosity increased. High polymer concentrations, such as 5% xanthan gum and 2% guar gum, increased liquid limit and viscosity. At lower concentrations, polymer solutions induced aggregation of clay particles, which actually decreased the liquid limit of kaolinite (Nugent et al. 2009). Aggregation of clay particles reduced the amount of surface area available for water absorption, which inherently decreased the liquid limit since less water was required to fully saturate the aggregate.

Due to the limited cation exchange capacity of kaolinite, xanthan gum induced more aggregation than guar gum. The absence of charge on guar gum allows it to form hydrogen bonds with clay particles, forming a highly linked clay-polymer network. More xanthan gum was required to achieve the same effect since its interaction with kaolinite is limited due to the net negative charge on both the polymer and the clay. Therefore, the liquid limit for a lower concentration of guar gum (i.e. 3%) was similar to the liquid limit for a higher concentration of xanthan gum (i.e. 10%) (Nugent et al. 2009).

The addition of cross-linking agents, such as monovalent or divalent cations, influenced liquid limits in different ways (Nugent et al. 2009). For example, when calcium was added to xanthan gum solutions, rigid cross-links occurred due to the strong affinity between negatively charged functional groups and positively charged ions. At low polymer concentrations, cross-linking between xanthan gum and calcium cations was not enough to raise the liquid limit and overcome the weakness of the electrical double layer in the clay from increased concentration of solutes. At higher concentrations, however, cross-linking of xanthan gum with cations increased viscosity and the liquid limit of clays to combat any negative effects of increased aggregation.

Sodium, another cross-linking agent, is a well-hydrated cation, thereby strongly absorbing to clay particle surfaces in preference to polymer molecules. Potassium, a poorly-hydrated cation, disturbs the layer of water around clay particles, which allowed guar gum to penetrate further and form

hydrogen bonds with clay particles. The increased adsorption of guar gum led to aggregation, which decreased liquid limit as described before.

In general, since undrained shear strength of a soil depends on moisture content, the polymers' effect of increasing the liquid limit of clays should inherently increase the undrained shear strength of an amended soil.

The current experiment focused on the importance of the relationship between physical properties of sediments and microbial activity in wetland sediments and how these properties affect the success of polymers in natural sediments. In general, physical properties of soils such as particle size affect the orientation of polymeric materials, which is one factor determining ease of microbial access to substrates. High content of hydrolysable carbon in the added substrates stimulated microbial activity to the point that polymers were entirely metabolized and removed from the soil, along with soil organic carbon. A closer look at experimental variables emphasizes the relationship between physical and microbial properties.

## **4.2 Principal Components Analysis**

Principal components analysis takes highly correlated response variables and groups them into new sets of uncorrelated variables known as principal components. Principal components analysis using a rotation method in SAS (2009) explained which response variables contributed the most variation to the data.

### **4.2.1 Principal Components Analysis on Entire Data Set**

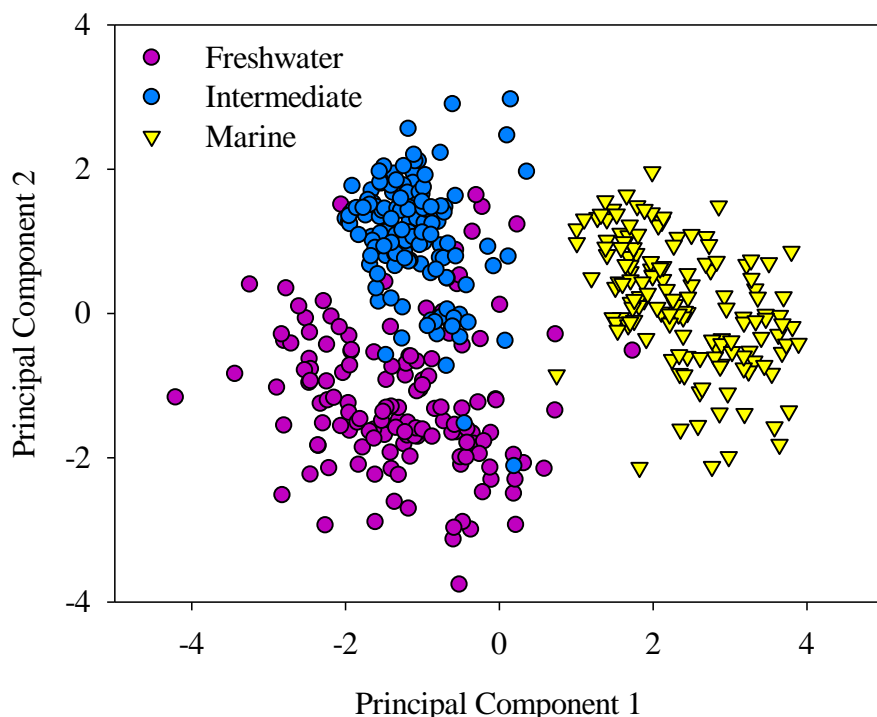
Principal components analysis of the entire experimental data set separated the three sediments based on physical properties. For example, the first principal components analysis looked for groupings based on pH, redox potential, microbial biomass, moisture content, total consolidation, amount of dewatering, percent volume of clay, percent volume of silt, percent volume of sand, and mean aggregate diameter (Table 4.1).

**Table 4.1 Principal Component Loadings for First Principal Components Analysis. The last row gives the percent of variation for which each principal component contributes to the experimental dataset.**

Response Variable	Principal Component 1	Principal Component 2
pH	-0.028	-0.226
Redox	0.066	0.130
Microbial Biomass	0.811	0.211
Moisture Content	0.877	-0.322
Total Consolidation	0.060	-0.087
Dewatering	-0.770	0.211
% Clay	0.683	-0.299
% Silt	0.517	-0.830
% Sand	-0.556	0.809
Mean Agg. Diameter	0.186	0.910
% of Variation	41	20

If the absolute value of a principal component loading [PC loading] was greater than 0.5, the corresponding response variable was considered significant in determining the principal component. For example, in principal component 1, the following variables had absolute values of PC loadings higher than 0.5: microbial biomass, moisture content, dewatering, percent clay, percent silt, and percent sand. The sign on the significant PC loadings helped further delineate groups in the analysis. For example, microbial biomass, moisture content, and percent clay have highly positive PC loadings. These results are congruent with previous statistical analyses showing that the freshwater sediment had the highest microbial biomass, highest moisture content, and highest percent clay.

In addition, the positive PC loading for moisture content and negative PC loading for dewatering supports the previous conclusion that sediments with higher moisture content exhibited less dewatering. Finally, the positive PC loading for percent silt and negative PC loading for percent sand show that as the volume of an aggregate fills more of the silt fraction, it inherently fills less of the sand fraction. Thus, principal component 1 appears to group sediments based on the amount of sample in the fine particle fraction (Figure 4.1); this component explains approximately 41% of the variation in the experimental dataset.



**Figure 4.1 Results of First Principal Components Analysis completed on wet sediment samples.**

Analysis of PC loadings for the second principal component emphasized percent silt, percent sand, and mean aggregate diameter as significantly contributing factors. As percent sand content and mean aggregate diameter increase, percent silt content decreases, which supports previous conclusions. As a greater proportion of a given sediment sample enters the sand fraction, pore space of the aggregate increases, giving an overall larger aggregate diameter. Similarly, a greater proportion of a sample in the sand fraction means that less of the sample is available for the silt fraction. The second principal component explains an additional 20% of the variation in the experimental data.

Together, the first and second principal components explain 60% of the variation in the experimental data set. Both principal components from this analysis group the sediments based on grain size. From left to right along principal component 1, samples have less fine particles (Figure 4.1). From bottom to top along principal component 2, samples have more sand particles and greater aggregate diameter.

#### 4.2.2 Principal Components Analysis on Ecophysiological Indices

Principal components analysis of the data set for which ecophysiological indices were calculated showed the separation of the three sediments based on physical properties and microbial activity. The principal components analysis looked for groupings based on pH, redox potential, microbial biomass, the  $C_{mic}:C_{org}$  ratio, basal respiration rate over specified 10-day periods, metabolic quotient  $qCO_2$ , moisture content, amount of dewatering, total consolidation, percent volume of clay, percent volume of silt, percent volume of sand, and mean aggregate diameter (Table 4.2).

**Table 4.2 Principal Component Loadings for Principal Components Analysis on Ecophysiological Indices. The last row gives the percent of variation for which each principal component contributes.**

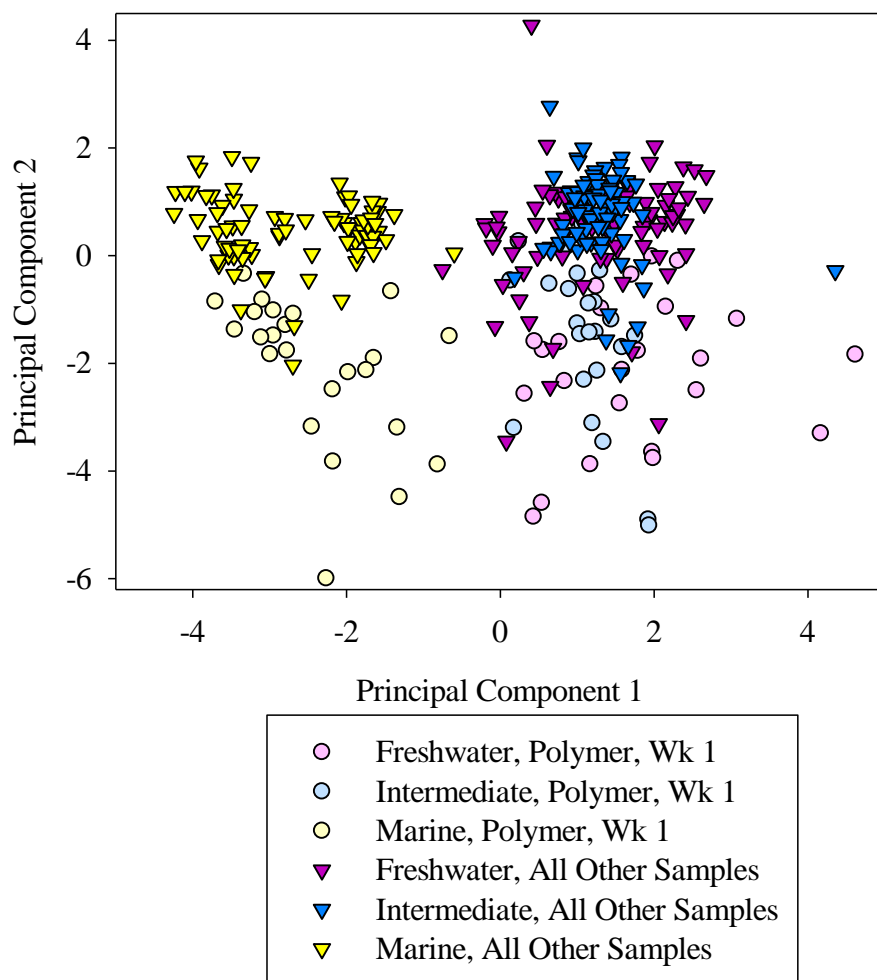
Response Variable	Principal Component 1	Principal Component 2	Principal Component 3
pH	-0.131	-0.374	-0.624
Redox Potential	0.036	0.745	-0.096
Microbial Biomass	-0.213	0.667	-0.133
$C_{mic}:C_{org}$	0.661	-0.066	-0.280
Basal Respiration	-0.207	-0.025	0.787
$qCO_2$	-0.119	-0.298	0.839
Moisture Content	-0.804	0.392	0.225
Dewatering	0.559	-0.070	-0.291
Total Consolidation	0.027	0.426	0.248
% Clay	-0.676	0.600	-0.009
% Silt	-0.974	-0.005	-0.051
% Sand	0.977	-0.060	0.047
Mean Aggregate Diameter	0.744	0.336	0.171
% of Variation	36	15	15

For principal component 1, the following response variables had absolute values of PC loadings greater than 0.5:  $C_{mic}:C_{org}$  ratio, moisture content, dewatering, percent clay, percent silt, percent sand, and mean aggregate diameter (Table 4.2). The trends indicated by the negative signs agree with conclusions from previous chapters. For example, as clay and silt decrease, the amount of sand and the mean aggregate diameter increase. In addition, as dewatering increases, moisture content of the



sediment decreases. The  $C_{mic}:C_{org}$  ratio is inversely related to moisture content and clay content, which is plausible because sediments with high moisture content and high clay content have a greater amount of organic matter than sediments with low moisture content and low clay content. Therefore, the  $C_{mic}:C_{org}$  ratio will be lower because there is more organic matter relative to the amount of microbial biomass.

In this experiment, the freshwater sediment had the highest moisture content, highest clay content, and lowest  $C_{mic}:C_{org}$  ratio, which is portrayed along the first principal component axis (Figure 4.2).



**Figure 4.2 Results of Principal Components Analysis on Ecophysiological Indices completed on wet sediment samples. Polymer indicates the 0.5% and 1% levels for week 1. All other samples indicates the 0% concentration level for week 1, and the 0%, 0.5%, and 1% levels for weeks 8 and 16.**

Therefore, the first principal component appears to group sediments based on particle size relative to organic matter content. From left to right, the particles become finer, which allows for more organic matter for the microbial community resulting in a decreasing  $C_{mic}:C_{org}$  ratio (Figure 4.2). Principal component 1 accounts for 36% of the variability.

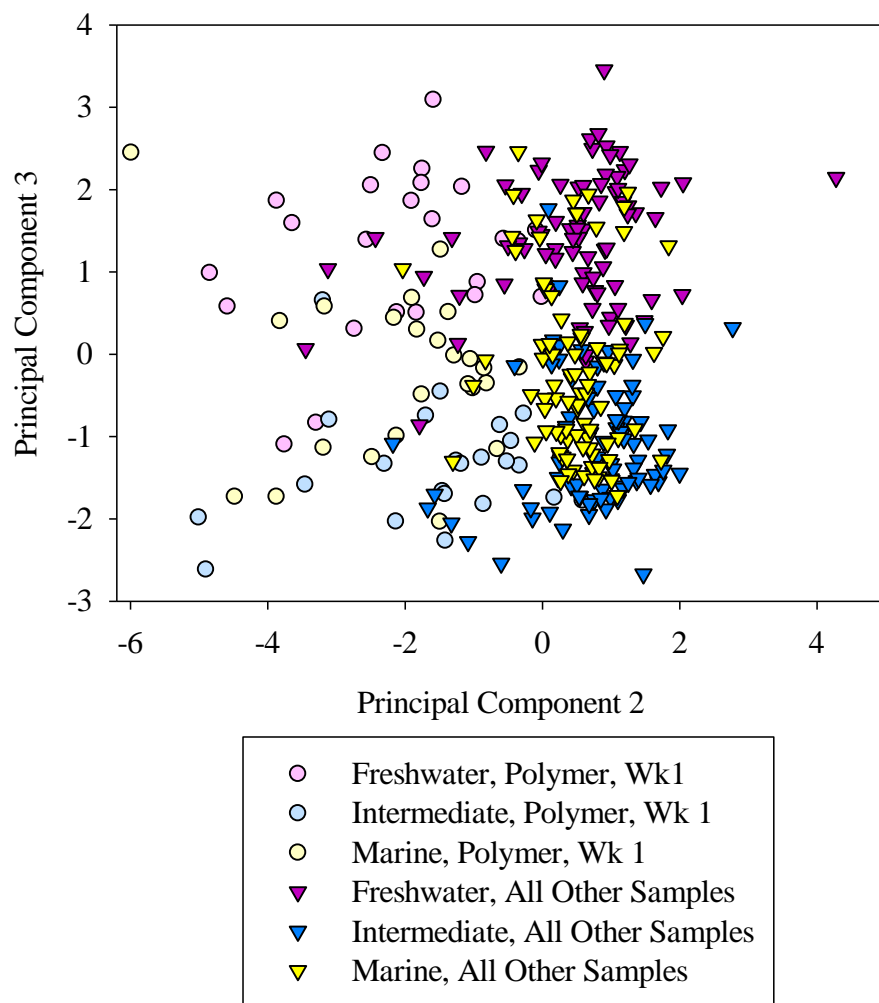
Analysis of PC loadings for the second principal component emphasized redox potential, microbial biomass, and percent clay as contributing factors to determining the component. All three of these response variables are positively related to each other, which supports previous data. Throughout this study, the freshwater sediment had the highest clay content, highest microbial biomass, and highest redox potential. In general, the redox potential for all three sediments increased over time, which suggests lower availability of soluble or labile carbon with time.

The second principal component seems to group sediments based on the availability of soluble carbon and the response from the microbial biomass over time (Figure 4.2). At the bottom of the second principal component axis, the data represents the samples with a polymer treatment after the first week. Moving up the axis, control samples and samples with polymer from weeks 8 and 16 clump together, which supports the result that the polymer had no significant influence on microbial biomass. Principal component 2 accounted for an additional 15% of the variability in the data.

Principal component 3 was also analyzed because it accounted for 15% of the variability. The highly positive PC loadings for basal respiration rate and  $qCO_2$  suggest that principal component 3 groups sediments based on metabolic efficiency. In general, the freshwater sediment had higher amounts of microbial biomass, which produced the most carbon dioxide. Plotting principal component 3 against principal component 2 further emphasized the result that the microbial consortia in the freshwater sediment respired more than the intermediate or marine sediments (Figure 4.3).

Once again, principal component 2 emphasized that the samples with polymer amendment at week one were different from the control samples at week 1 and all the samples at weeks 8 and 16.

Principal component 3 emphasized the differences in metabolic efficiency. Along axis 3, some marine sediment samples are mixed in with the freshwater sediment samples. Those marine sediment samples are from week 8, during which the marine sediment's microbial biomass peaked. This increase in microbial biomass is reflected in the metabolic quotient, which helps define principal component 3.



**Figure 4.3 Results of Principal Components Analysis on Ecophysiological Indices completed on wet sediment samples. Polymer indicates the 0.5% and 1% levels for week 1. All other samples indicates the 0% concentration level for week 1, and the 0%, 0.5%, and 1% levels for weeks 8 and 16.**

Overall, principal components analyses further clarify and support experimental data. For all three sediments, particle size distribution influenced the availability of carbon substrates. During the first week of the study, microbial populations responded to added carbon with increased activity. After the polymer was mineralized, organic matter content of sediments sustained microbial biomass growth

over time. Removal of the polymer and removal of soil organic carbon limited sediment stabilization, which was indicated by no evidence of an increase in aggregation. In addition, no significant sediment stabilization from the polymers was supported by findings of increased consolidation and dewatering over time.

#### **4.3 Reasons Why Polymers Did Not Increase Aggregation**

After considering the conclusions from the lab-based engineering study, several reasons exist to provide some insight into why the polymer solutions were not effective at increasing stability of actual dredged sediment from the field. First, the chosen concentrations of polymer solutions may not have been high enough. Nugent's results indicate that a 0.5% and 1% application rate of xanthan gum and guar gum may have actually decreased the liquid limit of the sediments. However, in the current study, measures of aggregation and stability of samples with polymer were never significantly different from the control samples. Since Nugent found that increased aggregation decreased the liquid limit of clays in some cases, no evidence of aggregation in samples with field sediments is a positive result. Once again, aggregation of samples with polymer was not significantly different from aggregation of control samples, which implies that even though the liquid limit was not increasing, it was neither decreasing.

To increase stability of hydraulically dredged sediments, more information about the amount and type of clay in the field soils is required. In Nugent's study, determination of polymer impacts on soil stability required the use of pure kaolinite clay with 89% clay content. At a field site with naturally produced sediments, environmental variables such as high moisture, highly variable clay content, mixtures of clay minerals, presence of organic matter, and microbial communities add abiotic and biotic components to the system.

Experimental results clearly indicate that the polymer was metabolized very quickly at the beginning of the experimental timeline. Reasons for such a quick metabolism of the polymers relates to

microbial activity in the sediments. Since both xanthan gum and guar gum are water-soluble, the polymers became more microbially available when mixed with saturated sediment. Increased availability of the polymers was indicated by changes in redox potential at the very beginning of the experiment.

Respiration results indicate that the microbial consortia in each sediment greatly increased activity when the ecosystem was inundated with polymer. Since the polymer is an additional carbon substrate, the microbes entered a phase of greatly increased activity at the beginning. When the polymer was fully metabolized, microbial communities continued to assimilate carbon into their biomass. The microbial community initially increased activity, but over time, the community increased its biomass through production of more individuals. The community continued to respire, which indicates metabolism of soil organic carbon and microbially transformed products from the degraded polymers. Unfortunately, for purposes of wetland building, removal of soil organic carbon prevents soil aggregation by eliminating material that holds clay particles together.

#### **4.4 Possible Approaches for Future Attempts at Dredged Material Stabilization from Polymer Application**

To increase stability of hydraulically dredged sediments, the applied polymeric material needs to be more resistant to microbial decomposition. An example of such a material is a geopolymer, which is a long, cross-linked polymer made of humic material that has three-dimensional structure. In sediments, organic compounds accumulate and transform to refractory carbon molecules that are difficult for microbes to metabolize. Carbohydrate chains react with proteins to form humic compounds, which are recalcitrant in the environment and initiate aggregate formation. Lipids wrap around the humic material to create an interpenetrating polymer network (Kim et al. 2006). Through geologic time, a geopolymer forms around the center of the interpenetrating polymer network. The chemical structure and bond strength of geopolymers increases resistance to microbial breakdown.

Another approach for a material that may be more stable in a sediment environment involves combining synthetic and natural polymers. In soils amended with just polyvinyl alcohol (PVA), the polymer resists degradation except when added to a soil amended with specific PVA-degrading microbes (Cinelli et al. 2008). When PVA is combined with natural polymers, carbon dioxide production rates in the soil are similar to soils with just natural polymers. The presence of synthetic material does not negatively affect the degradation of natural components, which still decompose without degrading the PVA. Soil microbes prefer to utilize natural polymers instead of synthetic polymers for carbon (Cinelli et al. 2008).

For fully saturated sediments, yet another alternative solution involves the use of cationic polymers. Negatively charged clay particles strongly attract cationic polymers. After initial attachment to clay, the cationic polymer chain collapses inward (Theng 1982). Any loops or tails extending into solution carry a positive charge, which increases adsorption to the soil. The portion of polymer adsorbed to clay particles neutralizes negative charges, increasing the anionic exchange capacity of the soil. As the anion exchange capacity of the soil-polymer complex increases, more cationic polymer adsorbs to the structure, which increases aggregation (Theng 1982).

#### **4.5 Conclusion**

To increase stability of hydraulically dredged sediments, an amendment that is water-insoluble and that resists microbial decomposition might be a better solution than two natural, water-soluble polymers. Further research is needed to investigate the interactions between synthetic polymers and fully saturated sediments. An amendment that does not stimulate microbial activity is an optimal solution. Natural polymers are more easily decomposed and may lead to more rapid microbial mineralization of soil organic carbon, where the removal of soil organic carbon will decrease natural sediment aggregation.

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## **APPENDIX A: TECHNICAL NOTE: DEWATERING ANALYSIS**

### **A.1 Introduction**

Calculation of the amount of dewatering provides information about how much water is being expelled to the surface as sediment consolidates (Reddy et al. 2006). If the polymer is increasing sediment stability, less dewatering will occur.

### **A.2 Methods**

Dewatering efficiency was calculated using the following formula (Reddy et al. 2006):

$$\text{Absolute value}[\text{Final moisture content (\%)} - \text{Initial moisture content (\%)}]$$

By subtracting the final moisture content from the initial moisture content, the amount of dewatering could be calculated for each sample over time.

### **A.3 Results and Discussion**

The presence of a polymer had a significant effect on the amount of dewatering for each sediment type (Tables A.1 and A.2). Overall, the amount of dewatering for the marine sediment was significantly greater than the amount of dewatering for the freshwater and intermediate sediments at all weeks and all concentration levels ( $P < 0.0001$ , Table A.3).

On average, the marine sediment dewatered approximately 14% of its water content. The freshwater and intermediate sediments dewatered less, and the amounts were not significantly different from each other. The freshwater sediment dewatered approximately 3.5% of its water content, and the intermediate sediment dewatered approximately 5% of its water content.

For the freshwater sediment, the 1% polymer treatment resulted in significantly less dewatering than the control ( $P = 0.0004$ ) and the 0.5% polymer treatment ( $P < 0.0001$ ). From week 1 to week 26, the amount of dewatering caused by the control and 0.5% polymer treatment in the freshwater sediment significantly increased ( $P = 0.0011$ ) from 2% to 7%. For the 1% polymer treatment, the amount of dewatering significantly increased from week 1 to week 26 with values of 0.2% to 3%.

**Table A.1 Type 3 Test of Fixed Effects for Dewatering Data.**

Effect	Num DF	Den DF	F Value	Pr > F
A	2	281	303.6	<0.0001
B	3	281	0.91	0.4347
C	1	281	1.3	0.2555
A*C	2	281	10.19	<0.0001
B*C	3	281	0.26	0.8516
D	2	281	79.23	<0.0001
A*D	4	281	12.4	<0.0001
B*C	6	281	1.59	0.1485
C*D	2	281	0.92	0.3985
A*C*D	4	281	4.7	0.0011
B*C*D	6	281	1.27	0.2722
E	3	281	31.62	<0.0001
A*E	6	281	2.96	0.008
B*E	9	281	0.94	0.4903
C*E	3	281	1.48	0.2203
A*C*E	6	281	1.34	0.2411
B*C*E	9	281	1.26	0.2583
D*E	6	281	0.98	0.4412
A*D*E	12	281	0.95	0.4978
B*D*E	18	281	0.85	0.6422
C*D*E	6	281	1.58	0.1529
A*C*D*E	12	281	2.15	0.0144
B*C*D*E	18	281	0.9	0.5748

A = Sediment, B = Salinity(Sediment), C = Polymer, D = Concentration, E = Week

**Table A.2 Type 3 Test of Fixed Effects after Stepwise Variable Selection for Dewatering Data.**

Effect	Num DF	Den DF	F Value	Pr>F
A	2	407	286.32	<0.0001
E	3	407	29.24	<0.0001
D	2	407	75.03	<0.0001
A*E	6	407	2.53	0.0203
A*E	4	407	10.77	<0.0001

A = Sediment, B = Salinity(Sediment), C = Polymer, D = Concentration, E = Week

Similarly, for the intermediate sediment, the 1% polymer treatment resulted in significantly less dewatering than the control ( $P<0.0001$ ) and the 0.5% polymer treatment ( $P=0.0199$ ). Over 26 weeks, control samples dewatered an average of 9% water content. Samples with 0.5% polymer treatment dewatered an average of 8% water content. Samples with 1% polymer treatment had significantly less

dewatering with an average value of 5% loss of water content. From week 1 to week 26, the amount of dewatering in the intermediate sediment significantly increased.

**Table A.3 Percent fluid dewatered from beginning to end of each time period for the Freshwater, Intermediate, and Marine sediments at the 0%, 0.5%, and 1% concentration levels. Values for polymer types and salinities have been averaged due to no significant differences. Letters indicate significant differences between time periods within sediment. Crosses indicate significant differences between concentration levels within sediment.**

Sediment	Week	0%	0.5%	1%
Freshwater	1	2.11 $\pm$ 0.745 a	2.83 $\pm$ 0.611 a	0.192 $\pm$ 0.895 a <sup>+</sup>
	8	3.69 $\pm$ 0.654 ab	5.78 $\pm$ 4.04 ab	0.106 $\pm$ 0.959 ab <sup>+</sup>
	16	5.47 $\pm$ 0.526 ab	4.72 $\pm$ 0.959 ab	0.732 $\pm$ 0.830 ab <sup>+</sup>
	26	7.34 $\pm$ 0.550 b	6.56 $\pm$ 0.622 b	3.26 $\pm$ 0.778 b <sup>+</sup>
Intermediate	1	7.15 $\pm$ 0.703 a	3.92 $\pm$ 0.806 a	0.493 $\pm$ 0.619 a <sup>+</sup>
	8	6.05 $\pm$ 0.708 a	4.21 $\pm$ 0.936 a	1.43 $\pm$ 0.952 a <sup>+</sup>
	16	7.29 $\pm$ 0.664 ab	5.64 $\pm$ 0.988 ab	4.03 $\pm$ 1.44 ab <sup>+</sup>
	26	9.12 $\pm$ 0.957 b	8.39 $\pm$ 0.698 b	5.13 $\pm$ 0.823 b <sup>+</sup>
Marine	1	15.3 $\pm$ 1.13 a	8.76 $\pm$ 1.34 a <sup>+</sup>	5.44 $\pm$ 1.79 a <sup>+</sup>
	8	17.4 $\pm$ 1.05 b	13.1 $\pm$ 0.945 b <sup>+</sup>	10.7 $\pm$ 0.994 b <sup>+</sup>
	16	21.0 $\pm$ 1.33 b	13.3 $\pm$ 1.24 b <sup>+</sup>	12.6 $\pm$ 1.15 b <sup>+</sup>
	26	22.9 $\pm$ 1.11 b	14.9 $\pm$ 0.663 b <sup>+</sup>	12.1 $\pm$ 1.40 b <sup>+</sup>

For the marine sediment, both the 0.5% and 1% polymer treatments resulted in significantly less dewatering than the control ( $P < 0.0001$ ). Over 26 weeks, the amount of dewatering was 23% for the control samples, 15% for the samples with 0.5% polymer treatment, and 12% for the samples with 1% polymer treatment. The difference between the 0.5% and 1% polymer treatment was not significant. From week 1 to week 8, the amount of dewatering from the sediments with polymer increased from 7% to 12%; this increase was significant.

Overall, the presence of a polymer did cause less dewatering over time, in comparison to control treatments; however, the calculation for dewatering must be considered. Initial moisture content values for all sediments with polymer were lower than the control samples, possibly due to polymer adsorption of water in the first 24 hours. In addition, final moisture content values at week 26



were the lowest values for all three sediments since moisture content decreased over time. Subtracting final moisture content values from initial moisture content values may give the misleading conclusion that the polymer caused less dewatering. With this awareness, patterns in dewatering reflect patterns in moisture content and sediment consolidation. Over 26 weeks, moisture content decreased and consolidation increased, which was reflected by increased dewatering.

Relationships between moisture content, sediment consolidation, dewatering, and particle size corroborate the indication that polymers did not increase sediment stability because they were not present in the sediment long enough due to microbial degradation (supporting data in chapter 3).

## APPENDIX B: ADDITIONAL TABLES AND FIGURES

### B.1 Chapter 2 Tables and Figures

**Table B.1 Type 3 Test of Fixed Effects for pH Data.**

Effect	Num DF	Den DF	F Value	Pr > F
A	2	287	141.1	<0.0001
B	3	287	5.78	0.0008
C	1	287	103.24	<0.0001
A*C	2	287	18.19	<0.0001
B*C	3	287	1.43	0.2354
D	2	287	9.24	0.0001
A*D	4	287	0.43	0.7888
B*C	6	287	0.69	0.6539
C*D	2	287	28.65	<0.0001
A*C*D	4	287	8.07	<0.0001
B*C*D	6	287	1.09	0.3666
E	3	287	256.51	<0.0001
A*E	6	287	5.84	<0.0001
B*E	9	287	1.9	0.0522
C*E	3	287	7.32	<0.0001
A*C*E	6	287	8.78	<0.0001
B*C*E	9	287	1.06	0.3913
D*E	6	287	23.5	<0.0001
A*D*E	12	287	4.58	<0.0001
B*D*E	18	287	1.24	0.2318
C*D*E	6	287	4.3	0.0004
A*C*D*E	12	287	4.12	<0.0001
B*C*D*E	18	287	1.37	0.1459

A = Sediment, B = Salinity(Sediment), C = Polymer, D = Concentration, E = Week

**Table B.2 Type 3 Test of Fixed Effects after Stepwise Variable Selection for pH Data.**

Effect	Num DF	Den DF	F Value	Pr>F
E	3	372	84.11	<0.0001
A	2	372	58.49	<0.0001
C	1	372	47.64	<0.0001
A*C	2	372	8.76	0.0002
A*E	6	372	2.9	0.0087
C*E	3	372	3.78	0.0107
A*C*E	6	372	4.8	<0.0001
D	2	372	5.13	0.0063
D*E	16	372	15.17	<0.0001
C*D	2	372	20.73	<0.0001
C*D*E	6	372	3.14	0.0052
B	3	372	4.31	0.0053

A = Sediment, B = Salinity(Sediment), C = Polymer, D = Concentration, E = Week

Upon closer analysis, salinity was only significant for the freshwater sediment; therefore, all pH values for different salinities were combined. In addition, concentration was only significant for three instances; therefore, all pH values for different concentrations were combined.

**Table B.3 Type 3 Test of Fixed Effects for Moisture Content Data.**

Effect	Num DF	Den DF	F Value	Pr > F
A	2	287	7254.07	<0.0001
B	3	287	0.24	0.8717
C	1	287	1.11	0.2922
A*C	2	287	7.68	0.0006
B*C	3	287	0.32	0.811
D	2	287	2.42	0.0907
A*D	4	287	5.98	0.0001
B*C	6	287	0.38	0.8934
C*D	2	287	0.42	0.6583
A*C*D	4	287	2.83	0.025
B*C*D	6	287	0.09	0.9969
E	3	287	81.43	<0.0001
A*E	6	287	3.61	0.0018
B*E	9	287	0.21	0.9925
C*E	3	287	3.14	0.0257
A*C*E	6	287	2.97	0.0079
B*C*E	9	287	0.37	0.9508
D*E	6	287	0.71	0.6403
A*D*E	12	287	0.72	0.7325
B*D*E	18	287	0.13	1.000
C*D*E	6	287	0.61	0.7202
A*C*D*E	12	287	1.21	0.2781
B*C*D*E	18	287	0.3	0.9978

A = Sediment, B = Salinity(Sediment), C = Polymer, D = Concentration, E = Week

**Table B.4 Type 3 Test of Fixed Effects after Variable Selection for Moisture Content Data.**

Effect	Num DF	Den DF	F Value	Pr>F
A	2	413	7657.21	<0.0001
E	3	413	86.98	<0.0001
D	2	413	2.37	0.0948
A*E	6	413	3.97	0.0007
A*D	4	413	6.05	<0.0001

A = Sediment, B = Salinity(Sediment), C = Polymer, D = Concentration, E = Week

**Table B.5 Type 3 Test of Fixed Effects for Total Sediment Consolidation Data.**

Effect	Num DF	Den DF	F Value	Pr > F
A	2	288	6.28	0.0021
B	3	288	0.44	0.7232
C	1	288	12.92	0.0004
A*C	2	288	0.85	0.4298
B*C	3	288	0.09	0.9675
D	2	288	3.77	0.0242
A*D	4	288	0.94	0.4383
B*C	6	288	0.25	0.9577
C*D	2	288	1.59	0.2049
A*C*D	4	288	0.09	0.9868
B*C*D	6	288	0.05	0.9994
E	3	288	51.8	<0.0001
A*E	6	288	13.73	<0.0001
B*E	9	288	0.78	0.638
C*E	3	288	1.54	0.2051
A*C*E	6	288	2.12	0.0509
B*C*E	9	288	0.53	0.8494
D*E	6	288	0.25	0.9592
A*D*E	12	288	1.25	0.2473
B*D*E	18	288	0.21	0.9998
C*D*E	6	288	0.51	0.7975
A*C*D*E	12	288	0.95	0.5016
B*C*D*E	18	288	0.16	1.000

A = Sediment, B = Salinity(Sediment), C = Polymer, D = Concentration, E = Week

**Table B.6 Type 3 Test of Fixed Effects for Repeated Measures Consolidation Data Over 26 Weeks.**

Effect	Num DF	Den DF	F Value	Pr > F
A	2	1284	3.21	0.0405
B	3	1284	0.37	0.7723
C	1	1284	1.57	0.21
A*C	2	1284	0.52	0.593
B*C	3	1284	0.76	0.514
D	2	1284	0.31	0.7362
A*D	4	1284	0.5	0.7328
B*C	6	1284	0.12	0.9944
C*D	2	1284	2.09	0.1239
A*C*D	4	1284	1.01	0.4037
B*C*D	6	1284	0.35	0.9087
E	19	1284	68.49	<0.0001
A*E	38	1284	7.85	<0.0001
B*E	57	1284	0.8	0.8587
C*E	19	1284	4.17	<0.0001
A*C*E	38	1284	1.16	0.2327
B*C*E	57	1284	1.01	0.4534
D*E	38	1284	1.88	0.001
A*D*E	76	1284	1.48	0.0053
B*D*E	114	1284	0.99	0.5138
C*D*E	38	1284	2.04	0.0002
A*C*D*E	76	1284	1.45	0.0083
B*C*D*E	113	1284	0.84	0.8895

A = Sediment, B = Salinity(Sediment), C = Polymer, D = Concentration, E = Flooding Event

**Table B.7 Type 3 Test of Fixed Effects after Stepwise Variable Selection for Repeated Measures Consolidation Data Over 26 Weeks.**

Effect	Num DF	Den DF	F Value	Pr>F
A	2	1961	1.63	0.1968
C	1	1961	1.4	0.2372
E	19	1961	66.51	<0.0001
C*E	19	1961	3.46	<0.0001

A = Sediment, B = Salinity(Sediment), C = Polymer, D = Concentration, E = Flooding Event

**Table B.8 Type 3 Test of Fixed Effects for Clay Aggregate Size Data.**

Effect	Num DF	Den DF	F Value	Pr > F
A	2	281	365.29	<0.0001
B	3	281	0.27	0.8475
C	1	281	0	0.9983
A*C	2	281	0.08	0.9258
B*C	3	281	2.25	0.0827
D	2	281	4.59	0.0109
A*D	4	281	0.19	0.9443
B*D	6	281	0.34	0.9131
C*D	2	281	0.13	0.8805
A*C*D	4	281	0.17	0.954
B*C*D	6	281	0.04	0.9998
E	3	281	13.2	<0.0001
A*E	6	281	1.77	0.1049
B*E	9	281	0.37	0.9485
C*E	3	281	0.11	0.955
A*C*E	6	281	0.08	0.9983
B*C*E	9	281	0.57	0.8188
D*E	6	281	0.41	0.869
A*D*E	12	281	0.17	0.9993
B*D*E	18	281	0.22	0.9998
C*D*E	6	281	0.45	0.8414
A*C*D*E	12	281	0.21	0.9978
B*C*D*E	18	281	0.17	1.000

A = Sediment, B = Salinity(Sediment), C = Polymer, D = Concentration, E = Week

**Table B.9 Type 3 Test of Fixed Effects after Stepwise Variable Selection for Clay Aggregate Size Data.**

Effect	Num DF	Den DF	F Value	Pr>F
A	2	411	475.23	<0.0001
E	3	411	16.38	<0.0001
D	2	411	5.75	0.0034
A*E	6	411	2.61	0.0172

A = Sediment, B = Salinity(Sediment), C = Polymer, D = Concentration, E = Week

Upon closer analysis, the differences between concentration levels were between different sediments with different concentration levels. The sediment\*concentration\*week interaction was not significant when tested (Table B.10).

**Table B.10 Type 3 Test of Fixed Effects after Stepwise Variable Selection for Clay Aggregate Size Data with the Sediment\*Concentration\*Week Interaction.**

Effect	Num DF	Den DF	F Value	Pr>F
A	2	389	456.78	<0.0001
E	3	389	15.91	<0.0001
D	2	389	5.46	0.0046
A*E	6	389	2.48	0.0231
A*D*E	22	389	0.27	0.9997

A = Sediment, B = Salinity(Sediment), C = Polymer, D = Concentration, E = Week

**Table B.11 Type 3 Test of Fixed Effects for Silt Aggregate Size Data.**

Effect	Num DF	Den DF	F Value	Pr > F
A	2	280	885.22	<0.0001
B	3	280	0.03	0.993
C	1	280	0.08	0.7804
A*C	2	280	0.03	0.9738
B*C	3	280	2.17	0.0923
D	2	280	2.18	0.1147
A*D	4	280	0.71	0.5861
B*C	6	280	0.18	0.9828
C*D	2	280	0.37	0.6877
A*C*D	4	280	0.2	0.9402
B*C*D	6	280	0.13	0.9926
E	3	280	19.88	<0.0001
A*E	6	280	7.74	<0.0001
B*E	9	280	0.17	0.9971
C*E	3	280	0.19	0.9021
A*C*E	6	280	0.13	0.992
B*C*E	9	280	1.64	0.1034
D*E	6	280	0.17	0.9852
A*D*E	12	280	0.42	0.9544
B*D*E	18	280	0.14	1.000
C*D*E	6	280	0.41	0.8693
A*C*D*E	12	280	0.24	0.9961
B*C*D*E	18	280	0.58	0.9134

A = Sediment, B = Salinity(Sediment), C = Polymer, D = Concentration, E = Week



**Table B.12 Type 3 Test of Fixed Effects after Stepwise Variable Selection for Silt Aggregate Size Data.**

Effect	Num DF	Den DF	F Value	Pr>F
A	2	410	1078.62	<0.0001
E	3	410	23.35	<0.0001
A*E	6	410	10.24	<0.0001
D	2	410	2.77	0.0641

A = Sediment, B = Salinity(Sediment), C = Polymer, D = Concentration, E = Week

**Table B.13 Type 3 Test of Fixed Effects for Sand Aggregate Size Data.**

Effect	Num DF	Den DF	F Value	Pr > F
A	2	280	759.17	<0.0001
B	3	280	0.02	0.9958
C	1	280	0.05	0.8284
A*C	2	280	0.02	0.9824
B*C	3	280	2.14	0.095
D	2	280	1.02	0.3629
A*D	4	280	0.64	0.6358
B*C	6	280	0.2	0.9769
C*D	2	280	0.34	0.7108
A*C*D	4	280	0.14	0.9666
B*C*D	6	280	0.12	0.9943
E	3	280	19.18	<0.0001
A*E	6	280	6.74	<0.0001
B*E	9	280	0.13	0.9987
C*E	3	280	0.17	0.9196
A*C*E	6	280	0.12	0.9939
B*C*E	9	280	1.49	0.1527
D*E	6	280	0.16	0.9867
A*D*E	12	280	0.37	0.9724
B*D*E	18	280	0.12	1.000
C*D*E	6	280	0.39	0.887
A*C*D*E	12	280	0.21	0.9978
B*C*D*E	18	280	0.52	0.9467

A = Sediment, B = Salinity(Sediment), C = Polymer, D = Concentration, E = Week

**Table B.14 Type 3 Test of Fixed Effects after Stepwise Variable Selection for Sand Aggregate Size Data.**

Effect	Num DF	Den DF	F Value	Pr>F
A	2	411	939.41	<0.0001
E	3	412	22.83	<0.0001
A*E	6	412	9.06	<0.0001

A = Sediment, B = Salinity(Sediment), C = Polymer, D = Concentration, E = Week

**Table B.15 Type 3 Test of Fixed Effects for Mean Aggregate Size Data.**

Effect	Num DF	Den DF	F Value	Pr > F
A	2	269	207.87	<0.0001
B	3	269	0.19	0.902
C	1	269	0.55	0.4591
A*C	2	269	0.25	0.7803
B*C	3	269	1.46	0.2244
D	2	269	0.21	0.8123
A*D	4	269	0.61	0.6583
B*C	6	269	0.51	0.7976
C*D	2	269	0.2	0.8187
A*C*D	4	269	0.02	0.9995
B*C*D	6	269	0.12	0.9936
E	3	269	21.3	<0.0001
A*E	6	269	28.37	<0.0001
B*E	9	269	0.02	1.000
C*E	3	269	1	0.395
A*C*E	6	269	0.6	0.727
B*C*E	9	269	2.35	0.0145
D*E	6	269	0.67	0.6758
A*D*E	12	269	0.28	0.9916
B*D*E	18	269	0.38	0.9907
C*D*E	6	269	0.89	0.5049
A*C*D*E	12	269	0.49	0.9191
B*C*D*E	18	269	0.41	0.986

A = Sediment, B = Salinity(Sediment), C = Polymer, D = Concentration, E = Week

**Table B.16 Type 3 Test of Fixed Effects after Stepwise Variable Selection for Mean Aggregate Size Data.**

Effect	Num DF	Den DF	F Value	Pr>F
A	2	401	251.59	<0.0001
E	3	401	25.86	<0.0001
A*E	6	401	35.49	<0.0001

A = Sediment, B = Salinity(Sediment), C = Polymer, D = Concentration, E = Week

## B.2. Chapter 3 Tables and Figures

**Table B.17 Type 3 Test of Fixed Effects for Redox Potential Data.**

Effect	Num DF	Den DF	F Value	Pr > F
A	2	287	101.59	<0.0001
B	3	287	1.37	0.2522
C	1	287	1.17	0.2806
A*C	2	287	0.87	0.421
B*C	3	287	0.02	0.9969
D	2	287	66.67	<0.0001
A*D	4	287	11.82	<0.0001
B*C	6	287	1.44	0.2005
C*D	2	287	0.91	0.404
A*C*D	4	287	5.2	0.0005
B*C*D	6	287	0.59	0.7395
E	3	287	19	<0.0001
A*E	6	287	3.47	0.0025
B*E	9	287	1.15	0.3255
C*E	3	287	0.35	0.7882
A*C*E	6	287	1.04	0.4008
B*C*E	9	287	0.24	0.9877
D*E	6	287	2.14	0.0495
A*D*E	12	287	0.93	0.5122
B*D*E	18	287	0.49	0.9616
C*D*E	6	287	0.65	0.6926
A*C*D*E	12	287	2.28	0.0087
B*C*D*E	18	287	0.61	0.8904

A = Sediment, B = Salinity(Sediment), C = Polymer, D = Concentration, E = Week

**Table B.18 Type 3 Test of Fixed Effects after Variable Selection for Redox Potential Data.**

Effect	Num DF	Den DF	F Value	Pr>F
A	2	407	101.25	<0.0001
D	2	407	66.76	<0.0001
E	3	407	18.46	<0.0001
A*D	4	407	11.73	<0.0001
A*E	6	407	3.33	0.0033
D*E	6	407	2.04	0.0599

A = Sediment, B = Salinity(Sediment), C = Polymer, D = Concentration, E = Week

**Table B.19 Type 3 Test of Fixed Effects for Microbial Biomass Data.**

Effect	Num DF	Den DF	F Value	Pr > F
A	2	288	215.45	<0.0001
B	3	288	1.79	0.1491
C	1	288	0.4	0.5267
A*C	2	288	2.24	0.1088
B*C	3	288	0.73	0.5357
D	2	288	0.77	0.465
A*D	4	288	0.43	0.785
B*C	6	288	0.4	0.8806
C*D	2	288	0.03	0.968
A*C*D	4	288	0.35	0.8453
B*C*D	6	288	0.54	0.778
E	3	288	21.56	<0.0001
A*E	6	288	8.31	<0.0001
B*E	9	288	0.41	0.9319
C*E	3	288	0.72	0.5402
A*C*E	6	288	0.51	0.8013
B*C*E	9	288	0.28	0.9791
D*E	6	288	0.38	0.8914
A*D*E	12	288	0.28	0.9925
B*D*E	18	288	0.33	0.996
C*D*E	6	288	0.05	0.9993
A*C*D*E	12	288	0.15	0.9996
B*C*D*E	18	288	0.26	0.9991

A = Sediment, B = Salinity(Sediment), C = Polymer, D = Concentration, E = Week

**Table B.20 Type 3 Test of Fixed Effects for  $C_{mic}:C_{org}$  Ratio Data for Weeks 1, 8, and 16.**

Effect	Num DF	Den DF	F Value	Pr > F
A	2	216	83.03	<0.0001
B	3	216	0.66	0.5755
C	1	216	0.4	0.5273
A*C	2	216	0.12	0.8896
B*C	3	216	0.01	0.9983
D	2	216	0.01	0.9927
A*D	4	216	0.19	0.9431
B*C	6	216	0.04	0.9997
C*D	2	216	0.16	0.85
A*C*D	4	216	0.17	0.9536
B*C*D	6	216	0.21	0.973
E	2	216	11.81	<0.0001
A*E	4	216	11.28	<0.0001
B*E	6	216	0.06	0.9992
C*E	2	216	0.58	0.5633
A*C*E	4	216	0.61	0.6592
B*C*E	6	216	0.1	0.9965
D*E	4	216	0.05	0.9961
A*D*E	8	216	0.03	1.000
B*D*E	12	216	0.08	1.000
C*D*E	4	216	0.15	0.9619
A*C*D*E	8	216	0.17	0.9948
B*C*D*E	12	216	0.05	1.000

A = Sediment, B = Salinity(Sediment), C = Polymer, D = Concentration, E = Week

**Table B.21 Type 3 Test of Fixed Effects for Basal Respiration Data.**

Effect	Num DF	Den DF	F Value	Pr > F
A	2	69.9	58.67	<0.0001
B	3	69.8	0.86	0.4683
C	1	71.1	2.27	0.1364
A*C	2	69.9	1.56	0.2168
B*C	3	69.8	3.42	0.0219
D	2	71	130.45	<0.0001
A*D	4	69.6	5.96	0.0003
B*C	6	69.8	1.04	0.4094
C*D	2	71	3.61	0.0322
A*C*D	4	69.6	1.77	0.1454
B*C*D	6	69.8	0.91	0.4915
E	9	413	4.21	<0.0001
A*E	17	412	3.19	<0.0001
B*E	26	412	0.63	0.9193
C*E	9	413	0.88	0.5434
A*C*E	17	412	0.6	0.8897
B*C*E	26	412	0.79	0.7566
D*E	18	413	2.31	0.0019
A*D*E	34	412	1.3	0.1278
B*D*E	52	412	1.05	0.3828
C*D*E	18	413	0.5	0.9599
A*C*D*E	34	412	0.57	0.9774
B*C*D*E	52	412	0.62	0.9835

A = Sediment, B = Salinity(Sediment), C = Polymer, D = Concentration, E = Day

**Table B.22 Type 3 Test of Fixed Effects after Variable Selection for Basal Respiration Data.**

Effect	Num DF	Den DF	F Value	Pr>F
D	2	93	120.18	<0.0001
A	2	87.9	55.05	<0.0001
A*D	4	87.9	5.76	0.0004
C	1	87.7	3.34	0.071
C*D	2	87.7	4.45	0.0145
A*C	2	87.7	1.7	0.189
A*C*D	4	87.7	1.88	0.1208
D*E	27	699	3.17	<0.0001

A = Sediment, B = Salinity(Sediment), C = Polymer, D = Concentration, E = Day

**Table B.23 Type 3 Test of Fixed Effects for Metabolic Quotient Data for Weeks 1, 8, and 16.**

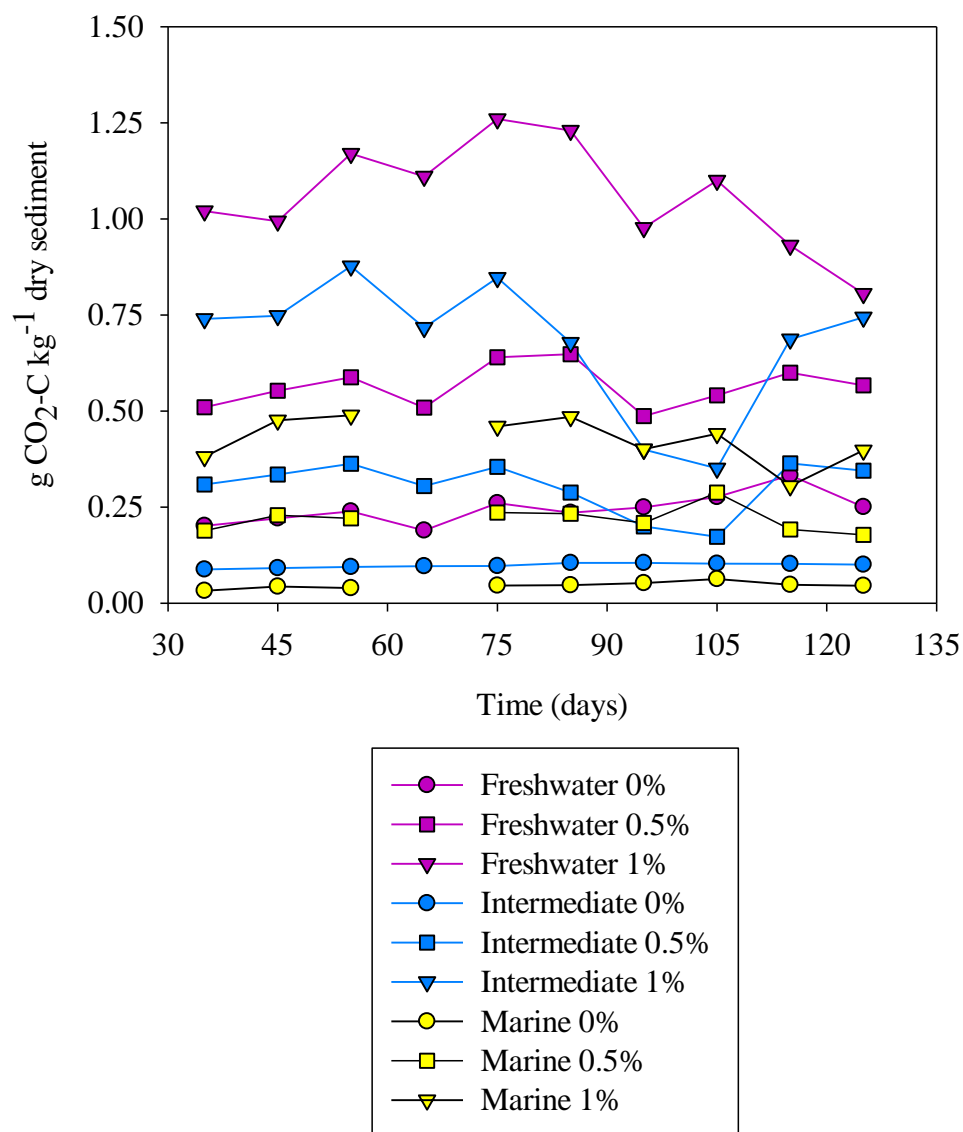
Effect	Num DF	Den DF	F Value	Pr > F
A	2	140	2.86	0.0605
B	3	140	0.56	0.6426
C	1	140	0.26	0.6121
A*C	2	140	1.5	0.2274
B*C	3	140	1.06	0.3673
D	2	140	15.16	<0.0001
A*D	4	140	2.23	0.0688
B*C	6	140	1.26	0.279
C*D	2	140	0.15	0.8572
A*C*D	4	140	0.42	0.7911
B*C*D	6	140	0.91	0.4902
E	2	140	37.04	<0.0001
A*E	4	140	0.73	0.5707
B*E	6	140	0.77	0.5975
C*E	2	140	0.96	0.3855
A*C*E	4	140	0.63	0.6443
B*C*E	6	140	0.4	0.8766
D*E	4	140	9.77	<0.0001
A*D*E	8	140	1.13	0.35
B*D*E	12	140	1.29	0.2312
C*D*E	4	140	1.33	0.2617
A*C*D*E	8	140	0.93	0.4958
B*C*D*E	12	140	0.35	0.9764

A = Sediment, B = Salinity(Sediment), C = Polymer, D = Concentration, E = Week

**Table B.24 Mean soil respiration values (g C-CO<sub>2</sub> kg<sup>-1</sup> dry soil) for Bayou Chevreuil, Leeville, and Grand Isle sediments for control, 1%, and 2% concentration levels at each measurement. Values for the polymer types have been averaged due to no significant differences. Letters indicate significant differences between sediment\*concentration values.**

	Freshwater			Intermediate			Marine		
Day	0%	0.5%	1%	0%	0.5%	1%	0%	0.5%	1%
35	0.201 a	0.51 a	1.02 c	0.088 a	0.309 a	0.74 d	0.032 a	0.189 a	0.381 e
45	0.221 a	0.553 a	0.994 c	0.103 a	0.335 a	0.748 d	0.043 a	0.229 a	0.476 e
55	0.239 a	0.588 a	1.17 c	0.115 a	0.363 a	0.877 d	0.039 a	0.221 a	0.489 e
65	0.189 a	0.509 a	1.11 c	0.092 a	0.305 a	0.717 d	-	-	-
75	0.26 a	0.64 b	1.26 c	0.124 a	0.355 a	0.847 d	0.046 a	0.236 a	0.46 e
85	0.235 a	0.648 b	1.23 c	0.094 a	0.288 a	0.678 d	0.047 a	0.233 a	0.485 e
95	0.249 a	0.487 a	0.977 c	0.12 a	0.2 a	0.4 d	0.052 a	0.209 a	0.401 e
105	0.276 a	0.541 a	1.1 c	0.114 a	0.173 a	0.351 d	0.063 a	0.288 a	0.441 e
115	0.333 a	0.6 a	0.931 c	0.161 a	0.364 a	0.687 d	0.048 a	0.192 a	0.305 e
125	0.25 a	0.567 a	0.805 c	0.148 a	0.345 a	0.744 d	0.045 a	0.178 a	0.398 e





**Figure B.1** Cumulative respiration curves for the Freshwater, Intermediate, and marine sediments at the 0%, 0.5%, and 1% concentration levels. Values for polymer types and salinities have been averaged due to no significant differences. (N=12)

## **APPENDIX C: TECHNICAL NOTE: GRAIN SIZE ANALYSIS OF PRE-TREATED SAMPLES**

### **C.1 Introduction**

In reducing sediments and wetland soils, organic matter decomposes slower than in aerobic soils and may increase aggregation. Water-stability of soil aggregates depends on available organic materials (Tisdall and Oades 1982; Martens et al. 1992). Free organic matter provides a substrate for microbial production and decomposition of extracellular polymeric substances (EPS).

As soil organisms decompose organic materials, microbial secretions bind soil particles and small organic particles together (Tiessen & Stewart 1988; Zhang et al. 2005). An increase in the amount of fine pores associated with organic matter improves water retention and makes conditions more suitable for microbial activity. Higher microbial activity leads to more EPS production, which leads to greater aggregation as polymers trap soil particles (Zhang et al. 2005). Even after microbial growth stops, microbial by-products remain in the soil and continue to aggregate soil particles (Frey 2005; Martens et al. 1992). In general, as the amount of organic matter declines, EPS production and the number of stable macroaggregates also declines.

Microbial secretions eventually produce a biofilm. A biofilm is the accumulation of microbes, EPS, multivalent cations, biogenic particles and dissolved compounds (Rillig 2005). Biofilm formation increases soil aggregation. Microbes transport themselves to soil-organic-matter complexes by diffusion, convection, sedimentation, or self motility. After single organisms adhere to soil and organic matter particles, microbes aggregate together and anchor themselves by producing EPS. Soils exhibit a positive correlation between concentrations of EPS and aggregate stability (Rillig 2005). As a result, changes in biota can have impacts on sediment erodability. As the EPS content of intertidal soils increases, critical erosion velocity also increases, suggesting that microbial biofilms increase sediment cohesiveness and sediment stability (Widdows et al. 2006).

The overall purpose of the experiment was to find increased aggregation of wet sediments from polymer addition, which was anticipated to increase particle aggregation as a process to enhance stability of newly deposited dredged sediment. Compared to aggregate size analysis, grain size analysis of samples pre-treated to remove organic matter emphasized the importance of soil organic matter in natural aggregation processes.

## **C.2 Methods**

Sediment samples were analyzed on a Beckman Coulter LS 13 320 Series Multi-Wavelength Laser Diffraction Particle Size Analyzer with an aqueous liquid module and sonicator system made available through Dr. Alex Kolker, LUMCON. With 116 size channels and 132 detectors, the LS 13 320 can measure particles from 0.017  $\mu\text{m}$  – 2000  $\mu\text{m}$ . The LS 13 320 uses Mie scattering, Fraunhofer diffraction, and PIDS (Polarization Intensity Differential Scattering) technology. Two sediment samples from each experimental unit were analyzed, following two separate protocols: one for grain size analysis and one for aggregate size analysis.

For grain size analysis, samples were analyzed following steps of pre-treatment. Several studies emphasize the importance of pre-treatment to obtain accurate grain size analysis (Matthews 1991; Beuselinck et al. 1998; Muggler et al. 1997). An aliquot of wet sediment (i.e. just a few grams) was taken from each experimental unit and given one mL of 30% hydrogen peroxide to destroy organic matter overnight. After at least eight hours, each sample received 10 mLs of a dispersing agent, 0.05 M sodium hexametaphosphate. Samples were sonicated for 60 seconds before being analyzed by the clay and silt protocol on the LS 13 320 (developed in the Kolker lab at LUMCON).

To find differences in aggregation due to the polymer, aggregate size analysis was completed on samples of wet sediment from each experimental unit. Samples for aggregate size analysis did not receive any pre-treatment in an effort to maintain natural aggregates (Matthews 1991). These samples

did not receive sonication and were analyzed by the sands standard operating protocol on the LS 13 320 (developed in the Kolker lab at LUMCON).

For all analyses, the LS 13 320 reported the percent volume of the sample falling into several size fractions: less than 2  $\mu\text{m}$ , greater than 2  $\mu\text{m}$ , less than 63  $\mu\text{m}$ , greater than 63  $\mu\text{m}$ , and greater than 1000  $\mu\text{m}$ . The LS 13 320 also reported mean particle size diameter for each sample. Particle size classes were assigned according to the following parameters: 0-2  $\mu\text{m}$  for the clay fraction, 2-63  $\mu\text{m}$  for the silt fraction, and greater than 63  $\mu\text{m}$  for the sand fraction.

SAS 9.1 software (2009) was used to analyze the data. SigmaPlot 11.0 software (2008) was used to graph the data. For grain size analysis, an ANOVA with a test of Type III fixed effects was run to find any significant differences. The least squares means analysis was evaluated to look for differences between significant effects for dependent variables. An alpha value of 0.05 was used for analyses.

### **C.3 Results and Discussion**

Comparing the size classes for the pre-treated and wet sediment samples reveals significant differences for the sediments at several weeks (Table C.1).

For the freshwater sediment, the percent volume in the clay fraction was significantly lower in the wet sediment samples than the pre-treated samples at all weeks ( $P<0.0001$ ), suggesting that clay particles form aggregates in larger size classes. The percent volume in the sand fraction was significantly higher in the wet sediment samples than the pre-treated samples at week 1 ( $P<0.0001$ ), week 16 ( $P<0.0001$ ), and week 26 ( $P=0.0019$ ).

The intermediate sediment exhibited similar comparisons. The percent volume in the clay fraction was significantly lower in the wet sediment samples than the pre-treated samples at all weeks ( $P<0.0001$ ). The percent volume in the sand fraction was significantly higher in the wet sediment

samples than the pre-treated samples at week 1 ( $P<0.0001$ ), week 8 ( $P=0.0038$ ), week 16 ( $P<0.0001$ ), and week 26 ( $P=0.0002$ ).

**Table C.1. Mean percent volume of clay, silt, and sand fractions of pre-treated and wet sediment samples for the A) Freshwater, B) Intermediate, and C) Marine sediments at weeks 1, 8, 16, and 26. Values for the 0%, 0.5%, and 1% concentration levels, polymer types, and salinities have been averaged due to no significant differences. Crosses indicate significant differences between pre-treated and wet sediment samples within a week.**

A	Clay (<2 $\mu\text{m}$ )		Silt (2-63 $\mu\text{m}$ )		Sand (>63 $\mu\text{m}$ )	
Wk	Pre-Treated	Wet Sediment	Pre-Treated	Wet Sediment	Pre-Treated	Wet Sediment
1	20.1 $\pm$ 0.623	7.04 $\pm$ 0.205 <sup>+</sup>	56.9 $\pm$ 1.46	47.8 $\pm$ 1.62 <sup>+</sup>	22.5 $\pm$ 2.02	45.3 $\pm$ 1.77 <sup>+</sup>
8	17.7 $\pm$ 0.616	7.03 $\pm$ 0.240 <sup>+</sup>	45.3 $\pm$ 1.51	50.2 $\pm$ 1.21	37.0 $\pm$ 2.00	42.7 $\pm$ 1.42
16	22.9 $\pm$ 0.778	7.41 $\pm$ 0.180 <sup>+</sup>	53.8 $\pm$ 1.65	49.1 $\pm$ 0.595	24.8 $\pm$ 2.74	43.5 $\pm$ 0.697 <sup>+</sup>
26	17.8 $\pm$ 0.534	7.00 $\pm$ 0.154 <sup>+</sup>	42.8 $\pm$ 1.17	43.9 $\pm$ 0.844	39.3 $\pm$ 1.70	49.1 $\pm$ 0.974 <sup>+</sup>

B	Clay (<2 $\mu\text{m}$ )		Silt (2-63 $\mu\text{m}$ )		Sand (>63 $\mu\text{m}$ )	
Wk	Pre-Treated	Wet Sediment	Pre-Treated	Wet Sediment	Pre-Treated	Wet Sediment
1	11.7 $\pm$ 0.331	4.97 $\pm$ 0.059 <sup>+</sup>	65.9 $\pm$ 1.24	54.9 $\pm$ 0.618 <sup>+</sup>	22.5 $\pm$ 1.52	40.1 $\pm$ 0.672 <sup>+</sup>
8	13.0 $\pm$ 0.377	5.21 $\pm$ 0.042 <sup>+</sup>	62.5 $\pm$ 1.03	61.0 $\pm$ 0.322	24.5 $\pm$ 1.27	33.8 $\pm$ 0.349 <sup>+</sup>
16	16.4 $\pm$ 0.504	5.66 $\pm$ 0.043 <sup>+</sup>	67.5 $\pm$ 1.00	62.0 $\pm$ 0.287	16.4 $\pm$ 1.41	32.4 $\pm$ 0.285 <sup>+</sup>
26	14.7 $\pm$ 0.227	5.78 $\pm$ 0.037 <sup>+</sup>	63.2 $\pm$ 0.443	61.5 $\pm$ 0.205	22.1 $\pm$ 0.620	32.7 $\pm$ 0.216 <sup>+</sup>

C	Clay (<2 $\mu\text{m}$ )		Silt (2-63 $\mu\text{m}$ )		Sand (>63 $\mu\text{m}$ )	
Wk	Pre-Treated	Wet Sediment	Pre-Treated	Wet Sediment	Pre-Treated	Wet Sediment
1	6.29 $\pm$ 0.424	3.66 $\pm$ 0.131 <sup>+</sup>	30.3 $\pm$ 2.10	27.0 $\pm$ 1.01	63.4 $\pm$ 2.51	69.3 $\pm$ 1.31
8	5.32 $\pm$ 0.291	4.06 $\pm$ 0.110	22.4 $\pm$ 1.44	32.2 $\pm$ 0.890 <sup>+</sup>	72.2 $\pm$ 1.71	63.7 $\pm$ 0.989 <sup>+</sup>
16	4.69 $\pm$ 0.206	4.72 $\pm$ 0.121	18.4 $\pm$ 0.865	35.1 $\pm$ 0.961 <sup>+</sup>	76.9 $\pm$ 1.07	60.2 $\pm$ 1.07 <sup>+</sup>
26	4.19 $\pm$ 0.199	4.40 $\pm$ 0.126	16.3 $\pm$ 0.941	34.7 $\pm$ 0.826 <sup>+</sup>	79.5 $\pm$ 1.13	60.9 $\pm$ 0.934 <sup>+</sup>

For the marine sediment, the percent volume in the clay fraction was significantly lower in the wet sediment samples than the pre-treated samples only at week 1 ( $P<0.0001$ ). The percent volume in the silt fraction was significantly higher in the wet sediment samples than the pre-treated samples at weeks 8, 16, and 26 ( $P<0.0001$ ). The percent volume in the sand fraction was significantly lower in the

wet sediment samples than the pre-treated samples at week 8 ( $P=0.0140$ ), week 16 ( $P<0.0001$ ), and week 26 ( $P<0.0001$ ).

In general, the three sediments showed a significant decrease in percent volume of the clay size class between the pre-treated and the wet sediment samples. The freshwater and intermediate sediments showed a significant increase in the percent volume of the sand size class, possibly due to aggregation of clay and silt particles from natural processes. The marine sediment showed a significant increase in the percent volume of the silt size class, corresponding with a significant decrease in the percent volume of the sand size class. The increase in the silt size class may result from increased clay aggregates in the pore spaces between sand grains.

In general, the presence of a polymer did not have any significant effect on increasing aggregation in the clay, silt, or sand fractions of any of the three sediments. The polymer addition was anticipated to increase particle aggregation as a process to enhance stability of newly deposited dredged sediment. Differences in size classes between initially characterized sediments and sediments for aggregate analysis result from natural processes and naturally occurring soil organic matter. In both the freshwater and intermediate sediments for aggregate analysis, the increase in the sand fraction compared to initially characterized sediments suggests aggregation of silt and clay particles from organic materials. In the marine sediments for aggregate analysis, the increase in the silt fraction compared to initially characterized sediments suggests that clay and silt particles filled the pore spaces between sand grains and formed aggregates. Once again, differences between size classes of initially characterized sediments and wet sediment samples reinforce the importance of naturally occurring soil organic matter in natural aggregation processes.

## **VITA**

Lauren Elizabeth Land was born in Maryland in October of 1986. She grew up with a younger sister and brother in Catonsville, Maryland, and graduated from Catonsville High School in 2004. For her undergraduate work, she went to the University of Maryland at College Park and participated in the College Park Scholars Environmental Studies Program. In 2008, she graduated with a Bachelor of Science in Environmental Science and Policy with a citation in Geographic Information Systems. Lauren moved to Baton Rouge, Louisiana, in July of 2008 to begin her graduate work in the School of the Coast and Environment at Louisiana State University. She is currently a candidate for the degree of Master of Science in the Department of Oceanography and Coastal Sciences, which will be awarded December 2010. She is a finalist for the Sea Grant Knauss Marine Policy Fellowship and will begin work in Washington, D.C., in February 2011.