Final Report

SEAGRASS MITIGATION SITE MODELING AND ASSESSMENT

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DISCLAIMER

The opinions, findings, and conclusions expressed in this publication are those of the author and not necessarily those of the Steate of Florida Department of Transportation

METRIC CONVERSION TABLE

Into Metric

If you know	Multiply by	To Get
Length		
inches	2.54	centimeters
foot	30	centimeters
yards	0.91	meters
miles	1.6	kilometers
Area		
sq. inches	6.5	sq. centimeters
sq. feet	0.09	sq. meters
sq. yards	0.8	sq. meters
sq. miles	2.6	sq. kilometers
Mass (Weight)		
ounces	28	grams
pounds	0.45	kilograms
short ton	0.9	metric ton
Volume		
teaspoons	5	milliliters
tablespoons	15	milliliters
fluid ounces	30	milliliters
cups	0.24	liters
pints	0.47	liters
quarts	0.95	liters
gallons	3.8	liters
cubic feet	0.03	cubic meters
cubic yards	0.76	cubic meters
Temperature		
Fahrenheit	Subtract 32, then multiply by 5/9ths to get	Celsius

Out of Metric

If you know	Multiply by	To Get
Length		
millimeters	0.04	inches
centimeters	0.4	inches
meters	3.3	feet
kilometers	0.62	miles
Area		
sq. centimeters	0.16	sq. inches
sq. meters	1.2	sq. yards
sq. kilometers	0.4	sq. miles
hectares	2.47	acres
Mass (Weight)		
grams	0.035	ounces
kilograms	2.2	pounds
metric tons	1.1	short tons
Volume		
milliliters	0.03	fluid ounces
liters	2.1	pints
liters	1.06	quarts
liters	0.26	gallons
cubic meters	35	cubic feet
cubic meters	1.3	cubic yards
Temperature		
Celsius	Multiply by 9/5ths, then add 32 to get	Fahrenheit

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16. Abstract

Spatiotemporal analysis of Lake Surprise and SL-15 has allowed for a robust assessment of successuful FDOT activities. Project results showed that bridge construction in Lake Surprise did not cause negative imapcts to seagrass. In fact, it is arguable that the bridge has improved conditions in the lake. We expect seagrass to continue expanding and becoming more dense. At SL-15, it is clear that natural recruitment will produce excellent densities and heights in target mangrove species. Initial planting is most likely unnecessary if the design can amplify natural recruitment. Seagrass recruitment, however, may not occur as easily. Improving exchange and possibly shifting resources towards seagrass transplanting should improve success provided soils are analyzed to ensure ample soil phosphorus is available to transplants and recruits. Larger or wider flushing channels would most likely preserve tidal exchange and improve recruitment of seagrass

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EXECUTIVE SUMMARY

BACKGROUND AND RATIONALE

Because of Florida's extensive coastline, planned construction must be carefully planned in order to properly accommodate the coastal ecosystems that are vital to the region's environmental health and often the reason that construction at a particular site is desirable. Coastal ecosystems, such as mangrove forests and seagrass flats, are easily impacted both directly and indirectly. Direct impacts include physical disturbance or removal. Indirect impacts include decline resulting from shading and impaired water quality. Restoring these areas and/or mitigating these impacts is challenging because often insufficient knowledge exists to effectively design the restoration/mitigation and because monitoring is often based on cumulative statistics for the entire restoration project rather than looking at the spatial distribution of success and failure of the design.

A spatial component to monitoring is useful because it allows for visualization of the spatial patterns at the target site. When this is conducted over time, the spatiotemporal data provide insights to the natural processes occurring of the site.

In addition to spatial modeling, an examination of the soil properties at these sites can provide an additional facet to the information gained through monitoring. Little is known about soil/vegetation relationships in coastal ecosystems so collecting these data is currently a research tool rather than a diagnostic tool.

The research presented in this report is presented as a series of studies that all fall within the scope of better understanding the spatial and temporal changes occurring at a spoil island in St. Lucie County and Lake Surprise in Monroe County.

SL-15: SPOIL ISLAND AS A SEAGRASS AND MANGROVE MITIGATION SITE

In 2004-2005, the Florida Department of Transportation partially removed the 15th spoil island in St. Lucie County (SL-15) to create seagrass and mangrove habitat. This was required mitigation for damages incurred during the widening of two portions of the causeway south of Ft. Pierce, FL. For the next five years, the island was monitored for permit compliance (80% survivorship of in the mangrove planter area and 10% seagrass recruitment in the seagrass embayment). At the end of the permit period in 2010, it was determined that the island had met permit requirements, and the island was released to the Florida Department of Environmental Protection. The permit-compliance monitoring did not assess which areas of the mangroves and seagrass were successful and which, if any, were not. Without spatial data, it is not possible to assess the design of the sites. Future mitigation and restoration efforts could benefit from understanding which portions of the site design contributed to success and which did not.

FIRST STUDY: SPATIOTEMPORAL DYNAMICS OF SL-15

The spatial patterns of mangroves and seagrass were monitored semiannually (winter and summer) from winter 2008 through summer 2011. The mangrove planter has experienced considerable natural recruitment in the back (North) portion where seeds and propagules have collected at the edge of the upland berm. The front (South) portion of the mangrove planter was originally planted with marsh grass (*Spartina alterniflora*). In 2009, the marsh grass became thick enough to trap all incoming mangrove seeds and propagules. As a result, this area had the highest concentrations of mangroves in 2011. Spatial patterns of mangrove heights mirrored these spatial patterns of density. The cause of these greater heights was not directly tested, but it is hypothesized that natural recruitment produces a healthier plant. Also, it is possible that areas of natural recruitment are also areas of particulate accumulation, which provides nutrients for these plants. The interior of the mangrove planter also supported large plants and greater densities than originally planted, but these areas were not as dense and robust as the areas of enhanced recruitment. No further action is recommended for the mangrove planter.

The seagrass recruitment area has not experienced the same success. While the permit-compliance monitoring was used to determine whether the seagrass embayment had recruited a sufficient amount of seagrass, the spatial monitoring documented less seagrass. Part of the disparity between the findings is that the permit-compliance monitoring uses the Braun-Blanquet scoring system while the spatial monitoring was a direct assessment of percent cover. The spatial analysis suggested that the long-term outlook of the embayment is uncertain.

Although the flushing channels were not a focus of this study, anecdotal observations of these channels at extreme low tide in the last field trip of 2011 suggested that they may have filled in following oyster recruitment across some channels. It is possible that this is creating a pool of water at low tide. Unlike the neighboring seagrass flats which drain on low tide, this pool of water could have greatly elevated temperatures and turbidity at low tide and in summer months. We recommend a focused analysis of the embayment area and seagrass transplant experiments to determine feasibility of seagrass growth.

SECOND STUDY: SPATIOTEMPORAL DYNAMICS OF LAKE SURPRISE

The removal of the US 1 causeway and the construction of a low-lying bridge across Lake Surprise was a reasonable cause for concern. The peat soils were expected to cause nutrient loading resulting in an algal bloom and seagrass decline. A semiannual spatial monitoring of Lake Surprise with an even distribution of sites across the lake was implemented to capture any changes. After three years of monitoring, there was no seagrass decline measured in Lake Surprise. Trends in water quality and soil properties were stable throughout the period of observation. The west portion of Lake Surprise experienced an increase in shoot density of *Thalassia testudinum* (turtle grass). This is likely attributable to the improved hydraulic connectivity following the causeway removal. Analysis of seagrass data suggested that slight seasonal fluctuations can occur in Florida Bay.

THIRD STUDY: INFLUENCE OF SUBAQUEOUS SOILS ON *HALODULE WRIGHTII*

Transplanting seagrass is sometimes required when restoring or creating seagrass habitat. Success of transplanting is generally low. To date, there has been little available information about transplant soil preferences. This study employed a randomized block design of small plastic containers of several different soils. At two locations, *Halodule wrightii* was transplanted into the buckets and measured over time.

A suite of soils that together composed wide ranges in soil physical and chemical properties were used in the experiment. Transplant shoot counts were recorded monthly to assess growth, and transplants were collected for vegetative analysis after five months. Our analysis suggested that soil types have a significant effect on transplant growth. In Key Largo, the effect of soil total phosphorus, total iron, and organic matter content on transplant growth was found to be highly significant ($p<0.0001$), while soil texture, total carbon, total nitrogen, and porewater sulfides were also found to significantly ($p<0.05$) influence transplant growth. In Fort Pierce, insufficient environmental conditions outside of soil properties diminished the influence of soil properties on transplants, yet soil Total phosphorus, Total nitrogen, organic matter content, and porewater sulfide were found to significantly ($p<0.05$) influence transplant growth. Transplants at the Key Largo site consistently exhibited greater growth responses relative to transplants within the same soil treatments at the Fort Pierce site. We suggested the soils of areas where seagrass recruitment is desired (e.g., Boca Chica) be analyzed to determine suitability.

Additionally, we suggested a more extensive experiment at multiple sites with more replications and over a longer period of time. This would be necessary to establish threshold values for soil properties. It is plausible that soil amendments of iron and/or phosphorus could be very beneficial to seagrass recruitment. An assessment of this possibility could be included in these experiments.

FOURTH STUDY: ASSESSMENT OF SL-15 RESTORATION TRAJECTORIES

Coastal ecosystems are significant natural carbon sinks. If constructed coastal ecosystems can obtain the same carbon sink capacity as their natural counterparts, then construction and restoration of these systems has the potential to become a tool for reducing atmospheric CO_2. In this study, sediment organic carbon (OC) of a recently constructed mangrove and seagrass system in the Indian River Lagoon, Florida, was compared with sediment OC of nearby mature, reference systems. Total OC, extractable OC, and microbial biomass C pools were measured to compare C storage. Organic C lability in the constructed and reference sites was also measured. The main sediment OC sources were determined using ^{13}C isotopes, and C:N ratios and were compared among systems. Organic C pools were generally larger in sediments of reference systems than in sediments of the constructed systems, but differences in pool sizes were much greater between the constructed and reference mangrove systems. Organic C lability was greater in the constructed systems, indicating their sediments could not store OC for as long as the references. Seston was a major source of sediment OC in all systems. Other main sources of OC were higher-plant-derived in constructed and reference mangrove and reference seagrass sediments,

but were algal-derived in constructed seagrass sediments. After one year, the C sink capacity of the constructed systems is less than the capacity of the reference systems, but the constructed seagrass system is functioning more like its reference than the constructed mangrove system. In the long term, however, the potential C sink capacity of the constructed mangrove system is greater.

PARTICLE SIZE DISTRIBUTION ANALYSIS IN MARINE SOILS

Prior to 2008, we observed high correlations between silt content and organic matter content in marine soils. Unpublished investigations of the mineralogy of the silt fraction revealed unusually high concentrations of calcium oxalate. We suspect that the formation of this silt-sized mineral occurs during the organic matter removal pretreatment of particle size distribution analysis (PSDA). To avoid creating silt in samples analyzed in these studies, we developed an alternative method of removing organic matter from twin samples. The literature review supporting this is presented in this section of the report. It was not within the scope of this project to implement an experiment to refine this method. We have included an experimental design as part of the findings of our investigations. We recommend pursing this experiment so that future PSDA efforts can benefit from a well-studied and developed method.

CONCLUSIONS

Spatiotemporal analysis of Lake Surprise and SL-15 has allowed for a robust assessment of successful FDOT activities. Project results showed that bridge construction in Lake Surprise did not cause negative imapcts to seagrass. In fact, it is arguable that the bridge has improved conditions in the lake. We expect seagrass to continue expanding and becoming more dense. At SL-15, it is clear that natural recruitment will produce excellent densities and heights in target mangrove species. Initial planting is most likely unnecessary if the design can amplify natural recruitment. Seagrass recruitment, however, may not occur as easily. Improving exchange and possibly shifting resources towards seagrass transplanting should improve success provided soils are analyzed to ensure ample soil phosphorus is available to transplants and recruits. Larger or wider flushing channels would most likely preserve tidal exchange and improve recruitment of seagrass

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CHAPTER 1: INTRODUCTION

BACKGROUND

The destruction of seagrass and mangrove habitat necessitates the creation of mitigation sites. The design and monitoring of these sites is hindered by a lack of soils knowledge. Monitoring these sites is usually conducted for a few years following the completion of the site. However, monitoring usually focuses on determining whether target vegetative cover has been met. This typically involves using quadrants or transects to assess vegetative cover. In contrast, more spatially explicit methods of assessing vegetative patterns can employ geostatistics to understand spatiotemporal patterns of vegetative cover, thereby allowing additional insight into the vegetative trajectory of the area. Also, a similar analysis of soil properties can provide insight into the development of the benthic habitat conducted such analyses on a spoil island that had been partially removed to create a seagrass and mangrove mitigation site.

A NOVEL MITIGATION APPROACH: SPOIL ISLAND REMOVAL

Typically, suitable seagrass mitigation sites are difficult to locate. A potential site would be one that does not currently support seagrass, but will after alteration or planting. Spoil islands offer excellent alternatives to altering more natural sites. These islands were created when shallow areas were dredged to deepen navigable waters. The resultant spoil was typically deposited on nearby shallow flats. Often the shallow flats have supported seagrass, therefore removal of the spoil island can result in the creation of a seagrass and/or mangrove mitigation site. Near Ft. Pierce Inlet in St. Lucie County, FL, a spoil island was removed in 2006. This site has thus far recruited seagrass and mangroves in excess of requirements. Additionally, the dominant seagrass at SL-15 is *Halophila johnsonii* (Johnson's seagrass) which is the only aquatic plant listed as an endangered species.

LAKE SURPRISE CAUSEWAY REMOVAL

In a manner similar to the removal of SL-15, Lake Surprise Causeway will be removed to create seagrass habitat. Based on unpublished observations of subaqueous soils, this area is unique due to the organic soils that occur beneath a thin veneer of recent marine sediments. Also, the enclosed nature of Lake Surprise affords the seagrass in the lake little water exchange with the surrounding marine habitat. Finally, recent algal blooms have created a heightened awareness of the health of Lake Surprise. Both governmental agencies and the public have expressed concern over the health of the lake.

OBJECTIVES AND STUDIES

GOAL

The goal of this project it to better understand the equilibrium states that SL-15 and Lake Surprise will be approaching over the next few years. This objective will be accomplished via spatiotemporal

modeling of the SL-15 and Lake Surprise benthic habitats. To achieve this objective, sites will be sampled and the vegetation, water, and soils accessed via a spatial sampling scheme, through time, and modeled geostatistically.

SPECIFIC OBJECTIVES AND STUDIES

To support this goal, the research was divided into four studies:

- Spatiotemporal Dynamics of SL-15
- Spatiotemporal Dynamics of Lake Surprise
- The Influence of Subaqueous Soils on the Subtropical Seagrass *Halodule wrightii*
- Assessment of SL-15 Restoration Trajectories
-

ORGANIZATION OF THIS REPORT

This report is presents the research in the following chapter format:

CHAPTER 1: INTRODUCTION

Presented in this chapter is the research, which outlines the objectives, and presents the organization of the report.

CHAPTER 2: SL-15 SPOIL ISLAND AS A SEAGRASS AND MANGROVE MITIGATION SITE

Presented in this chapter is a review of the design and permit-compliance monitoring of SL-15. Results are summarized and presented to provide a context for the parallel spatial monitoring presented in Chapter 3.

CHAPTER 3: SPATIOTEMPORAL DYNAMICS OF SL-15

Presented in this chapter is the spatiotemporal monitoring of the mangrove planter and seagrass recruitment areas. The purpose is to track the growth and development of the planted mangroves and recruited seagrass. Design improvements and lessons learned will be presented for improving future mitigation/restoration sites.

CHAPTER 4: SPATIOTEMPORAL DYNAMICS OF LAKE SURPRISE

Presented in this chapter is the spatiotemporal monitoring of seagrass, water, and soil within Lake Surprise. The purpose is to determine whether post-construction changes to the lake occur. If they occur, the spatial sampling scheme is designed to isolate and track the extent of change.

CHAPTER 5: INFLUENCE OF SUBAQUEOUS SOILS ON THE SUBTROPICAL SEAGRASS *HALODULE WRIGHTII*

Presented in this chapter is an experiment to determine the affect of soil properties on seagrass transplants. The purpose is to provide supporting information for understanding areas of seagrass recruitment such as SL-15.

CHAPTER 6: ASSESSMENT OF SL-15 RESTORATION TRAJECTORIES

Presented in this chapter are the findings of an investigation to determine soil formation trajectories at SL-15. This provides foundational knowledge for understanding soil development at SL-15.

CHAPTER 7: PARTICLE SIZE DISTRIBUTION ANALYSIS OF MARINE SOILS

While analyzing soils in previous studies, it became clear that formation of Calcium Oxalate was enhancing the silt and clay fractions of the soil. An alternative method for determining particle size was explored. This literature review is the result of those investigations. It provides improved particle size distribution analysis used in the analysis of soils in the proceeding chapters.

CHAPTER 8: CONCLUSIONS

This section summarizes the four studies that constitute this research effort to better understand SL-15 and Lake Surprise.

CHAPTER 2: SL-15 SPOIL ISLAND AS A SEAGRASS AND MANGROVE MITIGATION SITE

INTRODUCTION

Pressure from coastal development has caused the destruction of ecologically important mangrove and seagrass habitats. These habitats are vital for coastal wildlife, storm surge protection, economically-important fish and shellfish nurseries, and biogeochemical processes (Alongi, 2002; Duarte, 2002; Zedler and Kercher, 2005). They also play an important role in Florida's economy. The Florida Department of Environmental Protection (FDEP) estimates Florida's seagrass and mangroves to provide over 40 billion dollars a year in ecosystem services. These habitats play a vital role in the state's recreational and commercial fisheries industries. To offset impacts to mangrove and seagrass habitats state and federal law requires mitigation when these habitats are destroyed.

Most restoration projects have been by regulatory agencies, or mitigation projects for wetland fill or excavation allowed by permits. In North America, mangrove restoration often involves re-establishment of natural hydrologic and tidal regimes, planting of mangrove propagules, or planting marsh plants as nurse species (Proffitt and Devlin, 2005). Florida has been the site of numerous coastal restoration or mitigation projects although most have never been assessed for more than a few years after project completion. Some examples of these projects include the Tampa Bay Shoreline Initiative project (Beever et al., 2004), the Marine Resource Council Shoreline Restoration project, and numerous bridge and urban developmental permitted mitigation projects.

The purpose of this manuscript is to explain a unique shoreline mitigation project in St. Lucie County, Florida in which a spoil island was converted into mangrove and seagrass habitat. This is the first time that this type of mitigation project has been attempted. The success of this project may lead to the use of several of Spoil Island as future mitigation sites. The environmental monitoring which was used to monitor this first time mitigation project will be compared and discussed throughout this manuscript.

BACKGROUND

The Indian River Lagoon (IRL) is a shallow barrier island lagoon which stretches 250 km along the Atlantic coast of Florida with an average depth of 1.7 m and a width of 3 km (Smith, 1987). The IRL includes a collection of three estuaries, the Mosquito Lagoon, Banana River, and Indian River, located along Florida's Atlantic coast. The IRL is separated from the Atlantic Ocean by a barrier island system that is interrupted by five inlets (Ponce de Leon, Sebastian, Fort Pierce, St. Lucie, and Jupiter) providing exchange with marine waters. The IRL stabilized over the past 6,000 years during a period of minimal sea level fluctuation, resulting in increased barrier island stability (Davis et al., 1992). All seven subtropical species of seagrass found in the western hemisphere occur in the IRL. In addition, the IRL is home to rich aquatic life including 397 species of fish (Gilmore, 1995). Dredging of the Atlantic Intracoastal Waterway (ICW) by the U. S. Army Corps of Engineers between 1953 and 1961 resulted in the creation of 137 spoil islands within the IRL. Dredge spoils were typically placed in very shallow

seagrass flats near the cuts during a time when wetland impacts were ignored. Over the past decades, some spoil islands have become colonized by native, threatened and endangered species and serve as bird rookeries, adding to the ecological diversity of the IRL. However, colonization by exotic and invasive species has also taken place. Islands dominated by exotic and invasive species are potential locations for mitigation efforts.

The Florida Department of Transportation (FDOT) received permits from the South Florida Water Management District (SFWMD) and the U. S. Army Corps of Engineers for the construction of the Frank A. Wacha Bridge in 2001 and the Ernest Lyons Bridge in 2004 (E Sciences Inc., 2008). Both projects spanned the IRL and resulted in the destruction of sea grass and mangrove habitats, requiring mitigation. St. Lucie (SL-15) (27° 28' 40'' N, 80° 19' 23" W), a 5.6 ha spoil island in Ft. Pierce, located approximately 27 km north of the Wacha Bridge and 33 km north of the Lyons Bridge, was selected as the mitigation site. This was one of the first mitigation projects to utilize a spoil to offset the destruction of seagrass and mangrove habitats due to coastal development. Prior to mitigation, SL-15 had a maximum elevation of +9 ft (NGVD) and was primarily vegetated in the island's interior by Australian pine (*Casuarina equisetifolia*) and Brazilian pepper (*Schinus terebinthifolius*), with a fringe dominated by red mangroves (*Rhizophora mangle*), black mangroves (*Avicennia germinans*), and white mangroves (*Laguncularia racemosa*) (Figure 1; Marcus et al., 2006). Extensive seagrass covered sub-tidal flats were present surrounding the island and up to the ICW to the west.

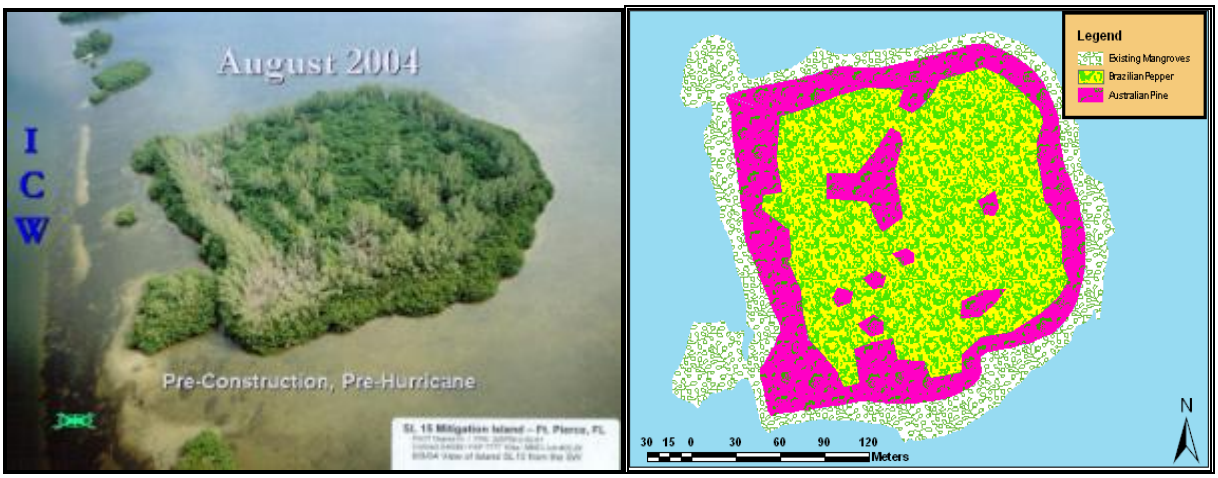

Figure 1. Aerial photograph and species map of spoil Island SL-15 prior to mitigation (Marcus et al., 2006).

Restoration efforts began in March 2005 contracting firm Misener Marine with the removal of exotic vegetation and excess spoil material, the preservation of the mangrove dominated fringe, and the reshaping of the island to create areas for seagrass and mangrove habitat. In total, the mitigation resulted in the creation of 3.38 acres of seagrass habitat, 4.89 acres of mangrove habitat, and the improvement of 2.43 acres of upland berm or transitional habitat. In order to facilitate restoration of SL-

15 a temporary trestle was built on the west side of the island on which a conveyor belt transported material off the island onto a barge. The trestle was constructed to minimize impacts to existing seagrass beds in the area which were known to contain Johnson's seagrass (*Halophila johnsonii*). This trestle remained in place for ten months, during which time approximately 77,000 yd^3 of spoil material were removed. Exotics were removed though clearing and burning. The 1.24 ha island fringe, dominated by red, white, and black mangroves, and the 2.38 acre of uplands forming a berm (+4.0 NGVD) along SL-15's western, northern, and eastern sides were preserved (Figure 2).

Figure 2. Transformation of SL-15. Aerial photos taken in (A) June 2004, (B) May 2005, (C) November 2005, and (D) December, 2005. The constructed seagrass, mangrove and upland habitats are shown by the yellow polygons (Fischler, 2006).

A mangrove planting area within the upland berm was leveled to an elevation of +1.0 NGVD. A seagrass recruitment area was created with a maximum depth of -1.5 NGVD and connected to the IRL via the creation of seven flushing channels separated by six small, islands containing preexisting vegetation including red mangroves (Figure 3). Approximately 23,000 red mangrove seedlings were planted in December 2005 within the mangrove planting zone on 1 m centers, while the upland berm, was planted with native vegetation including button woods (*Conocarpus erectus*), sea grapes (*Coccoloba uvifera*), coco plums (*Chrysobalanus icaco*), and myrsines (*Myrsine guianensis*) (Marcus et al., 2006). Bare-root cordgrass (*Spartina alterniflora*) was planted in the transition zones between the mangrove planting area and seagrass recruitment area, and sea oxeye daisy (*Borrichia arborescens*) and seashore paspalum (*Paspalum distichum*) were planted between the upland berm area and the mangrove planting area

(Marcus et al., 2006). The seagrass embayment was not planted, allowing for recruitment by seagrass species from the naturally surrounding IRL. An additional 8,000 red mangrove seedlings were planted in September 2007 (E Sciences Inc., 2008).

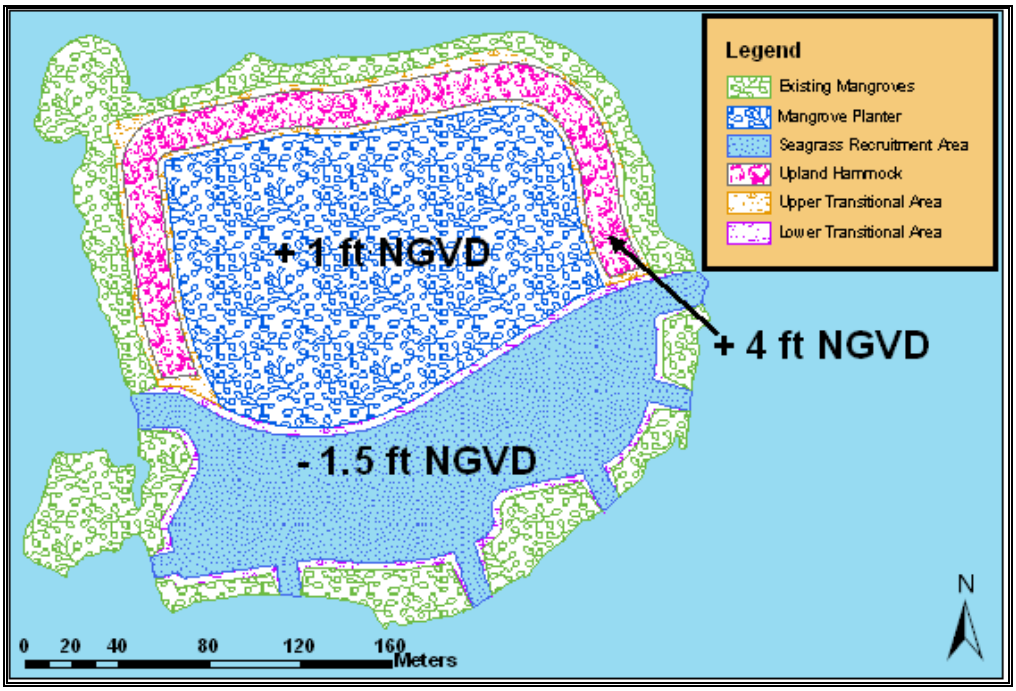

Figure 3. Elevations of the constructed seagrass, mangrove, and upland habitats with the flushing channels (Marcus et al., 2006).

ECOLOGICAL MONITORING METHODS

E Sciences Inc. was contracted to conduct monitoring of the mangrove planting area and seagrass embayment. The schedule for monitoring included a baseline (time zero) event in January 2006 followed by four quarterly events (April, July, October 2006, January 2007) to be followed by two semiannual monitoring events (July 2007, January 2008) and two annual monitoring events (July 2008, July 2009), as set forth by the permits (Marcus et al., 2006). National Marine Fisheries Service aided in determining the monitoring methodologies for survivorship in the mangrove planter area and recruitment in the seagrass area. Since the upland hammock purpose was to reduce erosion and not required by permit, there was no survivorship requirement. However the upland berm was monitored using three permanent 100 m^2 plots to quantify vegetation coverage by species. The mangrove planting area was monitored on a semiannual basis using four randomly placed transects (50 m x 2 m) to quantify survivorship (Figure 4). Success criteria within the mangrove planting area was established as 80% or more survivorship (cover) for planted or recruited mangroves and 5% or less coverage of exotic species. The seagrass embayment was monitored quarterly for the first year after completed construction,

semiannually for the second year after completed construction, and annually for the third, fourth and fifth years after completed construction. Monitoring was carried out by quantifying seagrass shoot counts and coverage using the Braun-Blanquet Classification system within 20 randomly placed paired 1 m^2 quadrates (Figure 4). Twenty additional randomly placed paired 1 m^2 quadrates were also conducted outside of the seagrass recruitment area within the surrounding IRL. Success criteria in the seagrass embayment were set at 3% (approximate Braun-Blanquet cover class of 0.5) or more coverage by year three, 6% (approximate Braun-Blanquet cover class of 2.0) or greater coverage by year four, and 10% (approximate Braun-Blanquet cover class of 2.0) or more coverage by year five with supplemental plantings required if these criteria were not met by year five. Observations of fauna were quantified within the mangrove planting area and along three randomly established transects within the seagrass embayment. Additionally, fiddler crab burrow counts were conducted in 50 randomly established 1 m^2 quadrates within the mangrove planting area. Permit also required the documentation of any other wildlife such as birds, invertebrates, or fish.

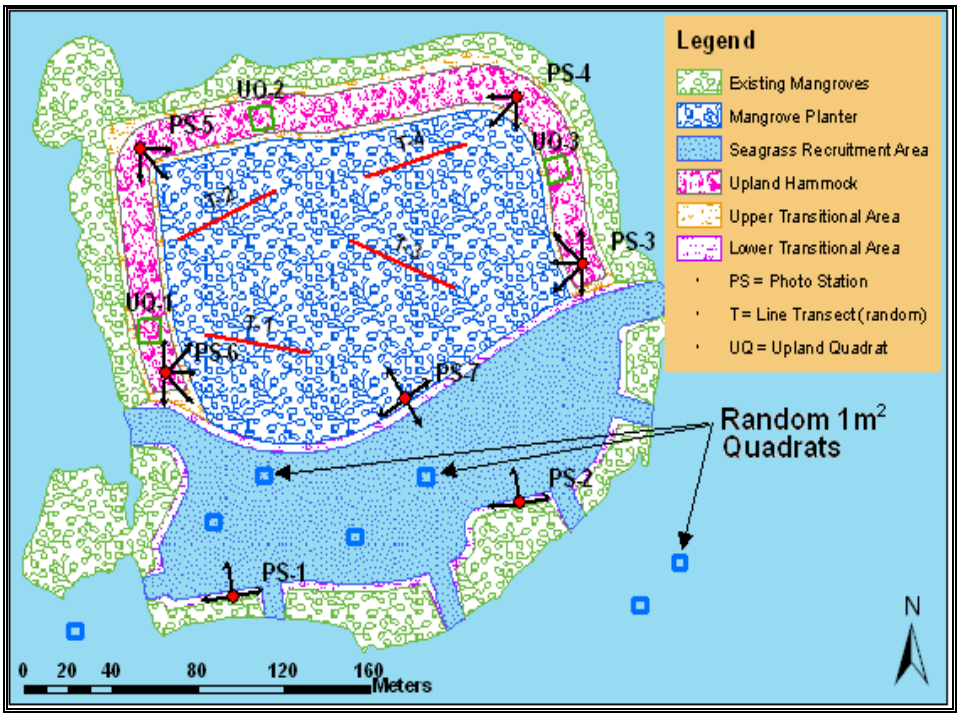

Figure 4. SL-15 post construction mangrove and seagrass monitoring locations (Marcus et al., 2006).

RESULTS

Results reported during by E Sciences during the first year (2006) indicated that the mangrove planting area contained an average mangrove survivorship of 41% (.41 trees/m^2)(Figure 5). During this time a supplemental mangrove planting was scheduled in order to facilitate permit requirements. The seagrass

embayment was reported to have recruited 1.5% coverage (Braun-Blanquet cover class of 0.2) while the coverage in the control area outside the seagrass recruitment area was to have 56% seagrass coverage (Figure 6). The species composition of year one was determined to be 12% *H. wrightii*, 42.5% *H. johnsonii*, 5% *S. filiforme*, 10% *H. decipiens*, and the remainder being bare substrate (Figure 7).

For year two (2007) results reported by E Sciences indicated that the mangrove planting area contained an average mangrove survivorship of 84% (.84 trees/m^2). During this time the mangrove planting area was in compliance with the required survivorship set forth by the permits. The seagrass embayment was reported to have recruited 1.7% coverage (Braun-Blanquet cover class of 0.3) while the coverage of seagrass outside the embayment was 32%. The species composition of year two was determined to be 5% *H. wrightii*, 35% *H. johnsonii*, 7.5% *S. filiforme*, and the remainder being bare substrate.

E Sciences year three (2008) results indicated that the mangrove planting area contained an average mangrove survivorship of 108% (1.08 trees/m^2). During this time the mangrove planting area exceeded the permitted required survivorship set forth by the permits and no additional mangroves seedlings were planted. The seagrass embayment was reported to have recruited 7.5% coverage (Braun-Blanquet cover class of 0.8) while the coverage of seagrass of the control area outside the embayment was 24.5%. The 7.5% seagrass coverage in the seagrass embayment exceeded the permit requirement of 3% coverage after the first three years. The species composition of year three was determined to 25% *H. wrightii*, 67.5% *H. johnsonii*, 37.5% *S. filiforme*, and the remainder being bare substrate.

Results reported by E Sciences for year four (2009) indicated that the mangrove planting area contained an average mangrove survivorship of 87% (.87 trees/m^2). The seagrass embayment was reported to have recruited 10% coverage (Braun-Blanquet cover class of 1.0) while the coverage of seagrass of the control area outside the embayment was 77.6%. Mangroves did not meet survivorship criteria during this time but percent cover appeared to be adequate (>80%). During this time the seagrass embayment exceeded and met the required mangrove survivorship set forth by the permits. The species composition of year four was determined to 50% *H. wrightii*, 7.5% *T. testudinum*, 20% *H. johnsonii*, and the remainder being bare substrate.

E Sciences year five (2010) final results indicated that the mangrove planting area contained an average mangrove survivorship of 72% (.72 trees/m^2). The seagrass embayment was reported to have recruited 16.9% coverage (Braun-Blanquet cover class of 1.4) while the coverage of seagrass of the control area outside the embayment was 87.5%. During this time the mangrove planting area and the seagrass embayment exceeded and met the required mangrove survivorship set forth by the permits. The species composition of year five was determined to 5% *H. wrightii*, 2.5% *T. testudinum*, 52.5% *H. johnsonii*, and the remainder being bare substrate. Once permit requirements were met management of the island was transferred over to the FDEP.

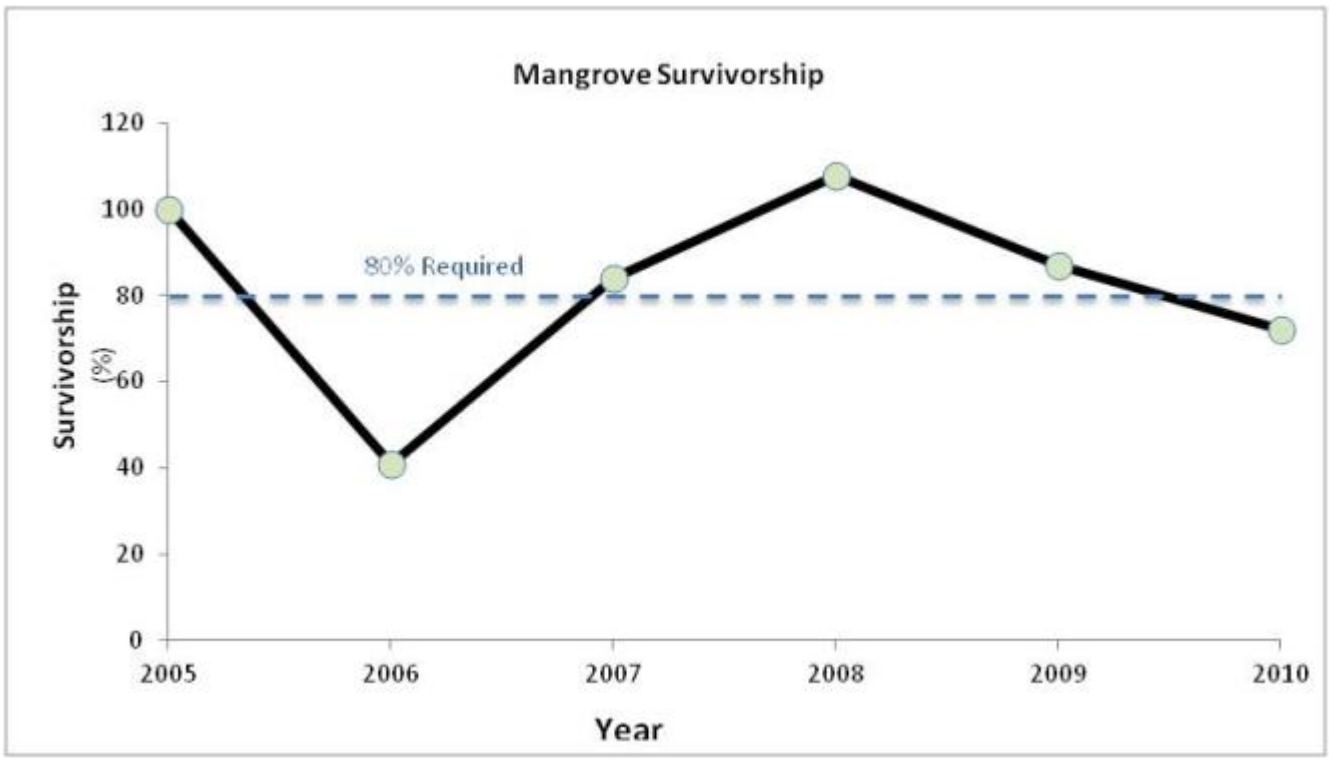

Figure 5. Mangrove survivorship from planting to year 5 (2010).

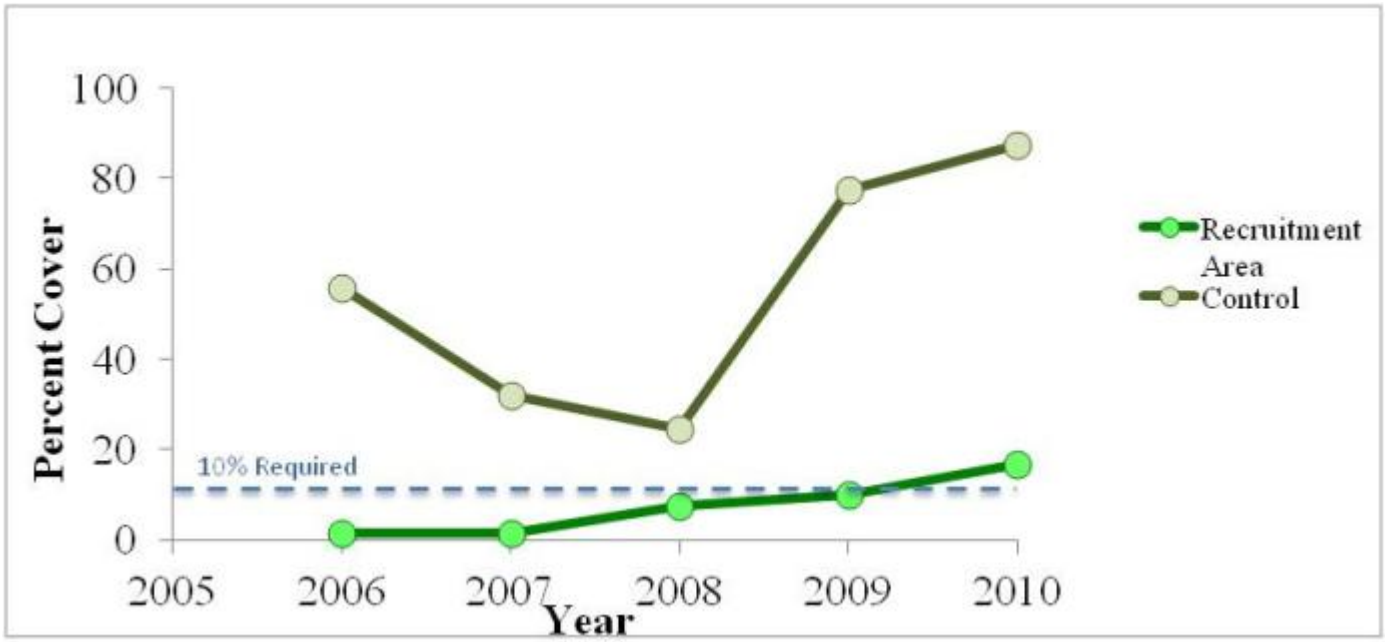

Figure 6. Percent cover of seagrass in recruitment area versus control plots for years 2005-2010.

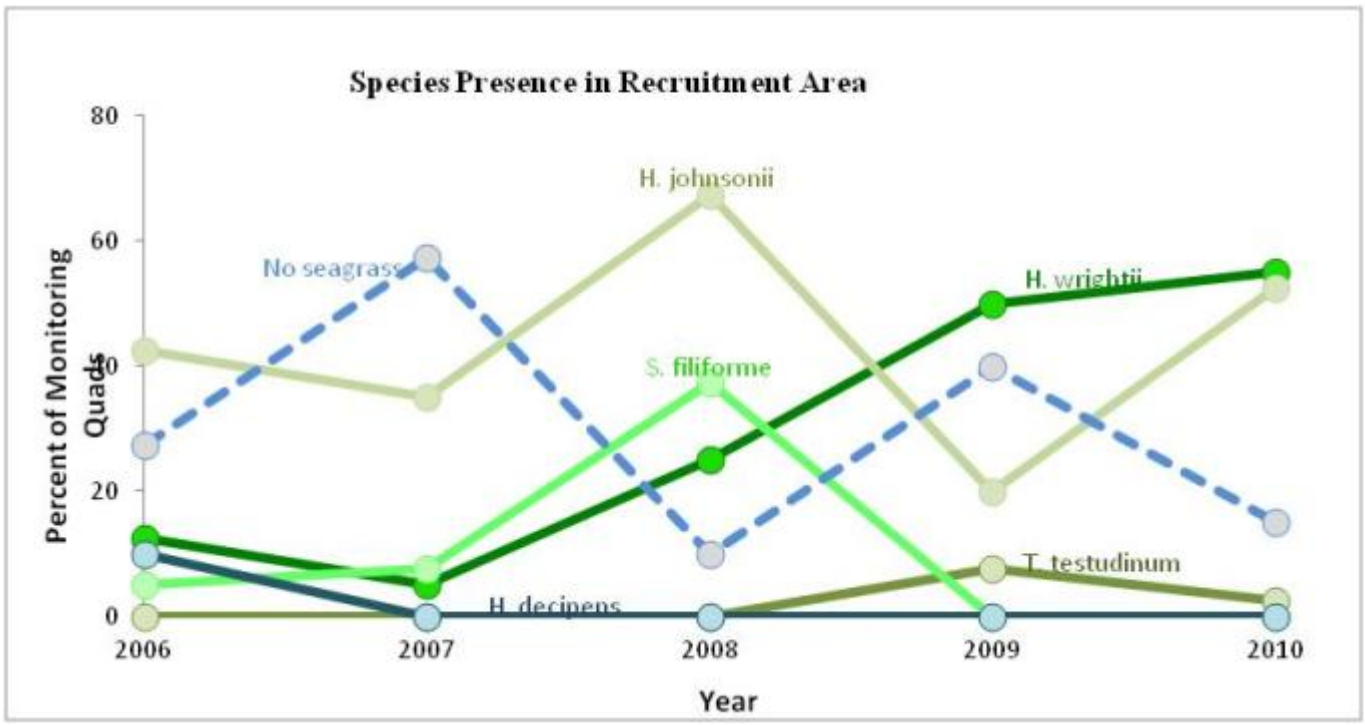

Figure 7. Seagrass species presence in recruitment areas for years 2005-2010.

DISCUSSION

Restoration success suggested that spoil islands in the Indian River Lagoon can be utilized for future mitigation sites. The restoration of SL-15 surpassed the required 80% survivorship for mangroves and met the 10% coverage of seagrass for the five year period set forth by the SFWMD. The restored spoil island was also observed to have suitable habitat for fiddler crabs and other marine wildlife including juvenile fish, birds, and insects. Since there are few natural areas with the sufficient conditions to promote seagrass growth, this mitigation method may serve as a useful way to offset future impacts to seagrass habitats. The success of this project showed that spoil island restoration can be a useful mitigation tool in the IRL, which is considered one of the most diverse estuaries in the United States (Gilmore et al., 1983).

Although this project was deemed a success, valuable lessons can be learned from the results and observations which could be used to improve future similar projects. The random transects utilized to calculate mangrove survivorship and seagrass coverage did not allow for accurate spatial analysis. Spatial analysis can give project managers a better understanding of which areas where successful and unsuccessful, which can lead to more efficient project management. In future projects, monitoring transect methods may be altered to give more detailed spatial analysis data. In addition to mangrove and seagrass monitoring, other biological and chemical parameters linked to the health of these two key species could have also been monitored, such as soil properties. Since utilizing spoil islands as mitigation sites is a relatively new approach to offset coastal habitat destruction, monitoring efforts continued to

ensure the success of this project was ongoing. The second part of this manuscript addresses soil properties, spatial patterns, and post permit monitoring of the mitigation site SL-15.

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CHAPTER 3: SPATIOTEMPORAL DYNAMICS OF SL-15

INTRODUCTION

The data presented in the previous chapter indicate SL-15 has maintained the minimum required mangroves and seagrass. The spatiotemporal patterns of this vegetation were not assessed. These patterns would provide important an understanding about the vegetative succession that should take place within the newly created mitigation areas of SL-15.

OBJECTIVES

Where do the mangroves grow tallest? Are there areas of mangrove decline? Does natural recruitment occur? Are untargeted species of mangroves present and if so, where and in what concentrations? Where are seagrass accumulating? What interactions can be observed between different species of plants and ecological succession occurs?

These questions were unanswered by the design in the previous chapter, so a spatiotemporal approach was chosen to answer these questions. The goal of this project it to capture the spatial patterns of SL-15 vegetation changes through time. This objective was being accomplished via spatiotemporal modeling of the SL-15 mangrove and seagrass mitigation areas. To achieve this objective, sites were sampled semiannually: winter 2008, summer 2009, winter 2009, summer 2010, winter 2010, and summer 2011. The data collected were modeled geostatistically to create maps of the vegetative patterns.

SPECIFIC OBJECTIVES

Additional and Post Permit monitoring was to establish whether the success criteria set forth by the previous permits continued after post construction monitoring. Since this approach to mitigate for coastal development is relatively new the previous monitoring may not have been long enough to determine whether this project had a long term success and survey techniques may need to be modified. Post monitor sampling methods differed from post construction methods in order to better assess spatial patterns and more intensely survey the study sites. Samples were taken from a total of eighty-three mangrove sites and ten seagrass sites. Soil sample analysis was not a post construction permit requirement however these analysis can be compared to healthy mangrove forest and seagrass bed soil analysis in order to give more detailed perspective to the success of this project.

METHODS

MANGROVES

From summer 2009 to summer 2011, 83 sites within the mangrove planting area were sampled biannually (Figure 8). At each site the number of mangroves, species, and height was recorded within a two meter circle. In order to obtain mangroves per square meter the following formula was used:

$$\text{Observed number of mangroves}/(4*\pi) = \text{Mangroves}/m^2 \text{ or mangrove density}/m^2$$

For each site average and maximum height was calculated for each species of mangrove. In summer 2011 the shortest canopy diameter length, longest canopy diameter length, and the height to the start of the canopy was recorded (Figure 9). Canopy area and volume were calculated using the formulas shown in Figure 10 for each tree. These values were then added together to obtain total calculated are and volume for each site. In order to obtain calculated area and volume per square meter the following formulas were used:

$$\text{Calculated total mangrove area } m^2/(4*\pi) = \text{Calculated total mangrove area } m^2/m^2$$

$$\text{Calculated total mangrove volume } m^3/(4*\pi) = \text{Calculated total mangrove volume } m^3/m^2$$

The spatial data for mangrove density, average height, maximum height, calculated total canopy area, and calculated total canopy volume were analyzed using the Geostatistical Analyst extension of ArcGIS 9.3. Ordinary Kriging was used to interpolate the data and estimate measurements and calculations over the mangrove planting area. The resultant models were converted to 1 m raster files in ESRI GRID format for display and spatial analysis.

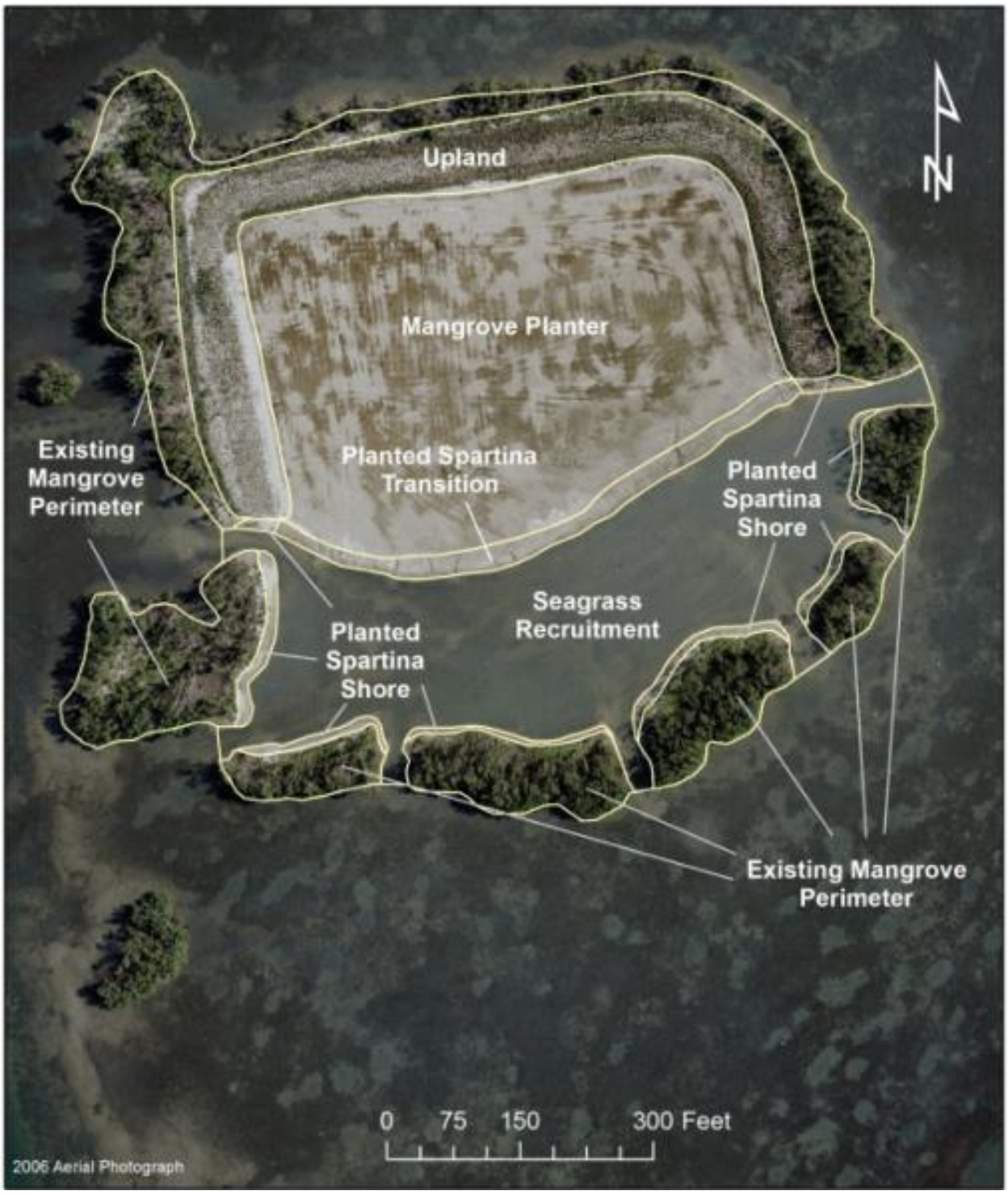

Figure 8. Study areas, existing mangrove perimeter, upland, and planted S. alterniflora zones for SL-15 additional and post permit monitoring.

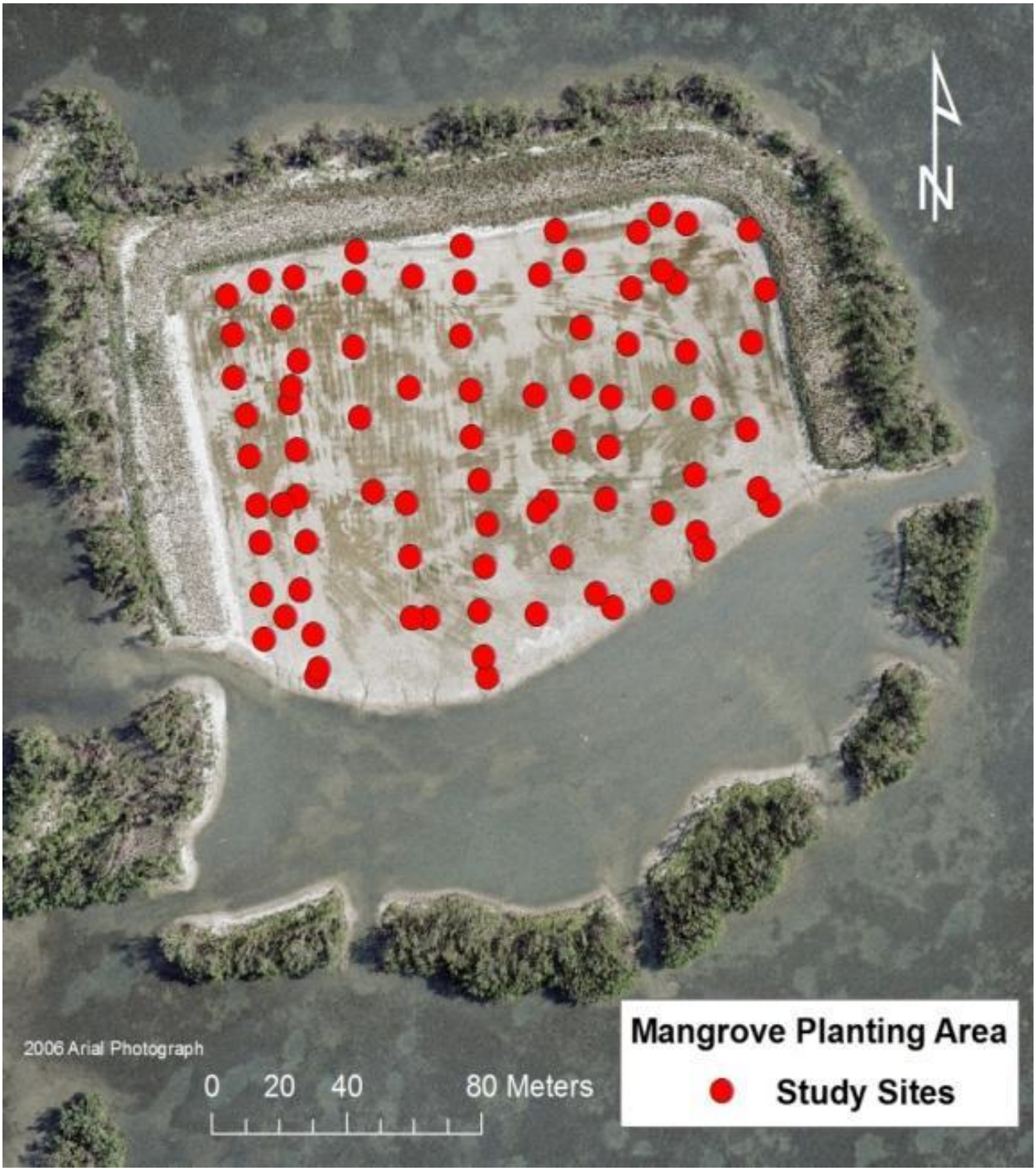

Figure 9. SL-15 post monitoring mangrove planting area study site locations.

Figure 10. Additional mangrove metrics taken in summer 2011.

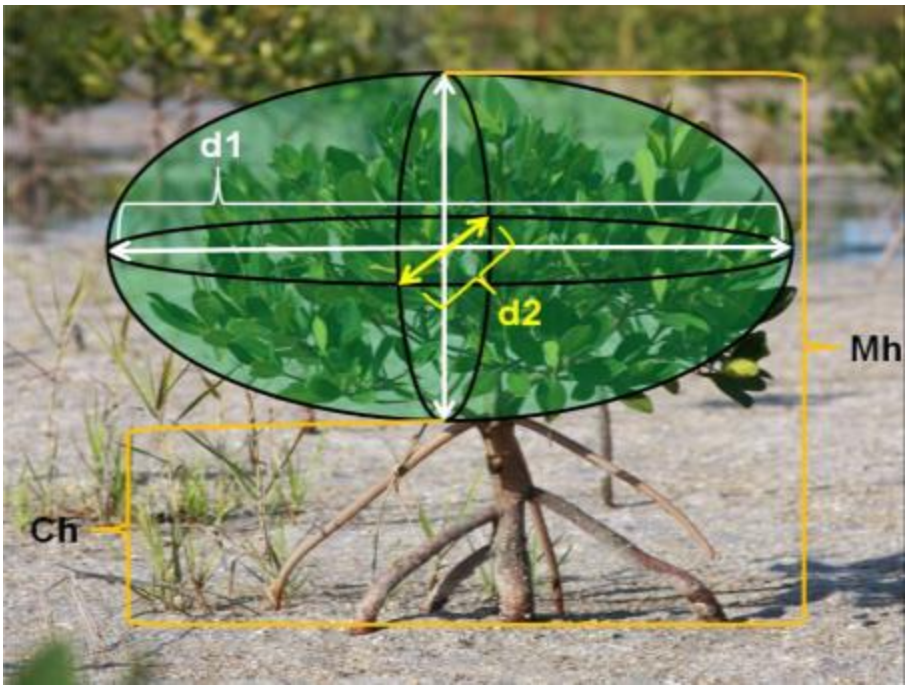

Figure 11. Calculated mangrove canopy area was derived from using the following equation: $\text{Calculated Area} = \pi * \left(\frac{d1}{2}\right) * \left(\frac{d2}{2}\right)$. Calculated mangrove volume was derived from using the following equation: $\text{Calculated Volume} = \pi * \left(\frac{d1}{2}\right) * \left(\frac{d2}{2}\right) * \frac{Mh-Ch}{2}$.

SEAGRASS

From winter 2008 to summer 2011 seagrass coverage, algae coverage, seagrass density, and species composition was recorded semiannually for ten study sites within the seagrass embayment (Figure 12). From winter 2008 to winter 2010 data was also collected biannually from the three control sites located directly east-northeast of SL-15. Seagrass density was determined by quantifying seagrass shoot counts within the ten study sites and three control sites using one m^2 quadrates. Seagrass coverage was recorded as percent ground cover and using the Braun-Blanquet Classification system in order to compare data to post construction seagrass coverage results. In addition to seagrass coverage algae coverage was also recorded within the one m^2 quadrates. Average seagrass coverage, algae coverage, seagrass density, and species composition was calculated for the seagrass embayment and the control sites.

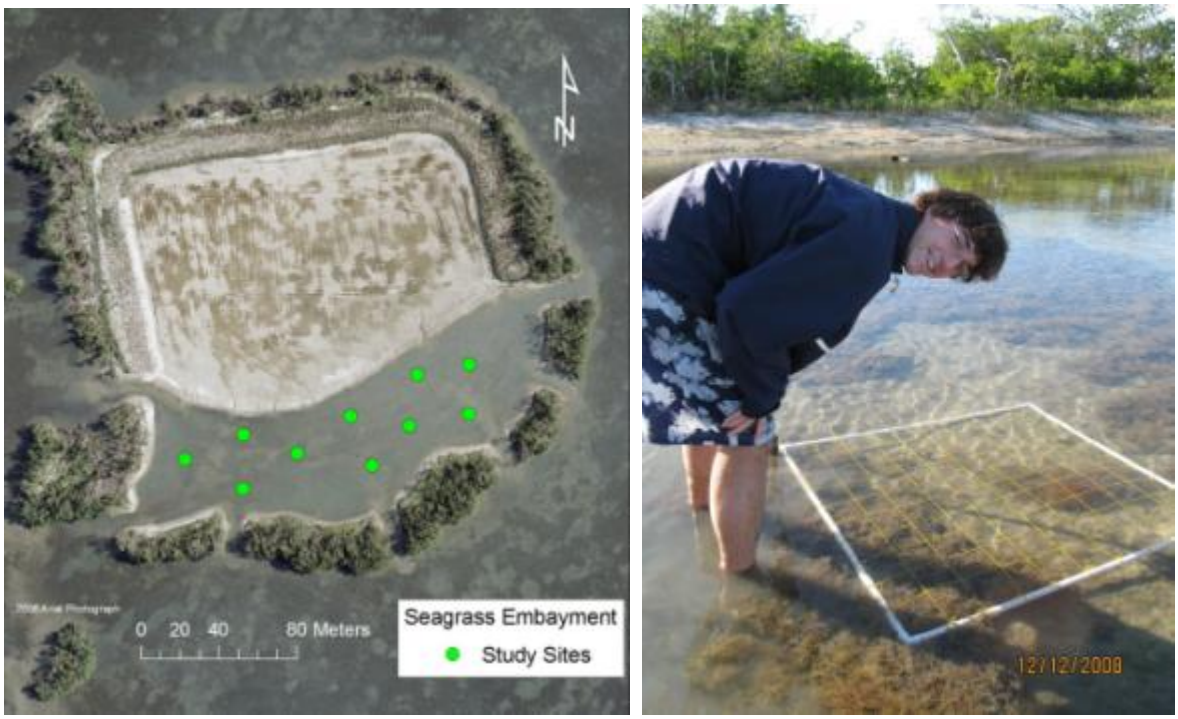

Figure 12. SL-15 seagrass spatiotemporal monitoring study site location (A). One meter quad used for visualizing percent cover (B)

The spatial data for seagrass coverage, algae coverage and seagrass density were analyzed using the Geostatistical Analyst extension of ArcGIS 9.3. Ordinary Kriging was used to interpolate the data and estimate coverage and density within the seagrass embayment. The resultant models were converted to 1 m raster files in ESRI GRID format for display and spatial analysis.

SOILS

From winter 2008 to summer 2011 soil samples were obtain biannually from the mangrove planting area. For comparison soil samples were taken from the upland control biannually from winter 2008 to winter 2010. Within the seagrass embayment and its control sites, soil samples were taken biannually from winter 2008 to winter 2010. The first 0-5 cm portion of the soil at all sites were sampled using polycarbonate core tubes (Figure 13). These soils were characterized by analyzing for particle-size distribution (Day, 1965), bulk density (Blake and Hartge 1986), and organic matter content (Heiri *et. al*, 2001). Total phosphorus within each soil sample was determined by HCl extraction (Reddy et al., 1998). The spatial data for particle size, organic matter, and total phosphorus were analyzed using the Geostatistical Analyst extension of ArcGIS 9.3. Ordinary Kriging was used to interpolate the data. The resultant models were converted to 1 m raster files in ESRI GRID format for display and spatial analysis.

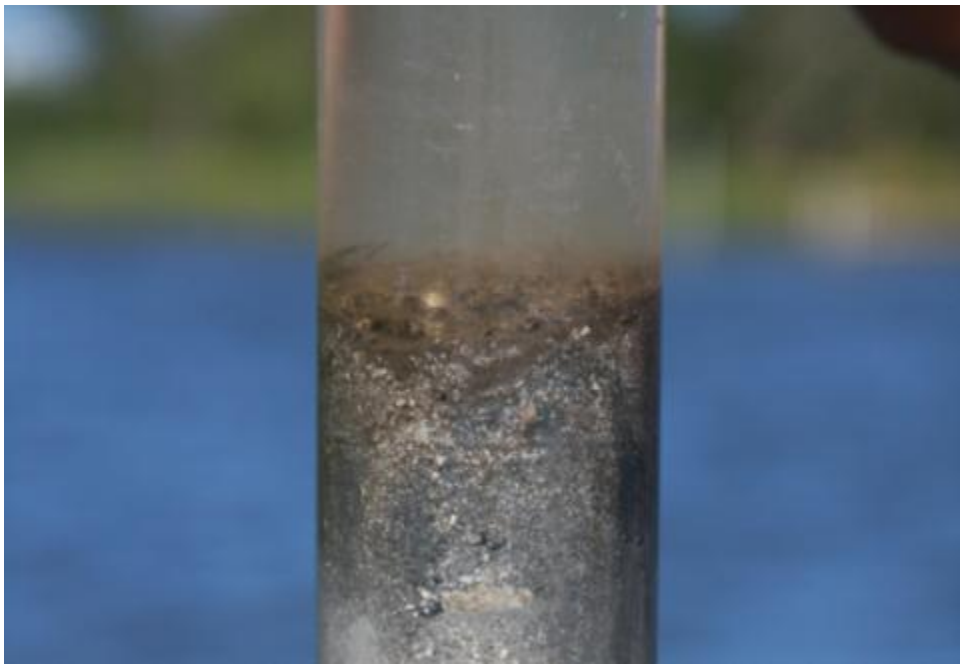

Figure 13. Soil sample obtained from the seagrass embayment area.

RESULTS

A complete set of full-page maps is presented in Appendix A of this report.

MANGROVES

In summer 2009 the mangrove planting area contained an average *R. mangle* survivorship of 150% or a density of 1.5 trees/m^2 (SD=1.7). During this time the mangrove planting area surpassed the survivorship compliance requirement for *R. mangle* set forth by the previous permits. The average height of *R.*

mangle in the planting area was 57.0 cm (SD=29.8). The average maximum height for *R. mangle* in the planting area was 71.0 cm (SD=26.2). The densest places for *R. mangle* were along the transition area and the back north section of planting area. The tallest *R. mangle* trees were concentrated in the northwest corner of the planting are and the southeast corner of the transition area. During this time, *A. germinans* had a density of 0.4 trees/m^2 (SD=0.6) in the planting area. The average height within the planting area for *A. germinans* was 37.3 cm (SD=47.8) with an average maximum of 42.1 cm (SD=53.2). The middle of the transition area was where the highest density of *A. germinans* could be found. The tallest *A. germinans* were located on the eastern side of the planting area. During this sampling event, *L. racemosa* density was found to be 0.5 trees/m2 (SD=1.3) within the planting area. The average height within the planting area for *L. racemosa* was 30.9 cm (SD=38.1) with an average maximum of 37.2 cm (SD=47.2). The eastern portion of the transition area was where the highest density of *L. racemosa* could be found. The tallest *L. racemosa* were also located in the eastern portion of the transition area and the northeast corner of the planting area.

In winter 2009 the mangrove planting area contained an average *R. mangle* survivorship of 150% or a density of 1.5 trees/m^2 (SD=2.5). During this time the mangrove planting area surpassed the survivorship compliance requirement for *R. mangle* set forth by the previous permits. The average height of *R. mangle* in the planting area was 65.6 cm (SD=29.8). The average maximum height for *R. mangle* in the planting area was 84.6 cm (SD=39.2). The densest places for *R. mangle* were along the transition area and the back north section of planting area. The tallest *R. mangle* trees were concentrated in the northwest corner of the planting are and the southeast corner of the transition area. During this time *A. germinans* had a density of 0.5 trees/m^2 (SD=0.7) in the planting area. The average height within the planting area for *A. germinans* was 39.3 cm (SD=47.2) with an average maximum of 48.6 cm (SD=56.3). The middle of the transition area and the northwest corner of the planting area was where the highest density of *A. germinans* could be found. The tallest *A. germinans* were located on the eastern portion of the transition area. During this sampling event *L. racemosa* density was found to be 0.4 trees/m2 (SD=1.1) within the planting area. The average height within the planting area for *L. racemosa* was 39.3 cm (SD=51.3) with an average maximum of 44.3 cm (SD=57.4). The eastern portion of the transition area was where the highest density of *L. racemosa* could be found. The tallest *L. racemosa* were also located in the eastern portion of the transition area and the northeast corner of the planting area.

In summer 2010, the mangrove planting area contained an average *R. mangle* survivorship of 170%, a density of 1.7 trees/m^2 (SD=2.8). During this time, the mangrove planting area surpassed the survivorship compliance requirement for *R. mangle* set forth by the previous permits. The average height of *R. mangle* in the planting area was 69.4 cm (SD=24.9). The average maximum height for *R. mangle* in the planting area was 86.6 cm (SD=36.5). The densest places for *R. mangle* were along the transition area, the northwest corner, and the northeast section of planting area. The tallest *R. mangle* trees were concentrated in the northwest corner of the planting are and the southeast corner of the transition area. During this time *A. germinans* had a density of 1.0 trees/m^2 (SD=1.6) in the planting area. The average height within the planting area for *A. germinans* was 32.8 cm (SD=39.2) with an average maximum of 55.2 cm (SD=59.4). The middle of the transition area, the western portion and eastern portion of the planting area was where the highest density of *A. germinans* could be found. The

tallest *A. germinans* were located in the transition area, western and eastern corners of the planting area. During this sampling event *L. racemosa* density was found to be 0.8 trees/m2 (SD=1.8) within the planting area. The average height within the planting area for *L. racemosa* was 33.3 cm (SD=42.6) with an average maximum of 50.0 cm (SD=59.2). The eastern portion of the transition area and the northwest corner of the planting area was where the highest density of *L. racemosa* could be found. The tallest *L. racemosa* were also located in transition section of the planting area.

In winter 2010 the mangrove planting area contained an average *R. mangle* survivorship of 140% or a density of 1.4 trees/m^2 (SD=1.3). During this time the mangrove planting area surpassed the survivorship compliance requirement for *R. mangle* set forth by the previous permits. The average height of *R. mangle* in the planting area was 76.9 cm (SD=30.0). The average maximum height for *R. mangle* in the planting area was 97.3 cm (SD=38.0). The densest places for *R. mangle* were along the transition section of planting area. The tallest *R. mangle* trees were concentrated in the transition area and the northwest corner of the planting are. During this time *A. germinans* had a density of 1.0 trees/m^2 (SD=1.8) in the planting area. The average height within the planting area for *A. germinans* was 48.1 cm (SD=41.7) with an average maximum of 71.9 cm (SD=65.9). The northeast corner of the planting area was where the highest density of *A. germinans* could be found. The tallest *A. germinans* were located in the transition section of the planting area. During this sampling event *L. racemosa* density was found to be 0.5 trees/m2 (SD=0.8) within the planting area. The average height within the planting area for *L. racemosa* was 44.5 cm (SD=55.1) with an average maximum of 57.8 cm (SD=74.2). The eastern portion of the transition area was where the highest density of *L. racemosa* could be found. The tallest *L. racemosa* were also located in transition area, northeastern and northwestern corners of the planting area.

In summer 2011 the mangrove planting area contained an average *R. mangle* survivorship of 210% or a density of 2.1 trees/m^2 (SD=4.2). During this time the mangrove planting area surpassed the survivorship compliance requirement for *R. mangle* set forth by the previous permits. The average height of *R. mangle* in the planting area was 73.9 cm (SD=18.7). The average maximum height for *R. mangle* in the planting area was 112.6 cm (SD=32.2). The average canopy area and volume for *R. mangle* within the planting area was 0.3 m^2 leaf area/m^2 (SD=0.2) and 0.1 m^3 of leaf volume/m^2 (SD=0.1), respectively. The average density of *R. mangles* new recruits during this time was 0.5 new recruits/m^2 (SD=1.2). The densest places for *R. mangle* trees were along the transition section of planting area. The tallest *R. mangle* trees were concentrated in the transition area and the northwest corner of the planting area. *R. mangle* canopy area and volume were the highest in the east transition area and in the northwest section of the planting area. *R. mangle* new recruits were most abundant within the eastern portion of the transition section of the planting area. During this time *A. germinans* had a density of 1.6 trees/m^2 (SD=2.1) in the planting area. The average height within the planting area for *A. germinans* was 52.7 cm (SD=36.2) with an average maximum of 112.1 cm (SD=60.6). The average canopy area and volume for *A. germinans* within the planting area was 0.1 m^2 leaf area/m^2 (SD=0.2) and 0.1 m^3 of leaf volume/m^2 (SD=0.2), respectively. The average density of *A. germinans* new recruits during this time was 1.3 new recruits/m^2 (SD=2.0).The northwest and northeast corners of the planting area was where the highest density of *A. germinans* could be found. The tallest *A. germinans* were located in the transition area and northeastern corner of the planting area. *A. germinans* canopy area was greater in the eastern half of

the planting area, while canopy volume was the highest in the middle of transition area and in the northeast corner of the planting area. *A. germinans* new recruits were concentrated within the east and west corners the planting area. During this sampling event *L. racemosa* density was found to be 1.2 trees/m2 (SD=3.3) within the planting area. The average height within the planting area for *L. racemosa* was 51.2 cm (SD=45.7) with an average maximum of 84.9 cm (SD=61.2). The average canopy area and volume for *L. racemosa* within the planting area was 0.1 m^2 leaf area/m^2 (SD=0.4) and 0.1 m^3 of leaf volume/m^2 (SD=0.4), respectively. The average density of *L. racemosa* new recruits during this time was 0.7 new recruits/m^2 (SD=2.1).The eastern portion of the transition area, the northeastern and northwestern corners was where the highest density of *L. racemosa* could be found. The tallest *L. racemosa* were also located in transition area and the upper northern portion of the planting area. *L. racemosa* canopy area and canopy volume was greatest in the east part of the transition area and the northeast corner of the planting area. *L. racemosa* new recruits were concentrated within the east and west corners the planting area.

SEAGRASS

In winter 2008 the seagrass embayment was reported to have 3.5% (SD=6.5) average seagrass ground coverage while the outside seagrass control sites were found to have 54.0% (SD=5.3). Average Braun-Blanquet cover class was found to be 0.6 (SD=0.7) for the seagrass embayment and 4.0 (SD=0.0) for the control sites. Average Braun-Blanquet percent coverage for the study area was found to be 2.5% (SD=5.6) while the control was reported to have 62.5% (SD=0.0) for the control site. Average seagrass density was determined to be 34.4 shoots/m^2 (SD=41.0) for the seagrass embayment and 71.7 shoots/m^2 (SD=25.7) for the outside control sites. The species density within the seagrass embayment during this sampling period was composed of 3.7 shoots/m^2 (SD=7.9) for *H. wrightii*, 30.6 shoots/m^2 (SD=40.8) for *H. johnsonii*, 0.0 shoots/m^2 (SD=0.0) for *S. filiforme*, and 0.0 shoots/m^2 (SD=0.0) for *T. testudinum*. The species density for the seagrass control sites during this sampling period was composed of 0.0 shoots/m^2 (SD=0.0) for *H. wrightii*, 0.0 shoots/m^2 (SD=0.0) for *H. johnsonii*, 58.0 shoots/m^2 (SD=20.4) for *S. filiforme*, and 13.7 shoots/m^2 (SD=23.7) for *T. testudinum*. Average algae coverage during this time for the seagrass embayment was found to be 71.8% (SD=24.4) and 17.3% (SD=14.2) for the seagrass control sites. Seagrass coverage was at the lowest in the middle of the seagrass embayment at this time. Seagrass density was the highest towards the southeastern portion of the seagrass embayment near a flushing channel. Algae coverage was uniform over the study area during this time.

In summer 2009 the seagrass embayment was reported to have 1.0% (SD=1.2) average seagrass ground coverage while the outside seagrass control sites were found to have 73.7% (SD=45.6). Average Braun-Blanquet cover class was found to be 0.2 (SD=0.2) for the seagrass embayment and 4.0 (SD=1.7) for the control sites. Average Braun-Blanquet percent coverage for the study area was found to be 2.0% (SD=1.1) while the control was reported to have 63.3% (SD=41.9) for the control site. Average seagrass density was determined to be 0.0 shoots/m^2 (SD=0.1) for the seagrass embayment and 710.0 shoots/m^2 (SD=255.9) for the outside control sites. The species density within the seagrass embayment during this sampling period was composed of 0.0 shoots/m^2 (SD=0.1) for *H. wrightii*, 0.0 shoots/m^2 (SD=0.1) for *H. johnsonii*, 0.0 shoots/m^2 (SD=0.0) for *S. filiforme*, and 0.0 shoots/m^2 (SD=0.0) for *T. testudinum*. The

species density for the seagrass control sites during this sampling period was composed of 0.0 shoots/m^2 (SD=0.0) for *H. wrightii*, 0.0 shoots/m^2 (SD=0.0) for *H. johnsonii*, 698.0 shoots/m^2 (SD=203.9) for *S. filiforme*, and 12.0 shoots/m^2 (SD=20.8) for *T. testudinum*. Average algae coverage during this time for the seagrass embayment was found to be 25.7% (SD=11.7) and 0.0% (SD=0.0) for the seagrass control sites. Seagrass coverage was highest in the eastern portion of the seagrass embayment at this time. Seagrass density was the uniform throughout the seagrass embayment. Algae coverage was highest in the western portion of the seagrass embayment during this time.

In winter 2009 the seagrass embayment was reported to have 0.3% (SD=0.5) average seagrass ground coverage while the outside seagrass control sites were found to have 36.7% (SD=23.1). Average Braun-Blanquet cover class was found to be 0.5 (SD=0.2) for the seagrass embayment and 2.7 (SD=0.6) for the control sites. Average Braun-Blanquet percent coverage for the study area was found to be 1.2% (SD=0.8) while the control was reported to have 30.0% (SD=13.0) for the control site. Average seagrass density was determined to be 0.0 shoots/m^2 (SD=0.1) for the seagrass embayment and 24.0 shoots/m^2 (SD=24.8) for the outside control sites. The species density within the seagrass embayment during this sampling period was composed of 0.0 shoots/m^2 (SD=0.1) for *H. wrightii*, 0.0 shoots/m^2 (SD=0.1) for *H. johnsonii*, 0.0 shoots/m^2 (SD=0.0) for *S. filiforme*, and 0.0 shoots/m^2 (SD=0.0) for *T. testudinum*. The species density for the seagrass control sites during this sampling period was composed of 0.0 shoots/m^2 (SD=0.0) for *H. wrightii*, 0.0 shoots/m^2 (SD=0.0) for *H. johnsonii*, 15.0 shoots/m^2 (SD=10.0) for *S. filiforme*, and 9.0 shoots/m^2 (SD=15.6) for *T. testudinum*. Average algae coverage during this time for the seagrass embayment was found to be 36.8% (SD=29.5) and 13.3% (SD=23.1) for the seagrass control sites. In winter 2009 seagrass coverage and density were the uniform throughout the seagrass embayment. Algae coverage was concentrated near the west flushing channels of the seagrass embayment during this time.

In summer 2010 the seagrass embayment was reported to have 8.3% (SD=10.7) average seagrass ground coverage while the outside seagrass control sites were found to have 43.3% (SD=49.3). Average Braun-Blanquet cover class was found to be 1.2 (SD=1.1) for the seagrass embayment and 3.0 (SD=1.7) for the control sites. Average Braun-Blanquet percent coverage for the study area was found to be 10.3% (SD=11.9) while the control was reported to have 39.2% (SD=41.9) for the control site. Average seagrass density was determined to be 95.0 shoots/m^2 (SD=144.7) for the seagrass embayment and 483.3 shoots/m^2 (SD=144.3) for the outside control sites. The species density within the seagrass embayment during this sampling period was composed of 7.5 shoots/m^2 (SD=16.9) for *H. wrightii*, 87.5 shoots/m^2 (SD=143.5) for *H. johnsonii*, 0.0 shoots/m^2 (SD=0.0) for *S. filiforme*, and 0.0 shoots/m^2 (SD=0.0) for *T. testudinum*. The species density for the seagrass control sites during this sampling period was composed of 0.0 shoots/m^2 (SD=0.0) for *H. wrightii*, 0.0 shoots/m^2 (SD=0.0) for *H. johnsonii*, 450.0 shoots/m^2 (SD=86.6) for *S. filiforme*, and 33.3 shoots/m^2 (SD=57.7) for *T. testudinum*. Average algae coverage during this time for the seagrass embayment was found to be 14.5% (SD=13.0) and 0.0% (SD=0.0) for the seagrass control sites. Seagrass coverage and density were the uniform throughout the seagrass embayment, while algae coverage was concentrated in the west half of the seagrass embayment during this time.

In winter 2010 the seagrass embayment was reported to have 0.7% (SD=1.1) average seagrass ground coverage while the outside seagrass control sites were found to have 71.7% (SD=36.2). Average Braun-Blanquet cover class was found to be 0.2 (SD=0.2) for the seagrass embayment and 4.3 (SD=1.2) for the control sites. Average Braun-Blanquet percent coverage for the study area was found to be 1.0% (SD=1.3) while the control was reported to have 70.1% (SD=28.9) for the control site. Average seagrass density was determined to be 10.0 shoots/m^2 (SD=31.6) for the seagrass embayment and 700.0 shoots/m^2 (SD=204.6) for the outside control sites. The species density within the seagrass embayment during this sampling period was composed of 10.0 shoots/m^2 (SD=31.6) for *H. wrightii*, 0.0 shoots/m^2 (SD=0.0) for *H. johnsonii*, 0.0 shoots/m^2 (SD=0.0) for *S. filiforme*, and 0.0 shoots/m^2 (SD=0.0) for *T. testudinum*. The species density for the seagrass control sites during this sampling period was composed of 0.0 shoots/m^2 (SD=0.0) for *H. wrightii*, 0.0 shoots/m^2 (SD=0.0) for *H. johnsonii*, 683.3 shoots/m^2 (SD=200.5) for *S. filiforme*, and 16.7 shoots/m^2 (SD=28.9) for *T. testudinum*. Average algae coverage during this time for the seagrass embayment was found to be 70.5% (SD=26.5) and 0.0% (SD=0.0) for the seagrass control sites. In winter 2010 seagrass coverage and density were highest in the eastern half of the seagrass embayment. Algae coverage was concentrated in the western three quarters of the seagrass embayment during this time.

In summer 2011 the seagrass embayment was reported to have 0.5% (SD=1.6) average seagrass ground coverage. Average Braun-Blanquet cover class was found to be 0.1 (SD=0.2) and average Braun-Blanquet percent coverage for the study area was found to be 0.3% (SD=0.8). The outside seagrass control sites were not monitored during this sampling event. During this time the seagrass embayment did not meet the seagrass coverage compliance requirement set forth by the previous permits. Average seagrass density was determined to be 5.0 shoots/m^2 (SD=15.8) for the seagrass embayment. The species density within the seagrass embayment during this sampling period was composed of 5.0 shoots/m^2 (SD=15.8) for *H. wrightii*, 0.0 shoots/m^2 (SD=0.0) for *H. johnsonii*, 0.0 shoots/m^2 (SD=0.0) for *S. filiforme*, and 0.0 shoots/m^2 (SD=0.0) for *T. testudinum*. Average algae coverage during this time for the seagrass embayment was found to be 68.7% (SD=27.3). Seagrass coverage and density were highest in the eastern half of the seagrass embayment, while algae coverage was uniform throughout the seagrass embayment during this time.

SOILS

In winter 2008 the average percent sand and fine soil particles in the mangrove planting area were found to be 87.8% and 12.2% (SD=10.8), respectively, compared to the upland control proportions of 91.8% and 8.2% (SD=2.6). Average percent organic matter was found to be 1.7% (SD=0.5) in the planting area and 3.6% (SD=1.2) in its control. Average bulk density and total phosphorus in the planter soil was found to be 1.1 (SD=0.2) and 247.4 mg P/kg soil (SD=131.9). The upland control was found to have a bulk density of 0.8 (SD=0.0) and 463.7mg P/kg soil (SD=378.4). During this time the average percent sand and fine particles in the seagrass embayment was found to be 80.4% and 19.6% (SD=7.8), respectively, compared to the seagrass control proportions of 77.1% and 22.9% (SD=9.8). Average percent organic matter was found to be 2.6% (SD=0.7) in the seagrass embayment and 2.7% (SD=1.4) in its control. Average bulk density and total phosphorus in the seagrass embayment soil was found to be 0.8 (SD=0.2) and 503.3 mg P/kg soil (SD=128.7). The seagrass control was found to have a bulk density of

0.6 (SD=0.2) and 560.6 mg P/kg soil (SD=172.6).). Percent fine particles in the soil were higher in the seagrass embayment than in the mangrove planting area and were relatively uniform in the seagrass embayment. Within the planting area fine particles were the highest on the east side. Organic matter was higher in the seagrass embayment and uniform. There were low percentages of organic matter in the northern border of the mangrove planting area during this time. There was more phosphorus in the soil in the seagrass embayment which was concentrated near the western flushing channels. Phosphorus in the mangrove planting area was concentrated in the middle portion of the planting area.

In summer 2009 the average percent sand and fine soil particles in the mangrove planting area were found to be 89.1% and 10.9% (SD=4.1), respectively, compared to the upland control proportions of 84.5% and 15.5% (SD=10.5). Average percent organic matter was found to be 1.4% (SD=0.5) in the planting area and 5.3% (SD=2.9) in its control. Average bulk density and total phosphorus in the planter soil was found to be 1.3 (SD=0.2) and 418.0 mg P/kg soil (SD=74.3). The upland control was found to have a bulk density of 1.1 (SD=0.1) and 636.0 mg P/kg soil (SD=98.4). During this time the average percent sand and fine particles in the seagrass embayment was found to be 77.9% and 22.1% (SD=6.1), respectively, compared to the seagrass control proportions of 86.2% and 13.8% (SD=6.1). Average percent organic matter was found to be 2.6% (SD=0.8) in the seagrass embayment and 2.2% (SD=1.0) in its control. Average bulk density and total phosphorus in the seagrass embayment soil was found to be 1.1 (SD=0.5) and 619.8 mg P/kg soil (SD=52.7). The seagrass control was found to have a bulk density of 1.0 (SD=0.1) and 706.7 mg P/kg soil (SD=25.9).). Percent fine particles in the soil were higher in the seagrass embayment than in the mangrove planting area and were relatively uniform in the seagrass embayment. Within the planting area fine particles were the highest on the east side during this sampling period. Organic matter was higher in the seagrass embayment and uniform. Organic matter was higher on the east half of the mangrove planting area. There was more phosphorus in the soil in the seagrass embayment which was relatively uniform. Phosphorus in the mangrove planting area did not showed a distinct spatial pattern.

In winter 2009 the average percent sand and fine soil particles in the mangrove planting area were found to be 92.2% and 7.8% (SD=3.5), respectively, compared to the upland control proportions of 93.3% and 6.7% (SD=4.6). Average percent organic matter was found to be 1.2% (SD=0.3) in the planting area and 3.5% (SD=3.4) in its control. Average bulk density and total phosphorus in the planter soil was found to be 1.6 (SD=0.1) and 348.7 mg P/kg soil (SD=137.6). The upland control was found to have a bulk density of 1.0 (SD=0.5) and 644.3 mg P/kg soil (SD=61.1). During this time the average percent sand and fine particles in the seagrass embayment was found to be 85.4% and 14.6% (SD=5.2), respectively, compared to the seagrass control proportions of 78.2% and 21.8% (SD=9.8). Average percent organic matter was found to be 1.6% (SD=0.5) in the seagrass embayment and 2.0% (SD=0.8) in its control. Average bulk density and total phosphorus in the seagrass embayment soil was found to be 1.3 (SD=0.3) and 572.2 mg P/kg soil (SD=52.8). The seagrass control was found to have a bulk density of 1.0 (SD=0.3) and 655.8 mg P/kg soil (SD=9.5). Percent fine particles in the soil were higher in the seagrass embayment than in the mangrove planting area and were relatively uniform in the seagrass embayment. During this time fine particles were the highest in the southern half of the planting area. Organic matter was higher in the seagrass embayment the highest percentage in the western middle

part of the embayment. Organic matter was relatively uniform in the mangrove planting area. There was more phosphorus in the soil in the seagrass embayment which did not exhibit any spatial pattern. Phosphorus in the mangrove planting area was highest in the southeast corner.

In summer 2010 the average percent sand and fine soil particles in the mangrove planting area were found to be 92.1% and 7.9% (SD=3.2), respectively, compared to the upland control proportions of 66.0% and 34.0% (SD=10.5). Average percent organic matter was found to be 1.0% (SD=0.2) in the planting area and 3.1% (SD=1.5) in its control. Average bulk density and total phosphorus in the planter soil was found to be 1.6 (SD=0.2) and 356.1 mg P/kg soil (SD=123.9). The upland control was found to have a bulk density of 0.4 (SD=0.2) and 519.3 mg P/kg soil (SD=261.8). During this time the average percent sand and fine particles in the seagrass embayment was found to be 82.7% and 17.3% (SD=6.3), respectively, compared to the seagrass control proportions of 82.1% and 17.9% (SD=1.5). Average percent organic matter was found to be 2.7% (SD=4.1) in the seagrass embayment and 1.0% (SD=0.3) in its control. Average bulk density and total phosphorus in the seagrass embayment soil was found to be 1.2 (SD=0.3) and 443.6 mg P/kg soil (SD=93.2). The seagrass control was found to have a bulk density of 1.2 (SD=0.2) and 496.2 mg P/kg soil (SD=175.9). Percent fine particles in the soil were higher in the seagrass embayment than in the mangrove planting area and were relatively uniform in the seagrass embayment. During this time fine particles were the highest in the southeastern corner of the planting area. Organic matter was higher in the seagrass embayment the highest percentage in the western quarter of the embayment. Organic matter was relatively uniform in the mangrove planting area. Phosphorus was the highest in the middle of the seagrass embayment and eastern half of the mangrove planting area.

In winter 2010 the average percent sand and fine soil particles in the mangrove planting area were found to be 94.0% and 6.0% (SD=3.0), respectively, compared to the upland control proportions of 93.8% and 6.2% (SD=1.7). Average percent organic matter was found to be 1.2% (SD=0.3) in the planting area and 2.0% (SD=1.2) in its control. Average bulk density and total phosphorus in the planter soil was found to be 1.4 (SD=0.2) and 463.4 mg P/kg soil (SD=117.4). The upland control was found to have a bulk density of 1.7 (SD=0.0) and 807.1 mg P/kg soil (SD=250.5). During this time the average percent sand and fine particles in the seagrass embayment was found to be 86.7% and 13.3% (SD=7.3), respectively, compared to the seagrass control proportions of 85.5% and 14.5% (SD=4.3). Average percent organic matter was found to be 1.8% (SD=0.7) in the seagrass embayment and 1.6% (SD=0.2) in its control. Average bulk density and total phosphorus in the seagrass embayment soil was found to be 1.2 (SD=0.3) and 649.9 mg P/kg soil (SD=128.7). The seagrass control was found to have a bulk density of 1.2 (SD=0.1) and 666.8 mg P/kg soil (SD=8.5). Percent fine particles in the soil were higher in the seagrass embayment than in the mangrove planting area. Fine particle were highest in the western edge and the middle section of the seagrass embayment. During this time fine particles were the lowest in the western edge of the planting area. Organic matter was higher in the seagrass embayment than in the mangrove planting area and relatively uniform in both areas. Phosphorus was the highest in the seagrass embayment and relatively uniform in both areas.

In summer 2011 control samples were taken only for the seagrass embayment and phosphorus analysis was not performed during this time. The seagrass embayment was only sampled for organic matter and

bulk density during this sampling period. The average percent sand and fine soil particles in the mangrove planting area were found to be 95.3% and 4.7% (SD=3.5), respectively. Average percent organic matter and bulk density was found to be 1.8% (SD=0.8) and 1.5 (SD=.1), respectively in the planting area. Average percent organic matter was found to be 1.8% (SD=0.7) in the seagrass embayment and 1.4% (SD=0.1) in its control. Average bulk density in the seagrass embayment soil was found to be 1.4 (SD=0.1). Percent fine particles in the soil were highest in the transition section of the mangrove planting area. Organic matter was also highest in the transition section of the mangrove planting area.

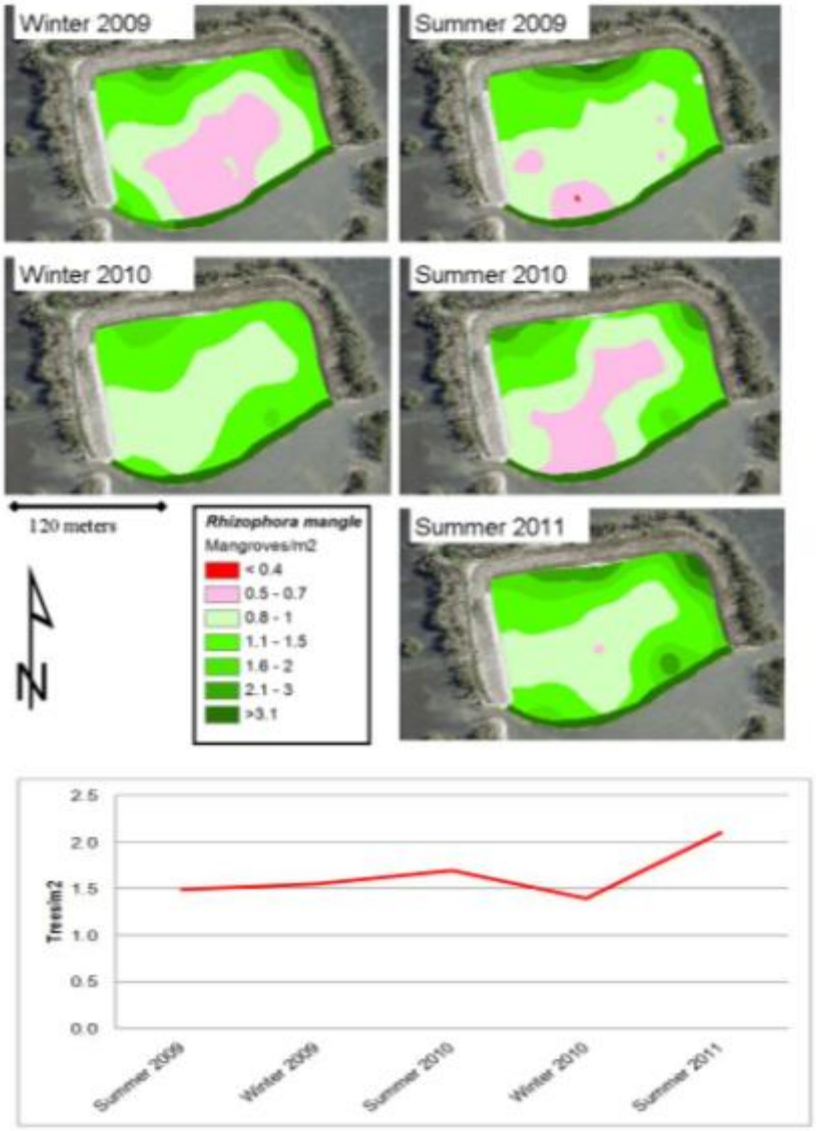

Figure 14. *R. mangle* density from summer 2009 to summer 2011.

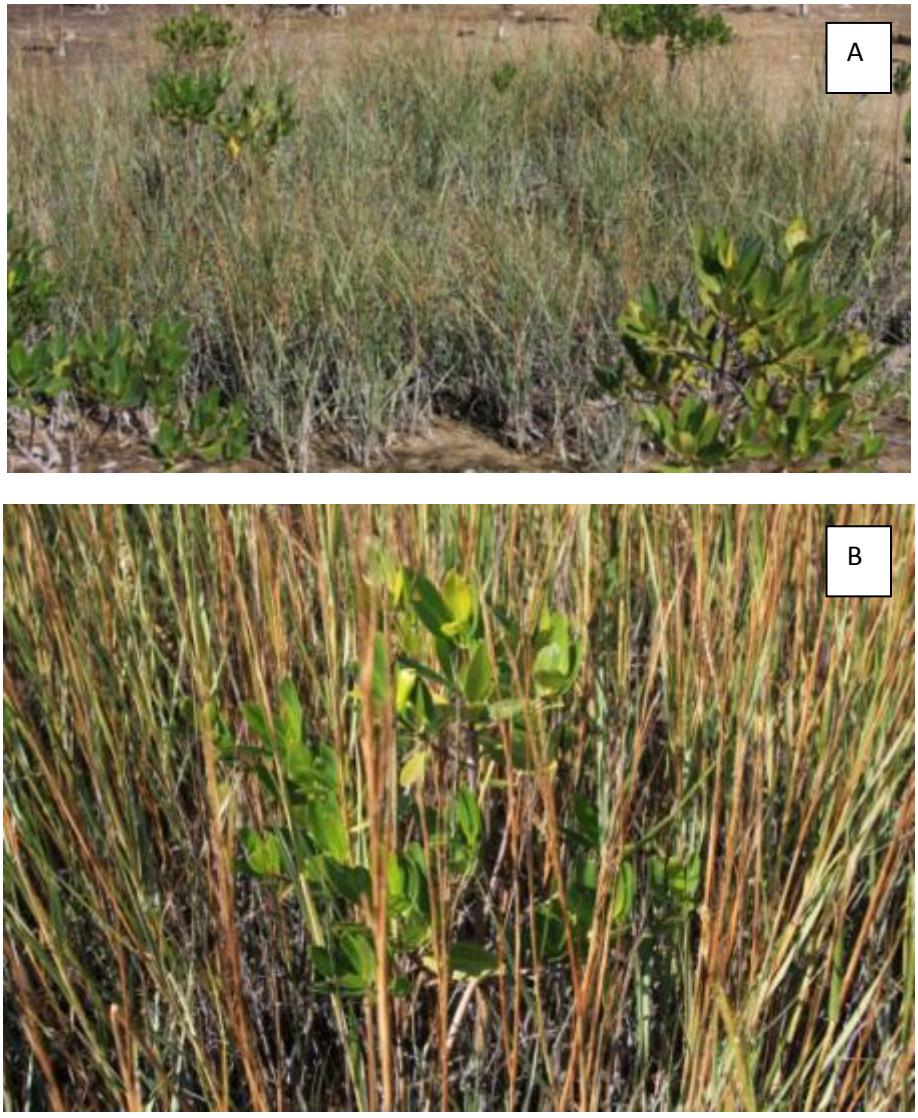

Figure 15. Patch of *Spartina alterniflora* (A) with a close-up view of *R. mangle* recruit within that patch (B).

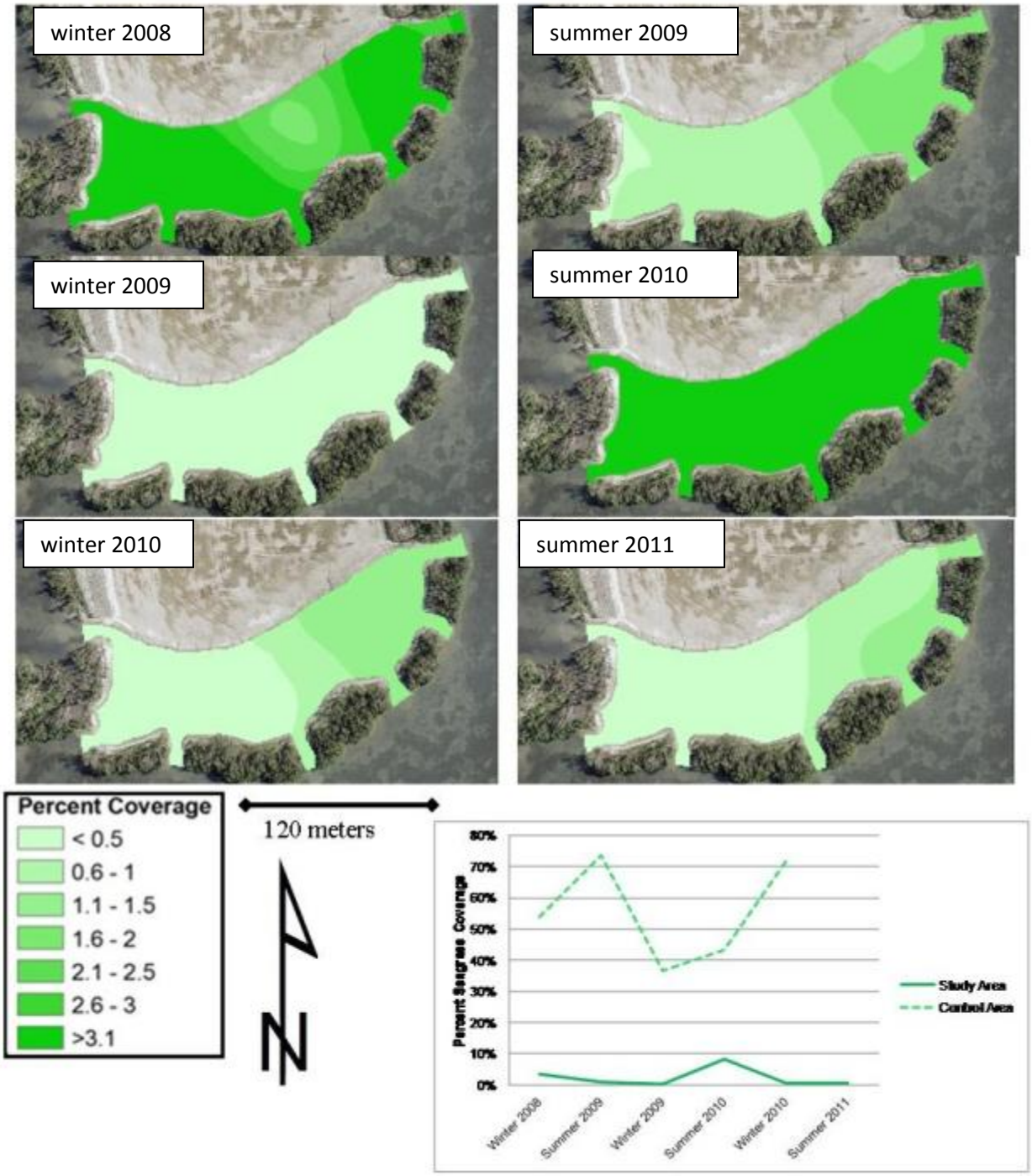

Figure 16. Seagrass ground coverage from winter 2008 to summer 2011.

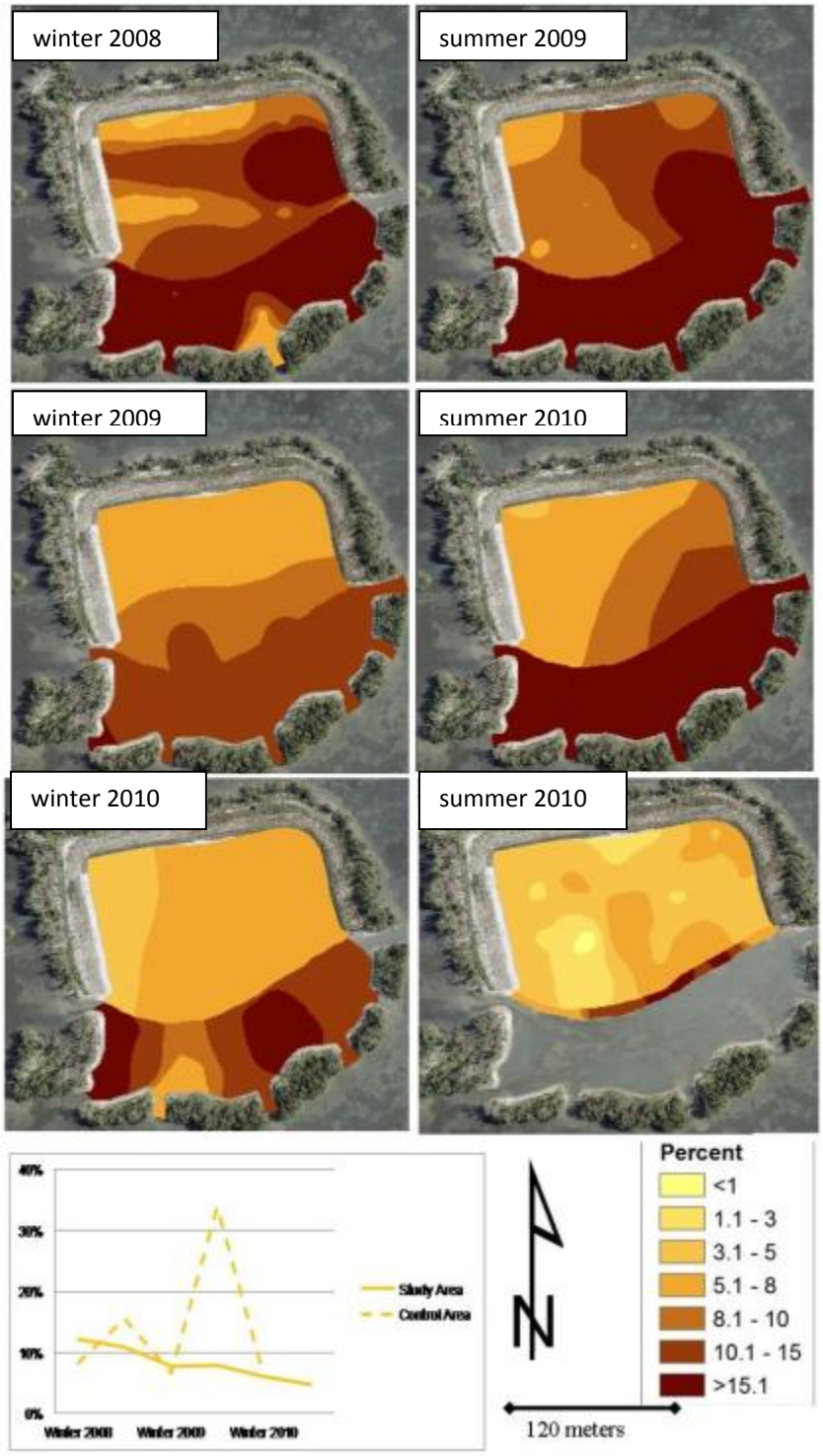

Figure 17. Percent fine particles within the seagrass recruitment area and the mangrove planting area from winter 2008 and summer 2011.

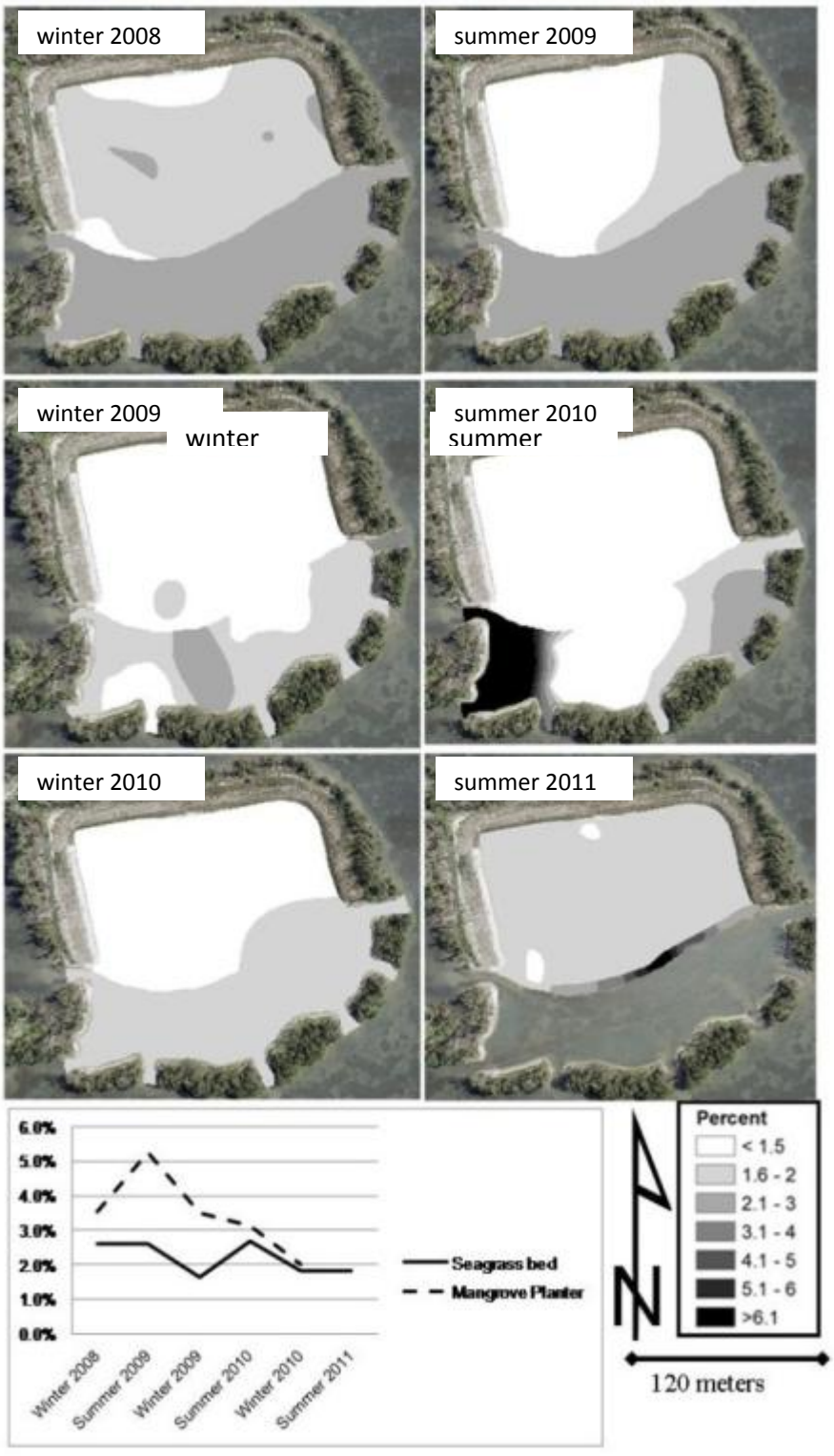

Figure 18. Percent organic matter within the seagrass recruitment area and the mangrove planting area from winter 2008 and summer 2011.

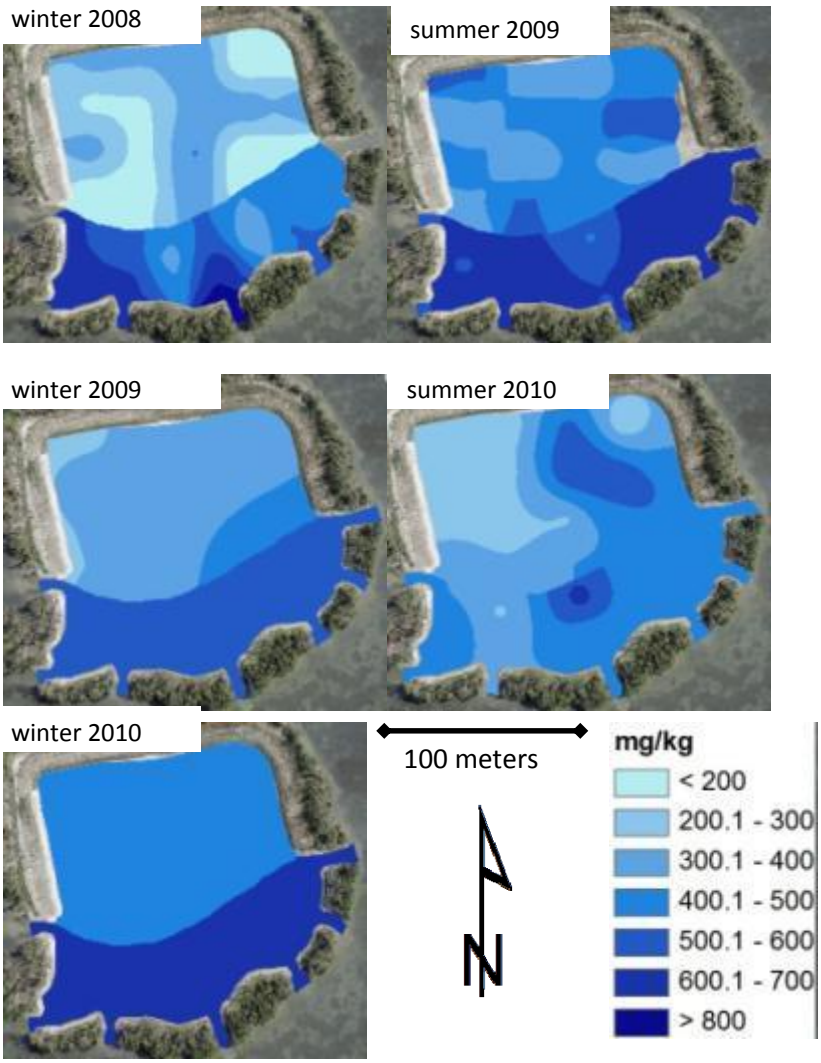

Figure 19. Total phosphorus within the seagrass recruitment area and the mangrove planting area from winter 2008 and summer 2011.

Figure 20. Ponded area on low tide.

DISCUSSION

MANGROVES

R. mangle trees exceeded permit expectations for the entire additional and post monitoring period with a maximum survivorship of 210% (including natural recruitment) in summer 2011. Spatial patterns produced in ArcGIS suggested the densest areas within the mangrove planting area occur in the transitional zone and along the borders of the planting area (Figure 14). In these areas mangrove density was observed to be larger than original planting density, which would imply natural recruitment. The transitional zone was found to have the most natural recruitment. The 2011 *R. mangle* new recruit interpolation (Appendix Figure 103) confirms this. Note in winter 2009 and summer 2010 there were large zones in the center of the planter with low *R. mangle* densities, less than permit requirements. However in winter 2010 *R. mangle* density within the center of the mangrove planting area increased to meet permit requirements, which again suggested natural recruitment. Average height and maximum height increased steadily through time. Spatial analysis of height can be found in the appendix of this manuscript. Incidental observations showed that recruitment seemed to be the highest in areas of dense *S. alterniflora* (Figure 15).

Post construction monitoring of the mangrove planter showed densities less than the calculated densities using the additional and post permit monitoring methods. Possible reasons for the difference may be due to the amount and location of study sites used in the two different sampling methods. Post construction monitoring and the additional and post permit monitoring results both illustrated that the mangrove planting area met and exceeded the permit requirements for survival. Additional and post permit monitoring methods were more adapted for spatial analysis and were more precise due to the larger number of sample sizes. If mangrove survivorship is the main metric to judge the success of a mitigated spoil island, then post construction monitoring methods may be sufficient in evaluating success. However, it may be useful to use information from a combination of both methods to improve the success of the island and help project managers more efficiently make additional planting decisions. Utilizing spatial data for the beginning years can help project managers decide where to plant additional mangroves, if a success criterion is not met.

This project not only met success criteria for *R. mangle* but also recruited other mangrove species, A. germinans and *L. racemosa*, as well. By summer 2011, both of these species exceeded one tree per square meter: 1.6 trees/m^2 and 1.2 trees/m^2 for A. germinans and *L. racemosa*, respectively. Spatial analysis of density, average height, and maximum height can be found in Appendix A. These two species of mangroves were not planted within the mangrove planting area and were all considered to be recruited to the island after construction. R. mangle recruitment is unknown because original planted mangroves were unmarked and recruitment could not be determined. Mangrove recruitment may play an important role in the mangrove survivorship metrics and should be further examined in future projects.

SEAGRASS

Seagrass coverage within the seagrass embayment did not meet permit requirements for all years, according to additional and post permit monitoring methods. Seagrass coverage did meet permit requirements according to the post construction methods for all years. Both methods did showed that *H. johnsonii* was the dominant seagrass and that control sites had larger percent seagrass coverage. The differences in percent coverage of the study sites were largely attributed to sampling locations and methods. Spatial analysis, of the seagrass embayment did not showed a strong spatial pattern and seemed to be variable throughout the years (Figure 16). Algae coverage was variable as well and did not appear to affect seagrass coverage. Shoot density and percent algae coverage interpolations can be found for the additional and post permit monitoring in Appendix A.

The random post construction monitoring sites may have included more of the seagrass beds then the additional and post permit monitoring sites. The additional and post permit monitoring sites were positioned for best spatial analysis. By positioning the study sites for ideal spatial analysis small seagrass beds along the shallower shoreline may have been missed. Silt and turbidity in the deeper areas of the seagrass embayment made it more difficult to obtain shoot count and percent coverage, which also may account for the difference in percent seagrass coverage. Methods utilized in the additional and post permit monitoring also recorded percent ground coverage for seagrass. This percent coverage was lower than that of the coverage calculated by the Braun-Blanquet classification system.

This project was deemed a success however due to the inconsistencies from the two monitoring methods additional monitoring may better illustrate continued successful seagrass mitigation. By using a combination of the two survey methods, a better spatial analysis can be done. It is recommended that the Braun-Blanquet classification system for coverage be used since this is the proffered method to assess seagrass density in previous studies.

SOILS

Percent fine particles of the soil samples within the mangrove planter and upland control both decreased over time. The mangrove planting area had decreased by 61.5% relatively steady over time. The upland control decreased by 25.4% but, was much more variable over time. Healthy mangrove forests normally have a large percentage of fine particles (Boto and Wellington, 1984). In general the fine particles seemed to decrease from the back of the mangrove planter towards the seagrass embayment (Figure 17).

Percent fine particles within the seagrass embayment and its study sites also decreased slightly, however with much more variability. Soils within healthy seagrass beds are typically fine in texture (Ellis, 2006).

Organic matter content within the mangrove planting area did not seem to change with respect to time. Organic matter averaged 1.4% (SD=0.3) between winter 2008 and summer 2011. At the upland control sites the organic matter content was approximately twice as much at any given sampling period. There did not seem to be any clear spatial pattern to organic matter content within the mangrove planting area (Figure 18). The percentage of organic matter within the mangrove planting area was found to be less than that of a healthy mangrove forest (Boto and Wellington, 1984). The mangroves at SL-15 are relatively new and organic matter content may continue to rise as time goes on. The organic matter content found within the seagrass embayment was closely related to the organic matter content found at the seagrass control sites at approximately 2%. A previous study showed that organic matter content of vegetated subaqueous soils is less than 5% (Koch, 2001). Soils found within the seagrass embayment meet this criterion. Organic matter content should not inhibit seagrass recruitment.

Total phosphorus increased both within the mangrove planting area, the seagrass embayment, and their respective controls. Both control sites had slightly higher total phosphorus within the soil samples than their respective study sites. There is not a clear overall spatial pattern however, looking at individual sampling events some spatial patterns do emerge (Figure 19). In the winter 2008, total phosphorus is highest in seagrass embayment near the west flushing channels. The higher amounts of phosphorus in the middle of the mangrove planter appear to follow the middle flushing channel and some of the observed ponding patterns on high tide (Figure 20). These patterns are amplified in the mangrove planter in summer 2009. The total phosphorus values found within the mangrove planting area were similar to values for from Chamber's and Pederson's 2006 study of soil properties in mangrove ecotones in the Shark River Slough basin.

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CHAPTER 4: SPATIOTEMPORAL DYNAMICS OF LAKE SURPRISE

INTRODUCTION

In 2007, the US1 causeway spanning Lake Surprise in Key Largo, FL was removed and replaced with a low spanning bridge (Figure 21). The original construction of US 1 required the digging of a barge canal to allow access to the lake interior. The causeway supporting US 1 hydrologically separated the lake in two portions, referred to in this report as the East Portion and West Portion. The East Portion is hydrologically connected to Jewfish Creek via the barge canal while the West Portion is connected via an access canal. Note that US 1 is not oriented directly North-South. The "east" and "west" names of the lake refer to the directional sides of US 1. Any other references to direction (e.g. Northern shoreline) will be relative to true north, not Northbound US 1.

Figure 21. Map of Lake Surprise and low-lying bridge (inset).

Until 2008, the East Portion has remained hydrologically isolated from the West Portion. Removal of the causeway and the subsequent construction of a low-lying bridge has allowed for water to flow between these two areas of the lake. The lake can now be considered a single water body with two points of connectivity to the outside waters.

The removal of the causeway in 2007 was anticipated to cause an algal bloom. The concern was that the organic matter in causeway soils would add nutrients to the water column once excavation occurred. The resultant algal bloom could then potentially cause a decline in light availability and seagrass converge.

OBJECTIVES

It was unknown whether a seagrass die off would occur and if it did, what would be the extent and severity of the event. Would a decline start in one location and spread or occur evenly across the lake? Would isolated areas of decline, if they were to occur, grow or move? A spatiotemporal approach was chosen to answer these questions.

The goal of this project it to capture the spatial patterns of Lake Surprise seagrass changes through time. This objective will be accomplished via spatiotemporal modeling of the Lake Surprise benthic habitats. To achieve this objective, sites will be sampled and the vegetation, water, and soils accessed via a spatial sampling scheme, through time, and modeled geostatistically.

METHODS

SAMPLE DESIGN

Site Locations

A stratified grid with columns and rows spaced 100 m apart was used to establish sample locations (Figure 22). Each location was semi-permanently marked with PVC. At each site, vegetation, soils, and water were inventoried. The entire set of sites was sampled twice each year for three years: winter 2008, summer 2009, winter 2009, summer 2010, winter 2010, and summer 2011.

Water

At each site, the following water quality was measured using a light meter and YSI 600: light, dissolved oxygen, pH, salinity as electrical conductivity. Light was converted to downward light extinction coefficient (Kd) using Beer's Law:

$$I = I_o \, e^{-Kd * D}$$

Where: I = light available at depth D
Io = light available at water surface
Kd = downward light extinction coefficient
D = water depth at which light was measured

Light extinction coefficient (Kd) is a commonly used parameter for quantifying the clarity of water.

Bathymetry

Bathymetry was not expected to change throughout the duration of the sampling. Rather than record water depth readings at each point, more points were collected at the beginning of the monitoring.

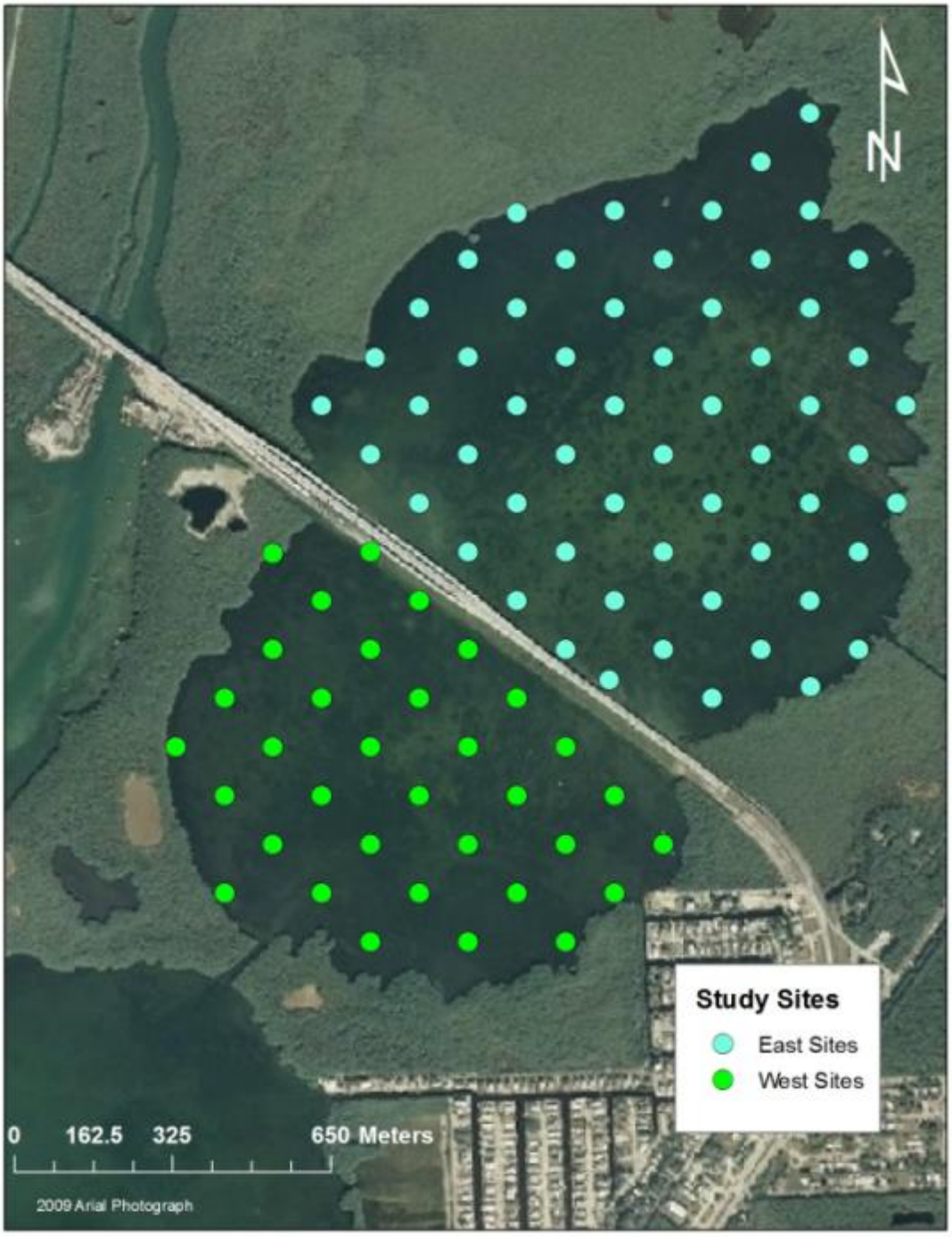


Figure 22. Soil and vegetation sampling design

Approximately 3000 depth soundings were recorded using a Garmin GPS sounder/chartplotter. This instrument recorded positional coordinates using Wide Angle Augmentation System GPS with a typical accuracy of 2 m. Depths were recorded with a high frequency sounder at an accuracy of 0.03 m. Depth soundings were collected by navigating a boat along North-South and East-West transects throughout the lake (Figure 23)

Figure 23. Bathymetric soundings at Lake Surprise.

Vegetation

At each site, vegetation was inventoried at ten random locations using a 1 m square grid. The following vegetation parameters were recorded: percent cover of each seagrass species, percent cover of algae, and shoot density of each seagrass species. These data were averaged to provide a single value at the monitoring site (Figure 24).

Soils

At each monitoring site, the upper 0-5 cm soil was sampled and analyzed for: Bulk density, organic matter, total phosphorus, and particle size distribution (Figure 24)

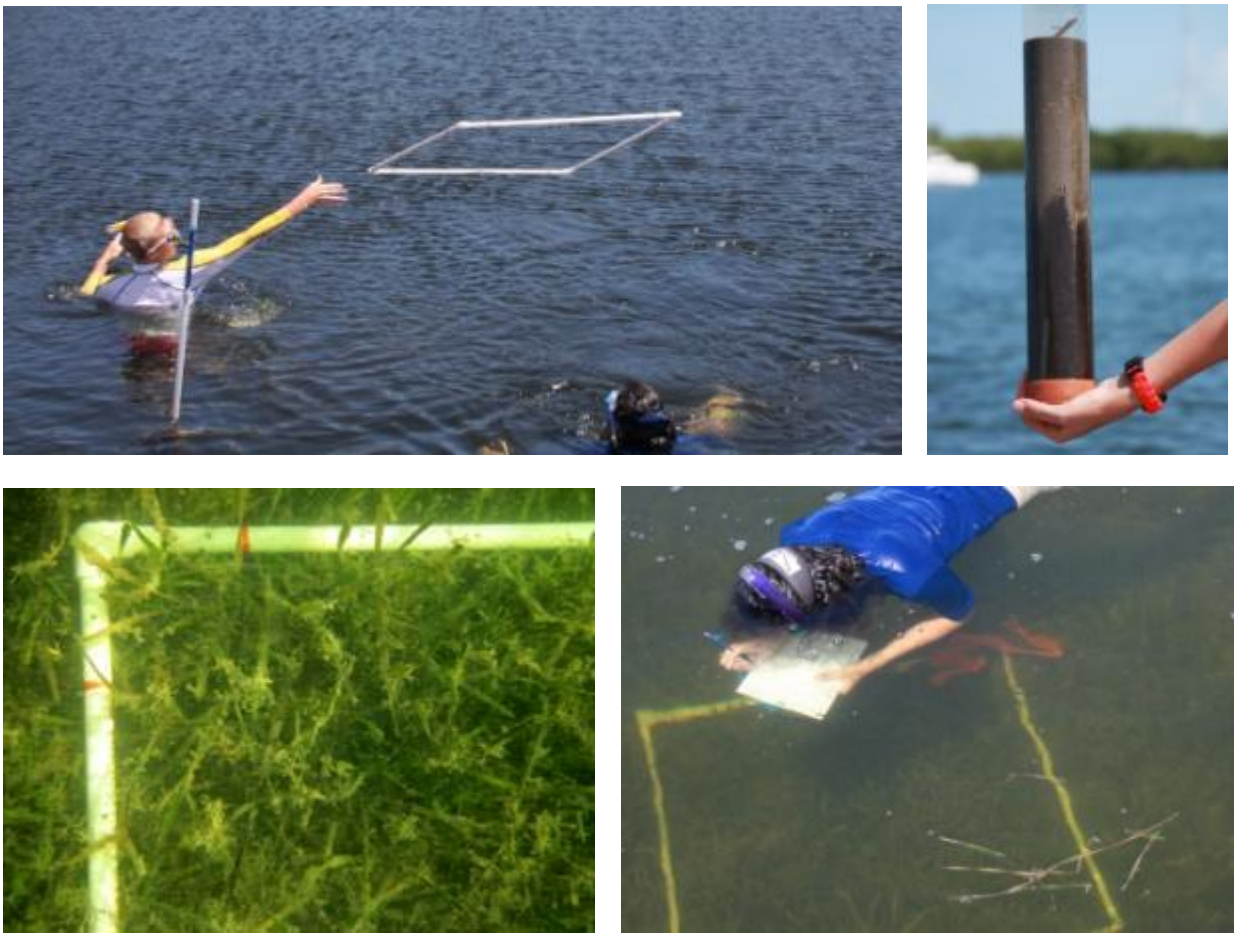


Figure 24. Vegetative and soil sampling at Lake Surprise.

RESULTS

A complete set of full-page maps is presented in Appendix B of this report.

WATER QUALITY

All water quality parameters showed slight improvements over time.

Light

The downward light attenuation coefficient (Kd) decreases slightly with time but was not significantly different from the outside control sites (Figure 25). The light attenuation was not significantly different between the east and the west sides of the lake. The average Kd was 0.46 m^{-1}. Spatial patterns of light attenuation were not consistent through time but suggested there may be greater light attenuation in the West portion at times.

Dissolved Oxygen

The average dissolved oxygen (DO) of Lake Surprise was not significantly different from the outside control sites, but did increase relative to winter 2008. Spatial patterns showed consistently lower DO along the northwestern side of Lake Surprise (Figure 26).

pH

The average water column pH was high (8.5) in winter 2008 but remained normal (8.1) for all remaining seasons (Figure 27). There were no consistent spatial patterns of pH.

Salinity

The water column salinity approached tolerances for *Thalassia* and *Halodule* (60-65 ppt) in the summer of 2009 and 2011. Salinity in all other seasons was below these thresholds. Salinity was not significantly different between the east and west portions of Lake Surprise and was not significantly different from the outside control sites. There were no consistent spatial patterns of salinity (Figure 28).

Bathymetry and Light Availability

The water in Lake Surprise ranges from 0.30 m to 3 m deep (Figure 29). The average depth of the east side is 0.3 m deeper, and the maximum depth is 1.2 m deeper than the west side of the lake (Table 1). Given constant water quality and therefore constant light attenuation with depth, deeper water results in less plant-available light at the soil surface.

SEAGRASS AND ALGAE

The average percent cover of all seagrass at the outside control sites did not significantly change with time. Similarly, the west and east portions of Lake Surprise did not change with time. The percent cover was consistently higher in the west portion than the east portion (Figure 30). *Thalassia* is the dominant seagrass at most sites. The spatial and temporal patterns of *Thalassia* match that of the total seagrass percent cover (Figure 31). *Halodule* was the other species present however the percent cover was constantly minor compared to *Thalassia* (Figure 32). Spatial patterns of *Halodule* were not consistent and most likely represent brief periods of colonization.

The shoot density of *Thalassia* was unchanged in the east portion but increased in the west. As a result, the total shoot density in the west increased with time (Figures 33 and 34). There was no change in *Halodule* shoot density through time (Figure 35).

Algae coverage remained below 20% for all seasons. Spatial patterns suggested higher algae in the east, but it is not statistically significant (Figure 36).

SOIL

Average values of soil properties such as organic matter (Figure 37), clay (Figure 38), sand (Figure 39), and total phosphorus (Figure 40) did not change though time.

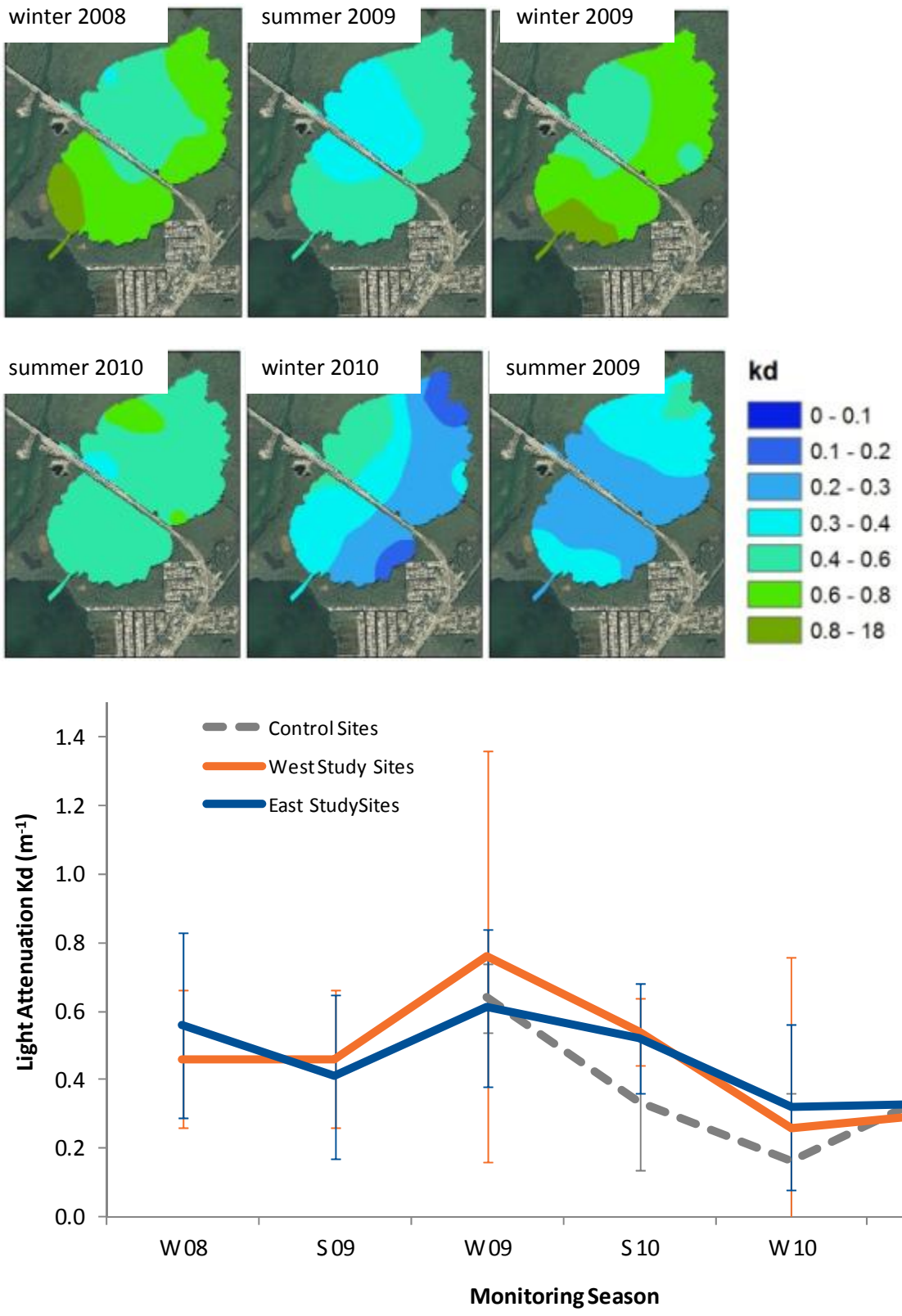
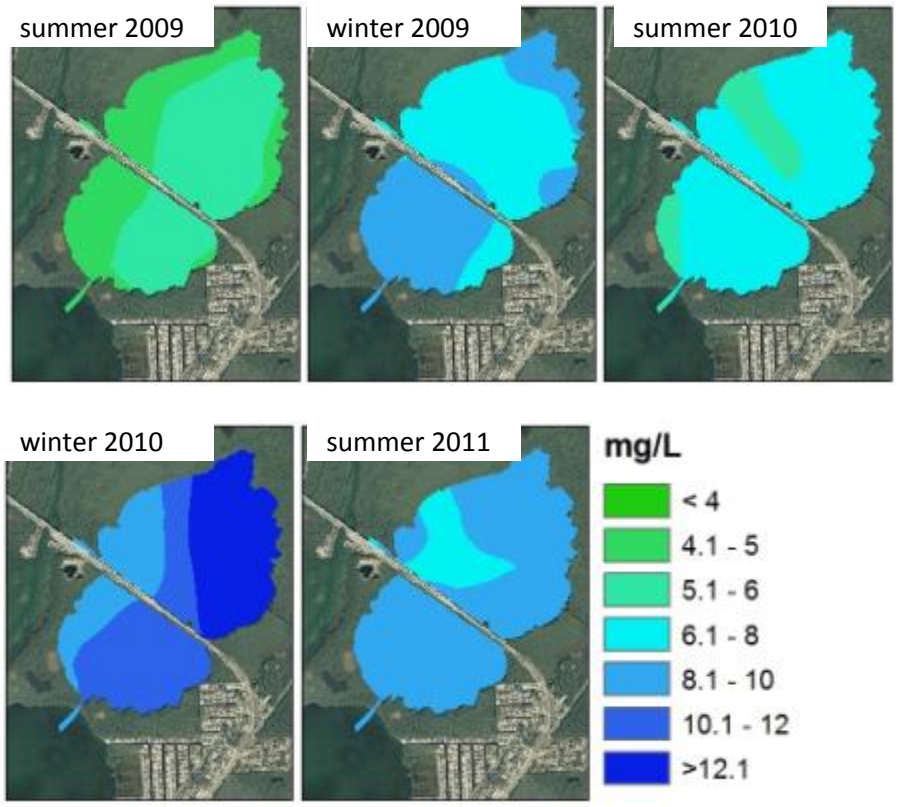


Figure 25. Water column downward light attenuation coefficient (Kd).



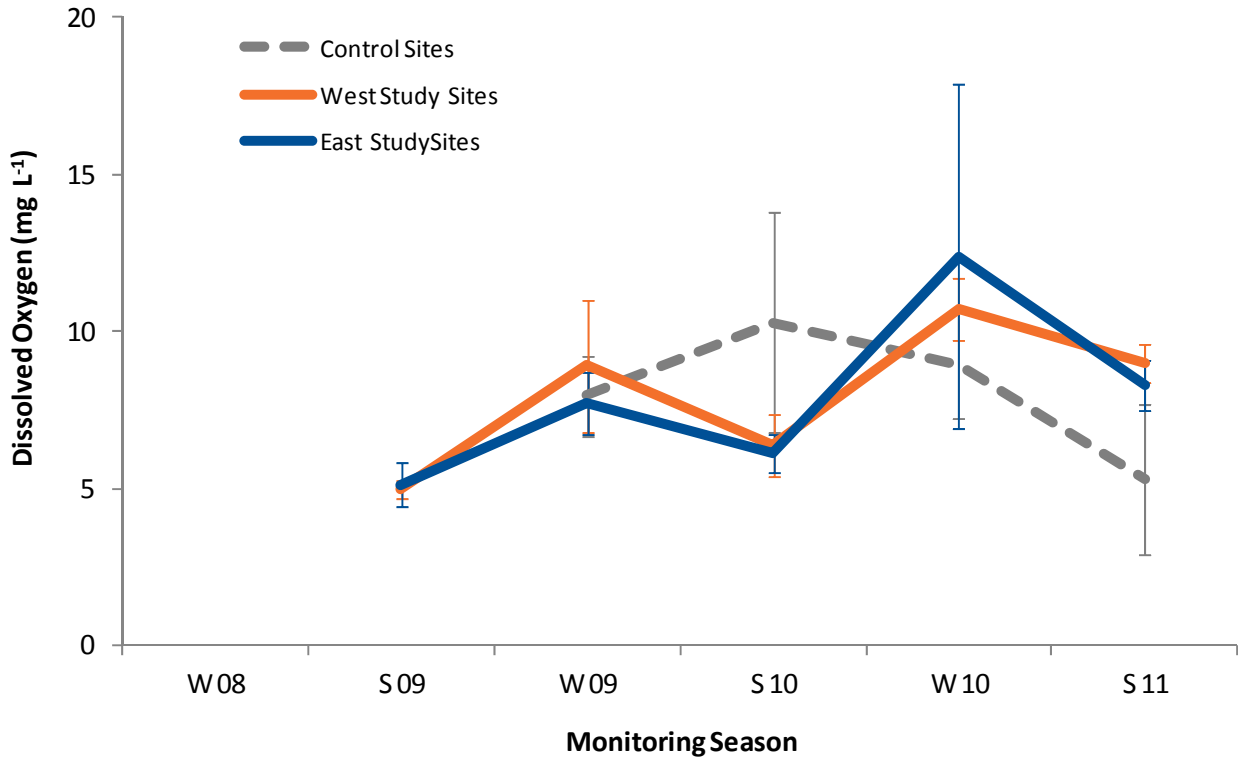
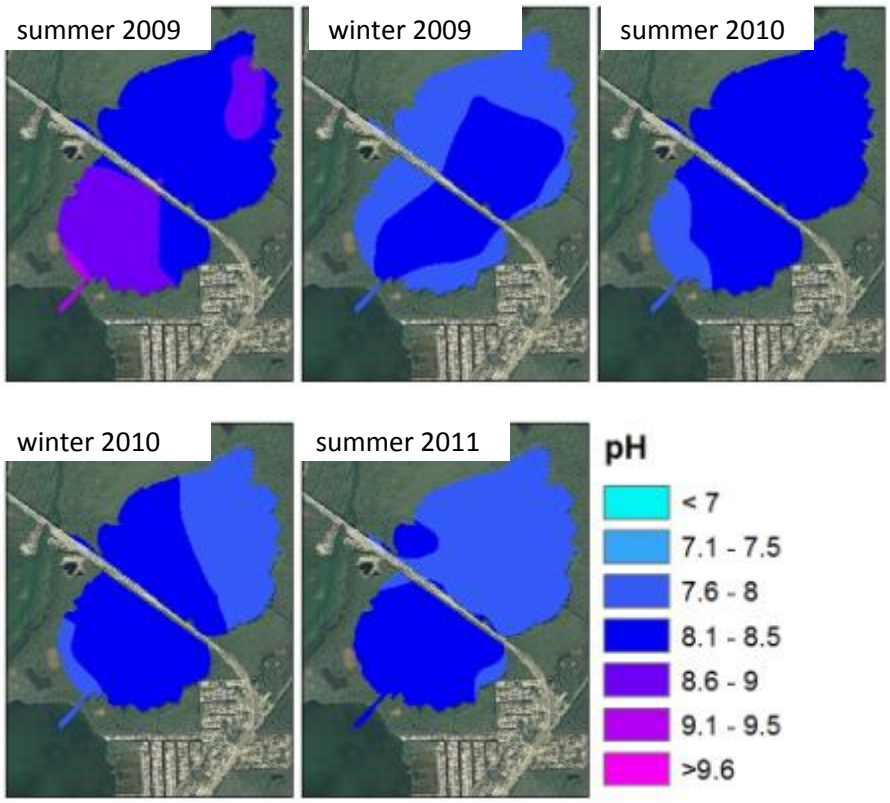


Figure 26. Water column dissolved oxygen



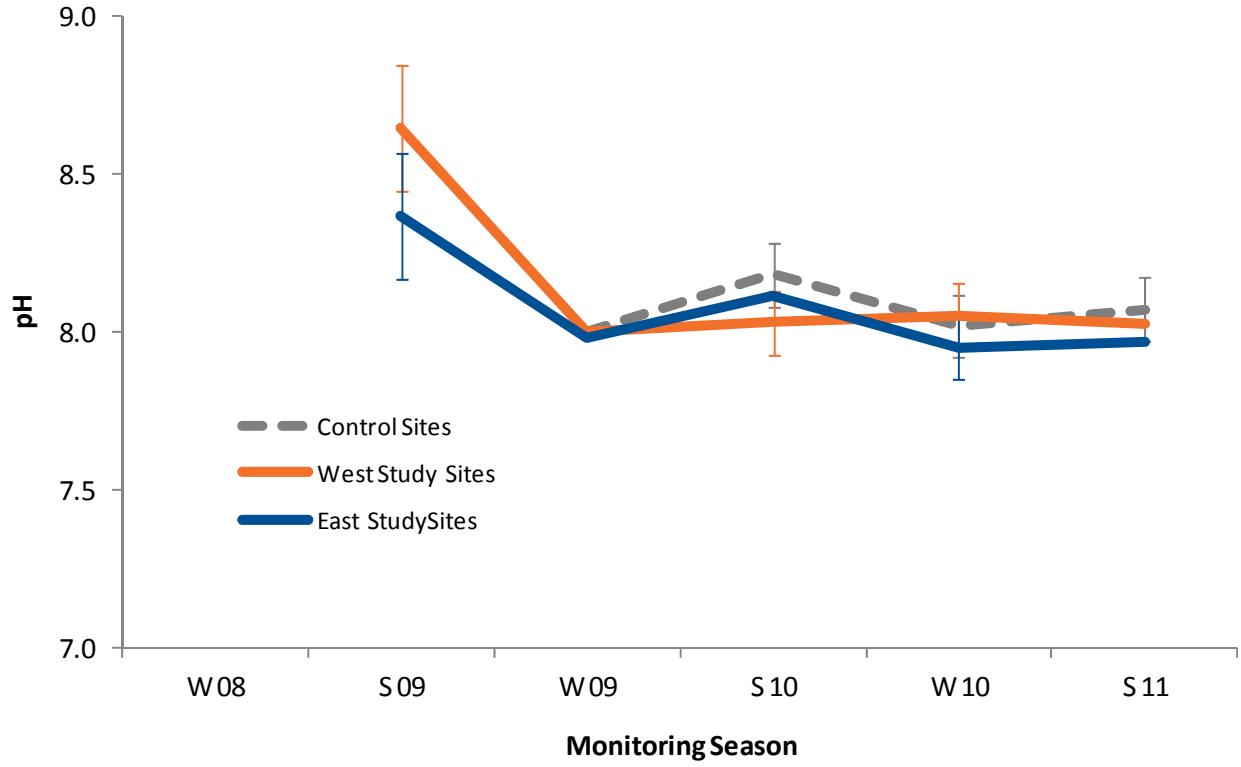
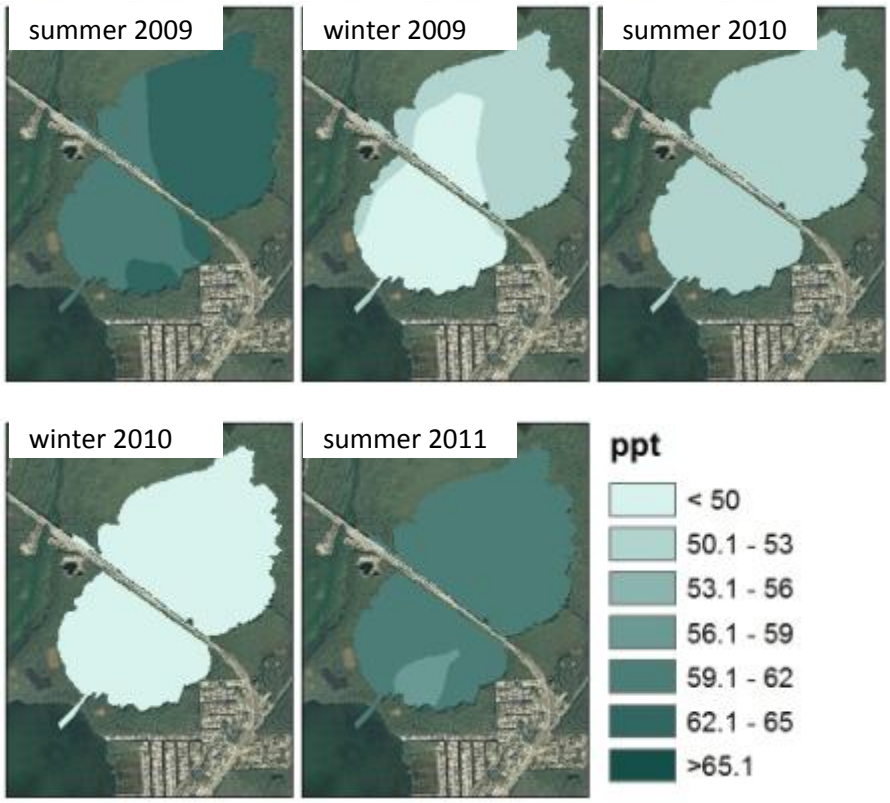


Figure 27. Water column ph.



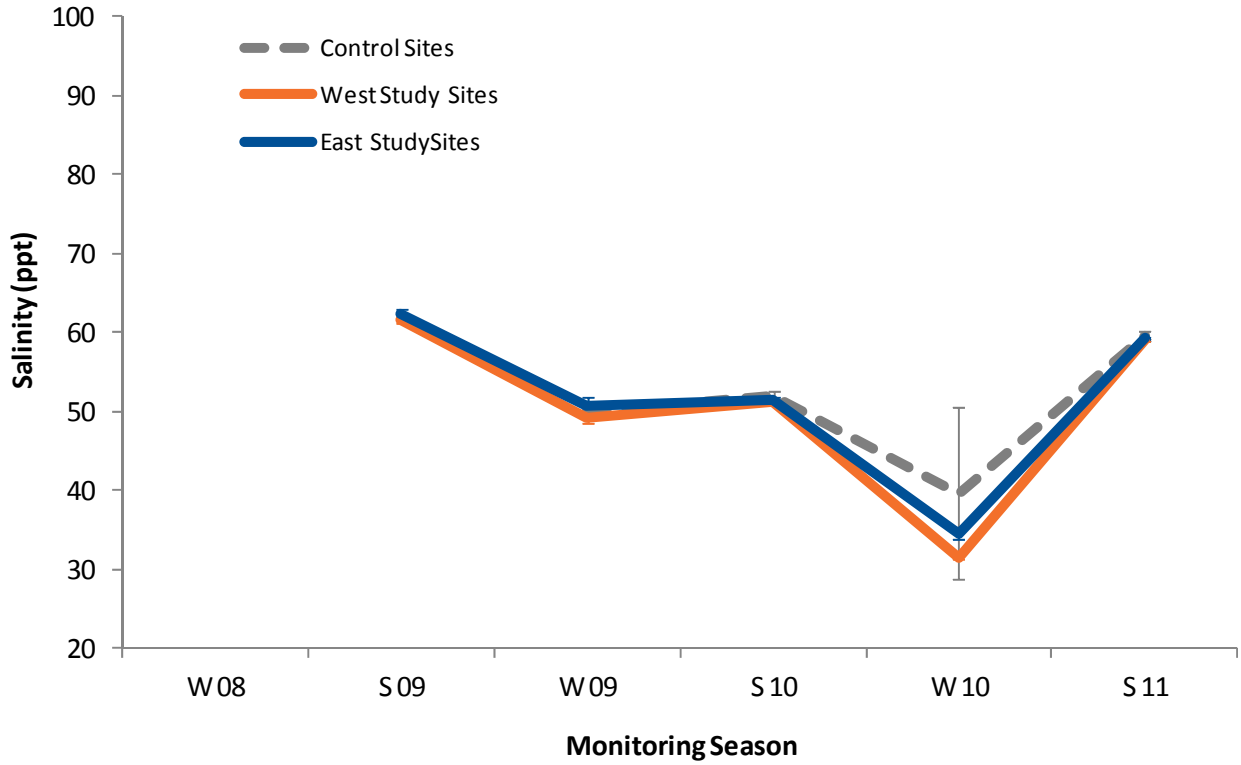


Figure 28. Water column salinity.

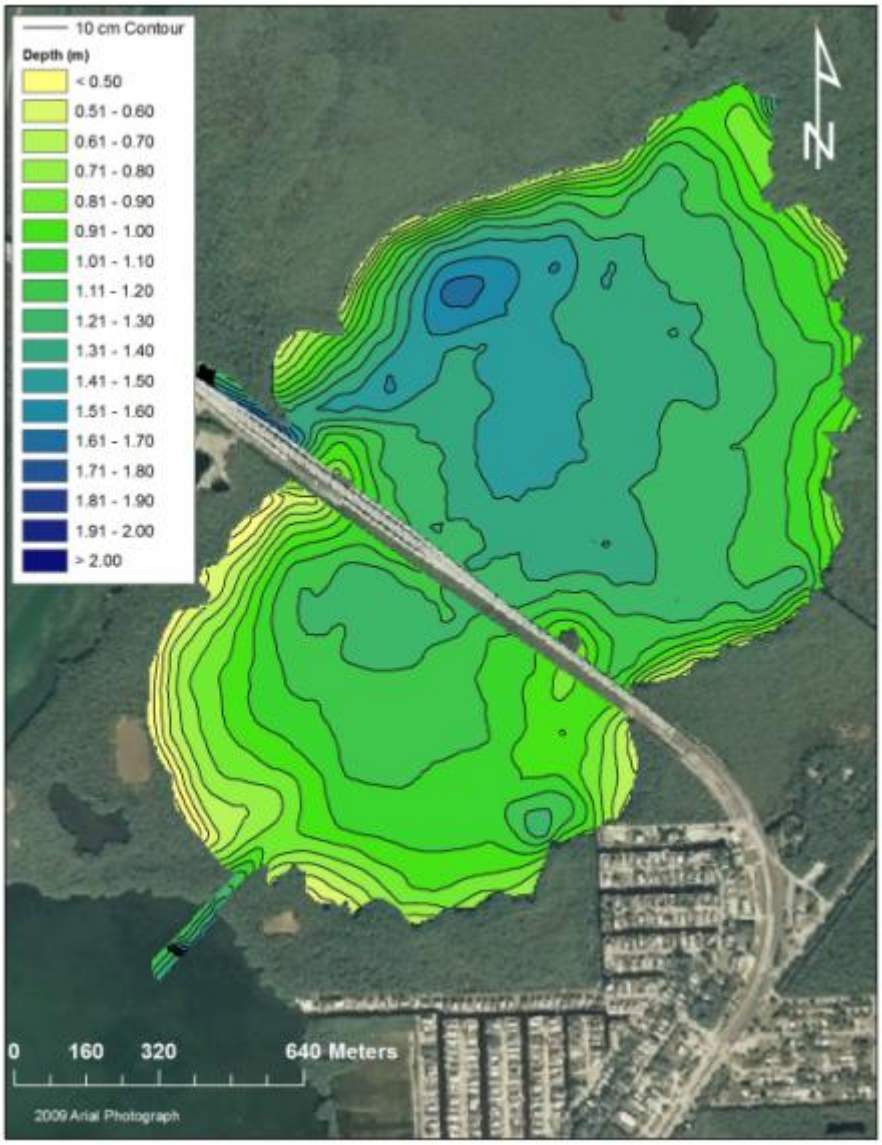


Figure 29. Bathymetry of Lake Surprise.

Table 1. Summary of bathymetry for East and West portions of Lake Surprise. Plant available light as a percentage of surface irradiance is given in parentheses and assumes Kd = 0.46 m^{-1}.

	East	West
Minimum Depth (m)	0.3 (87%)	0.3 (87%)
Mean Depth (m)	1.2 (58%)	0.9 (66%)
Maximum Depth (m)	2.9 (26%)	1.7 (46%)
Std Deviation (m)	0.3	0.3
n	1814	1276

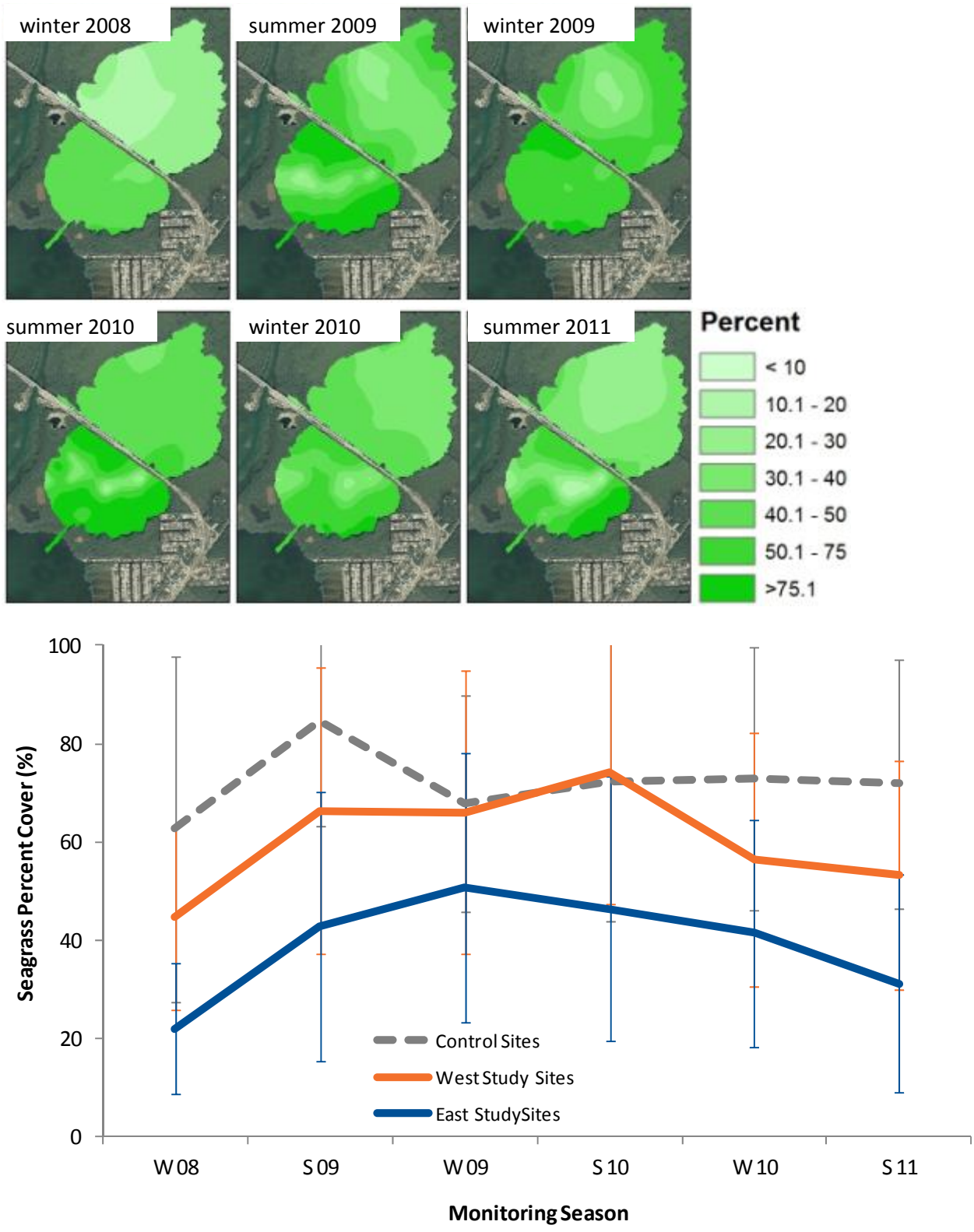


Figure 30. Average seagrass percent cover.

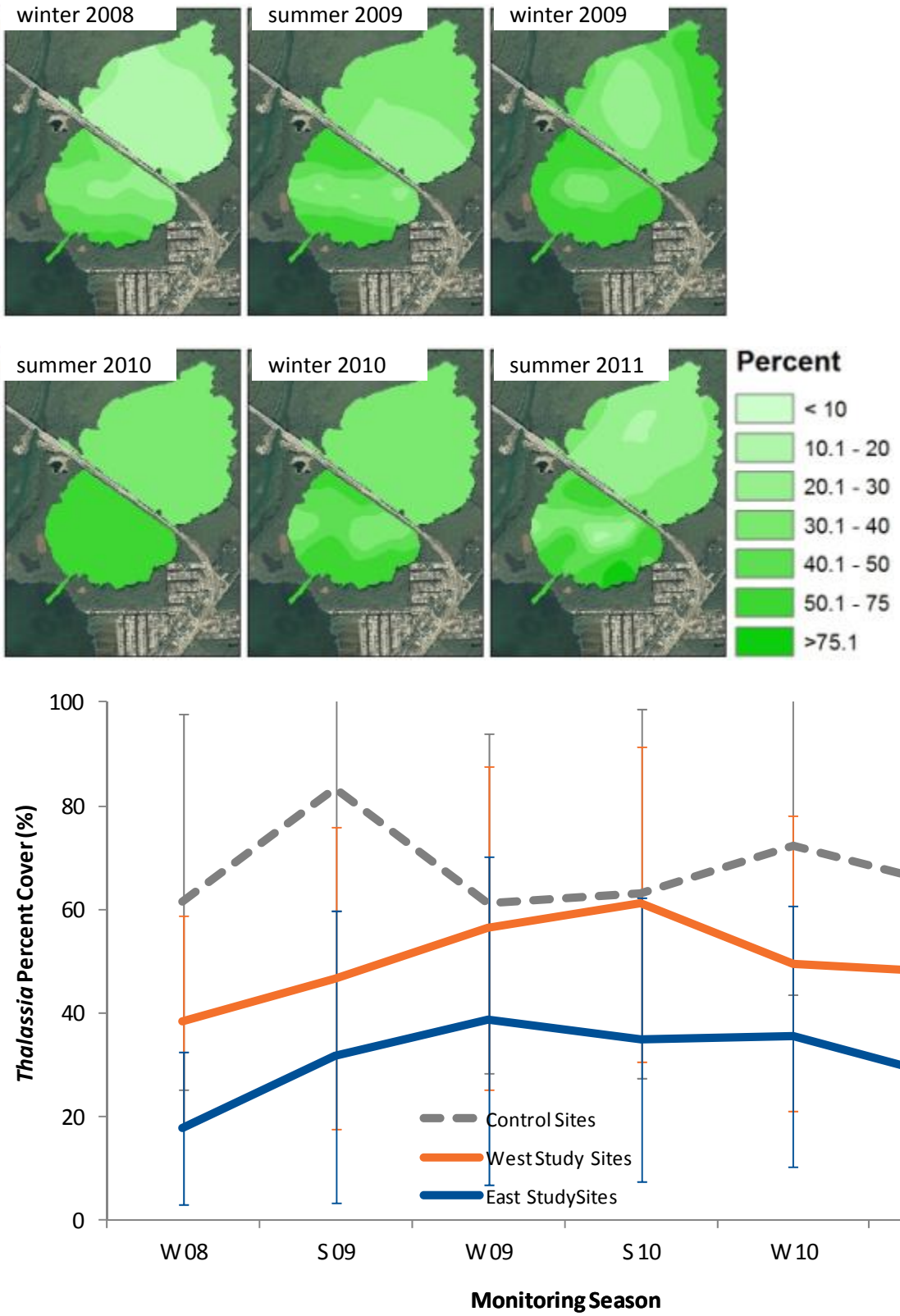
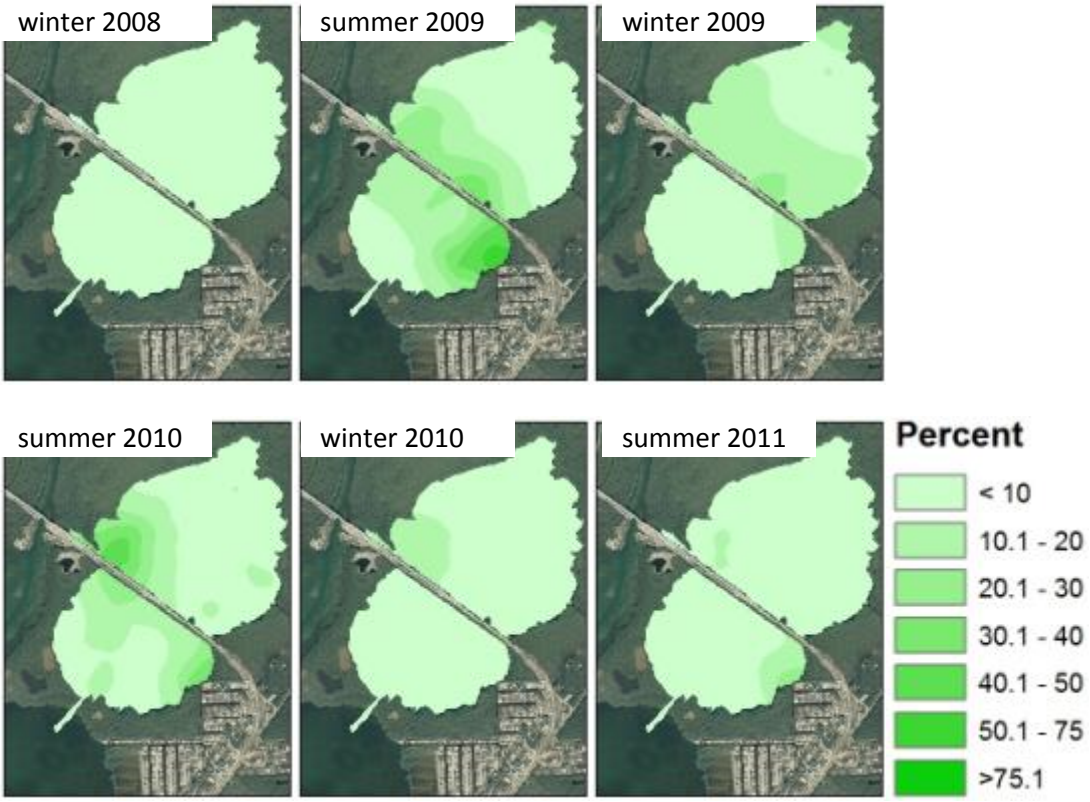


Figure 31. Average *Thalassia* percent cover.


winter 2008
summer 2009
winter 2009
summer 2010
winter 2010
summer 2011
Percent
< 10
10.1 - 20
20.1 - 30
30.1 - 40
40.1 - 50
50.1 - 75
>75.1

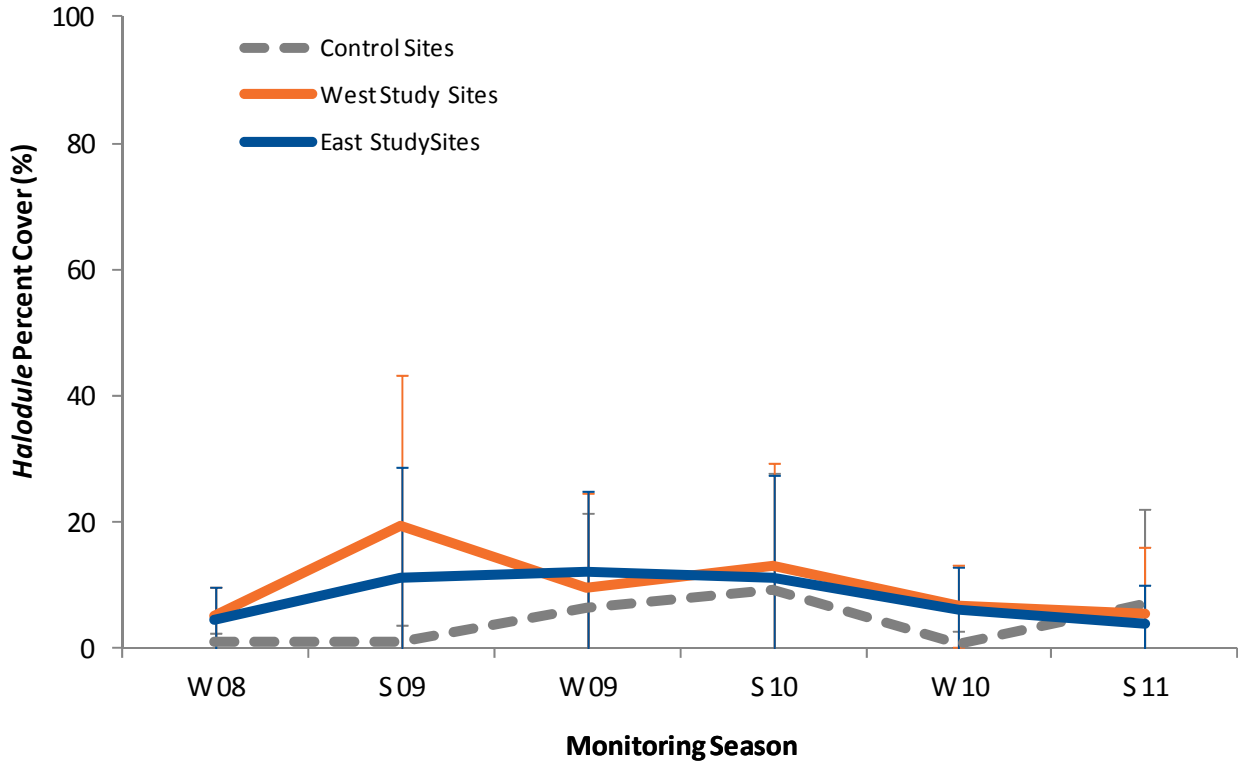
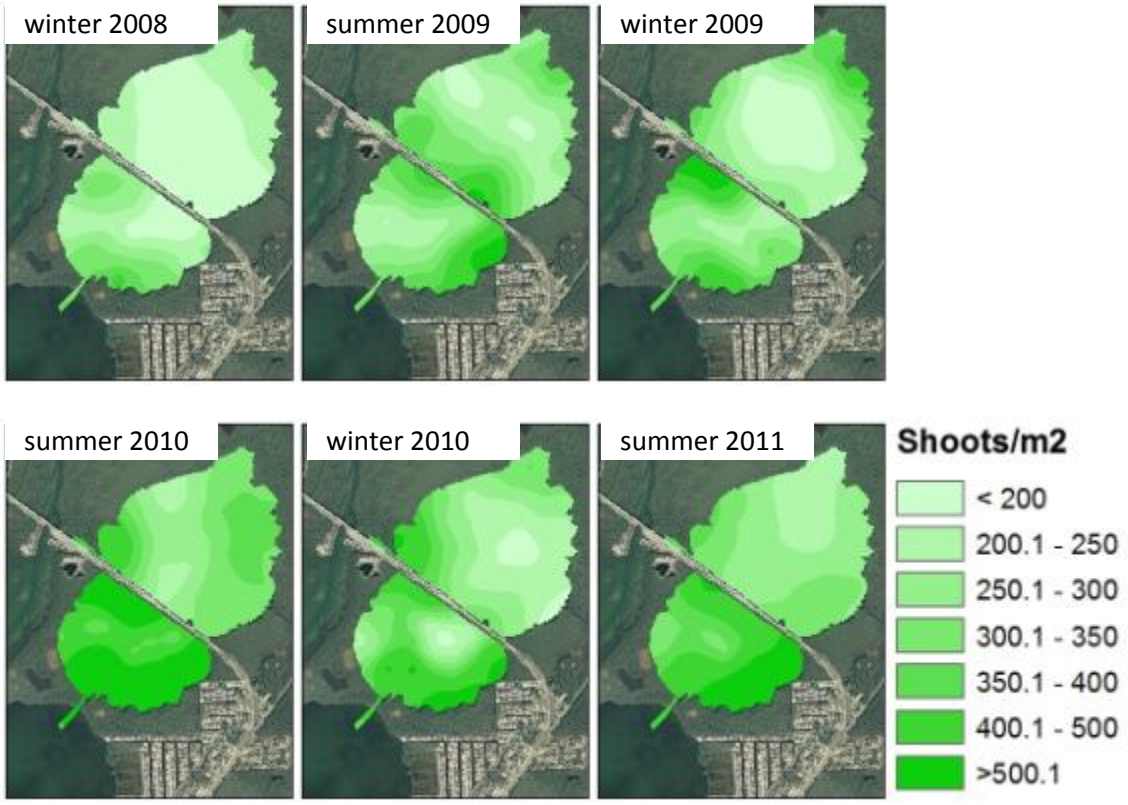

Control Sites
West Study Sites
East StudySites
Halodule Percent Cover (%)
100
80
60
40
20
0
W 08
S 09
W 09
S 10
W 10
S 11
Monitoring Season

Figure 32. Average *Halodule* wrightii percent cover.



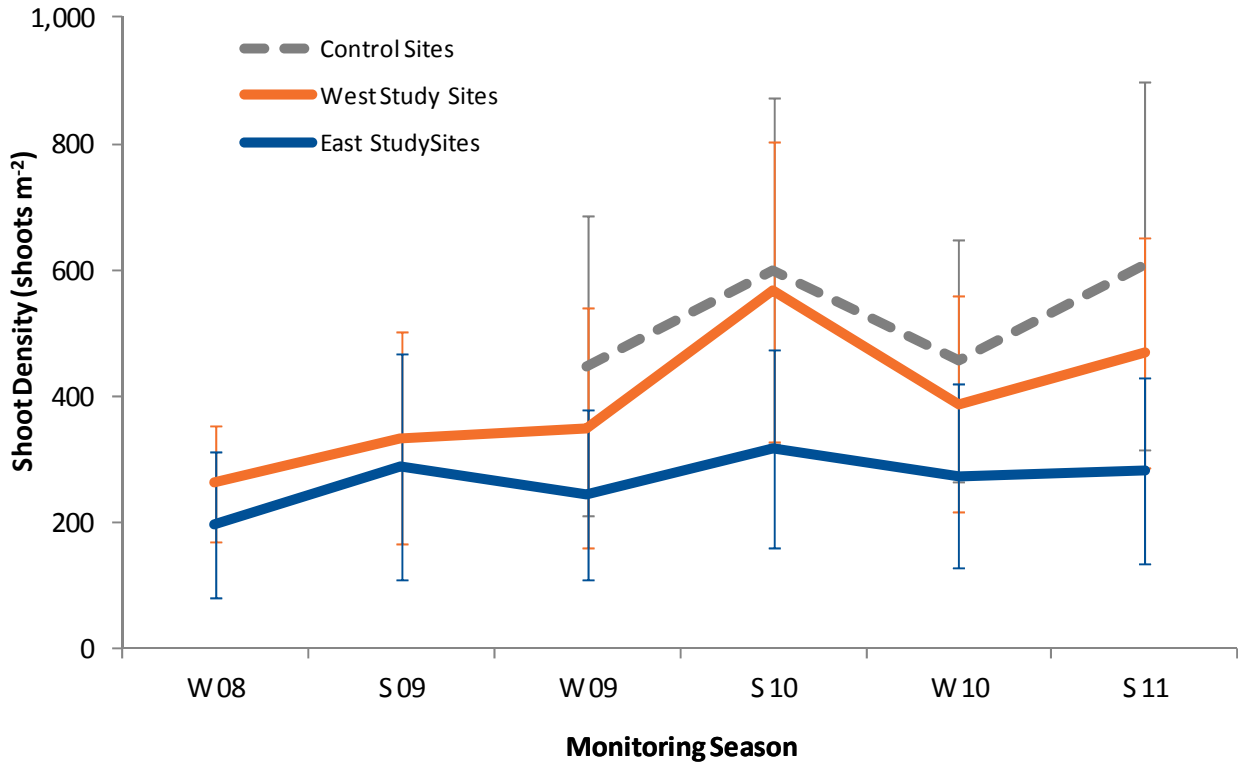


Figure 33. Average total seagrass shoot density

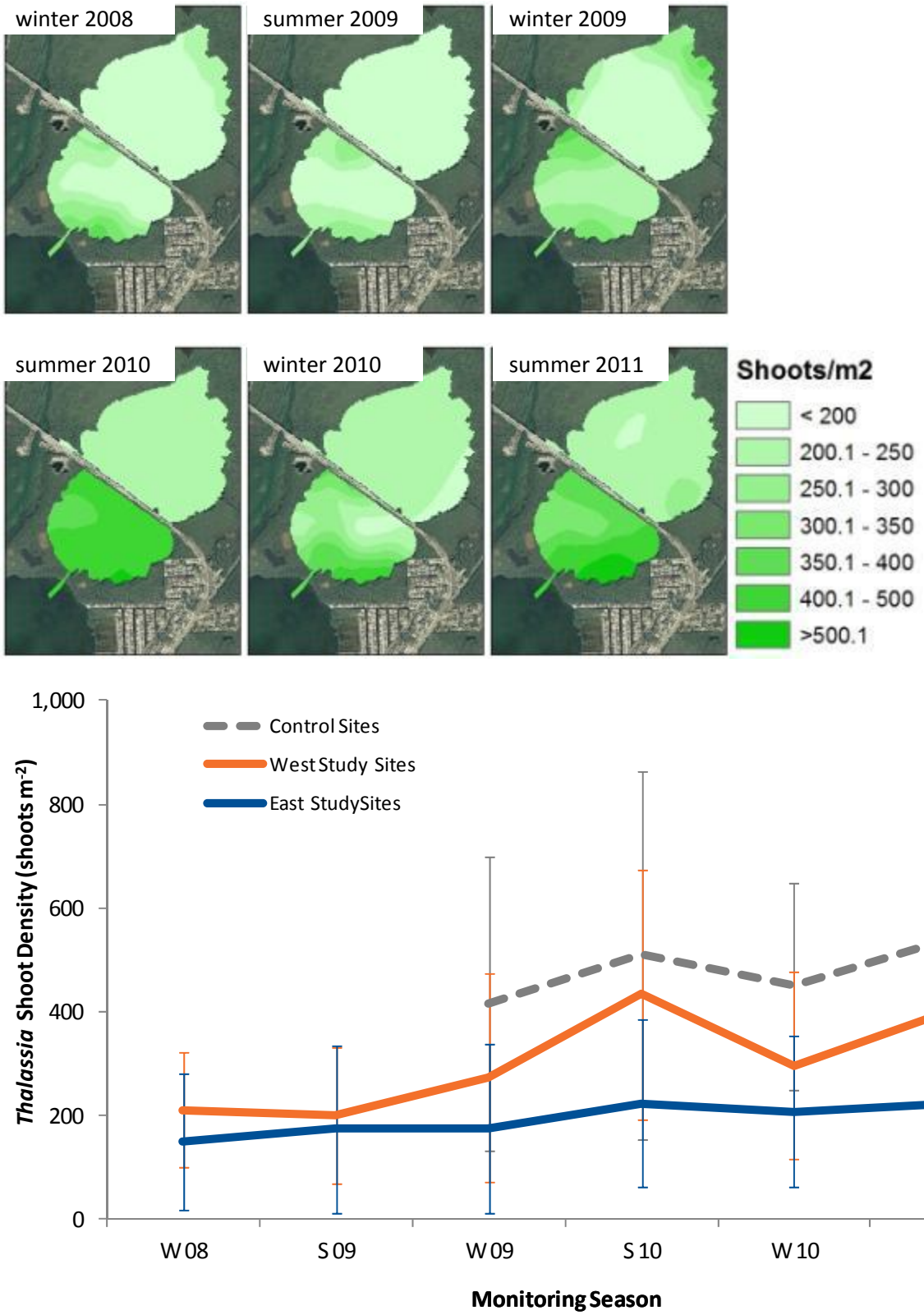


Figure 34. *Thalassia* shoot density and maps through time

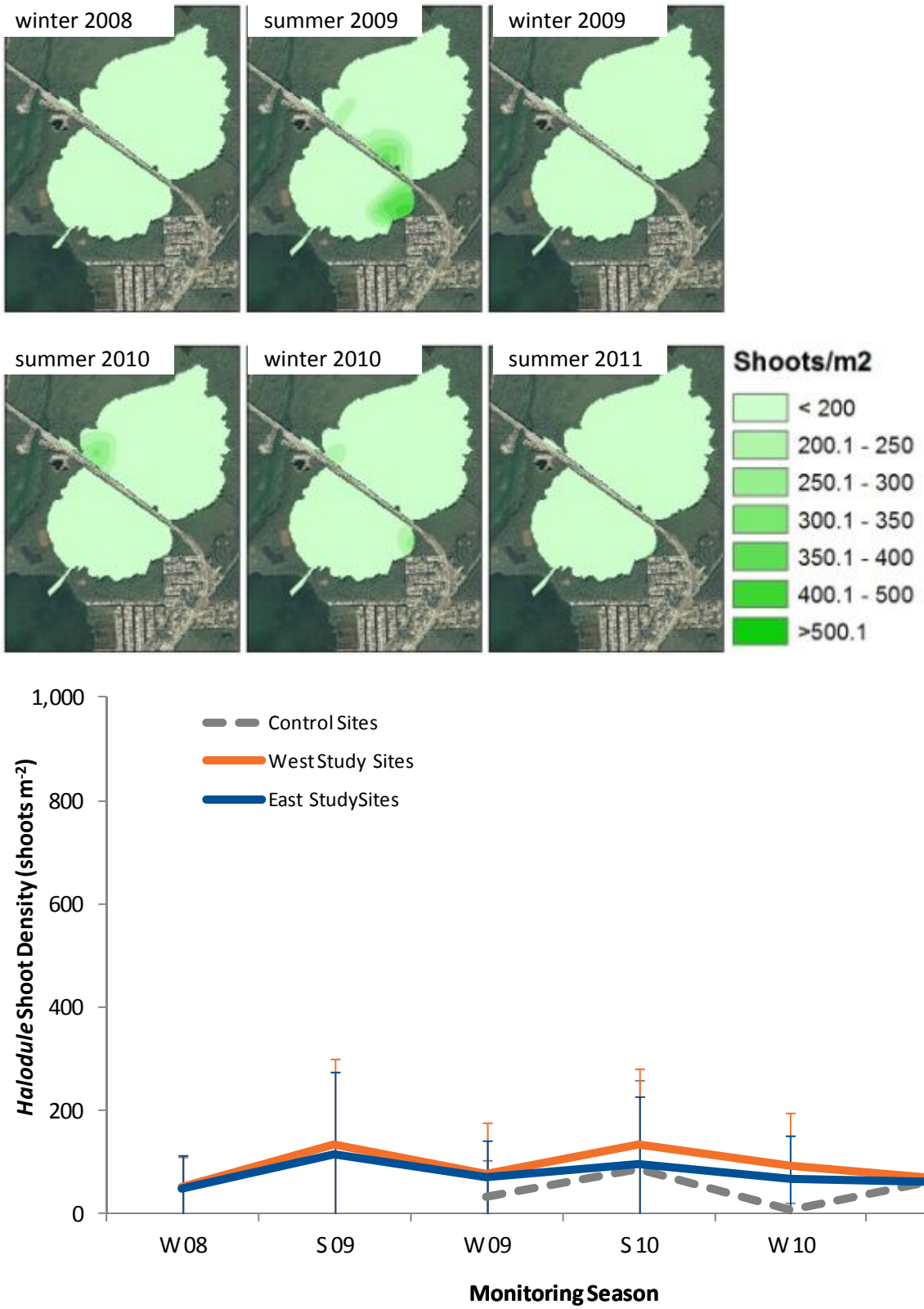
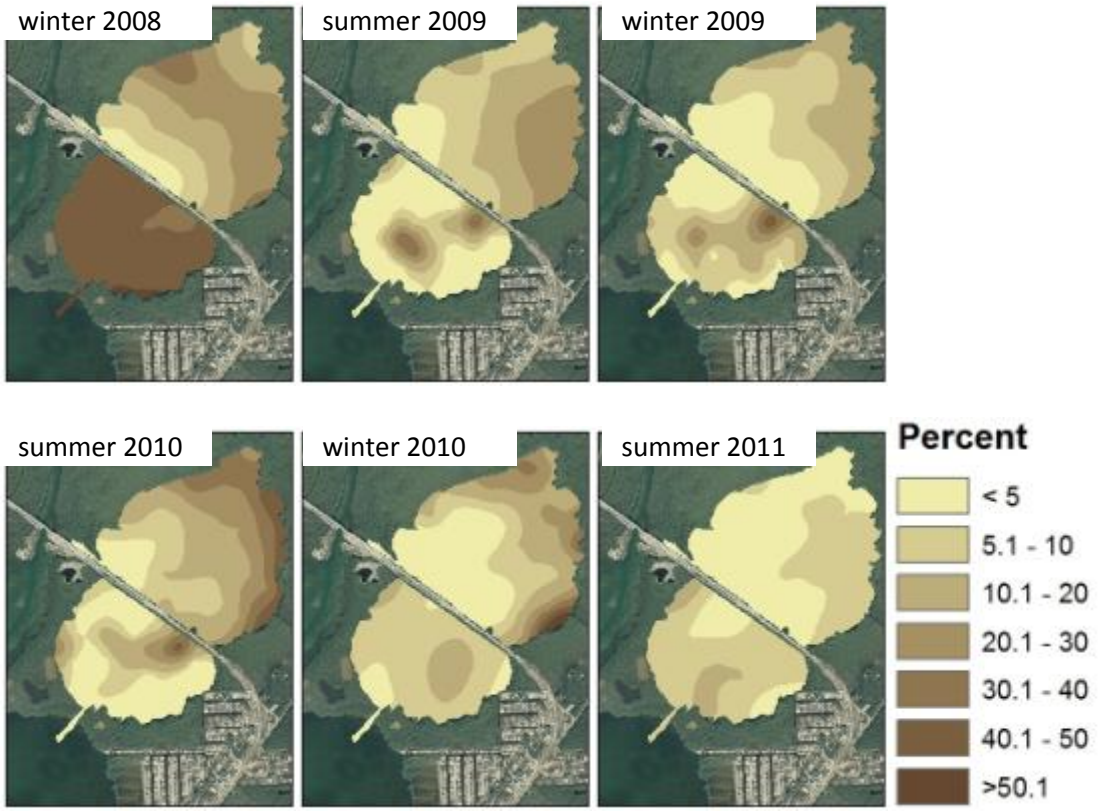


Figure 35. *Halodule* density and maps through time



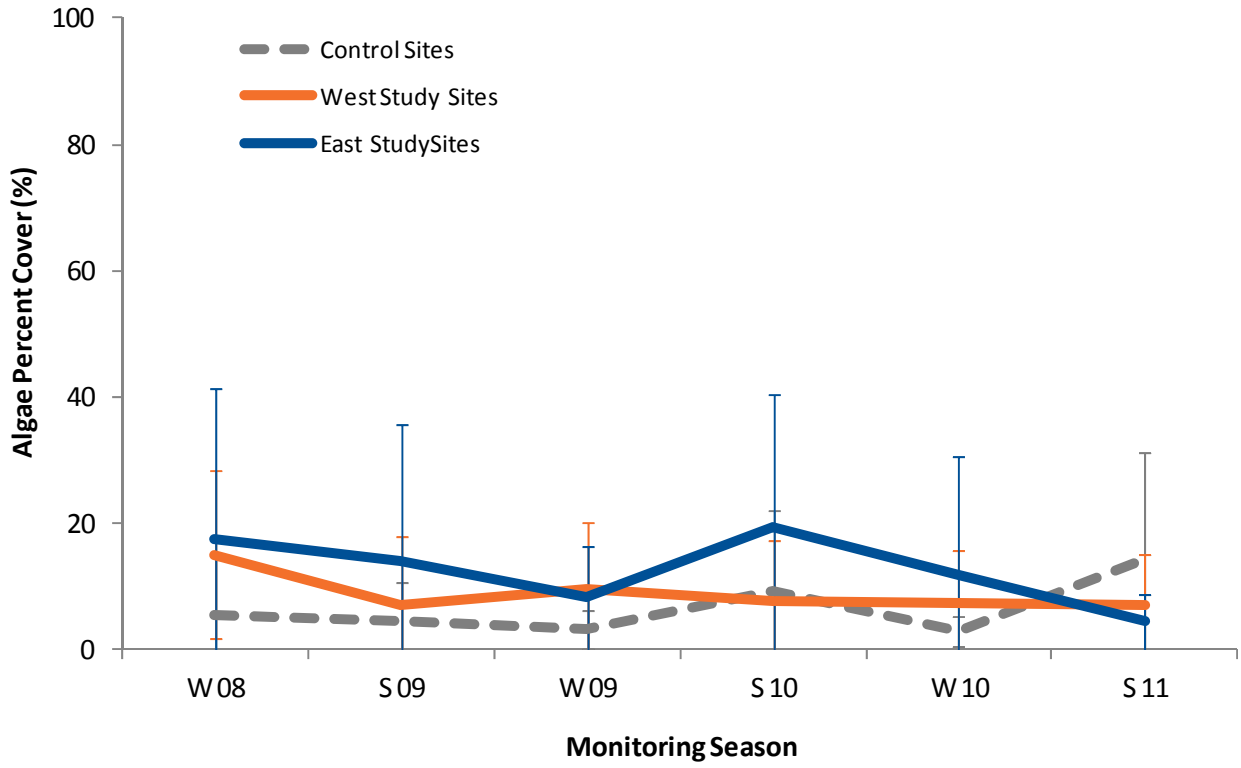


Figure 36. Algae cover and maps through time

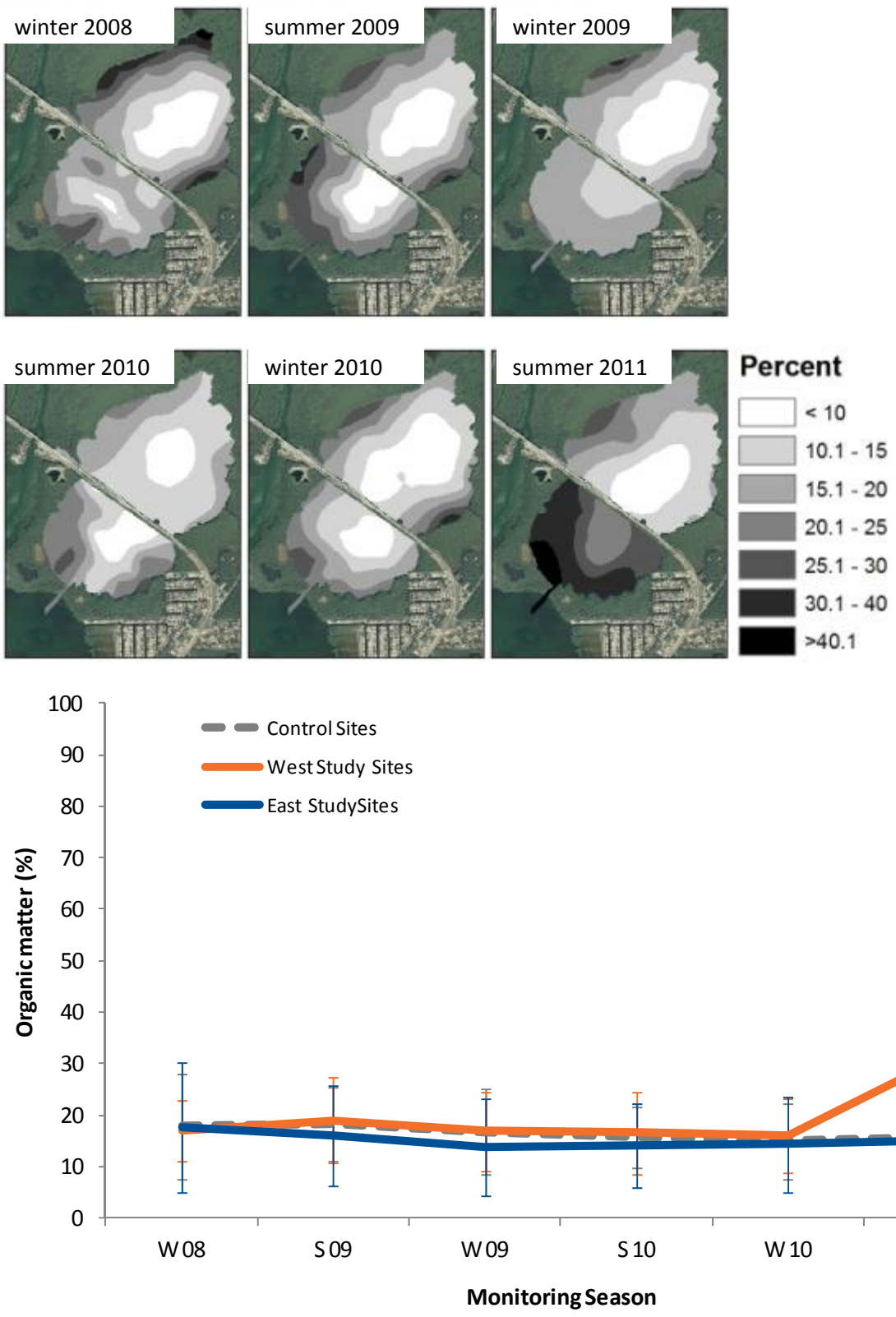
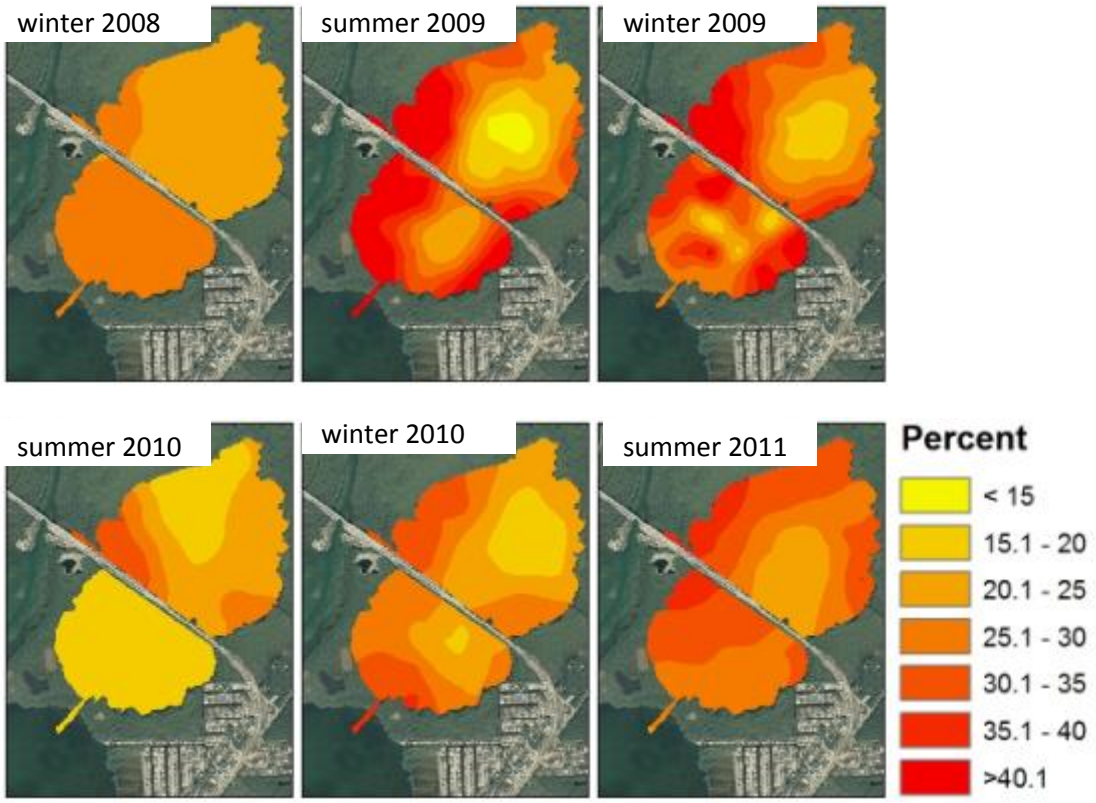


Figure 37. Average soil organic matter concentration.



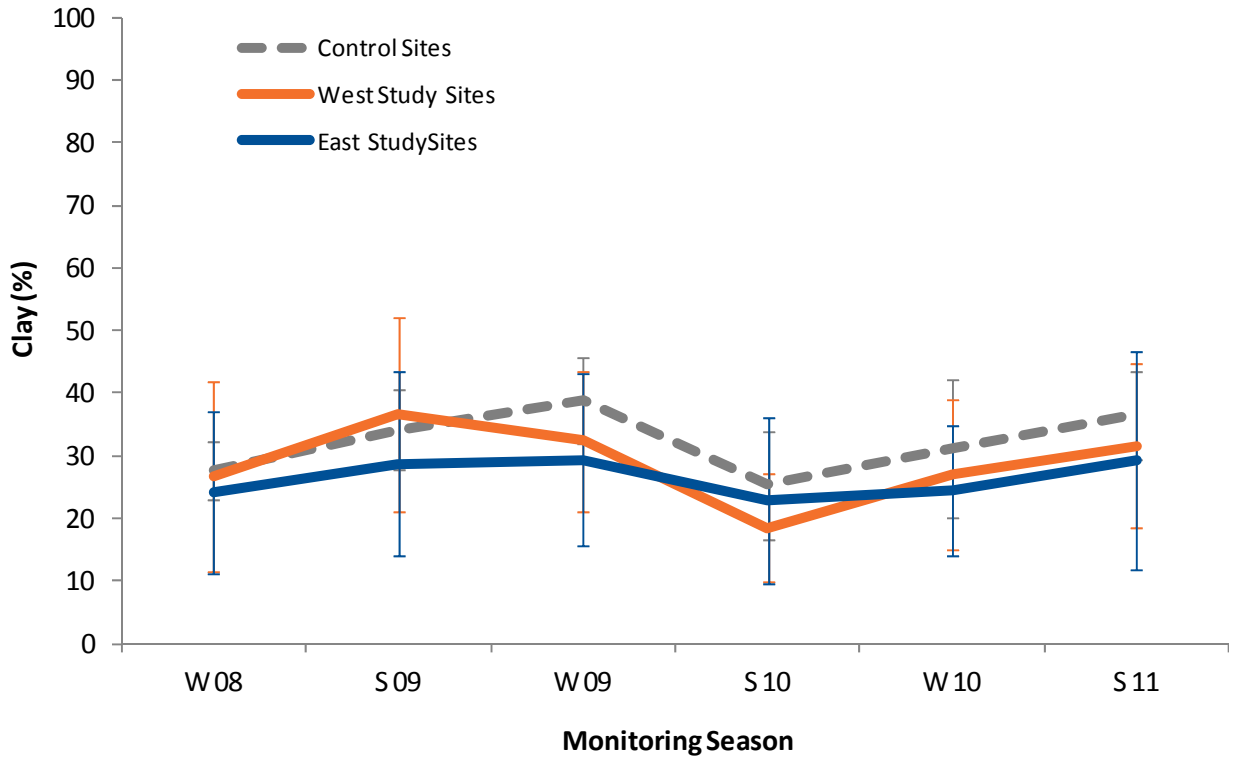


Figure 38. Average soil clay concentration.

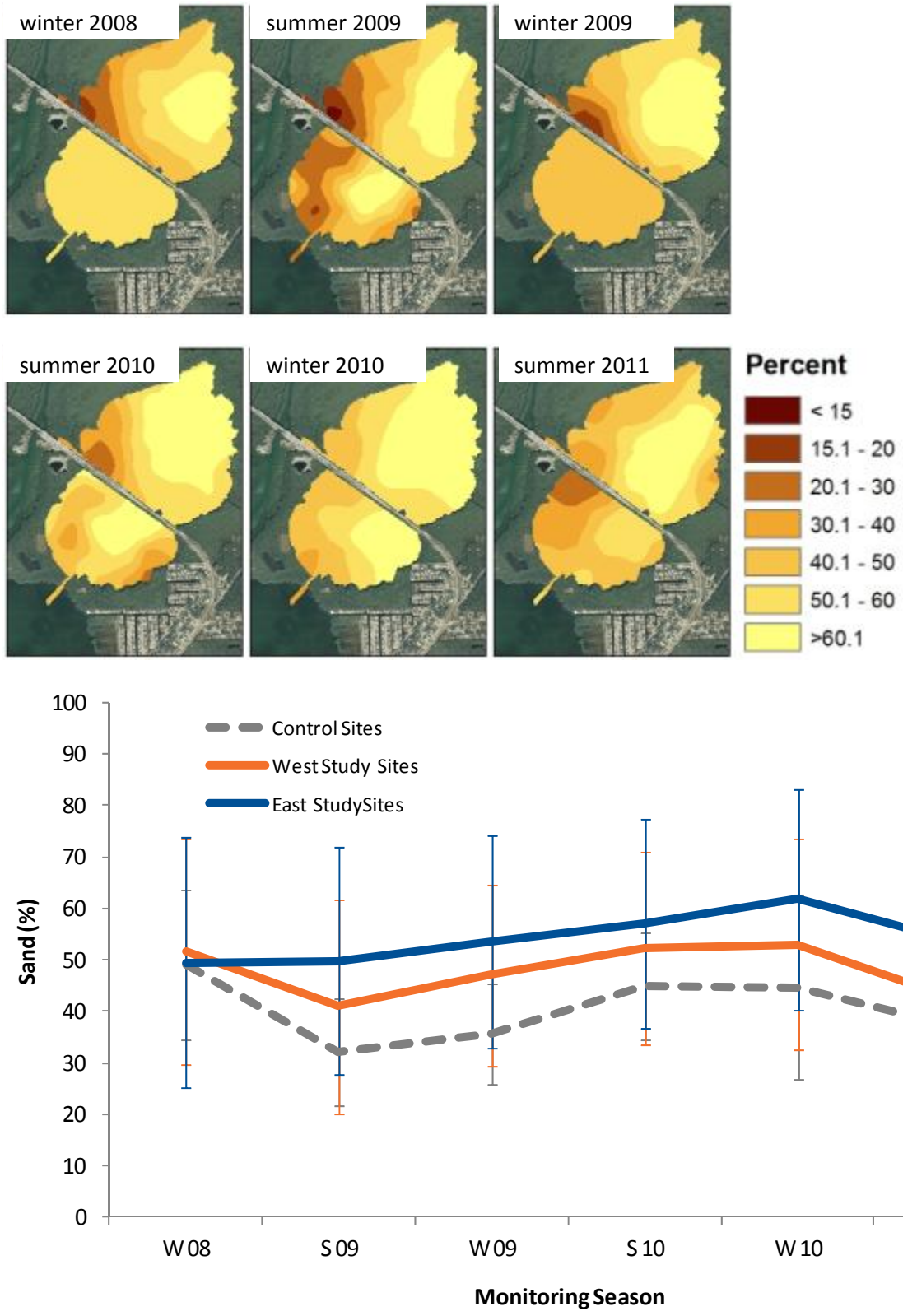
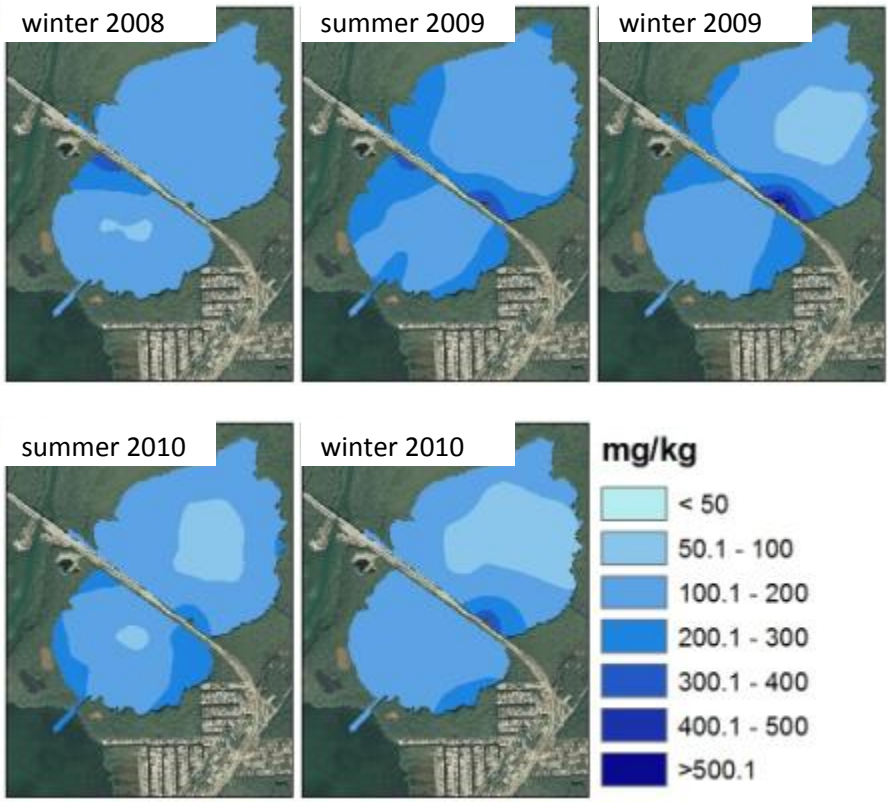


Figure 39. Average soil sand concentration.



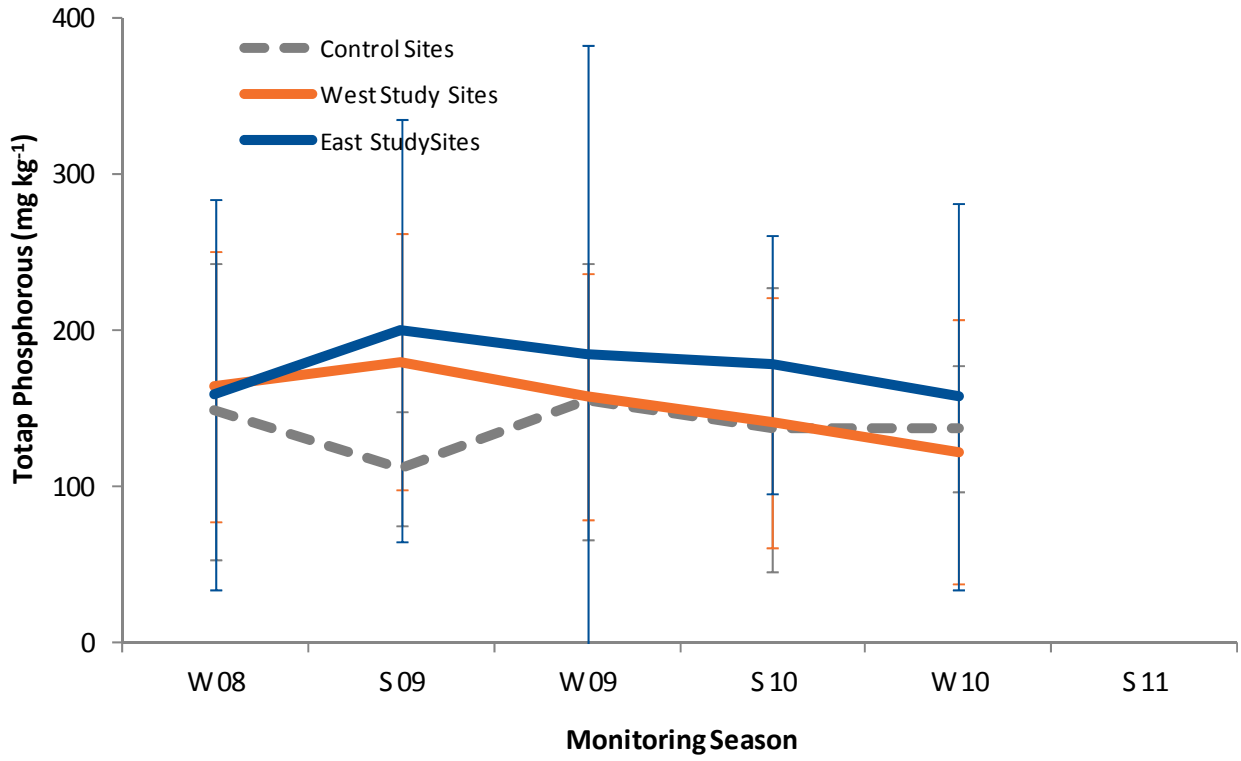


Figure 40. Average soil total phosphorus concentration.

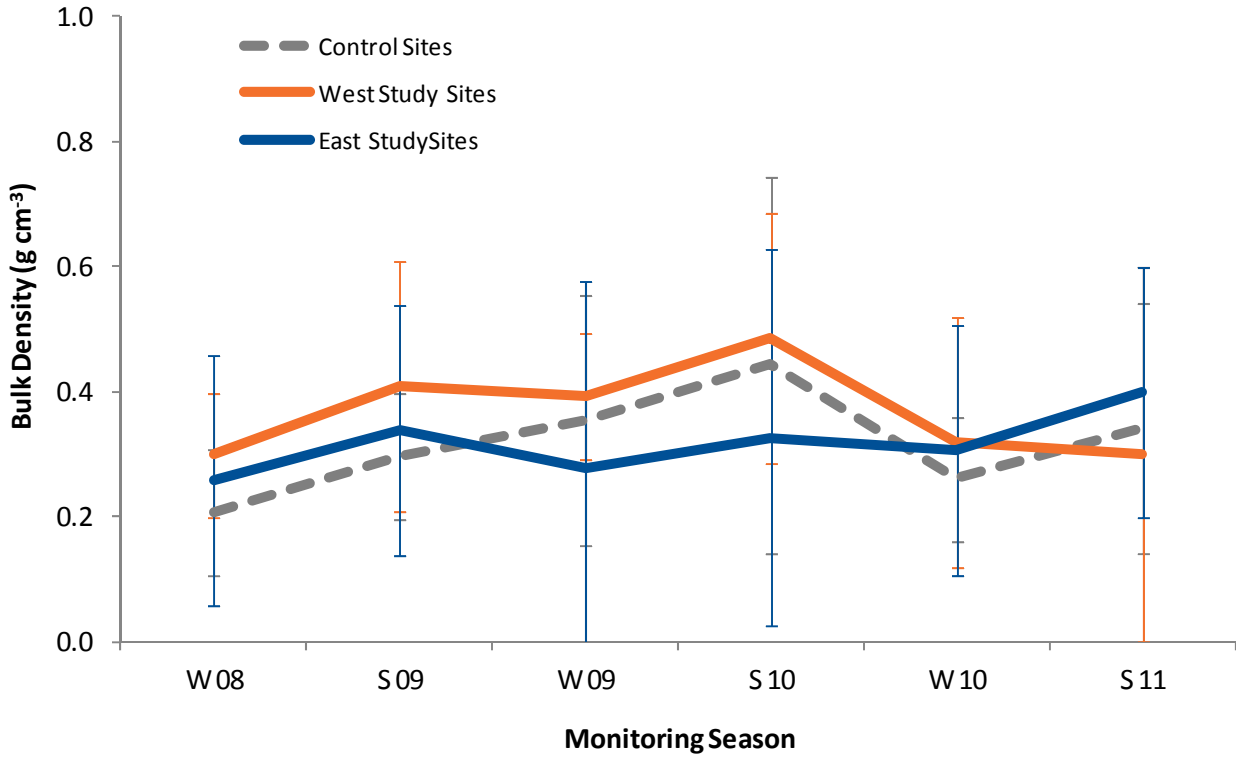


Figure 41. Average soil bulk density.

DISCUSSION

What can we conclude based on where seagrass collects? What is it related to and what does it seem to be unrelated to? How has the spatial modeling improved our understanding of the area? What can we say by virtue of the spatial modeling?

The original concern with Lake Surprise was that post-construction conditions would result in reduced water quality, sedimentation, and seagrass decline or die-off. This did not occur. During the six season of monitoring, water quality spatial trends indicate the East and West portions are sufficiently connected with each other. This can be clearly attributed to the causeway removal. The slight improvement in light attenuation over time and increase in DO is accompanied by an improvement in percent cover and shoot density of *Thalassia*. It is probable that the causeway removal has improved hydraulic exchange with the outside Florida Bay, meaning the bay now flushes more completely with outside water from the bay. The consequence of increased flushing would most likely be increased light availability (reduced Kd). Typical seagrass responses to greater available light is increases in percent cover (seagrass is spreading) and increases in shoot density (seagrass is getting thicker). Another positive development is that these increases have occurred in *Thalassia*. It is reasonable to forecast that since *Thalassia* is a slower growing climax species that these changes represent a stable trend in seagrass increase in Lake Surprise. Finally, the lack of change in soil properties reinforces the hypothesis that improve water quality is responsible for improved seagrass coverage.

CHAPTER 5: THE INFLUENCE OF SUBAQUEOUS SOIL ON *HALODULE WRIGHTII*

SUMMARY

Accelerating declines in seagrass systems worldwide have increased the need to optimize seagrass transplanting. Subaqueous soils support seagrass as a medium of attachment and source of nutrients. A number of soil properties have been documented to influence seagrass growth in both natural, mature seagrass meadows and during experimental transplanting efforts. Recently, interpretations regarding the suitability of subaqueous soils for seagrass restoration have been proposed for inclusion in future subaqueous soil surveys. However, several questions must be addressed before this interpretation is accepted. First, will seagrass grow equally upon a particular soil type across different locations? We hypothesized that seagrass response to soil type is spatially dependent as other environmental conditions influencing seagrass growth may vary between locations. Second, should interpretations regarding soil suitability for transplanting be derived from empirical observations of mature meadows, experimental assessments of transplanted seagrass, or are both methods acceptable? As natural seagrass meadows modify their soil environment, we hypothesized that seagrass-soil relationships obtained from mature meadows may not reveal growth limitations experienced by transplanted seagrass in colonizing environments. To assess our first question, we conducted a transplant experiment utilizing *Halodule wrightii* (shoal grass) at two locations, Fort Pierce and Key Largo, Florida in the summer of 2009 utilizing a suite of soils that together composed wide ranges in soil physical and chemical properties. Transplant shoot counts were recorded monthly to assess growth, and transplants were collected for vegetative analysis after five months. Our analysis suggested that soil types do have a significant effect on transplant growth and the influence of soil type on transplants is spatially dependent. In Key Largo, the effect of soil Total phosphorus, Total iron, and organic matter content on transplant growth was found to be highly significant ($p<0.0001$), while soil texture, Total carbon, Total nitrogen, and porewater Sulfides were also found to significantly ($p<0.05$) influence transplant growth. In Fort Pierce, insufficient environmental conditions outside of soil properties diminished the influence of soil properties on transplants, yet soil Total phosphorus, Total nitrogen, organic matter content, and porewater sulfide were found to significantly ($p<0.05$) influence transplant growth. Transplants at the Key Largo site consistently exhibited greater growth responses relative to transplants within the same soil treatments at the Fort Pierce site. To address our second question, we performed monitoring of natural *H. wrightii* meadows and collected subaqueous soil supporting a natural seagrass population which surrounded our Key Largo transplant site to obtain empirical soil limitations for comparison with our experimental findings. We observed that soil organic matter concentration was the only soil property found to significantly influence the coverage and density of natural *H. wrightii* meadows. In contrast to our transplant results, *H. wrightii* populations failed to exhibit significant response with soil total phosphorus, possibly due to low total phosphorus variability inherent at the site. This study suggested subaqueous soils can have a significant influence on the success or failure of seagrass restoration efforts, but cautions the use of soil interpretations, particularly when based on empirical

observations, without consideration of other environmental conditions that may preclude the influence of soils on transplants.

INTRODUCTION

SEAGRASS

Seagrass are a collection of angiosperm plant species that evolved to live in shallow, sub tidal and intertidal, marine and estuarine regions around the Earth excluding the Arctic and Antarctic poles. Seagrass are not "true grasses," (members of the terrestrial Poaceae family); rather, the sixty-seven species of seagrass currently recognized are divided into 6 taxonomic families: Zosteraceae, Cymodoceaceae, Posidoniaceae, Hydrocharitaceae, Ruppiaceae, and Zannichelliaceae (den Hartog and Kuo, 2006).

Seagrass are crucial components of the systems they inhabit. Though seagrass ecosystems are spatially limited, Net Primary Production rates range from ~300-1500 g C/m^2/year, placing them among the most productive ecosystems on Earth (Mateo et al., 2006). This production may be consumed by herbivores, detritivores, and microorganisms, transported to other adjacent ecosystems, or deposited to short-term or long-term storage within soils (Mateo et al., 2006). Seagrass are considered "Ecosystem Engineers," or organisms that modify their environment. Seagrass aboveground biomass exhibits drag on wave orbitals and water currents, promoting the deposition organic and inorganic particulate matter from the water column and inhibiting the suspension of particles and soils where hydrodynamic forces (e.g. water currents, wave exposure) are not excessive (Fonseca et al., 1998, Koch, 2001, Marba et al, 2006). When active, this process leads to coastal stabilization and improved water quality and clarity. Additionally, leakage of oxygen from seagrass roots produced during photosynthesis provides a source of oxygen to soils, and in addition to other leaked or senesced nutrients, promotes microbial activity and nutrient cycling within the soil. Seagrass ecosystems are recognized for their function as a physical habitat, which can serve as a nursery and feeding ground for numerous marine organisms, including many commercially important species. Finally, because seagrass colonize oligotrophic systems and require clear water conditions, these organisms are sensitive to environmental change. This characteristic has led to seagrass being referred to as "Coastal Canaries" that can serve as local and global indicators of ecosystem health (Orth et al., 2006). For these and other services, Costanza et al., (1997) valued seagrass meadows and associated algae beds at \$19,004 ha^{-1} yr^{-1}.

While an appreciation for the functions of seagrass has grown tremendously over the last century, worldwide declines in seagrass populations have concurrently been observed and are accelerating (Waycott et al., 2009). Impacts from anthropogenic sources (e.g. eutrophication, coastal construction and dredging, boating, contamination, and global climate change) and natural events (e.g. disease, subsidence, volcanic eruption, passage of severe storms, grazing, and bioturbation) have been cited as potential causes for these declines (Short and Wyllie-Echevierria, 1996, Fonseca et al., 1998, Hemminga and Duarte, 2002, Walker et al., 2006). These pressures may act independently or synergistically. For example, Blohm (2008) determined that increased water column nutrient concentrations led to accelerated disease spread in the seagrass *Zostera marina*. As human population continues to grow in

coastal areas, the threat of future detrimental anthropogenic impacts to seagrass populations is likely to increase without intervention (Orth et al., 2006). Most notably, because seagrass communities within tropical and subtropical latitudes share important trophic relationships with adjacent mangrove and coral reef systems, damage to either of these ecosystems will likely lead to detrimental effects outside of the damaged system itself (Odgen, 1980, Zieman, 1982, Short and Wyllie-Echevierria, 1996). Finally, the response of seagrass to climate change is largely unknown, but will likely influence rates of seagrass productivity and seagrass distributions (Edwards, 1995, Duarte, 2002, Orth et al., 2006, Blohm, 2008).

SEAGRASS TRANSPLANTING

With the realization that rates of natural seagrass recovery may be magnitudes slower than rates of seagrass loss, seagrass transplanting efforts are often attempted to accelerate seagrass ecosystem recovery utilizing a variety of planting techniques (Fonseca et al., 1998). In an effort to reestablish seagrass in coastal systems where losses or degradation cannot be avoided, United States legislation requires mitigation of seagrass meadows or "habitat of equivalent functional values" (Davis and Short, 1997). Similar legislation exists globally but is not ubiquitous (Hemminga and Duarte, 2000). While successful seagrass transplanting have been accomplished globally (Cambridge et al., 2002, Lewis, pers. comm.), results often meet with varying degrees of success; in a review of 53 published seagrass transplanting efforts, Fonseca et al., (1998) found median and mean values of survival rates of seagrass transplanting units of 35% and 42%. Additionally, seagrass restoration is an expensive endeavor; Fonseca (2006) estimated that restoring subtropical seagrass costs $240,000-$393,000/acre.

TRANSPLANT METHODOLOGY

Much research regarding seagrass planting attempts to optimize the methodology of transplanting by increasing survival and growth of transplant units. Planting units from the published literature fall along a wide range of plant development and size, including broadcast or buried seeds, plant segments with a single vertical shoot, sprigs consisting of a segment of rhizome containing an apical meristem and multiple vertical shoots typically anchored to the transplant surface, and plugs (sods) of seagrass which include associated soils with the transplant unit (Lewis, 1987).

Not all transplanting methods are suitable for all seagrass species (Lewis, 1987). In addition, all transplant methods face limitations that have been noted to diminish survival rates. For instance, sprigs are typically rinsed of rooted soils, which can damage roots and rhizomes and lead to limited nutrient uptake at the transplant site. Additionally, sprigs often require partial burial or anchoring and are more likely to become detached from the transplanted surface than larger planting units, such as plugs. While plugs are often highly viable transplants, it has been documented that bare patches created in donor meadows where plugs were obtained are susceptible to erosion lasting years, particularly in meadows of slow growing species, such as *Thalassia testudinum* (Fonseca et al., 1994). It has been suggested that harvesting of seagrass plugs should be limited to areas destined for destruction, such as prior to dredging activities. The collection and distribution of seagrass seeds are notoriously unreliable with low germination rates (McMillan, 1981) and may also negatively impact population dynamics at the donor site (Hemminga and Duarte, 2000).

The density at which planting units are established has also been noted to influence transplant success; generally, more dense arrangements of transplants yields greater success rates (Sheridan et al., 1998, Worm and Reusch, 2000, Bos and van Katwijk, 2007). Also variation in transplant growth has been traced back to environmental and genetic characteristics of the donor bed. However, these influences are inconsistent between studies (Kenworthy and Fonseca, 1977, Fonseca et al., 1979, Hämmerli and Reusch, 2002).

Recent technological advances aim to improve the efficiency and effectiveness of transplanting methods, although often neither goal is attained (e.g. Bell et al., 2008). However, in Australia and the United States, transplanting using submersible technology and "habitat enhancement" has been documented to improve transplant survival rates, particularly in areas that are historically difficult to transplant due to high water energy (Campbell and Paling, 2003, Paling et al., 2001, Uhrin et al., 2009).

THE IMPORTANCE OF SITE SELECTION IN SEAGRASS TRANSPLANTING

If transplanting is attempting to restore damaged or lost seagrass meadows, identification and removal of the originating stressor(s) from the transplant's environment must first be addressed (Fonseca et al., 1998, Meehan and West, 2002). In fact, natural recolonization by seagrass has been observed without transplanting in areas that had lost seagrass coverage once originating stressors were removed (Campbell, 2003, Lewis, pers. comm.). If transplanting is to be conducted at sites previously uninhabited with seagrass, knowledge of conditions limiting seagrass colonization at these sites should be acquired before transplanting is attempted.

Seagrass growth can be limited by numerous environmental conditions, including insufficient light availability, excessive hydrodynamics, insufficient or excessive salinity, desiccation, and inadequate soils (Hemminga and Duarte, 2000). A literature review pertaining to the influence of soil on seagrass is discussed in Chapter 2. Additionally, biotic interactions such as smothering by drifting macroalgae, bioturbation, competition from invasive species, and grazing may limit seagrass growth (Bos and van Katwijk, 2007, Short et al., 2002, Bando, 2006, Hauxwell et al., 2004). Environmental conditions also influence reproduction in seagrass, thus the long-term durability of a seagrass restoration effort, despite initial success, may be limited if environmental conditions are not considered (Harwell and Rhode, 2007). Increased complexity arises as environmental conditions are species-specific and may not be known for the species to be transplanted, potentially leading to inadequate site selection (Balestri et al., 1998). More research is necessary to determine species-and-site-specific environmental conditions to enhance transplant success.

Several guidelines have been developed to improve seagrass restoration; a reoccurring theme within these guides suggested inappropriate site selection has contributed to the failure of many seagrass transplanting efforts (Fonseca et al., 1987, Fonseca et al., 1994, Fonseca et al., 1998, Short et al., 2002, Campbell, 2002, van Katwijk et al., 2009). For example, Lewis (1987) and Bell et al. (2008) discuss that many previous restoration projects did not consider the potential of a site for supporting seagrass (e.g. sufficient soil depth, excessive bioturbation or suspended sediments, sufficient light, and protection

from human and boating disturbances were not considered or ignored) before transplanting was conducted.

SUBAQUEOUS SOILS

Soil, as defined in *Soil Taxonomy, A Basic System of Soil Classification for Making and Interpreting Soil Surveys*, 2nd Edition is:

> "composed of solids (minerals and organic matter), liquid, and gases …and is characterized by one or both of the following: horizons, or layers, that are distinguishable from the initial material as a result of additions, losses, transfers, and transformations of energy and matter or the ability to support rooted plants in a natural environment"
> (Soil Survey Staff, 1999)

An ecosystem's soil is invariably one of the most critical components of its functioning as it contains the mineral and organic nutrients necessary for autotrophic growth and provides a medium for plant stability. In turn, autotrophic organisms convert solar radiation into the chemical energy that drives the development of and sustains all other trophic levels in the ecosystem. Coincidentally, one of the first requirements for soil development is the presence of vegetation (Crocker and Major, 1955). Only after vegetation becomes established can a soil accumulate organic soil nutrients and fine particles through plant senescence and increased soil stability (Bradshaw, 1997).

Beginning with the pioneering work of Dr. George Demas (Demas et al., 1996, Demas and Rabenhorst, 1999) in the estuaries of Maryland, coastal sediments that undergo pedogenic processes or support rooted vegetation can also be considered, and may be more aptly defined as, subaqueous soils. Pedogenic processes noted to take place in subaquatic environments include additions of Calcium Carbonate and organic matter, losses and translocation of matter and oxygen via bioturbation, and transformation of redox sensitive elements including iron and sulfur (Demas et al., 1996, Demas and Rabenhorst, 1999). For the purposes of soil survey, an arbitrary water depth limit of 2.5 meters has been established (Soil Survey Staff, 1999). Subaqueous soils can be mapped utilizing soil/landscape relationships, similar to the method utilized in terrestrial soil survey (Bradley and Stolt, 2006).

FACTORS OF SUBAQUEOUS SOILS FORMATION

Jenny (1941) listed five factors that influenced terrestrial soil development: climate, organisms, relief, parent material, and time. Variations in these factors across space result in morphological differences between soils; this concept forms the foundation of soil-landscape relationships. Demas and Rabenhorst (2001) note that Folger (1972) developed a model stating that parent material, hydrology, and water depth (bathymetry) were variables influencing the development of coastal sediments located in estuaries. Demas and Rabenhorst (2001) united the frameworks of Jenny and Folger and added additional concepts in their model of subaqueous soil development, which stated that subaqueous soils are a function of climatic temperature, organisms, bathymetry, flow regime, parent material, time, water column characteristics, and catastrophic events.

DEVELOPING SUBAQUEOUS SOIL INTERPRETATIONS

The influence of each soil forming factor on a soil's development varies across space, which results in variations in soil morphologies across landscapes. A soil survey is a document composed by pedologists that provides information relating to the soils present within a geographic area of interest – typically across a county – for use among residents, farmers, natural resource managers, soil professionals, and academics, among others. Soil surveys include maps outlining the extent of soil map units, where each map unit represents a particular soil-landscape relationship. The soil survey includes taxonomic descriptions and laboratory analyses of the physical and chemical characteristics of each soil map unit. This information provides the basis for making interpretations assessing the potential uses a soil can provide for humans and ecosystems. For example, terrestrial soil surveys include interpretations for a particular soil map unit's suitability to support playgrounds, camping and picnic sites, natural habitats for wildlife, buildings, roads, septic tanks, and landfills.

The National Cooperative Soil Survey (NCSS) is a collection of public and private groups under the leadership of the United States Department of Agriculture National Resource Conservation Service. The objective of the NCSS is to advance the science of pedology in the United States, including maintaining an inventory of soils and developing interpretations for soils (U.S. Department of Agriculture, NRCS, 2010). Over the past decade, the NCSS has begun to conduct research with the intent of broadening the scope of soil survey to include subaqueous soils.

Currently, developing meaningful interpretations for subaqueous soils is a NCSS research priority as it will be necessary for subaqueous soil survey to develop interpretations relevant to aquatic environments. For example, the presence of sulfidic materials may influence a subaqueous soil's suitability to serve as dredge or spoil material, as high concentrations of reduced sulfide may oxidize under aerobic conditions, resulting in soils that are highly acidic (Demas and Rabenhorst, 1999, Bradley and Stolt, 2006). Interpretations regarding the subaqueous soil suitability for mooring structures are also being identified (Suraban, 2007). The aquaculture industry may also benefit from an improved understanding of the suitability of subaqueous soils for farming practices (Demas and Rabenhorst, 1999, Bradley and Stolt, 2006).

Another potential interpretation that has been suggested is a subaqueous soil's suitability as a site for seagrass restoration (Demas and Rabenhorst, 1999, Bradley and Stolt, 2006). Fonseca et al., (1979) stated, "The most serious problem facing managers of seagrass restorations is locating appropriate planting sites." Could surveys of subaqueous soils resolve this issue? It has been stated that the development of such interpretations "can only be beneficial" to seagrass restoration (Bradley and Stolt, 2006). Bradley and Stolt (2006) collected empirical data suggested *Zostera marina* distributions across a Rhode Island estuary, Ninigret Pond, were largely explained by subaqueous soil characteristics including texture, carbonate content, rock fragment content, and soil salinity. The authors continued that if suitable soils are located without seagrass present, these areas are sites where seagrass restoration could be attempted.

Yet, past efforts attempting to transplant seagrass into bare or sparsely vegetated areas (except those made bare by "acute events" such as coastal construction) are largely unsuccessful (Fonseca et al., 1979). It is important to consider that the presence of a soil is only one of a suite of several environmental requirements necessary to support seagrass growth, and there are several reasons why a barren sea bottom may not support seagrass. There is evidence that environmental conditions, such as light availability and hydrodynamic forces, may exclude the survival of seagrass at a site regardless of the presence of suitable soils.

NEED FOR RESEARCH

Appropriate site selection of seagrass transplanting efforts has been identified as a limitation of successful survival and growth of transplants. Advances in species-specific environmental conditions relating to subaqueous soils are necessary to enhance site selection criteria, but may be complicated by other site-specific environmental limitations.

Additionally, it is commonly assumed that the soil relationships of mature seagrass meadows are identical to soil relationships of colonizing seagrass. However, post-establishment processes within mature seagrass meadows modify the soil environment; therefore relationships derived from this environment may not be appropriate to apply in colonizing settings.

RESEARCH OBJECTIVES

An analysis of empirical and experimental soil relationships for *Halodule wrightii* in two subtropical systems will be conducted. The following objectives were proposed:

- Determine if the influence of subaqueous soils on *H. wrightii* is site specific
- Determine if empirical or experimental approaches are more appropriate to characterize *H. wrightii*-soil relationships for future interpretations

We hypothesize that the influence of subaqueous soils on *H. wrightii* transplant growth is site specific, and that experimental determinations are more appropriate than empirical observations for determining *H. wrightii*-soil relationships.

FIELD SITES

Key Largo

The seagrass communities of South Florida compose one of the largest populations in the northern hemisphere, spanning over 17,600 km^2 (Fourqurean et. al, 2001). Within this system lies Lake Surprise, a ~1.8 km^2 body of water located less than a kilometer northeast of Key Largo, Florida. Encompassed by a fringing mangrove forest dominated by *Rhizophora mangle* and *Avicennia germinans*, Lake Surprise maintains hydraulic connection to both eastern Florida Bay and southern Biscayne Bay. Since 1912, a causeway (formerly the foundation of Henry Flagler's Florida East Coast Overseas Railroad and more recently, U.S. Highway 1) had bisected Lake Surprise, hydraulically separating the lake into northeastern and southwestern halves (Thorhaug, 1983). As part of the CERP initiative to restore historical freshwater

flow in South Florida, and also to reduce water stagnation leading to potential algal blooms, a multi-million dollar project was undertaken to restore hydraulic connectivity between East and West Lake Surprise. This included the construction of a low level bridge and partial removal of the causeway, completed in late 2008. Fill material included a mixture of limestone rubble, mangrove peat, and calcareous mud, and was used to create a submerged berm that extends across the lake where the causeway once existed.

Seagrass restoration has already been attempted at Lake Surprise. In early 1981, a water pipeline was installed across Lake Surprise, paralleling the western edge of U.S. 1, to meet the growing water demands of the Florida Keys. Thorhaug (1983) conducted initial *Halodule wrightii* transplants in 1981 as opposed to the contracted *Thalassia testudinum* transplants after observing that the backfill operation failed to cover the newly installed pipe with soil, instead leaving only limestone rubble as a transplanting substrate that was believed doubtful to support *T. testudinum*. After 10 months, algal species including *Acetabularia crenulata* and *Batophoria oestedii* composed the dominant autotrophic community of the rubble zone, with *H. wrightii* surviving where a thin veneer of soil was present. To assess the suitability of moderately and severely impacted areas (totaling ~2 ha) for seagrass growth, Derrenbacker and Lewis (1983) preformed a series of transplant experiments. *H. wrightii* aerial runners were collected from adjacent beds and anchored to the sediment or rock with staples. In addition, both *T. testudinum* seedlings and mature transplants were collected and studied for growth in impacted areas. After 7 months, *H. wrightii* increased in coverage from 5% to at least 98% and in density from less than 15 shoots/m^2 to over 150 shoots/m^2 in moderately and severely impacted plots with non-rocky substrates. Concurrently, *T. testudinum* seedlings and rhizome transplants had ~50% and 75% survival, respectively. Transplants upon rocky substrates (using only *H. wrightii*) increased to 18%. Despite initial success, however, a return to the study site by Lewis et al. (1994) revealed the failure of sustained revegetation upon the severely impacted rocky substrates.

Fort Pierce

The Indian River Lagoon (IRL) includes a collection of three estuaries, the Mosquito Lagoon, Banana River, and Indian River, located along Florida's Atlantic coast. These estuaries are separated from the Atlantic Ocean by a 260 km barrier island system that is interrupted by five inlets (Ponce de Leon, Sebastian, Fort Pierce, St. Lucie, and Jupiter). The IRL stabilized over the past 6,000 years during a period of minimal sea level fluctuation, resulting in increased barrier island stability (Davis et al., 1992). Freshwater flows enter the IRL via canals, rivers, and overland runoff. Limited tidal flushing takes place in the northern IRL, where tidal residence times can exceed several months (Smith, 1993). The IRL averages between 1 and 3 meters in water depth (Dawes et al., 1995).

All seven subtropical species of seagrass found in the western hemisphere occur in the IRL. In addition, the IRL is home to rich aquatic life including 397 species of fish (Gilmore, 1995). This great diversity is thought to result from the IRL's geographic location between temperate and tropical climatic regions (Dawes et al., 1995).

Fishler (2006) conducted seagrass transplanting in the Indian River Lagoon utilizing *Halodule wrightii.* One transplant site was selected within an embayment created during the mitigation of a spoil island and another transplant site was selected exterior to the mitigation site within natural seagrass meadows. All sites experienced declines in shoot counts after approximately four months, with two of the three replicates within the mitigation embayment experiencing loss of all transplant units. It was suggested that desiccation or light limitation arising from smothering by drift algae led to these declines.

Seagrass Transplanted

Halodule wrightii is a native seagrass species to the Southeastern United States and Caribbean. It has relatively broad exposure, salinity, and temperature tolerances relative to other species of seagrass. *Halodule wrightii* growth has been described as “opportunistic” and “pioneering” as it is able to quickly colonize an area if sufficient environmental parameters are present (Uhrin et al., 2009). Due to these features, the technique of "compressed succession" is often utilized whereby *H. wrightii* is initially planted at restoration sites to promote a more stable environment for climax species, including *Thalassia testudinum*, to later colonize into (Durako and Moffler, 1984).

BACKGROUND INFORMATION - LITERATURE REVIEW

SUBAQUEOUS SOILS AS A SEAGRASS GROWTH REQUIREMENT

Seagrass require a suite of acceptable environmental conditions to survive and grow, including acceptable ranges in light availability, salinity, temperature, inundation, nutrient and phytotoxin concentrations, rooting depth, soil stability, and hydrodynamic regimes. In addition, ecological pressures present in the seagrass environment (e.g. herbivory, competition, bioturbation) can also have an influence on seagrass growth. Tolerance to growth limitations, however, varies between seagrass species. For example, studies of minimal light requirements reveal that *Thalassia testudinum* requires less surface irradiance (~14%) relative to *Halodule wrightii* (between 24-37%) (Dunton, 1996, Kenworthy and Fonseca, 1996).

Subaqueous soils have direct and indirect connections to many of the environmental requirements listed above, and as a result, have been noted to influence seagrass. There have been two primary methods to relate soil properties with seagrass growth. Empirical studies attempt to study this relationship by documenting the physical, chemical and biological characteristics of vegetated and unvegetated subaqueous soils to better understand the relationship between seagrass and their rooted substrates. However, seagrass are well known to directly influence subaqueous soils through a number of processes, including deposition of organic matter, oxidation of the rhizosphere, limiting the resuspension of settled organic and inorganic particulate matter, and enhancing nutrient recycling. As stated by Barko et al., (1991), “sediment physical and chemical properties are considered as a product of macrophyte growth as well as potential delimiters of growth.” Hence, such empirical seagrass-soil relationships derived from mature seagrass meadows may not accurately depict required soil properties of colonizing (transplanted) seagrass.

Alternatively, many studies have also been conducted to test the effect of soils on seagrass experimentally via transplanting. While generally smaller in scale than empirical observations, experimentation offers improved control that can better elucidate the influence of soils relative to other environmental factors (Short, 1987). Despite an increased awareness regarding the influence of site selection on seagrass transplanting efforts, site-specific environmental parameters, including subaqueous soils, are not always assessed during transplanting efforts.

The following literature review summarizes such studies that noted significant relationships between subaqueous soils and seagrass, beginning with early perspectives and continuing with modern observations and experimentation of the seagrass-soil relationship.

EARLY PERSPECTIVES RELATING SEAGRASS GROWTH AND SUBAQUEOUS SOILS

A review by Pond (1905) notes that many of the earliest accounts of Submerged Aquatic Vegetation (SAV)-soil relationships held that the substrate was merely a site of attachment for aquatic plants. However, later observational accounts found that the presence and abundance of freshwater SAV appeared to be dependent on their rooted soils; as noted in Pond (1905), Seligo (1890) observed more abundant aquatic plants along shorelines adjacent to fertile terrestrial soils, while Forel (1902) observed a reduction in growth of *Elodea canadensis* with time after it invaded a new water body, presumably after it depleted previously abundant soil resources. Documentation of species-specific soil preferences were noted in Pieters' (1901) observations of SAV in Lake Erie.

As a rule, the soils on which the plants occurred in abundance were composed largely of sand and very fine sand, and contained relatively little silt, fine silt, and clay, while the soils on which few or no plants occurred, although the depth of the water and other physical conditions were favorable, were composed largely of silt, fine silt, and clay, and were poor in fine sand and very fine sand. (Pond, 1905).

Pond (1905) conducted a series of experiments utilizing the freshwater species Vallisneria spiralis, Potamogeton perfoliatus, Myriophyllum spicatum, Elodea canadensis, Chara, Potamogeton obtusifolius, and Ranunculus aquatilis trichophyllus, as part of an economic analysis of the Great Lake's fishery. Pond concluded that these species were dependent on soils for both attachment and nutrient acquisition. Phillips (1960) conducted a review of the literature pertaining to the distribution of seagrass including *Thalassia testudinum*, *Halodule wrightii (Diplanthera wrightii)*, *Syringodium filiforme*, and *Ruppia maritime* along Florida coastlines. Phillips noted that while all species had been found growing upon soils ranging in texture from mud (dominated by fine clay and silt-sized particles) to coarse sand, *T. testudinum* growth appeared more limited, and *S. filiforme* occurred less frequently, in coarse sandy soils relative to *H. wrightii* and *R. maritima*.

Transplanting of seagrass in the United States has been conducted as early as 1947 (Phillips, 1960). Initial studies of seagrass transplanting had a tendency to ignore the suitability of a transplant site outside of wave and erosive forces (van Breedveld, 1975). Phillips (1974) and van Breedveld (1975) were several of the first researchers to highlight the importance of suitable soils for successful seagrass transplanting. Phillips (1974) noted the complete failure of *Thalassia testudinum* transplants during a project in Tampa Bay, Florida in 1960 due to erosion of soils beneath transplant units. Van Breedveld

(1975) conducted a series of transplant studies also near Tampa Bay, Florida utilizing *Thalassia testudinum* and *Syringodium filiforme*, noting that variations in methodology and soil type had drastic implications on transplant success.

MODERN FINDINGS

Research relating seagrass growth to subaqueous soils accelerated rapidly during the 1980's and 1990's. As a result of this work, is now well accepted that most seagrass are dependent on the presence of soil for both a source of nutrients and for stability; however, there are a number of exceptions to this rule, and include seagrass capable of colonizing rocky coastlines such as *Phyllospadix torreyi* and *Amphibolis antarctica* (Fonseca et al., 1998, Hemminga and Duarte, 2000, Bull et al., 2004). While seagrass as a collection of species are capable of colonizing a wide range of subaqueous soil types (Zieman, 1982), empirical relationships of natural seagrass meadows have revealed that edaphic controls can have a strong environmental control over submerged aquatic vegetation community structure (Pulich, 1989, Barko et al., 1991, Lee and Dunton, 1999, Koch, 2001, Bradley and Stolt, 2006) and experimental studies have related specific seagrass species growth parameters to soil properties (e.g. Short, 1987, Halun et al., 2002, Terrados et al., 1999).

Before this material is reviewed in-depth, it is important to note that many subaqueous soil properties are dependent on hydrodynamic conditions; for instance, stronger hydrodynamic forces favor the deposition of coarser sediments with lower nutrient contents and diminished soil stability (Demas and Rabenhorst, 2001; Fonseca and Bell, 1998; Frederickson et al., 2004). Many studies have documented decreased seagrass abundance or seagrass transplant survival with increased hydrodynamic exposure (Eleuterius, 1987, van Katwijk et al., 2009). In a meta-analysis of over forty attempted seagrass transplanting in the Wadden Sea, van Katwijk et al., (2009) observed hydrodynamic exposure was the most significant factor relating to transplant growth and survival, and went on to state that the "sediment composition (texture) seems not to be vital for seagrass transplantations and is probably not a habitat requirement." Even the role of seagrass as "ecosystem engineers" has been questioned in regions of particularly high hydrodynamics (Paling et al, 2003). Therefore, while the soils are a requirement of seagrass growth, the relative influence of soil properties on seagrass can vary considerably across locations.

SOIL NUTRIENTS AND ANAEROBIOSIS

While studies have demonstrated the ability of seagrass to uptake nutrients through their leaves, many nutrients – including the majority of plant-available Nitrogen and Phosphorus – are obtained from the sediment (Brix and Lyngby, 1985; Boon, 1986; Erftemeijer and Middelburg, 1995; Lee and Dunton, 1999; Hemminga and Duarte, 2000). Several review papers have suggested that sediment nutrition is of utmost importance to submerged aquatic vegetation community structure (Short, 1987, Barko et al., 1991, Herbert and Fourqurean, 2008). For example, Pulich (1989) found that *Halodule wrightii* and *Ruppia maritima* populations along the Gulf of Mexico coast of Texas appeared to be regulated by soil nutrient and organic matter contents, where *H. wrightii* was found to outcompete *R. maritima* in soils enriched in nitrogen and sulfides, while both species co-occurred in less fertile or sulfide-rich soils.

The parent material of a subaqueous soil can have drastic implications on nutrient availability available for seagrass (Marba et al., 2006). Soils of terrestrial origin are typically dominated by highly weathered, siliceous components, such as quartz, that have low nutrient availabilities. As a result, terrestrial sediments are often nitrogen or both nitrogen and phosphorus limited (Short, 1987, Hemminga and Duarte, 2000). Carbonate soils are typically found to have phosphorus and iron limitations due to the strong affinities of these soils for these ions and the generally limited terrestrial and anthropogenic inflows into these systems (Short, 1987; Duarte, 1995). However, studies have also observed nitrogen limitations in carbonate systems (e.g. Ferdie and Fourqurean, 2004).

The texture (particle size distribution) of a soil also has an influence on soil nutrient status. For example, submerged aquatic vegetation typically exhibit greater root biomass and productivity in coarse soils, which is often presumed to be a response to lower nutrient availabilities (Fonseca et al., 1979). Additionally, it has been demonstrated that coarse grained carbonate sediments have lower phosphate absorption relative to fine grained carbonate soils, due to relative proportions of the surface areas between these particles (Erftemeijer and Middleburg, 1993; Erftemeijer et al., 1994).

Despite the strong association between soil texture and soil nutrients, many studies that have observed significant seagrass growth correlations with soil texture did not assess the effects of soil nutrient status from soil texture per se. For example, Kenworthy and Fonseca (1977) transplanted *Zostera marina* into a coarse sand, sandy loam, and silty loam soils, documenting significantly greater leaf production by transplants with increasing silt content after one month. A more recent study by Park and Lee (2007) transplanted *Zostera marina* off of the southern coast of South Korea utilizing three transplanting methods across three sites of differing subaqueous soil particle size distributions. After approximately 6 months, it was observed that transplants showed the highest survival (93.8%) at a site with loam textured soils, while seagrass transplanted into a sand textured site had the lowest survival (77.1%). Understanding of the drivers of successful seagrass growth would be improved in these studies with statistical isolation of these factors.

Nutrient amendments in the form of fertilizers have been applied during seagrass transplant efforts to elevate nutrient availability, which is thought to result in enhanced seagrass growth and survival. In addition, it has been reasoned that nutrient additions will minimize stress experienced by transplants whose rhizosphere has been significantly altered during the transplanting process (Kenworthy and Fonseca, 1992).

The results of these fertilization efforts have been variable. Several studies have observed clear increases in seagrass growth, biomass, and transplant survival after fertilization (Peralta et al., 2003, Sheridan et al., 1998). Other studies have noted that fertilization was demonstrated to be effective at one site, yet was ineffective at another (Fonseca et al., 1994; Duarte et al., 1995), or found the effect of fertilization depended on the season of application (Erftemeijer et al., 1994; Pulich, 1985). The effect of fertilization was also found to vary by seagrass species within the same location (Kenworthy and Fonseca, 1992, Ferdie and Fourqurean, 2004); in one case, a two-year fertilization project lead to a shift in dominant community structure in which *Halodule wrightii*, a colonizing species, overgrew and

displaced a meadow dominated by *Thalassia testudinum* and nutrient concentrations remained elevated at these sites over two decades after fertilization was terminated (Herbert and Fourqurean, 2008).

Many studies, however, have observed few, or no, improvements in seagrass growth or survival after fertilization (Cambridge and Kendrick, 2009; Bulthuis and Woelkerling, 1981; Worm and Reusch, 2000; Erftemeijer et al., 1994; Armitage and Fourqurean 2006). These and other observations led Lepoint et al., (2004) to hypothesize that fertilization may be ineffective after transplanting as planting unit root structure is damaged during removal from the donor bed; instead, Lepoint instead suggested applying hormones rather than fertilizer to stimulate root growth of transplants. Another study noted that fertilization resulted in increased abundance in macroalgae and microalgae, suggested the assumption that adding slow release fertilizer into the soil does not influence algal growth may be incorrect (Ferdie and Fourqurean (2004).

Seagrass growth and survival has been documented to be limited by elevated levels of soil organic matter; in a review by Koch (2001), soils with organic contents over 5% were generally detrimental to seagrass, but this may vary considerably by species. There are several mechanisms proposed for this detrimental influence. First, nutritional detriments may result from increased diffusional distances in the rhizosphere of soils with low density (Barko and Smart, 1986). Alternatively, while soil organic matter provides autotrophs with an additional source of nutrients, it also supplies microbial populations with sources of electrons to carry out redox-sensitive chemical reactions. When anaerobic conditions are present in subaqueous soils of marine environments, these reactions can result in the formation of phytotoxins including sulfide and ammonium.

Accumulation of sulfide has been documented to diminish photosynthetic rates and photosynthetic efficiency, potentially leading to diminished oxygen release into the rhizosphere and enhanced sulfide toxicity inhibition (Goodman et al., 1995). Soil porewater sulfide levels in seagrass meadows can vary across several orders of magnitude. For example, Calleja et al., (2007) observed sulfide values as low as 4.6 µM in Mediterranean subaqueous soils while Carlson et al., (1994) measured values exceeding 10 mM in Florida Bay *Thalassia testudinum* meadows during a fatality event spanning eight years. Sulfide tolerance varies by species, but reviews have determined seagrass generally become limited by sulfide concentrations ranging from 0.1 – 2.0 mM, although sulfide resistance has been found to be reduced for transplants acclimating to new growing conditions (Terrados et al., 1999; Koch, 2001; Halun, et al., 2002). Seagrass growing in soils elevated in iron are less susceptible to sulfide toxicity as iron can mineralize with sulfide, thereby reducing sulfide porewater concentrations (Calleja et al., 2007).

Ammonium, despite being considered the most abundant nitrogen source in soils for seagrass (Short, 1987), may also be a seagrass phytotoxin if present at elevated concentrations. Ammonium toxicity has been documented in most plant species, and may result from cytosolic cation imbalance, photo protection inhibition, or metabolically-demanding active export of excess ammonium (Britto and Kronzucker, 2002). For example, Kaldy et al., (2004) studied *Halodule wrightii* transplant survival on two dredged deposit sites in the Lower Laguna Madre in Texas over one year. Transplants at both sites failed, and it was observed that soil porewater ammonium concentrations at these sites were significantly greater (up to 900 µM) than nearby control sites (< 50 µM), suggested potential ammonium

toxicity. Brun et al., (2008) found phosphate availability significantly decreased the susceptibility of *Zostera noltii* transplants to be inhibited by toxic levels of ammonium, suggested that the phosphate anions aided in preventing cytosolic charge imbalance while maintaining sufficient N/P ratios necessary for healthy transplant growth.

As noted above, soil texture has an influence on soil nutrient concentrations. Additionally, subaqueous soils dominated by coarse particles have greater porewater (and oxygen) exchange between the water column and soil pores relative to soils dominated by finer (silt and clay sized) particles. Thus, coarsely textured subaqueous soils typically have a thicker aerobic surface layer and less phytotoxin availability than soils dominated by fine textures (Kenworthy et al., 1982; Koch, 2001).

Anaerobiosis has also been noted to influence seagrass seed germination rate. Moore et al., (1993) observed increased *Zostera marina* seed germination under laboratory soil anoxia conditions while seeds exposed to the oxygenated water column demonstrated delayed germination. The field observations of Van Katwijk and Wijgergangs (2004), observing increased germination of *Zostera marina* seeds in finer textured soils relative to sandy soils appear to provide additional support for this finding.

SOIL DEPTH AND DISTURBANCE

Most seagrass require the presence of a stable soil environment for establishment. Therefore, both insufficient soil depths and unstable or disturbed soil environments have been documented to limit seagrass growth.

Insufficient soil depth is often observed in carbonate environments where limestone outcrops may be present at or below a shallow soil surface. For example, Lewis et al., (1994) documented patchy cover of both *Halodule* and *Thalassia* on shell hash and silty substrates and absent cover over rocky areas of Lake Surprise, near Key Largo, Florida. These observations led the authors to suggested,

Failure to completely restore the original level of sediment over the rocky substrate encountered during construction will probably limit the degree of final recovery in these areas of sparse to dense *Halodule* with sparse *Thalassia*, if present at all.

Soil depth is in part a function of a location's hydrodynamic characteristics. Elevated hydrodynamics may result in erosional events that have been noted to destroy seagrass meadows (e.g. Meehan and West, 2002) and uproot seagrass transplants (e.g. Phillips, 1960). Soil erosion may also result in increased turbidity, diminishing seagrass light availability (Kaldy et al., 2004). Similarly, burial of seagrass has also occurred after the passage of severe storms or during construction projects. Seagrass tolerance to burial varies seasonally and by species since seagrass growth rates are most rapid during warm, summer months. Typically, faster growing species and those with large rhizomes (larger carbon storage ability) are better suited to survive short-term burial (Hemminga and Duarte, 2000).

Competition amongst native and invasive seagrass has also been observed, particularly in soils that were recently disturbed. For example, Bando (2006) experimentally determined using reciprocal experiments that in undisturbed areas, *Zostera marina*, a native seagrass to the study area in northwest Willapa Bay,

Washington, outcompeted the exotic seagrass *Zostera japonica*. However, when both seagrass were transplanted to sites where all prior above and belowground vegetation had been removed, after two years *Z. marina* exhibited significant declines in biomass, density, and reproduction, while *Z. japonica* demonstrated significant increases in biomass, density, and reproduction. Hence, Bando (2006) highlights the role of soil disturbance in the ability of this exotic species to invade.

SOIL ORGANISMS

The presence of subaqueous soil biota has also been documented to positively and negatively influence seagrass transplant survival and growth. Such biotic interactions include sheltering, toxicity, presence of a microbial community, and bioturbation.

Macrofauna, such as oysters, have been hypothesized to have both beneficial and detrimental influences on seagrass transplants. For example, Bos and van Katwijk (2007) documented transplanted *Zostera marina* within mussel beds of the Dutch Wadden Sea had significantly greater survival than transplants located 60 m seaward of mussel beds. These authors attributed this biotic facilitation to diminished hydrodynamic exposure experienced by meadows sheltered by mussel beds. Similarly, *Zostera marina* units transplanted seaward of oyster beds near Cortez Island, British Columbia, Canada were documented to also have significantly lower shoot densities (Kelly and Volpe, 2007). However, the authors of this study suggested increased sulfide production resulting from additions of organic matter via oyster metabolism was the cause of this diminished growth.

Microbial populations have a large impact on seagrass. As noted previously, both Sulfate Reducing Bacteria (SRB) and diazotrophs (Nitrogen fixing bacteria) inhabit anaerobic soils characteristic of soils just beyond the oxidized rhizosphere, and produce both plant-available nutrients and phytotoxins (Hemminga and Duarte, 2000). For example, *Thalassia testudinum* grown in axenic cultures fertilized with ammonium and organic nitrogen exhibited chlorosis after one month and increases in C/N ratios after 3 months, while plants grown in non-axenic cultures exhibited healthy growth for 10 months, leading Durako and Moffler (1987) to suggested *T. testudinum* may depend on soil microorganisms to meet their nutritional demands. Similarly, Milbrandt et al., (2008) quantified the influence of subaqueous soil microbial communities on seagrass growth within Tarpon Bay, Florida, in addition to assessing the effectiveness of plug and bare-root transplants. *Thalassia testudinum* bare-root transplants (rinsed of donor soils) were planted into either the naturally occurring subaqueous soils or autoclaved soils at the transplant site along with plugs from the donor site. This study found significantly greater mortality of transplants planted into autoclaved soils, while transplants planted into undisturbed soils grew similarly to controls.

The availability of soil nutrients which are redox-sensitive, including ammonium, nitrate, sulfide, and ferrous iron (linked to phosphorus availability) may also be influenced by bioturbation, or mixing caused by organisms living in the soil. Many studies have documented the negative influence of bioturbation by organisms including crabs, worms, polychaetes, and rays on seagrass transplants (Davis and Short, 1997; Fonseca et al., 1994; Hughes et al., 2000; Siebert and Branch, 2006).

It is important to note that knowledge of factors which control bioturbation activity, such as temperature and seasonality, can result in more or less successful transplanting efforts. This interaction was observed during a reciprocal transplant study of seagrass and burrowing shrimp conducted simultaneously during the United States spring and the New Zealand fall by Berkenbusch et al., (2007). The American *Zostera japonica* transplants appeared to exclude and displace borrowing *Neotrypaea californiensis*, while the New Zealand *Zostera capricorni* transplants declined when placed in plots pre-occupied by the shrimp *Callianassa filholi*. The authors reasoned one possible explanation for this result was that the American transplants were planted during a period of optimal growth, while the New Zealand transplants were planted as the growing season was nearing its end limiting these transplants' ability to cope with the bioturbation effects of the burrowing shrimp. Similarly, Fonseca et al., (2006) observed seasonal bioturbation effects that resulted in the loss of nearly half of seagrass planting units transplanted, leading to a suggestion that plantings should be conducted when bioturbation activity is minimal.

INFLUENCE OF TRANSPLANTED SEAGRASS ON SUBAQUEOUS SOILS

A typical goal regarding seagrass transplanting is for planting units to eventually attain a level of equivalent ecological functionality as natural seagrass meadows. Several studies have observed that seagrass transplants are capable of modifying their edaphic environment. For example, Kenworthy et al., (1980) quantified physical, chemical, and biological subaqueous soil parameters within a transplanted seagrass site in Back Sound, North Carolina over 16 months. The transplanted seagrass were documented to reduce current flow by 19.5% over the planted area. As a result, the upper centimeter of transplanted areas had elevated proportions of silt and clay relative to unplanted areas. Additionally, soil organic matter content increased towards the center of the planted area and porewater ammonium concentrations reached levels similar to surrounding natural meadows (20-330 µM). Biological development was also documented by increased faunal density, diversity, richness and evenness within soils of planted areas.

THE INFLUENCE OF ENVIRONMENTAL CONDITIONS ON SAV TRANSPLANTING

As noted above, soils clearly influence seagrass growth and development. Yet, one may question if particular soil properties have a greater influence on seagrass than others, and how frequently soil properties have been observed to influence seagrass transplanting relative to other environmental conditions. To answer these questions, a meta-analysis of 70 published studies (denoted with "*" in the List of References) documenting efforts to transplant seagrass and freshwater submerged aquatic vegetation was conducted utilizing several search engines with the text "seagrass," transplant*," and "sediment OR soil." Soil properties influencing seagrass transplanting were subdivided into texture, density, stability, disturbance, depth, composition, organic matter content, nutrient content (including fertilizer additions), nutrient toxicity, bioturbation, biotic associations, and microbial community presence. Environmental conditions influencing seagrass transplanting not related to soils were subdivided into categories including biotic associations (e.g. grazing, herbivory, competition), hydrodynamics (e.g. currents, storm activity, wave exposure), light availability, air or water temperature, water column nutrients, desiccation, and water column salinity.

A total of 64 of the 70 studies involved transplanting seagrass, and the remaining 9% of studies documented transplanting freshwater SAV. Only one species of SAV was transplanted in 67% of the studies, two species were transplanted in 23% of the studies, and three or more species were transplanted in 10% of the studies. A total of 24 seagrass species were included in the 64 seagrass transplant studies; however, the frequency of seagrass species utilized in transplanting varied widely with five species (*Zostera marina*, *Halodule wrightii*, *Thalassia testudinum*, *Posidonia australis*, and *Syringodium filiforme*) being included in 63.7% of the studies (Figure 42). Nearly half (43%) of the studies utilized for the meta-analysis were conducted in the United States. Eighteen percent of studies were conducted in Europe, fifteen percent of studies were conducted in Australia, eleven percent of studies were conducted in greenhouses or laboratories, another eleven of studies were conducted in Asia, and two percent of studies were conducted in either Canada or Africa.

Our analysis revealed that 64% of studies found that environmental conditions outside of soils influenced SAV growth or survival (Figure 43). The most frequently cited environmental condition to limit SAV growth, hydrodynamics, was observed to occur in twenty-one percent of studies. Light availability limited transplant growth in eleven percent of studies, water column temperature and biotic associations external to soils limited transplants in ten percent of studies each. Desiccation limited transplant growth in nine percent of studies, while water column nutrients and water column salinity each limited transplants in one percent of studies.

Seventy-four percent of studies observed environmental conditions related to soil properties influenced SAV transplants (Figure 44). Physical soil properties influenced a total of 33% of studies, chemical soil properties influenced 27% of studies, and biological soil properties influenced 15% of studies. The most common soil property to limit SAV transplanting was soil nutrients, which included the addition of fertilizers to the soil. This component was found to influence SAV transplanting in seventeen percent of studies; however, soil nutrients was also the most observed environmental condition to not influence seagrass transplanting in thirteen percent of studies. Soil texture was found to influence transplants in eleven percent of studies, soil stability in ten percent of studies, and bioturbation in nine percent of studies. All other soil properties assessed were found to influence transplants in six or less percent of studies.

In sum, over eighty-four percent of studies included in this analysis observed that environmental conditions (both related and unrelated to soil properties) influenced SAV transplants. This confirms the growing consensus stressing the importance of considering environmental conditions during SAV transplanting efforts. The finding that more studies observed soil-related influences relative to non-soils environmental influences is likely a result of our search criteria, which required "soil" or "sediment" to be included in the text of studies utilized for this survey. It is probable that higher percentages of non-soils related environmental conditions will be found to influence SAV transplanting in a more general survey of the SAV transplanting literature.

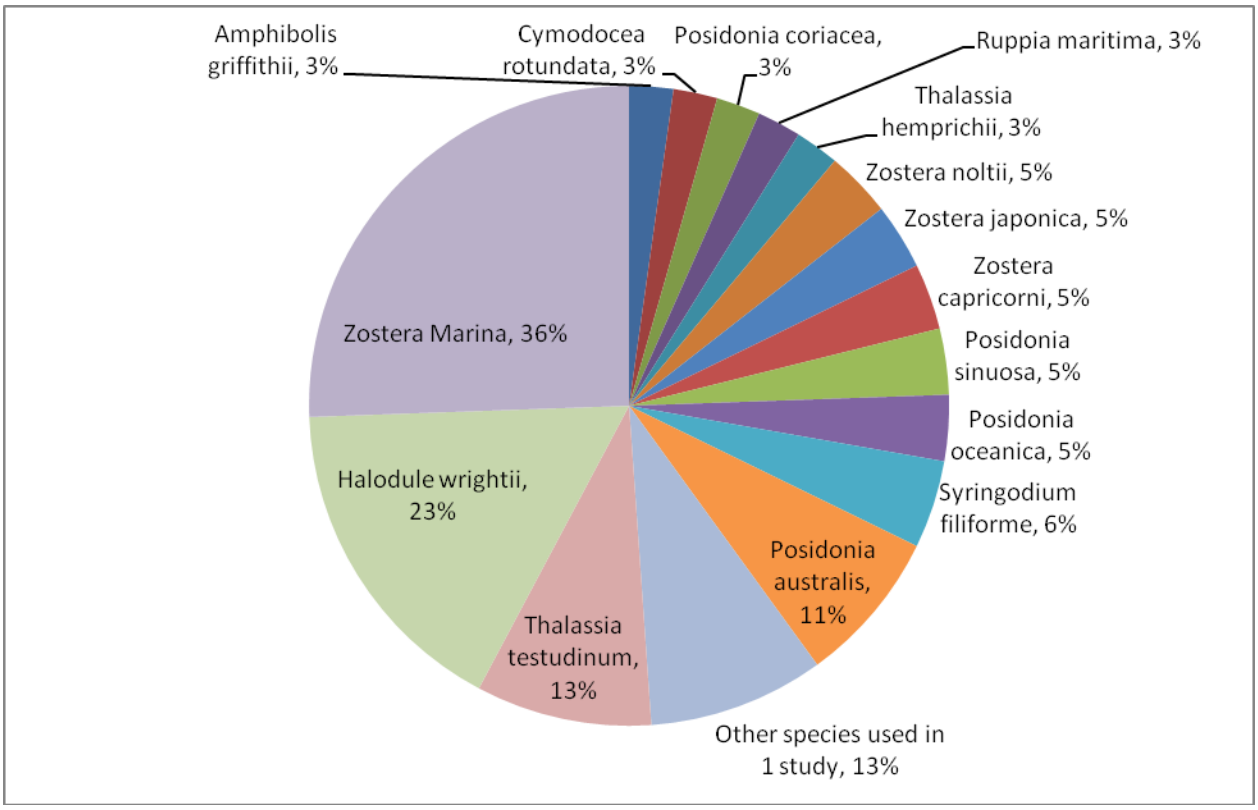


Figure 42. Frequency of utilization of seagrass species in the 64 seagrass transplanting studies analyzed.

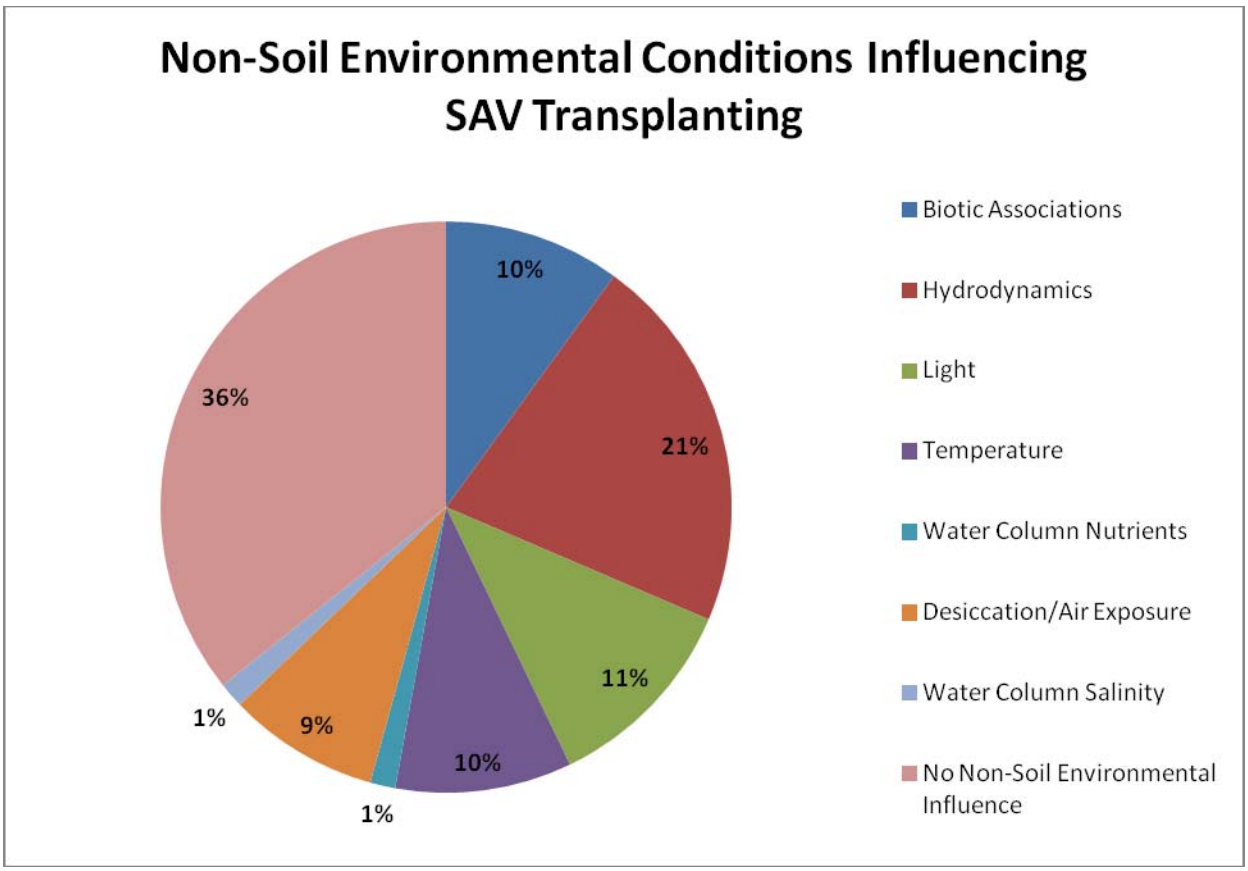


Figure 43. Percent of occurrence of environmental conditions limitations outside of soils in SAV transplant studies

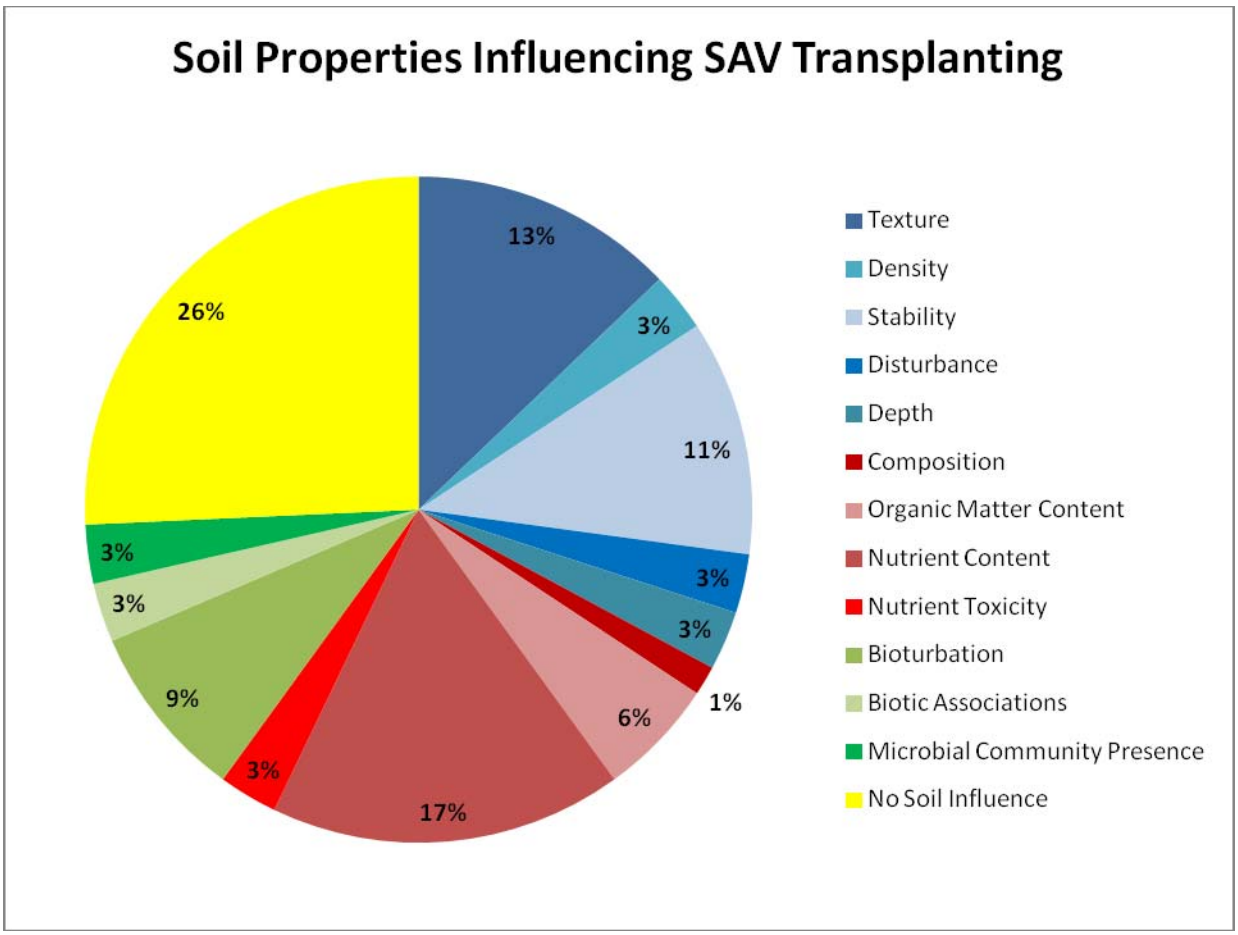


Figure 44. Percent of occurrence of soil-limitation in SAV transplant studies.

METHODS

TRANSPLANT SOIL COLLECTION AND PLACEMENT

Eleven soil samples from terrestrial, intertidal, and subaqueous environments across the Florida Peninsula, together composing a range of parent materials, soil textures, and organic matter and nutrient contents were collected in June 2009 (Figure 45). Each soil was placed into a 6-quart Sterilite® (34.3 cm x 21.0 cm x 12.1 cm) plastic containers to a depth of 7.6 cm and replicated 5 times at each of two sites, i.e. the Indian River Lagoon near Ft. Pierce, Florida (N 27° 28' 41", W 80° 19' 19") and Lake Surprise, near Key Largo, Florida (N 25° 11' 08", W 80° 22' 32"), in an incomplete randomized block design (Figure 46). To enhance stability, rope was drawn through experimental units of each row and tied to cement blocks placed at each end. Subaqueous soils (soils 1-6) were only placed at sites where corresponding parent materials were present (i.e. siliceous in Fort Pierce, carbonaceous in Key Largo). Soils (7-11) obtained from locations not supporting seagrass populations at the time of collection were placed at both transplant locations.

SEAGRASS TRANSPLANTATION

H. wrightii aerial runners were collected near Key Largo, FL., taking care to limit each planting unit to 4 vertical rhizomes with an apical meristem (Figure 47). To limit errors associated with individual planting units on success or failure of a transplanting, two *H. wrightii* planting units were anchored into each experimental unit in July 2009 utilizing four ~ 5 cm segments of plastic coated Hillman Twist Wire bent into a "V" shape (Figure 48).

TRANSPLANT FIELD MEASUREMENTS

Visual quantification of planting unit survivorship and vertical rhizome (shoot) counts were conducted to track seagrass transplant response. Coverage of experimental units by the drift algae *Gracilaria* sp. at the Fort Pierce block location was necessarily removed to conduct this effort in October 2009. Recruitment by other algal species (most notably *Acetabularia crenulata*, *Halimeda incrassata*, and *Penicillus capitata* at the Key Largo block location) presence was noted. The algae were not removed. Sulfide levels were obtained from experimental units beginning approximately 1 month after the transplantation date, and were measured using an Accumet ® Portable Laboratory device.

TRANSPLANT COLLECTION

In November 2009, shoot counts were conducted for a final time at Key Largo and Fort Pierce after 155 days in the field (Figure 49). Transplants were removed from soil treatments taking care to preserve transplant shoots, rhizomes, and roots and were brought back to the lab where they were rinsed of any remaining soil (Figure 50). Leaf and rhizome lengths were measured to the nearest 0.1 cm using a ruler and the number of leaves per vertical rhizome was recorded for each shoot. Canopy height was calculated as the 80th percentile of leaf length for each experimental unit. Transplants were then separated into shoots (vertical rhizomes), horizontal rhizomes, and roots and were dried in an oven at

70° C to quantify aboveground, belowground, and total plant biomass. Root to shoot ratio was calculated by dividing the sum of root and horizontal rhizome biomasses by the shoot (vertical rhizome) biomass.

NATURAL SEAGRASS MONITORING AND SUBAQUEOUS SOIL COLLECTION

Monitoring of seagrass vegetation was conducted within Lake Surprise, near Key Largo, Florida in June 2010. Seagrass percent cover and shoot density were recorded by species within ten 1 m^2 haphazardly placed plots within 5 meters of 98 fixed monitoring sites (Figure 51). One soil core varying in depth from 5-20 cm, depending on the depth to limestone bedrock, was collected from each plot and sectioned into 5 cm increments for laboratory analysis.

SOIL LABORATORY ANALYSIS AND CHARACTERIZATION

Soils utilized in the transplant study and obtained during monitoring were analyzed in the laboratory for organic matter content (LOI) as determined by the percent weight loss between oven drying overnight at 110° C and at 400°C for 16 hours. Dry bulk density was determined after obtaining an oven dry weight of samples. Total phosphorus (TP) was analyzed using the ashing technique (Anderson, 1976) with a Bausch and Lomb Spectronic 1001 spectrophotometer. Removal of soil organic matter while conserving soil carbonate material for particle size analysis was accomplished using $NaClO_3$ with centrifugation and Na_2CO_3 for dispersion, followed by the pipette method (Gee and Bauder 1986). Soil Total carbon (TC) and Total nitrogen (TN) were obtained using a Costech Elemental Combustion System (ECS) 4010. Soil and plant tissue Total iron (TFe) and Total nitrogen (TN) analysis were completed by Waters Agricultural Laboratories, Inc., in Camilla, Georgia.

Soil treatments were characterized based on their location relative to sea level (subaqueous, intertidal or terrestrial), dominant parent material (siliceous or carbonaceous), and texture. Three subaqueous soils collected from Fort Pierce, Florida, were characterized as subaqueous siliceous loamy sand 1 (SSLS1), subaqueous siliceous loamy sand 2 (SSLS2), and subaqueous siliceous sandy loam (SSSL). SSSL was not observed supporting seagrass at the time of sample collection. Three subaqueous soils supporting seagrass at the time of collection near Key Largo, Florida were characterized as subaqueous carbonaceous clay loam (SCCL), subaqueous carbonaceous loam 1 (SCL1), and subaqueous carbonaceous loam 2 (SCL2). A subaqueous soil treatment consisting of fill material utilized during the construction of the US Highway 1 bridge across Lake Surprise, near Key Largo, Florida was characterized as a subaqueous carbonaceous gravelly clay loam (SCGCL). A terrestrial siliceous sand (TSS) was collected near Fort Pierce, Florida. A subaqueous mucky peat (SMP) dominated by organic matter derived from mangroves was collected within Lake Surprise, near Key Largo, Florida. An intertidal siliceous sandy loam (ISSL) was collected near Titusville, Florida and a terrestrial carbonaceous sandy loam (TCSL) was collected near Ochopee, Florida.

STATISTICAL ANALYSIS

Least square means differences were calculated for each plant response variable measured by soil type and location using PROC GLM. A mixed model was first constructed to determine if soil treatment and

transplant location (block) had a significant effect on *H. wrightii* plant responses using SAS (version 9.2) software produced by the SAS Institute utilizing PROC GLIMMIX. Subsequently, individual mixed models were created for each transplant site and the natural setting to determine the effect of specific soil properties on *H. wrightii* growth parameters.

Table 2. Transplant soil collection locations and block placement.

Soil	Latitude	Longitude	Block
SSLS1	27.477	-80.325	FP
SSLS2	27.474	-80.309	FP
SSSL	27.486	-80.316	FP
SCCL	25.183	-80.382	KL
SCL1	25.194	-80.404	KL
SCL2	25.193	-80.399	KL
SCGCL	25.180	-80.382	FP, KL
TSS	27.462	-80.331	FP, KL
SMP	25.186	-80.382	FP, KL
ISSL	28.643	-80.749	FP, KL
TCSL	25.972	-81.316	FP, KL

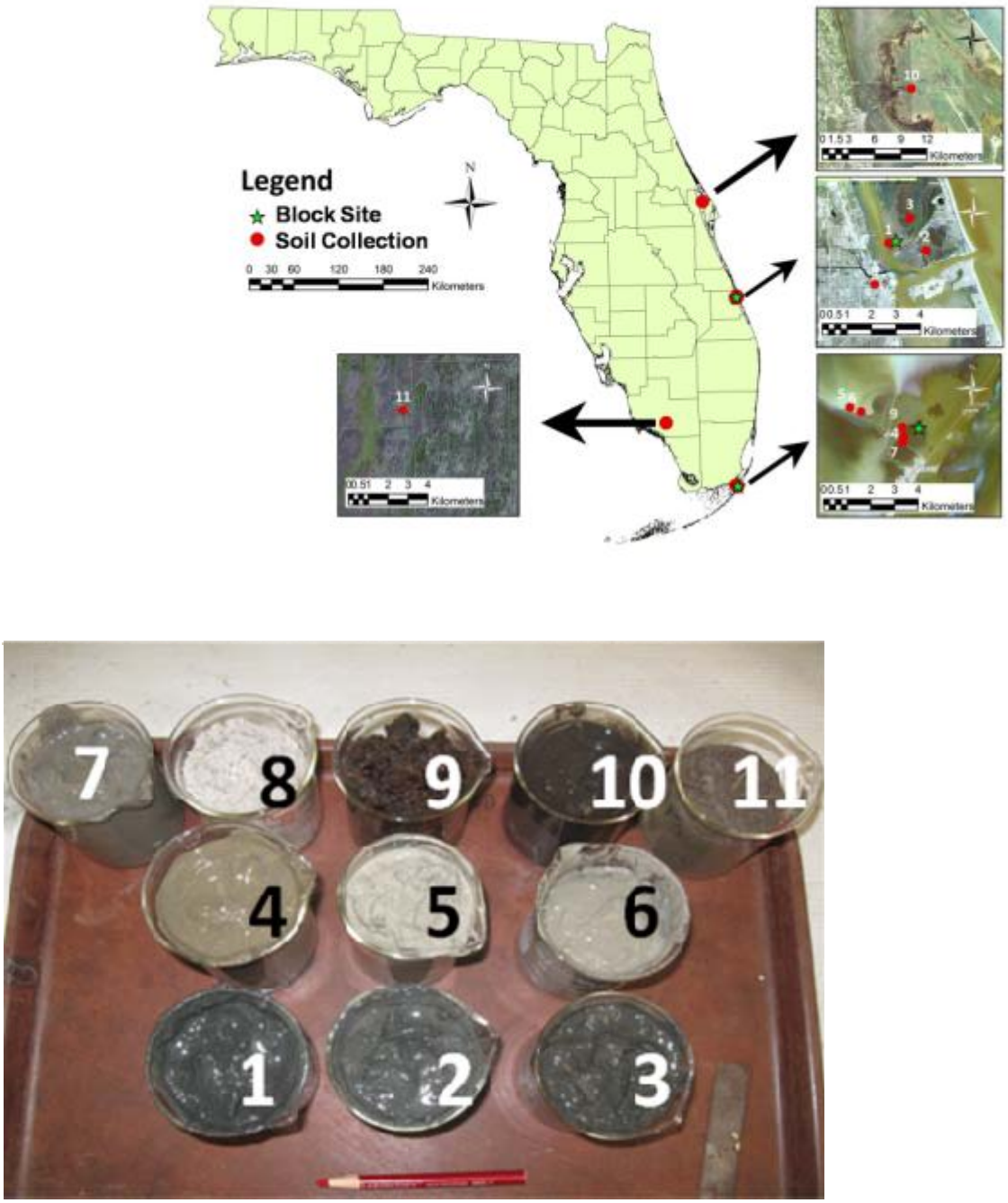


Figure 45. (Above) Locations of 11 sites where soil treatments were collected. Two transplant (block) sites were established near Fort Pierce and Key Largo, Florida. (Below) The eleven soil treatments utilized during transplanting.

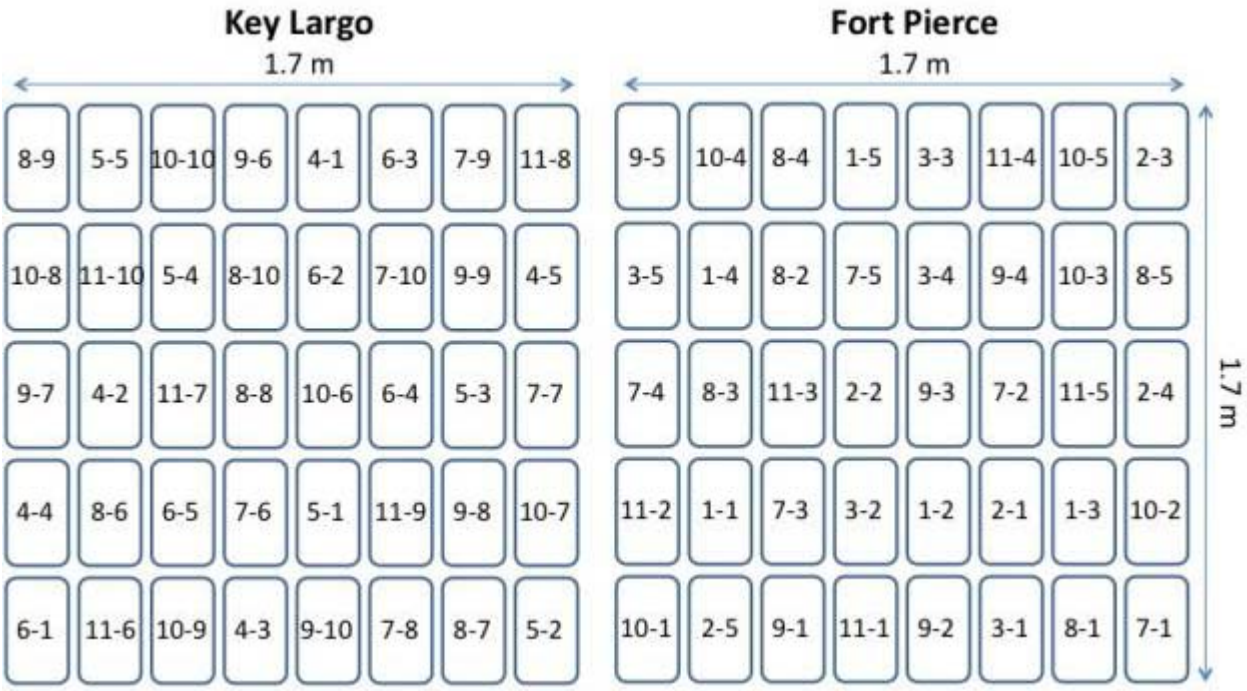


Figure 46. Randomized blocks of transplant pots located at the Key Largo site (left) and the Fort Pierce site (right).

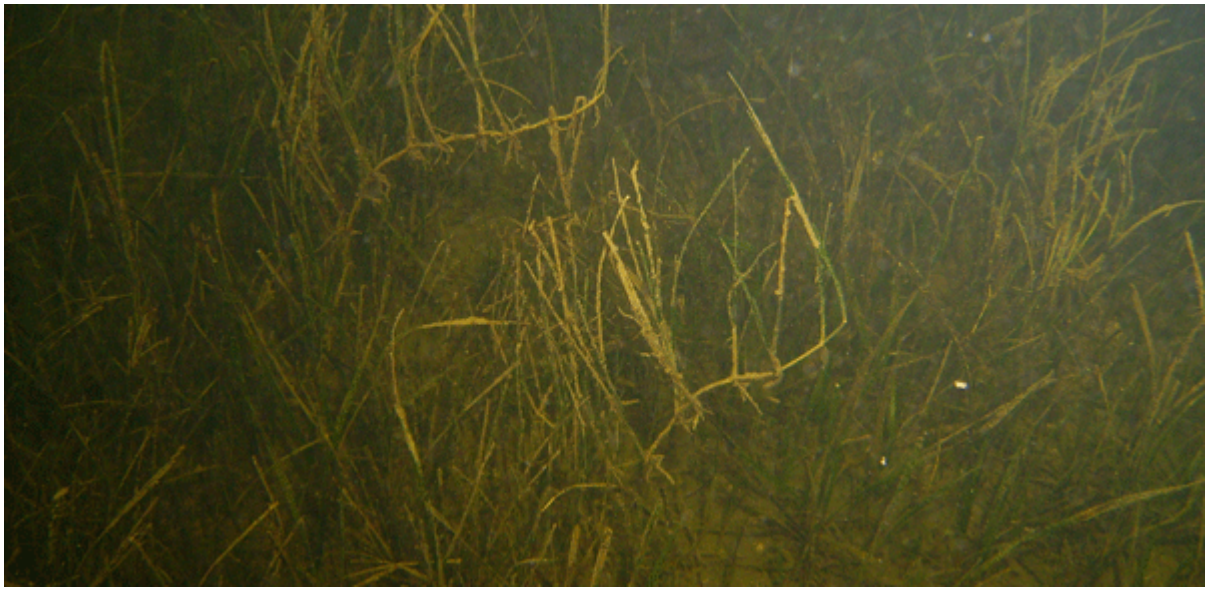
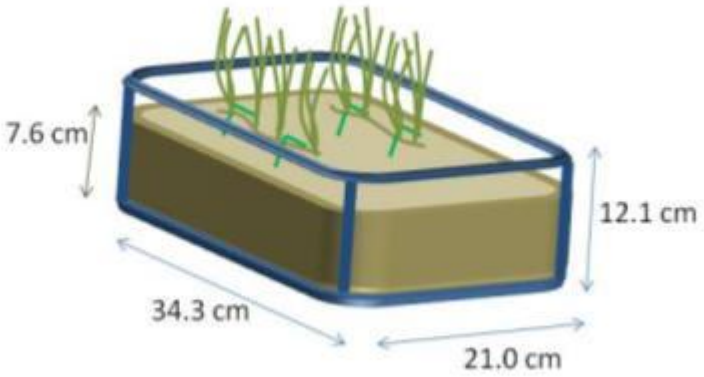


Figure 47. *H. wrightii* aerial runners collected in Lake Surprise, Key Largo, FL for transplantation. A planting unit is considered the last four vertical rhizomes attached to each horizontal rhizome. Note runners are free of parent soil to reduce possible contamination.



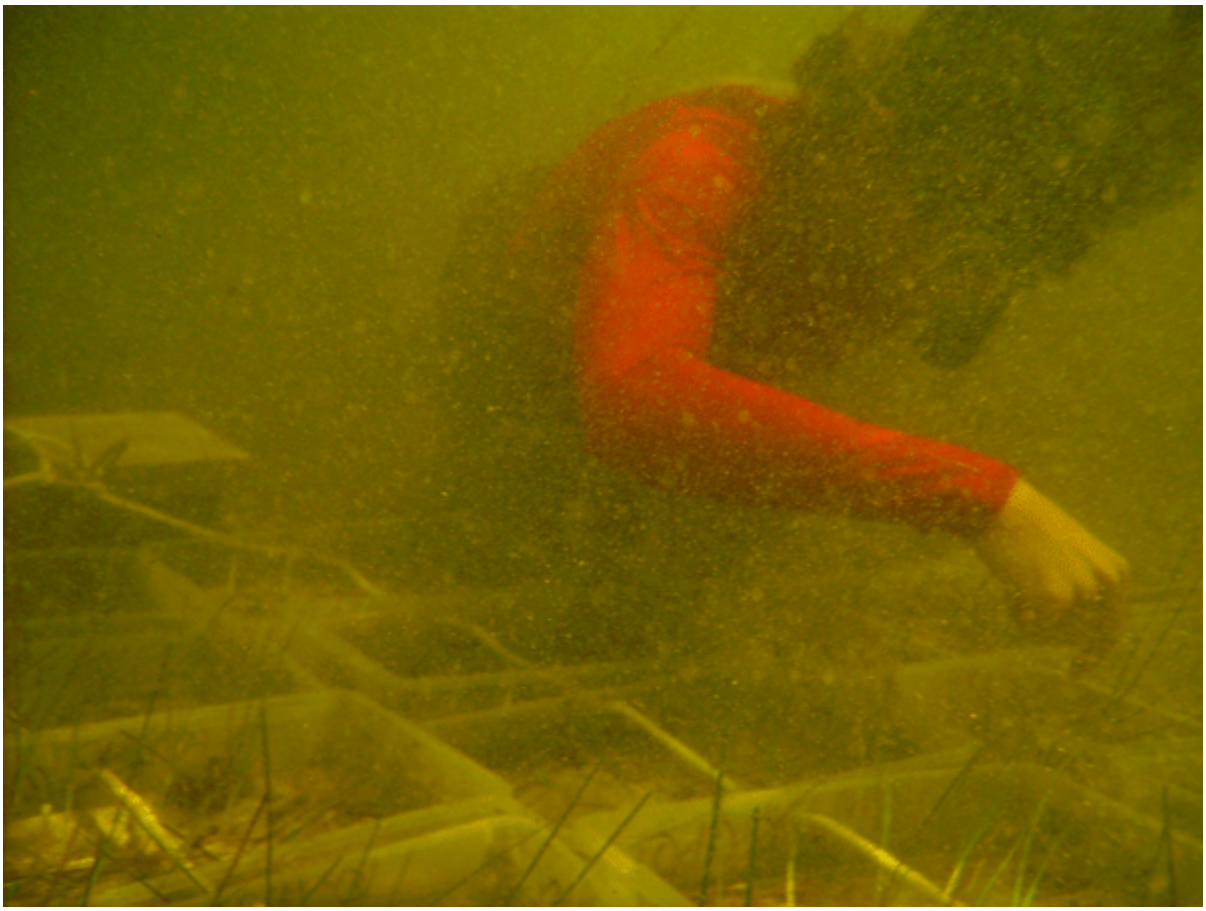


Figure 48. (Above) Experimental unit contains two planting units, each composed of a horizontal rhizome with four vertical rhizomes. (Below) Transplantation of *H. wrightii* planting units into experimental units.

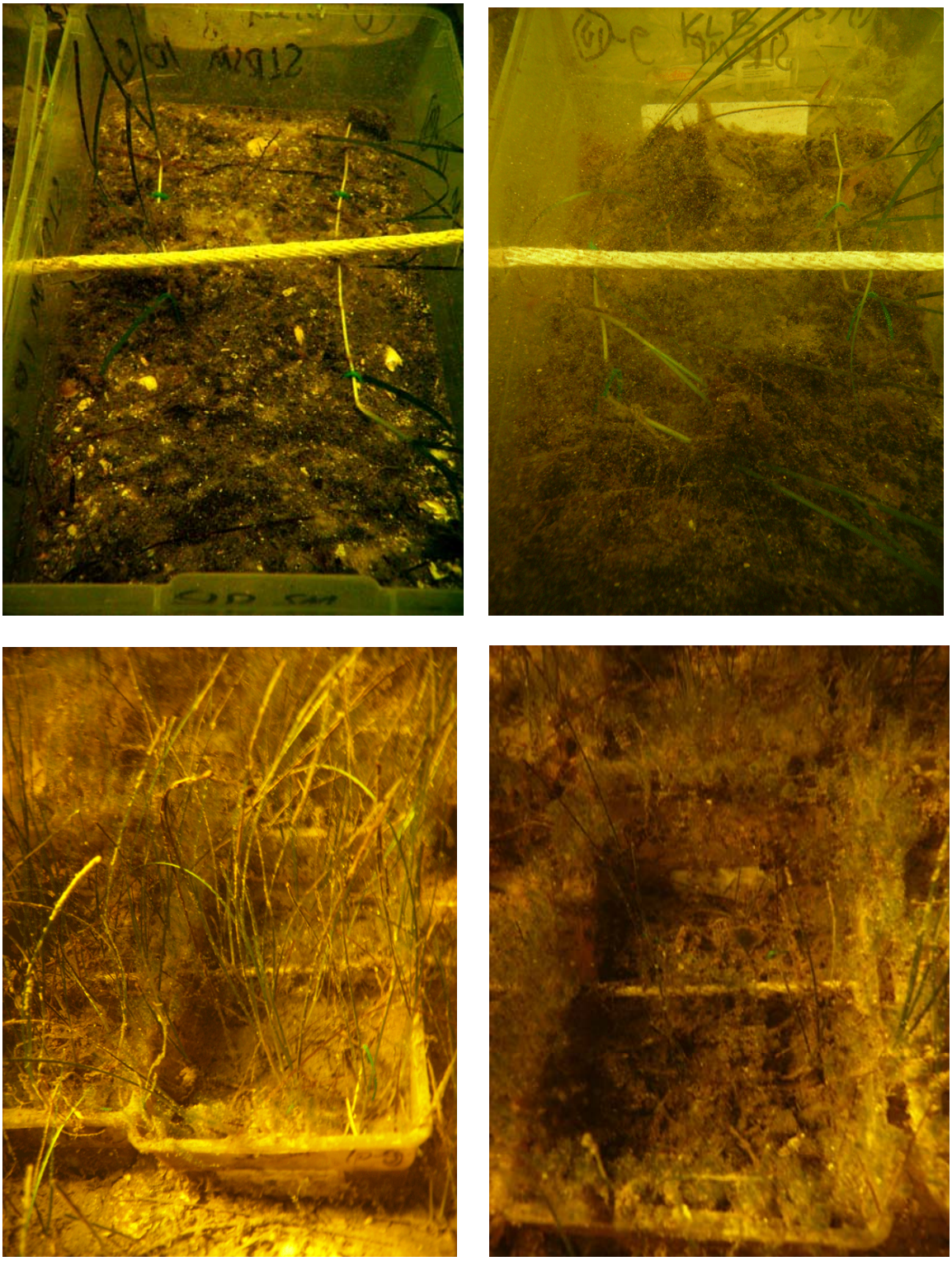


Figure 49. Close-up of an experimental unit of Soil 10 (ISSL) at Key Largo site after transplanting (above left) and after 5 months of growth (below left). Close-up of experimental unit of SMP (SMP) after transplanting (above right) and after 5 months (below right).

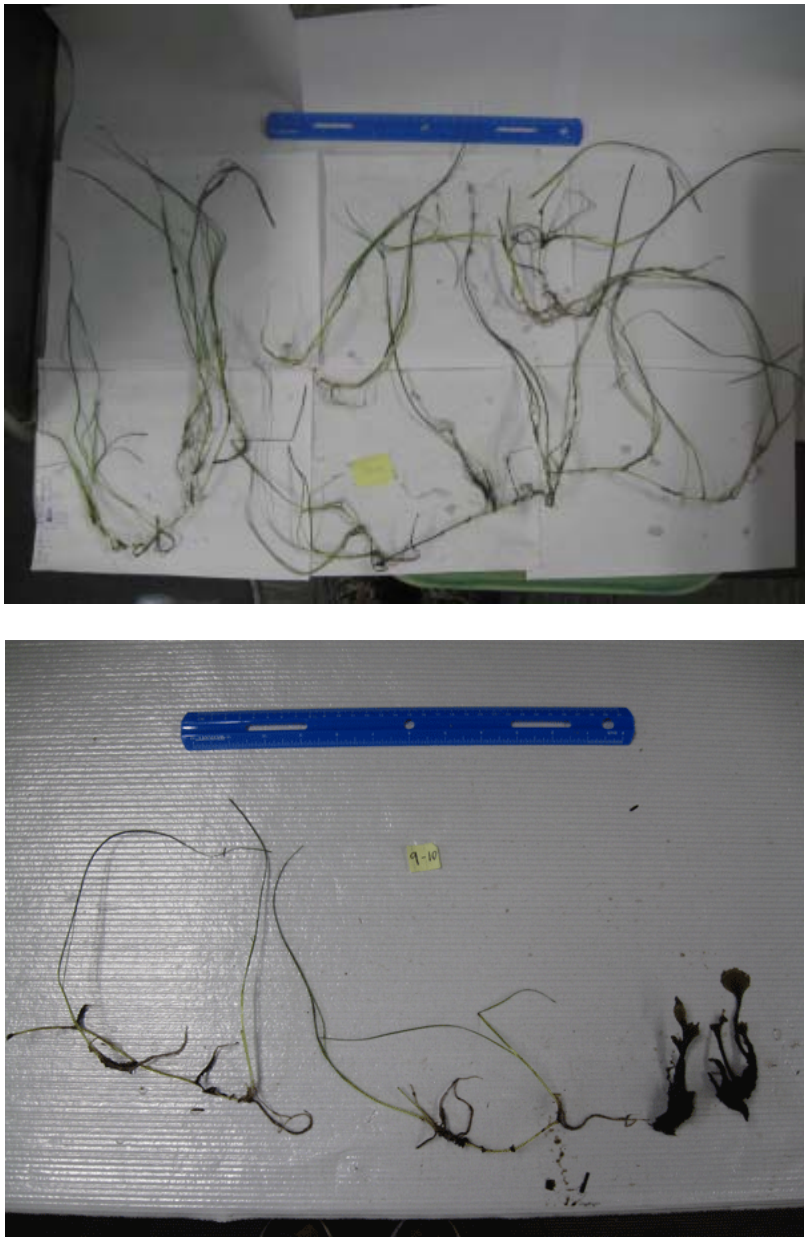


Figure 50. Plant biomass collected from experimental unit of soil type 10 (ISSL) pictured (above) and soil type 9 (SMP) pictured (below).

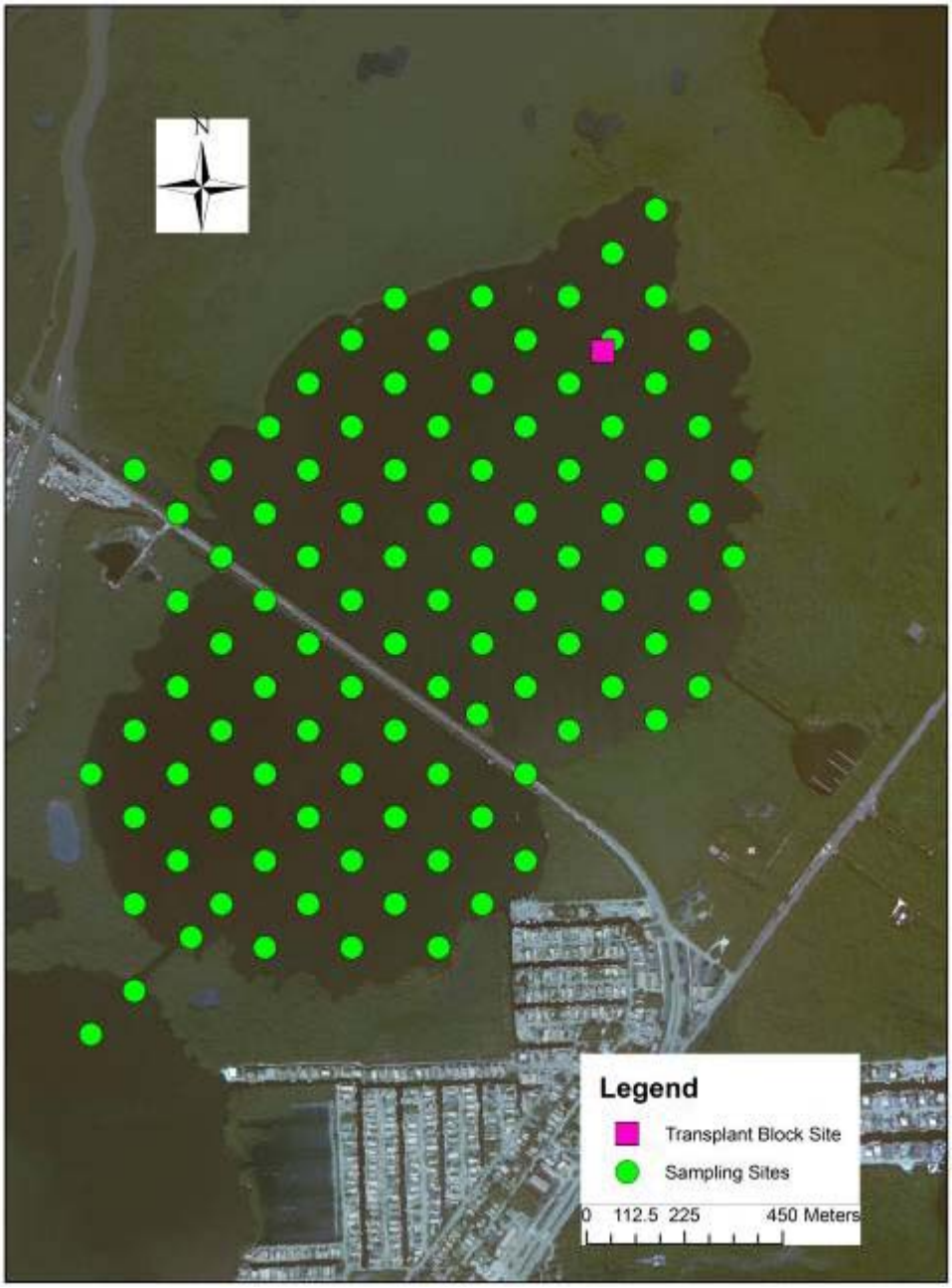


Figure 51. Locations of transplant site and sampling sites within Lake Surprise where natural vegetation and soil samples were collected. The city of Key Largo, Florida is visible in the lower center and right of the picture.

RESULTS

TRANSPLANT SOIL PROPERTIES

Soil treatments utilized for the transplant experiment contained significantly different physical and chemical properties (Table 3) at both blocks. Soil textures ranged from sand (TSS) to clay loam (SCGCL), soil organic matter from 0.0% (TSS) to 53.2% (SMP), and soil total phosphorus ranged from 24 mg/kg (TSS) to 248 mg/kg (TCSL) at the Key Largo site and from 24 (TSS) to 846 mg/kg (SSSL) at the Fort Pierce transplant site.

H. WRIGHTII TRANSPLANTS

Both soil type and transplant location had a significant effect on *H. wrightii* transplant growth responses including shoot counts (Figure 52), canopy height (Figure 53), leaves per shoot (Figure 54), aboveground biomass (Figure 55), horizontal rhizome length (Figure 56), belowground biomass (Figure 57), plant biomass (Figure 58), and root to shoot ratio (Figure 59). A summary of the influence of particular soil properties on these growth responses is listed in Table 32.

Table 3. Transplant soil physical and chemical properties (standard error in parenthesis).

Soil	Sand (%)	Silt (%)	Clay (%)	LOI (%)	TC (mg/kg)	TN (mg/kg)	TP (mg/kg)	TFe (mg/kg)	S (mM)
SSLS1	86.8 (2.7)	5.6 (2.0)	7.7 (0.8)	2.2 (0.1)	761 (36)	21.5 (0.9)	0.9 (0.0)	1.03	0.4 (0.2)
SSLS2	88.3 (2.1)	4.8 (2.2)	6.9 (0.9)	0.7 (0.0)	590 (16)	9.6 (0.3)	0.5 (0.0)	3.85	0.3 (0.2)
SSSL	67.1 (4.3)	19.8 (5.8)	13.1 (1.6)	5.8 (0.3)	846 (50)	27.0 (0.3)	1.7 (0.0)	3.99	0.1 (0.2)
SCCL	45.0 (2.0)	27.9 (3.8)	27.1 (2.5)	11.5 (0.5)	131 (11)	112.6 (1.5)	1.7 (0.1)	0.785	0.1 (0.1)
SCL1	44.2 (1.7)	33.7 (4.1)	22.0 (3.2)	4.7 (0.4)	52 (6)	113.8 (0.8)	1.9 (0.1)	0.845	0.2 (0.1)
SCL2	40.2 (2.4)	34.5 (4.1)	25.4 (3.3)	7.2 (0.2)	72 (10)	116.6 (0.2)	3.3 (0.0)	0.53	0.3 (0.2)
SCGCL	20.0 (1.7)	48.6 (3.6)	31.4 (4.4)	8.7 (0.8)	167 (9)	131.0 (0.2)	3.4 (0.0)	0.865	0.1 (0.0)/ 0.0 (0.0)
TSS	97.4 (0.6)	0.0 (0.9)	2.6 (0.6)	0.0 (0.0)	24 (7)	0.3 (0.1)	0.2 (0.0)	11.41	0.1 (0.0)/ 0.0 (0.0)
SMP	N/A	N/A	N/A	53.2 (1.2)	136 (9)	153.3 (2.0)	7.3 (0.2)	1.375	0.2 (0.1)/ 0.1 (0.1)
ISSL	79.7 (1.8)	7.0 (3.7)	13.3 (2.4)	5.4 (0.3)	148 (9)	48.4 (5.7)	2.8 (0.3)	28.57	0.1 (0.0)/ 0.3 (0.1)
TCSL	60.9 (2.7)	23.9 (0.5)	15.2 (2.4)	11.6 (0.2)	248 (15)	91.2 (1.7)	6.2 (0.2)	1.35	0.3 (0.1)/ 0.4 (0.1)

Table 4. Significance values of soil chemistry on transplant tissue chemistry. Values of p<0.05 are in italic bold type. Insufficient shoot biomass was available for TN shoot analysis in Fort Pierce.

	Transplant Part	TN	TFe
Fort Pierce	Rhizome	0.4958	0.7982
	Shoot	.	0.4949
Key Largo	Rhizome	0.6249	0.0305
	Shoot	0.5916	0.929

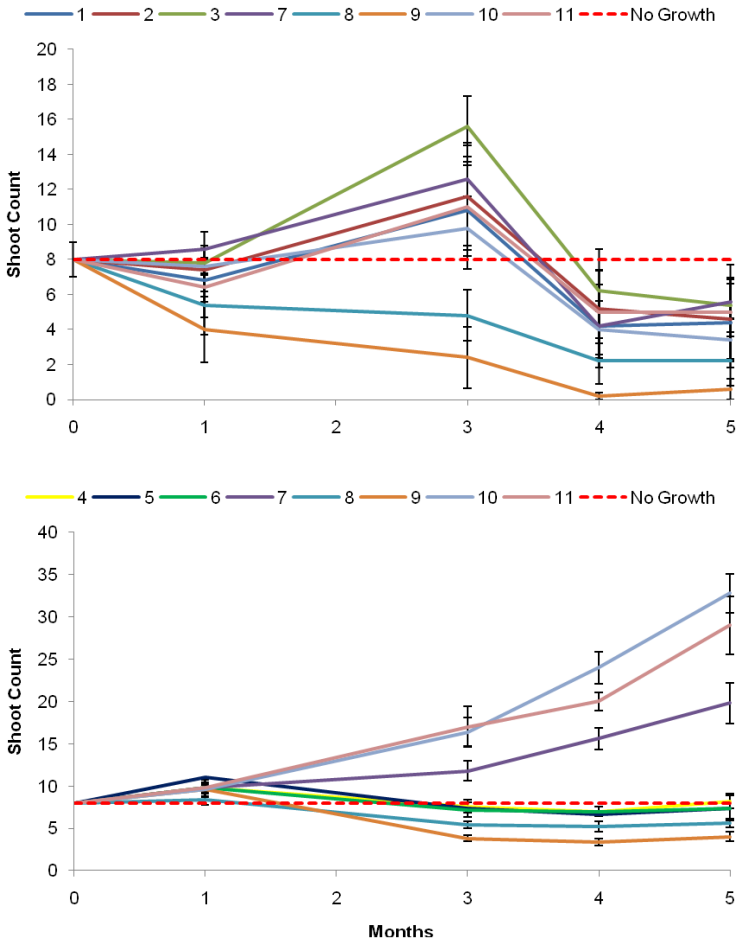


Figure 52. Transplant growth by soil type in Fort Pierce (Above) and Key Largo (Below). The dashed red line equals a shoot count of 8, the number of shoots transplanted at the start of the study, and represents zero net growth.

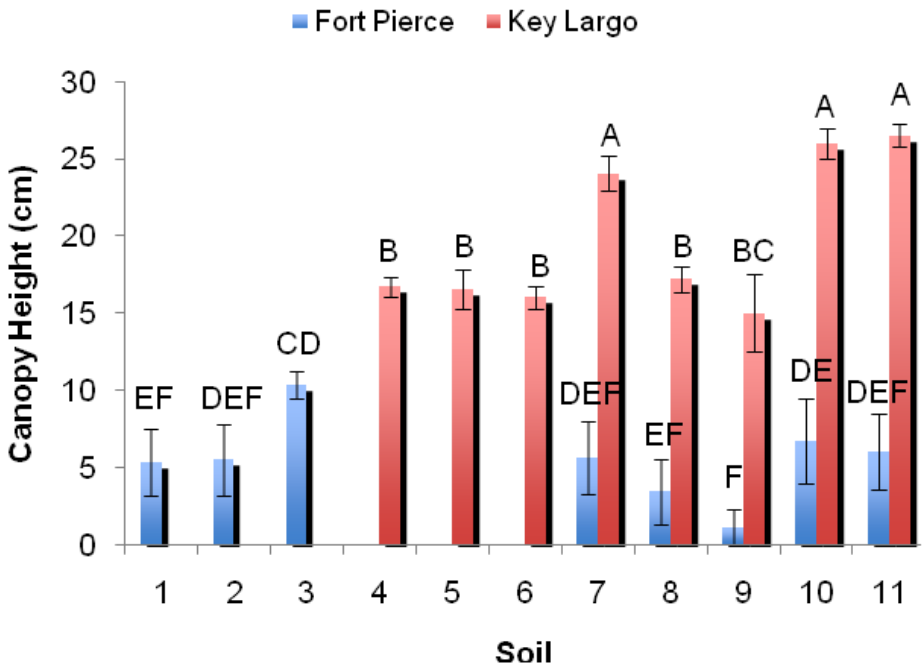


Figure 53. *H. wrightii* transplant canopy height varied significantly by soil type and location.

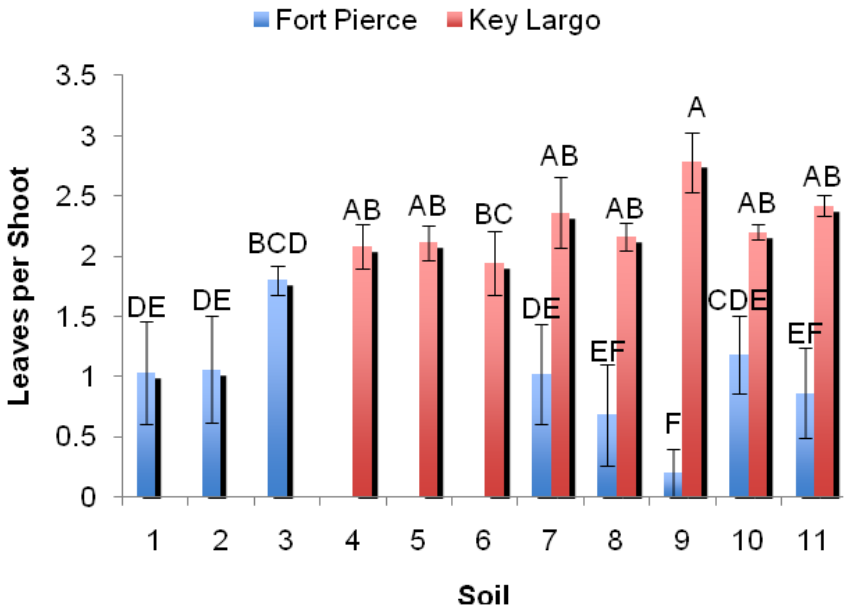


Figure 54. The number of leaves per shoot (vertical rhizome) of *H. wrightii* transplants varied significantly by soil type and location.

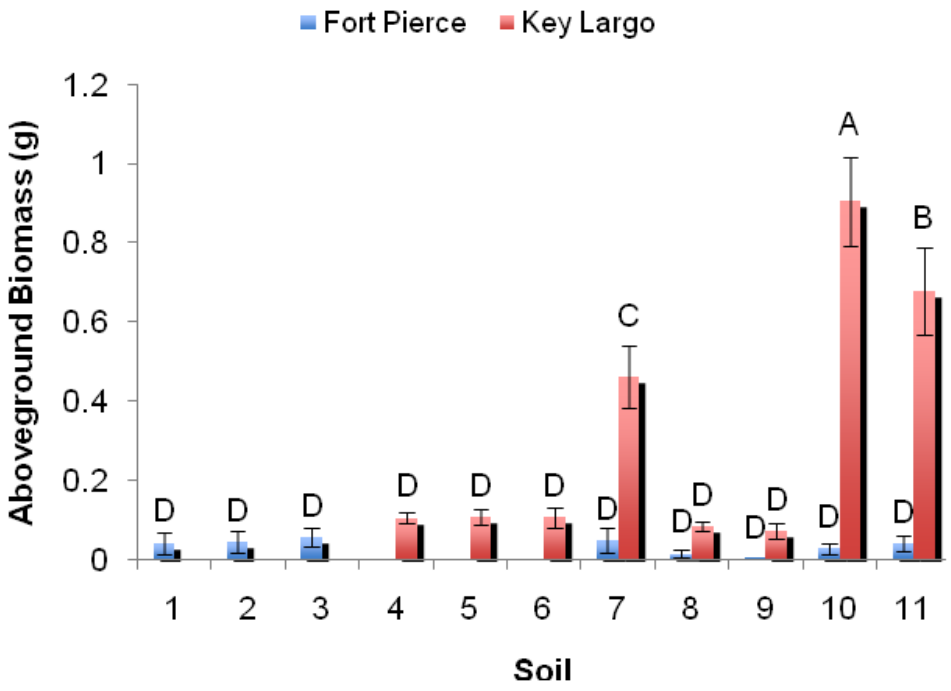


Figure 55. *H. wrightii* transplant aboveground biomass varied significantly by soil type and location.

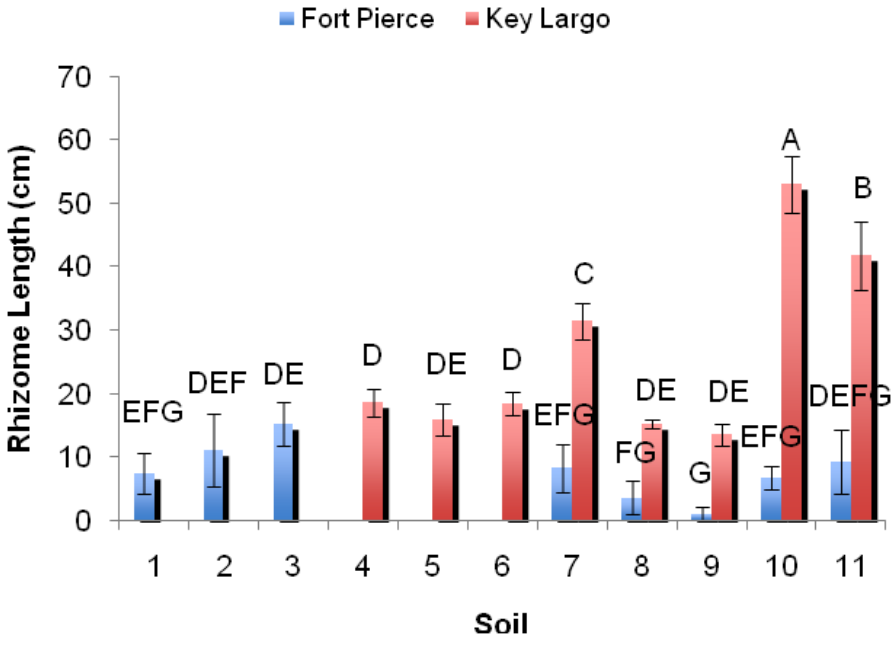


Figure 56. *H. wrightii* transplant (horizontal) rhizome length varied significantly by soil type and location.

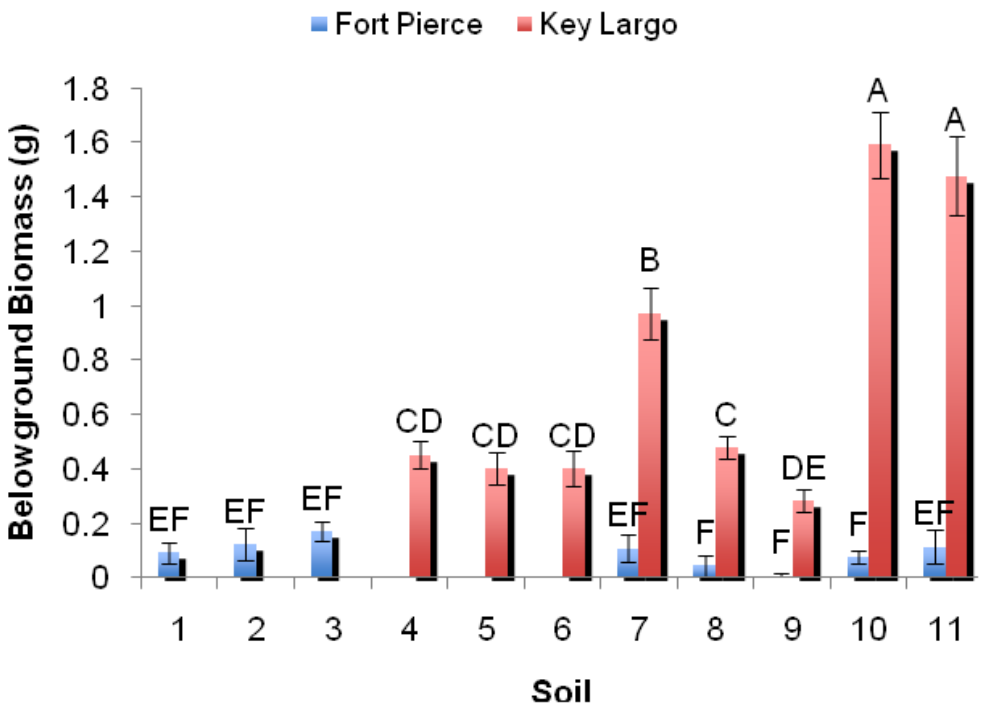


Figure 57. *H. wrightii* transplant belowground biomass varied significantly by soil type and location.

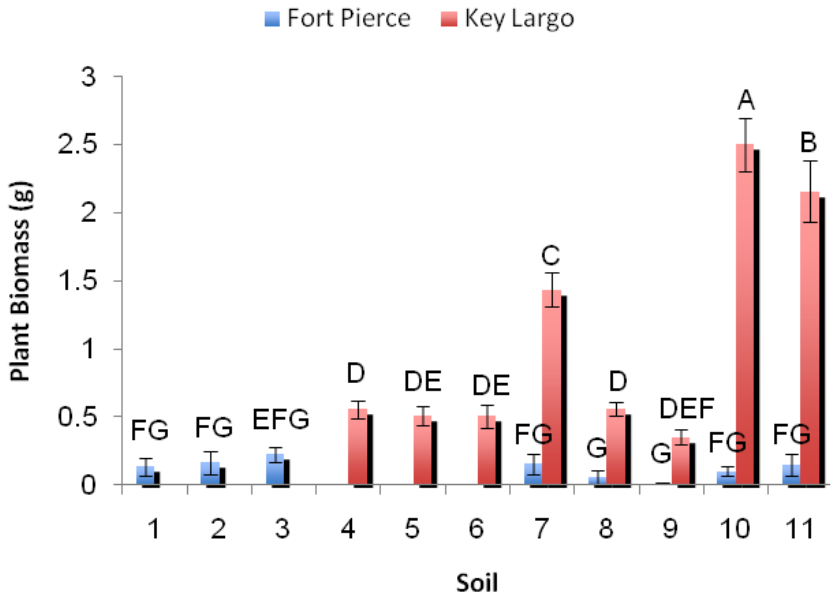


Figure 58. *H. wrightii* transplant plant biomass varied significantly by soil type and location.

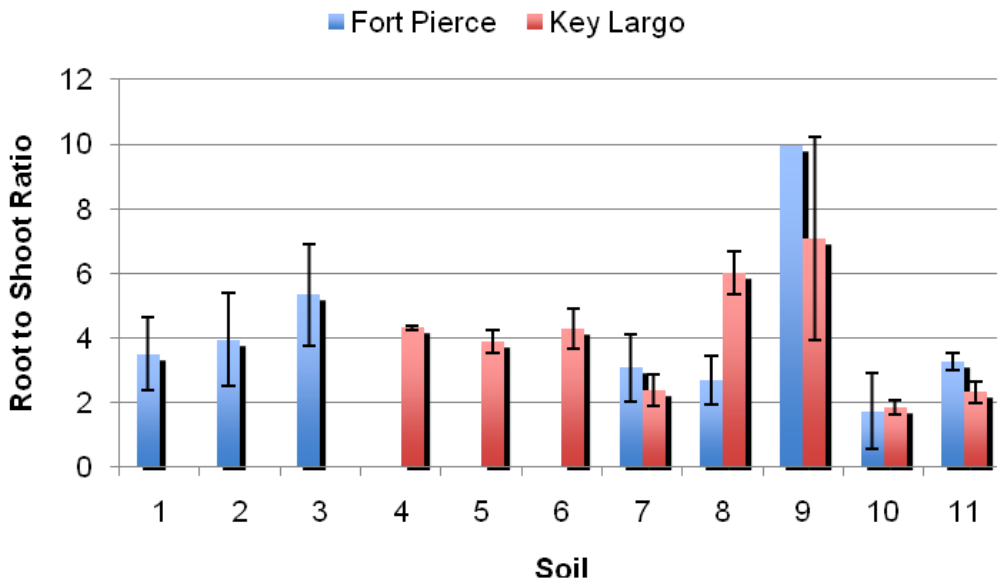


Figure 59. *H. wrightii* transplant root to shoot ratio varied significantly by soil type and location.

Table 5. Significance values of soil properties on transplant and natural seagrass characteristics.

		Soil (0-5 cm) Properties							
		Organic Matter (%)	Sand (%)	Clay (%)	Total carbon (%)	Total nitrogen (%)	Total phosphorus (mg/kg)	Total iron (mg/kg)	Sulfide (mM)
Seagrass Properties	Key Largo Transplants								
	Shoot Count	**** (-)	N/S	** (-)	*** (+)	N/S	**** (+)	**** (+)	N/S
	Canopy Height	N/S	N/S	N/S	N/S	N/S	*** (+)	*** (+)	*** (+)
	Leaves per Shoot	*** (+)	N/S	N/S	N/S	N/S	N/S	N/S	N/S
	Aboveground Biomass	*** (-)	** (-)	** (-)	N/S	N/S	**** (+)	**** (+)	N/S
	Rhizome Length	**** (-)	* (-)	N/S	N/S	* (+)	**** (+)	**** (+)	N/S
	Belowground Biomass	**** (-)	** (-)	** (-)	N/S	N/S	**** (+)	**** (+)	N/S
	Plant Biomass	**** (-)	** (-)	** (-)	N/S	N/S	**** (+)	**** (+)	N/S
	Root to Shoot Ratio	**** (+)	N/S	N/S	** (-)	N/S	**** (-)	** (-)	N/S
	Fort Pierce Transplants								
	Shoot Count	* (-)	N/S	N/S	N/S	N/S	N/S	N/S	N/S

table cont.

Canopy Height	N/S	N/S	N/S	N/S	N/S	* (+)	N/S	N/S
Leaves per Shoot	N/S	N/S	N/S	N/S	N/S	N/S	N/S	N/S
Aboveground Biomass	* (-)	N/S	N/S	N/S	N/S	N/S	N/S	N/S
Rhizome Length	N/S	N/S	N/S	N/S	N/S	** (+)	N/S	* (-)
Belowground Biomass	N/S	N/S	N/S	N/S	N/S	** (+)	N/S	* (-)
Plant Biomass	N/S	N/S	N/S	N/S	N/S	** (+)	N/S	* (-)
Root to Shoot Ratio	N/S	N/S	N/S	N/S	N/S	N/S	* (+)	N/S

Key Largo Natural Beds	Organic Matter (%)	Sand (%)	Clay (%)	Total carbon (%)	Total nitrogen (%)	Total phosphorus (mg/kg)	Bulk Density (g/cm3)	Depth (cm)
H. wrightii Shoot Density	* (-)	N/S	N/S	N/S	N/S	N/S	N/S	N/S
H. wrightii Percent Cover	* (-)	N/S	N/S	N/S	N/S	N/S	N/S	N/S

* p<.05, ** p<.01, *** p<.001, **** p<.0001, (+) positive effect, (-) negative effect

At the Fort Pierce site, the drift algae *Gracilaria sp*. was discovered resting atop all experimental units during the quantification of shoot counts four months after transplanting. After removal of the drift algae and another month's growth, the soil effect on *H. wrightii* shoot counts among soil types at the Fort Pierce was found to be significant at the conclusion of the study ($p=0.0434$). The only soil parameter found to significantly influence transplant shoot counts was soil organic matter ($p=0.0021$); increased concentrations were found to diminish transplant shoot counts. Soil organic matter also significantly diminished transplant aboveground biomass ($p=0.0442$). Soil TP concentrations were significantly correlated to increased transplant canopy height ($p=0.0119$), rhizome length ($p=0.0031$), belowground biomass ($p=0.0032$), and plant biomass ($p=0.0029$). Soil TFe was significantly related to increased transplant root to shoot ratio ($p=0.0290$), while porewater sulfide concentrations were significantly associated with diminished horizontal rhizome length ($p=0.0405$), belowground biomass ($p=0.0401$), and plant biomass ($p=0.0369$).

The soil effect was found to be highly significant ($p<0.0001$) among soil types present at the Key Largo location on *H. wrightii* shoot counts. Soil TP concentrations were highly significant ($p<0.0001$) for most transplant variables measured from Key Largo, including transplant shoot count, aboveground biomass, rhizome length, plant biomass, belowground biomass and transplant root to shoot ratio, and was also significant on transplant canopy height ($p=0.0002$). Soil TFe concentrations were also found to be highly significant ($p<0.0001$) on transplant shoot count, aboveground biomass, rhizome length, belowground biomass, and plant biomass. Additionally, soil TFe had a significant effect on transplant canopy height ($p=0.0001$) and transplant root to shoot ratio ($p=0.0011$). Soil organic matter content was highly significant ($p < 0.0001$) for transplant shoot count, rhizome length, belowground biomass, plant biomass, and root to shoot ratio, and was also significant on the number of leaves per shoot ($p=0.0009$) and aboveground biomass ($p=0.0002$). Soil TC had a significant effect on transplant shoot count ($p=0.0005$) and root to shoot ratio ($p=0.0035$) while soil TN had a significant effect on transplant rhizome length ($p=0.0166$). Porewater sulfide concentrations had a significant effect on transplant canopy height ($p=0.0008$). Soil clay content significantly influenced transplant shoot count ($p=0.0081$), aboveground biomass ($p=0.0079$), belowground biomass (0.0032), and plant biomass ($p=0.0017$), while soil sand content significantly influenced transplant aboveground biomass ($p=0.0013$), rhizome length ($p=0.0140$), belowground biomass ($p=0.0050$), and plant biomass ($p=0.0010$).

Transplant aboveground and belowground tissues were analyzed for TFe and TN. It was found that soil iron concentration had a significant effect ($p=0.0305$) on transplant rhizome tissue Fe concentrations at Key Largo (Table 3). Soil total nitrogen concentration had no effect on transplant Total nitrogen tissue nutrient content.

NATURAL SEAGRASS MONITORING AND SUBAQUEOUS SOIL SAMPLES

The seagrass sites near Key Largo were dominated by *H. wrightii* and *T. testudinum*. Shoot densities of *H. wrightii* ranged from 0-780 shoots/m^2. Surface soil textures ranged broadly from sand to clay across the 1.8 km^2 sampling site. Surface soil organic matter content also showed a large range across sites; approximately 25% of sites contained 0-10% organic matter, 45% of sites contained 10-20% organic matter, and 30% of sites ranged from 20-60% organic matter. *H. wrightii* percent cover and shoot

density were found to be negatively correlated ($p<0.0349$ and $p<0.0292$, respectively) to surface soil organic matter content. No other soil variables were found to be significantly related to *H. wrightii* cover or density.

DISCUSSION

Our hypothesis that soil properties would have a significant effect on *H. wrightii* growth provided other environmental parameters were sufficient was confirmed. In Key Largo, environmental conditions outside of soils, including light availability ($Kd < 1$, personal observation) and water currents (no observed currents and minimal wave action), provided ideal transplant growing conditions. This resulted in a highly significant soil effect at this site, and many soil properties were found to influence transplant growth. In sharp contrast, environmental conditions outside of soils were not optimal at our Fort Pierce transplant site. Here, our transplants were smothered by drift algae (*Gracilaria sp.*). The occurrence of the drift algae was associated with depressed transplant growth observed during the fourth month of our study. Such smothering by algae has also been observed to eliminate *Zostera marina* transplants off the Massachusetts coast (Short et al., 2002). However, because transplants were not completely destroyed by smothering in our study, soil properties at the Fort Pierce were still found to have an influence on *H. wrightii* growth, albeit diminished relative to the Key Largo site.

Soil nutrients including Total phosphorus and Total iron were found to significantly promote nearly all transplant growth responses at Key Largo. Additionally, transplant root to shoot ratios were significantly lowered by these soil properties, providing evidence that elevated levels of these nutrients led to additional growth in aboveground tissues relative to belowground tissues. Seagrass growing in environments characterized by carbonaceous soils, such as those in Key Largo, are often found to be either Phosphorus or Iron limited. These soils tend to form inorganic complexes with Phosphorus and Iron, diminishing the plant-available pools from the soil porewater (Duarte et al., 1995, Short, 1987). Additionally, ferrous Iron is able to bond with and remove sulfide, a seagrass phytotoxin, from soil porewater, seagrass growing in carbonaceous soils also have an increased risk to experience sulfide toxicity. For example, Calleja et al., (2007) observed increased sulfide toxicity in *Posidonia oceanica* meadows rooted in iron-deficient carbonaceous soils in the Mediterranean Sea. Our results from Key Largo provide additional support that these elements are limiting seagrass growth in the carbonaceous soils of the Florida Keys. However, sulfide levels were not observed to reach concentrations above 0.9 mM, which is below what are considered to be toxic levels (Koch, 2001); hence, the elevated transplant growth observed with elevated soil Total phosphorus and Total iron concentrations appear to document a plant nutrient limitation rather than a limitation associated with the accumulation of phytotoxins.

The Fort Pierce transplant site was characterized by siliceous subaqueous soils; generally, seagrass are nitrogen limited growing in these systems. Interestingly, soil Total phosphorus was found to significantly increase transplant canopy height, horizontal rhizome length, belowground biomass, and plant biomass, while soil Total nitrogen had no effect on transplants at the Fort Pierce site. Phosphorus limitation in siliceous substrates has been observed previously; for example, Cambridge and Kendrick (2009)

observed enhanced growth in *Posidonia australis* meadows after phosphorus fertilizer additions in Oyster Harbor, Southwestern Australia.

Studies analyzing the influence of soil texture on seagrass growth have found mixed results. Koch (2001) analyzed over 20 studies relating seagrass growth and particle size, with limitations generally occurring with soil clay and silt contents above 20% (Koch, 2001). This is hypothesized to be due to the limited porewater exchange between soil pores and the overlying water column in finer textured soils, which may result in phytotoxin accumulation or nutrient limitation. However, previous studies including Halun et al., (2002), Park and Lee (2007), Kenworthy and Fonseca (1977), and Short (1987) observed enhanced seagrass transplant growth in soils dominated by silty or "muddy" sediments; several authors suggested elevated nutrient levels associated with finer textured soils may have promoted seagrass growth. Our results from Key Largo document that *H. wrightii* transplant growth was negatively influenced by both elevated clay and sand contents suggested that soils dominated by silt-sized fractions may provide an optimal environment for seagrass growth.

Our analysis of empirically and experimentally derived soil limitations from Lake Surprise, Key Largo, Florida, documented that soil organic matter was the only soil property noted by both approaches to influence *H. wrightii*. Prior studies have observed seagrass typically grow in soils that have low soil organic matter contents. In a review 15 published studies of freshwater and marine submerged aquatic vegetation, the majority of observations found these plants are associated with soils that have less than 5% organic matter contents, although contents as high as 16% were noted (Koch, 2001). *H. wrightii* was not included in this review, however, the top performing treatments in our study had organic matter contents of 5.4% (Soil 10) and 11.6% (TCSL), while the poorest performing treatments had either extremely low (0.0% in TSS) or extremely high (53.4% in SMP) organic matter contents. This suggested that while an optimal soil organic matter content for *H. wrightii* transplanting may lie above what is considered optimal for many other seagrass species, this species is also limited by elevated soil organic matter contents. Prior work has hypothesized that mechanisms underlying organic matter limitation include phototoxic accumulation of sulfides or low soil nutrient contents (Barko and Smart, 1986). However, our results suggested that sulfide levels did not reach concentrations typical of those limiting seagrass growth in the published literature (Koch, 2001), nor were soils elevated in organic matter content low in nutrients such as Total phosphorus or Total nitrogen relative to other treatments. It has alternatively been suggested that low soil bulk densities typical of soils enriched in organic matter result in increased distance for nutrients to diffuse before they can be absorbed by seagrass roots (Barko and Smart, 1986). This mechanism may have resulted in a nutrient limitation leading to diminished transplant growth in our elevated organic matter treatment.

It is interesting to note that soil Total phosphorus had a highly significant effect on the growth of *H. wrightii* transplants, yet Total phosphorus was not significantly correlated with the coverage or density of *H. wrightii* based on our empirical observations. This may be due to the limited range in soil Total phosphorus inherent to the Lake Surprise study area. Our analysis revealed Total phosphorus averaged 168 +/- 5 mg/kg across the surface soils sampled during our empirical analysis.

To conclude, our analysis suggested that soil properties have the potential to limit the growth of seagrass during transplanting events. It is important to note that the influence of particular soil properties are site specific and that environmental conditions outside of soils, including light availability, hydrodynamics, and biotic interactions, can preclude the influence of soils on seagrass; thus, these findings lead us to caution the development of subaqueous soil interpretations regarding the suitability of a soil for seagrass transplanting without additional consideration of other environmental conditions. Additionally, empirical observations of soil-seagrass growth limitations failed to reveal several soil limitations determined experimentally. Therefore, it may be more suitable to determine optimal soil properties for transplanting seagrass using an experimental approach rather than rely on information obtained from mature meadows. These findings provide evidence that analysis of subaqueous soils before transplant efforts are initiated can reveal locations where transplanting efforts will be most effective and improve transplant performance.

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CHAPTER 6: ASSESSMENT OF SL-15 RESTORATION TRAJECTORIES

SUMMARY

Coastal ecosystems are significant natural carbon sinks. If constructed coastal ecosystems can obtain the same carbon sink capacity as their natural counterparts, then construction and restoration of these systems has the potential to become a tool for reducing atmospheric CO2. In this study, sediment organic carbon (OC) of a recently constructed mangrove and seagrass system in the Indian River Lagoon, Florida was compared with sediment OC of nearby mature, reference systems. Total OC, extractable OC, and microbial biomass C pools were measured to compare C storage. Organic C lability in the constructed and reference sites was also measured. The main sediment OC sources were determined using 13C isotopes and C:N ratios and were compared among systems. Organic C pools were generally larger in sediments of reference systems than in sediments of the constructed systems, but differences in pool sizes were much greater between the constructed and reference mangrove systems. Organic C lability was greater in the constructed systems indicating their sediments could not store OC for as long as the references. Seston was a major source of sediment OC in all systems. Other main sources of OC were higher plant-derived in constructed and reference mangrove and reference seagrass sediments, but were algal-derived in constructed seagrass sediments. After one year, the C sink capacity of the constructed systems is less than the capacity of the reference systems, but the constructed seagrass system is functioning more like its reference than the constructed mangrove system. In the long term, however, the potential C sink capacity of the constructed mangrove system is greater.

INTRODUCTION

Restoration and construction of coastal ecosystems may help mitigate the effects of climate change by reducing atmospheric carbon dioxide (CO2). Global climate change has become a major environmental concern over the past 50 years. The anthropogenic release of greenhouse gases is the major cause of global climate change (IPCC 2001). Atmospheric concentrations of CO2 and methane (CH4), the biggest contributors to climate change, are increased by fossil fuel burning and deforestation, and livestock production, respectively. Highly productive coastal ecosystems including salt marshes, seagrass beds, and mangrove forests are carbon (C) sinks. Salt marshes and mangroves store at least 44.6 Pg C in their sediments (Chmura et al., 2003). Seagrass beds, which make up only 0.15% of global marine area, account for 15% of global marine organic C (OC) storage (Hemminga and Duarte 2000). Sediment C storage values are even larger when stores of inorganic C like carbonates are taken into account (Zhu et al., 2002).

The C cycle in coastal ecosystems is an open cycle because OC is imported into and exported out of systems by water currents and tides. In a seagrass bed, seagrass and macroalgae (drift and epiphytic) take up CO_2 and HCO_3^- from the water column to produce OC through photosynthesis (Figure 60). When

seagrass fronds senesce or break, they (and their associated macroalgae) become litterfall on top of sediments or are exported out of the system. Litterfall OC is either decomposed by microbes or incorporated into the sediment OC (SOC) pool by leaching, bioturbation, or burial. Imported OC, which may include terrestrial and mangrove detritus, is trapped by seagrass fronds and settles on the sediment. Seston, composed of plankton, bacteria, and dissolved and particulate OC from in and outside the system, is also trapped by seagrass fronds. The fate of trapped OC is the same as litterfall. Seagrass root OC from exudates or dead tissue is immediately part of the SOC pool and can be used by microbes. SOC can be part of three pools—microbial biomass C, labile OC, or recalcitrant OC. Generally, microbes consume mainly labile OC, which they respire as CO_2 or incorporate into their biomass. When microbes die, their OC becomes part of labile and recalcitrant pools. The more OC is reworked by microbes, the more recalcitrant it becomes. The recalcitrant pool is where OC is sequestered long-term and where OC undergoes abiotic condensation into complex humic materials. In mangrove forests, the C cycle is basically the same except for the C sources (Figure 61). The inorganic C source used by mangroves is atmospheric CO_2, the litter is mangrove leaves, and imported OC trapped in litter by mangrove roots is seston and seagrass detritus.

The capacity of coastal ecosystems to sequester OC is greater than the capacity of terrestrial ecosystems. Coastal ecosystems are natural C sinks, while terrestrial systems reach an equilibrium where the net C fixed annually is about zero (Rabenhorst 1995). Constant accumulation of C in coastal ecosystems is due to their anoxic sediments. In these sediments, oxygen is depleted so electron acceptors that are not as efficient must be utilized by microbes to decompose OC. Coastal ecosystems also have a greater OC sequestration capacity than freshwater wetlands because they, unlike freshwater wetlands, do not use CO_2 as a terminal electron acceptor and therefore emit less CH_4 (Bridgham et al., 2006). In coastal ecosystems, sulfate is the terminal electron acceptor, and high sulfate levels inhibit methanogenesis (Capone and Kiene 1988). A study of mangrove forests did not detect CH_4 either dissolved in sediment porewater or fluxing out of sediments and 51 to 75% of OM oxidation was occurring through sulfate reduction (Alongi et al., 2004). Coastal ecosystems, like salt marshes, mangrove forests, and seagrass beds, may therefore be highly significant C sinks because they accumulate C in sediments without emitting CH_4.

Many salt marsh, mangrove, and seagrass ecosystems have been degraded or lost through disturbances such as dredging channels and developing coastlines for human habitation (Valiela et al., 2001; Kennish 2002; Zedler 2004). This degradation and loss affects the biogeochemical functioning of coastal systems including C sequestration. Loss and degradation of coastal systems therefore affects the global C cycle and may increase the effects of climate change (Duarte et al., 2005; Bridgham et al., 2006). Globally, 50% of wetlands (freshwater and coastal) have been lost (Moser et al., 1996). The continuous United States has lost 53% of its wetlands since the 1780's (Dahl, 1990). Since 1989, the United States has had a policy of no net wetland loss that includes coastal wetlands (Zedler 2004). In Florida, state policy applies this principle to seagrass systems as well. Therefore when mangrove and seagrass ecosystems are destroyed, their loss must be mitigated by restoring or creating these systems elsewhere. It is important to know whether mitigation of coastal ecosystems restores the accumulation and storage capacity of these important C sinks. Such research can indicate whether mitigation is truly effective and

whether coastal ecosystem restoration can become a policy tool for reducing CO_2 emissions, as was suggested by Connor et al., (2001). Functional trajectory studies of constructed systems and studies comparing constructed systems with natural systems are used to determine the effectiveness of mitigation in restoring ecosystem functions, like C storage.

Functional trajectories are used to monitor the development of ecological functions in constructed ecosystems over time. When constructed systems' functions equal those of reference systems, the constructed systems are said to be functionally equivalent. Studies that documented functional trajectories of OC in restored and constructed salt marshes concluded that it takes a long time for the restored/constructed marshes to develop SOC pools equal to their natural counterparts (Simenstad and Thom, 1996; Craft, 2001; Havens et al., 2002; Morgan and Short, 2002; Craft et al., 2003). Functional trajectory studies, with one exception (Evans and Short 2005), have been limited to temperate brackish and salt water marshes. There is a need to study functional trajectories in constructed mangrove forests and seagrass beds. Functional trajectory studies generally measure a suite of ecological functions, so SOC is usually the only variable measured that pertains to ecosystem C storage. Studies that examine multiple OC pools, OC lability, and OC sources are needed to more fully understand the recovery of C storage functioning in constructed systems. Studies that examine short term changes immediately following construction are also lacking. Short term trajectory studies are important because certain aspects of OC storage may recover quickly.

Whether constructed mangrove and seagrass ecosystems provide the same ecological services as their natural counterparts with respect to C storage, and whether restoration of these services follow a functional trajectory is currently unknown. In this thesis, the trajectories of SOC pools in constructed seagrass and mangrove systems were monitored during the first year after construction completion. SOC pools in the constructed systems were also compared with SOC pools in adjacent natural systems. Sediments were the focus of this research because they are the sites of long term C storage. Variables measured include the amount of OC in three pools (total OC, extractable OC, and microbial biomass C), the lability of SOC, and the C to nitrogen ratios and $\delta^{13}C$ of sediments and potential SOC sources. The constructed site was a former spoil island called SL-15 in the Indian River Lagoon, FL that was converted to a mangrove and seagrass ecosystem in November 2005. The reference sites were the natural seagrass beds that surround SL-15 and the nearby mangrove forests that occupy the edges of adjacent spoil islands.

The main research objectives were: 1) to determine short term trajectories of SOC pools in a constructed mangrove forest and seagrass bed; 2) to compare SOC pools in the constructed system with those in more natural, reference systems; 3) to compare the lability of SOC in the constructed and reference systems; 4) to determine and compare significant sources to the total SOC pool in the constructed and reference systems.

The hypotheses were: 1) in the short term, storage in the three OC pools studied would increase in the constructed systems, but would not reach the level of storage in the references' OC pools; 2) OC lability would be greater in sediments of constructed systems than in reference sediments; 3) SOC sources in

constructed systems would be macroalgae or plankton, while SOC sources in reference systems would be vascular plants, like mangroves and seagrass.

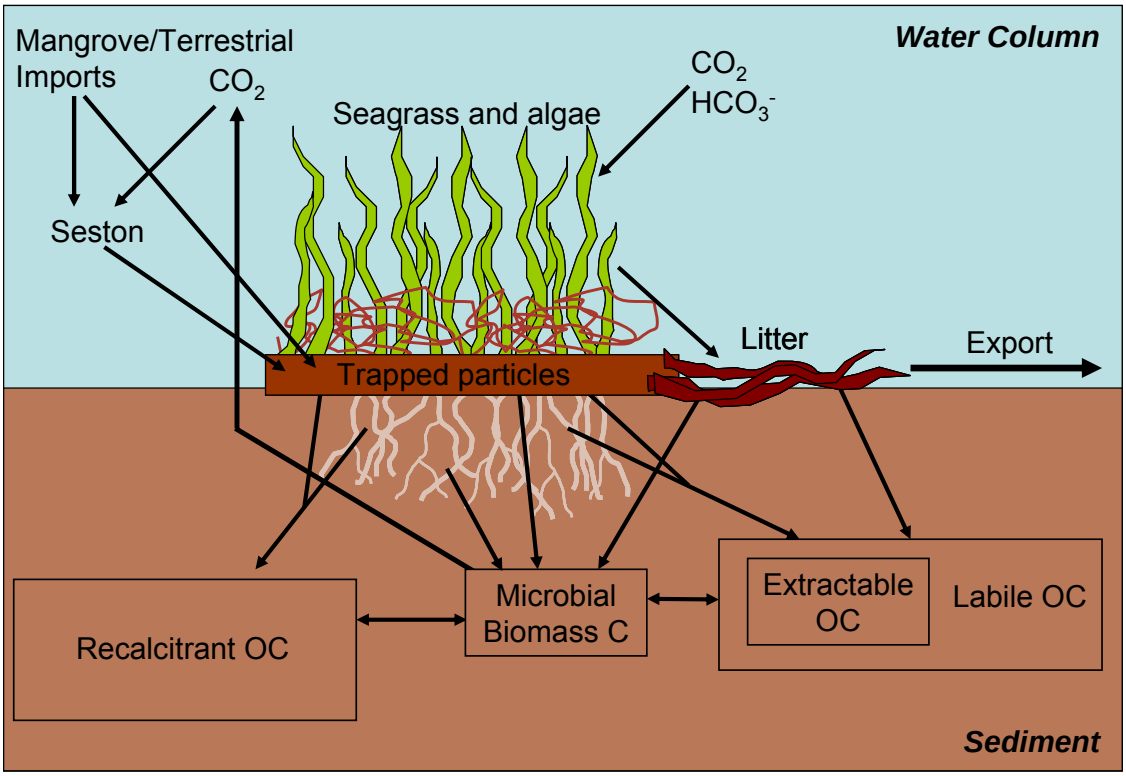


Figure 60. The carbon cycle in seagrass beds.

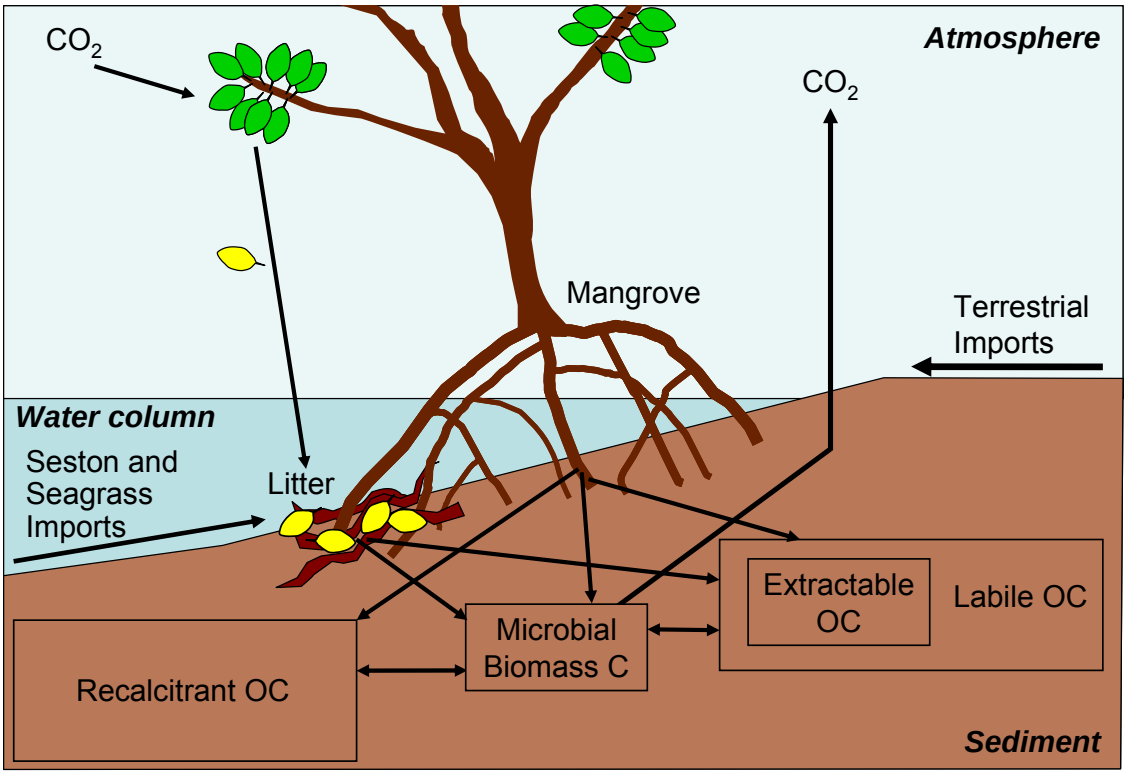


Figure 61. The carbon cycle in mangrove forests.

BACKGROUND INFORMATION

In this literature review, rates of organic carbon (OC) accumulation are compiled and compared for three coastal ecosystems—salt marshes, mangrove forests, and seagrass beds. Studies comparing sediment organic carbon (SOC) pools in restored or constructed salt marshes to SOC pools in natural salt marshes are then examined. This section does not discuss mangrove forests or seagrass beds because the literature on the functioning of restored or constructed coastal ecosystems is currently limited to salt marshes. Third, methods for determination of SOC sources are discussed for the three coastal ecosystems. These coastal ecosystems are dominated by vascular, halophytic macrophytes, with mangroves dominated by trees and salt marshes and seagrass beds dominated by grasses and other herbaceous species. Sediments in these systems are C sinks due to their high net primary production, trapping of material from the water column, and O_2 limited conditions.

These systems are globally distributed. Salt marshes and mangroves occupy non-rocky, sedimentary-driven intertidal zones of the world. Salt marshes predominate in temperate climates, while mangroves predominate in subtropical and tropical climates. Salt marshes are generally replaced by mangroves at a latitude of 25° N or S (Mitsch and Gosselink 2000). Seagrass systems are subtidal and are found from tropical through temperate climates where needs such as low light attenuation in the water column are met (Hemminga and Duarte 2000). Seagrass are often found adjacent to their intertidal counterparts—salt marshes or mangroves. Multiple estimates of global area covered by each system differ, but in each system the estimates are within the same order of magnitude. According to the average of the estimates, mangrove forests cover 220,000 km^2, salt marshes cover 350,000 km^2, and seagrass beds cover 450,000 km^2 (Table 2-1). Together these systems occupy only 0.8% of the global ocean area, but they contribute 30% of the total ocean C storage (Duarte and Cebrián 1996). This observation indicates that these systems play a significant role in global sequestration of C.

Coastal ecosystems are better C sinks than terrestrial systems and freshwater wetlands. Coastal ecosystems and freshwater wetlands can accumulate C indefinitely while terrestrial systems reach an equilibrium where C fixed equals the amount respired annually (Rabenhorst 2005). Coastal ecosystems and freshwater wetlands' abilities to continually accumulate C are due to their anoxic sediments where electron acceptors other than O_2 must be utilized to decompose OC. These electron acceptors yield less energy to microbes than O_2, slowing decomposition rates (Schlesinger 1997).

Coastal ecosystems are better C sinks than freshwater wetlands, because they release orders of magnitude less CH_4, a potent greenhouse gas (Bridgham et al., 2006). CH_4 has a higher radiative forcing capacity than CO_2, so its global warming potential (GWP), a measure of its radiative forcing capacity per one unit mass relative to the radiative forcing capacity of one unit mass of CO_2, is by definition greater than the GWP of CO_2 (IPCC 2001). CH_4 release occurs because methanogenesis, the process where CO_2 is reduced to CH_4 in order to breakdown organic matter (OM), is the dominant decomposition pathway in most freshwater systems. In coastal ecosystems high sulfate levels inhibit methanogenesis as sulfate is a more energetically efficient electron acceptor than CO_2 (Capone and Kiene 1988).

Rates of Organic Carbon Sequestration

Before any discussion on rates of C accumulation, terminology and resulting caveats must be addressed. Not all studies use the same terminology when reporting rates of C accumulation and often studies do not specifically define their rate terminology. Terms used in the literature that all essentially meant "rate at which OC builds up in soil" were "rate of OC accumulation," "rate of OC sequestration," "rate of POC burial," "rate of refractory accumulation," and "organic accumulation rate." Nuances of these terms could be gleaned from methodology. Some like "rate of C accumulation" referred to additions of both labile and refractory OC to the SOC pool (Craft et al., 2003). Others like "rate of refractory accumulation" referred to long-term burial of OC that is unlikely to decompose on a human time scale (Cebrián 2002), and others like "POC burial" were vague (Alongi et al., 2005). Some studies reported OM accumulation, not OC accumulation. Those rates were divided by two to obtain OC accumulation rates. Also it was assumed that "C accumulation rates" referred to accumulation rates of OC, not total C, because studies that used the term reported measuring OC. Lastly, for indirectly measured rates (modeled or based on mass balance equations) it was not always clear whether rates included amounts of OC from both autochthonous and allochthonous sources. This review reports all values as "C accumulation rates," which refers to the buildup of OC in sediments though there may be discrepancies in the lability of accumulating OC. Generally, the longer the timescale of a study, the more likely rates represent long-term burial. Only the term "C burial rates" definitively refers to long-term storage of refractory OC. There is a need for future studies to clearly define rate terminology and to be consistent in its use.

Global Rates

Many scientists have estimated global rates of C accumulation for coastal ecosystems due to their important role in the global C cycle (Table 2-2). Global rates of C accumulation for these systems are calculated in several ways. The most common way was averaging published accumulation rates for many sites (Duarte and Cebrián 1996; Chmura et al., 2003). Other methods included graphing frequency distributions of published accumulation rates (Cebrián 2002), scaling up from model-derived rates (Suzuki et al., 2003), or using mass balance equations derived from production and burial estimates (Jennerjahn and Ittekkot 2002). Despite the different ways of calculating global rates, accumulation rates for intertidal systems are basically in agreement (Table 2-2). Estimated global accumulation rates for mangrove forests range from 92 to 200 g C m^{-2} yr^{-1} and rates for salt marshes range from 50 to 175 g C m^{-2} yr^{-1} (ignoring the high estimate of Rabenhorst (1995). Rates for seagrass beds are more variable and range from 16.5 to 270 g C m^{-2} yr^{-1}. Higher variability for seagrass ecosystems is likely because C accumulation rates in seagrass sediments have been studied less than in mangroves and salt marshes. Suzuki et al., (2003) estimated that seagrass caused an accumulation of 1.2 g C m^{-2} yr^{-1} in deep ocean sediments due to export of their primary production to the open ocean and its subsequent burial. While this review concentrates on *in situ* accumulation, it is important to recognize that there are other ways these systems contribute to the global C sink.

Coastal ecosystems accumulate C at a rates several orders of magnitude greater than rates in terrestrial systems and the open ocean (Table 2). C cycling in terrestrial systems should reach a steady-state

condition, making them neither a C source nor C sink (Hussein et al., 2004). Disturbances such as fire, however, occur before climax stages causing terrestrial systems to become C sources to the atmosphere. Coastal ecosystem C accumulation rates are greater than open ocean rates because their primary producers differ. Open ocean phytoplankton have a much lower net primary production (NPP) per unit area, have a greater percentage of their NPP consumed by herbivores, and contain more easily decomposed OM than coastal macrophytes (Duarte and Cebrián 1996; Cebrián 2002).

Local Rates

Mean global rates of C accumulation (Table 2) were calculated using rates of C accumulation from numerous local studies (Table 3). The majority of C accumulation rates were measured in salt marshes while accumulation rates in seagrass beds were the least measured. Accumulation rates in mangrove forests ranged from 33 to 841 g C m^{-2} yr^{-1} with the lowest rate measured in Terminos Lagoon, Mexico (Gonneea et al., 2004) and the highest rate measured at the low marsh in Jiulonglijang Estuary in China (Alongi et al., 2005). Rates in salt marshes ranged from 2 to 300 g C m^{-2} yr^{-1} with the lowest rate measured at a natural site in Dell's Creek, North Carolina (Craft et al., 2003) and the highest rate measured behind a continuous canal in Lafourche Parish, Louisiana (Cahoon and Turner 1989). Rates in seagrass beds ranged from 19 to 191 g C m^{-2} yr^{-1}, a range measured offshore of Cala Culip, Spain (Romero et al., 1994).

Comparing the compiled rates for these systems, there were no trends of one system having consistently higher C accumulation rates than the other systems (Table 2-3). The system accumulating C at the highest rates even varied within the same region. For example, in Celestun Lagoon, Mexico, mangroves accumulated more carbon in their sediments than seagrass, but in Terminos Lagoon, Mexico, the reverse was true (Gonneea et al., 2004). This lack of a trend is supported by Chmura's (2003) review that found no significant differences between C accumulation rates in salt marshes and mangroves. It should be noted, however, that contributions of mangrove forests to C storage on an ecosystem scale may be greater than salt marshes and seagrass beds because large amounts of C are stored for decades in woody biomass of mangrove trees (Twilley et al., 1992).

Measuring Rates of Carbon Accumulation

Calculating rates of C accumulation typically involves three steps. The first step is to measure SOC pools. SOC pools for local rate studies were directly measured using either an elemental analyzer after acidification of the sample to get rid of carbonates (Gacia et al., 2002), a TOC analyzer (Brunskill et al., 2002; Alongi et al., 2004), or a mass spectrometer (Choi and Wang 2004). SOC pools were indirectly measured by using loss-on-ignition (LOI) values in regression equations describing the relationship between SOM and SOC (Connor et al., 2001). The second step is to age the sediment or measure rates of sediment accretion. Sediment age and accretion rates are less straightforward measurements than SOC pool measurements. Radioisotope dating of cores using either ^{210}Pb and ^{14}C activity or $\delta^{137}Cs$ and ^{14}C peaks from nuclear bomb fallout were the most commonly used methods to date sediments (e.g. Callaway et al., 1997; Connor et al., 2001; Choi and Wang 2004; Hussein et al., 2004; Alongi et al., 2005). Other methods of dating sediments were Romero's (1994) use of a shipwreck whose date was known

and that had been buried by seagrass over hundreds of years and Chmura et al.,'s (2001) use of pollen stratigraphy. Short term (1-3 years) accretion rates were measured with feldspar markers (Cahoon and Turner 1989; Cahoon 1994; Cahoon and Lynch 1997) or sediment traps (Gacia et al., 2002). The third step is to calculate C accumulation rates. The amount of OC in a unit of sediment is divided by the age of that sediment unit, or OC in a unit of sediment is multiplied by the rate at which that sediment unit accreted. Sediment ages in restored or constructed systems do not have to be determined because the site ages are known. C accumulation rates can be calculated by the difference between SOC content at the beginning of the restoration process and SOC content at subsequent points after the initial restoration, divided by site's age (Cammen 1975; Craft et al., 2003).

Calculated rates of C accumulation may be dependent on time scale, which is dependent on the method used. With a half-life of 5730 years, ^{14}C methods are suitable for measuring rates over many millennia, while with a half-life of 22.3 years, ^{210}Pb methods are suitable for measuring rates over a century (Bierman et al., 1998). Bomb fallout methods using $\delta^{137}Cs$ and ^{14}C peaks can only measure rates over the last 40 years as the peaks generally occur in 1963. Many of the highest rates of C accumulation were measured using the feldspar marker technique, which measures C accumulation rates over a year or two. These rates may be high because surface pools of SOC are relatively labile compared to deeper pools of SOC. Much of the surface SOC may be mineralized by the time it is buried deeper in the soil profile, where it would be measured if longer term methods were used. Long term rates calculated by ^{14}C dating were slower than rates measured over a decadal (Choi and Wang 2004) or an 100 year time scale (Hussein et al., 2004). Choi and Wang (2004) did not attribute this difference to methodology and speculated that greater C accumulation rates are due to increases in primary production over the last 100 years caused by increased CO_2 and nutrients.

Comparing Organic Carbon in Restored and Reference Coastal Marshes

Highly productive habitats like coastal ecosystems are C sinks as their high C accumulation rates demonstrate. In these coastal ecosystems atmospheric CO_2 becomes stored as OC for long periods of time. Restoration and construction of coastal ecosystems may therefore help mitigate the effects of climate change by reducing atmospheric CO_2 (Connor et al., 2001). If limits are placed on CO_2 emissions in the United States, coastal ecosystem restoration and construction may then become a viable option for C offsetting. C offsetting occurs when an industry needs to reduce its net CO_2 emissions but can or will not reduce their own emissions, so they invest in projects that reduce emissions elsewhere, such as tree planting. Anthropogenic release of greenhouse gases is the major cause of global climate change (IPCC 2001). CO_2 and CH_4 are greenhouse gases with the biggest effect on climate change due to their concentrations in the atmosphere and radiative forcing capacity (IPCC 2001). Humans increase atmospheric concentrations of CO_2 through fossil fuel burning and land use change and concentrations of CH_4 through livestock production. This section of the review focuses on salt marshes due to the dearth of literature on functional trajectories in restored or constructed mangrove and seagrass systems.

Despite the numerous important ecological functions coastal ecosystems and wetlands provide, which extend well beyond their function as C sinks, many were viewed as wastelands until recently (Broome et

al., 1988). These systems were seen as wasted space that could be utilized for agriculture or valuable development. Wetlands, including salt marshes, were summarily destroyed without much thought to the consequences of their destruction through the 1980's. The lower 48 U.S. states lost 53% of its wetlands from the 1870's to the 1980's (Dahl, 1990). Globally, it is estimated that 50% of the wetlands have been lost (Moser 1996). When these systems are lost, we lose a sink for anthropogenically-derived CO_2. For example, Connor et al., (2001) estimated that if 85% of the coastal marshes in the Bay of Fundy had not been altered for agricultural uses, $3.8 \times 10\delta^{13}$ g C could have been stored over the past 160 years. The loss of coastal ecosystems and wetlands therefore disrupts the global C cycle and may increase the effects of climate change.

Since 1989, the U.S. has had a policy of no net wetland loss that includes coastal marshes (Zedler 2004). The policy calls for mitigation if alternatives to destroying wetlands in the course of development projects are unavailable. This mitigation comes in the form of creating new wetlands onsite or nearby to the lost wetland, restoring an existing, degraded wetland, or buying into wetland mitigation banks (Zedler 2004). Because of coastal marshes' importance as C sinks and the widespread replacement of natural marshes with created marshes, it is important to know whether restored and constructed marshes have OC accumulation rates and storage capacities equivalent to those of natural marshes. Such research can indicate whether marsh creation can become a policy tool for reducing CO_2 emissions. Connor et al., (2001) suggested that restoring coastal marshes may help countries reduce their CO_2 emissions to the standards set by the Kyoto protocol. Monitoring functional trajectories of constructed marshes helps researchers understand if constructed marshes' OC storage can equal the storage of natural marshes.

Monitoring Constructed Coastal Marshes Using Functional Trajectories

Functional trajectories are used to track the progress of constructed ecosystems and to compare constructed and natural ecosystems (Simenstad and Thom 1996; Zedler and Callaway 1999; Morgan and Short 2002). Functional trajectory studies often examine a whole suite of "ecological attributes" (Craft et al., 2003) that act as indicators for more complex ecological functions (Simenstad and Thom 1996). Attributes are measured in the same constructed system over time, or in several different-aged constructed systems in the same region using a space-for-time substitution, to obtain a range of attribute values that can be plotted against time (Kentula et al., 1992). In coastal marshes, OC parameters are often just several of many attributes measured. Data are then fitted to a curve and compared with values from natural marshes. The resulting trajectory represents how the attribute develops in a restored or constructed marsh over time (Morgan and Short 2002). There are two main questions that functional trajectories studies seek to answer: 1) how long does it take for the attribute in the restored or constructed marsh to reach functional equivalence (i.e. the mean value of that attribute in a natural marsh); 2) is the mean value of an attribute in a natural marsh the correct endpoint for the development of that attribute in the restored or constructed marsh?

Not all attributes have the same trajectory, and trajectories of the same attribute may differ across different marshes and depending on the natural reference marsh used. There are many different

trajectories that attributes like SOC pools can follow (Kentula et al., 1992; Figure 62). Some attributes may not even follow a trajectory and instead stay relatively constant through time (Zedler and Calloway, 1999). Craft et al., (2003) proposed that different attributes follow one of three trajectories depending on whether they are part of hydrologic, biological, or soil development processes. OC pool formation is part of soil development, which in most cases is the slowest trajectory to reach functional equivalence (Craft et al., 2003). If a trajectory fits OC data well, it can be used to predict future levels of OC thereby helping agencies set standards for mitigation project monitoring or determine the amount of C emission credits a created marsh is worth. In theory, functional trajectories are a simple way to evaluate the current success and predict the future success of constructed marshes; data, however, do not always fit a smooth line. Often, there is high variability between constructed marshes (Craft et al., 2003) and between years in the same study (Zedler and Calloway, 1999). The reference marshes used can also influence the predicted success or failure of a constructed marsh as a result of their age, variability (Simenstad and Thom 1996), or stress level. Furthermore, predictions from functional trajectories should be considered with caution because they do not take into account disturbances that may alter the trajectory.

Functional Trajectory Case Studies

The studies reviewed here examine the equivalence in constructed and restored marshes that are one (Morgan and Short 2002) to 42 years old (Craft, 2001). The first prediction of functional equivalence for SOC in a salt marsh was made by Seneca et al., (1976) for one of the first salt marsh creation projects using dredge spoil, which is essentially devoid of OC. They predicted it would take 4 to 25 years for the constructed marsh to store as much C as the natural marsh. More recent studies showed that it probably takes *at least* 25 years for OC to reach functional equivalence (Table 2-4). SOC and the related attribute, sediment organic matter (SOM), seem to be one of the last attributes to reach functional equivalence in marshes after aboveground biomass, sedimentation rates, and diversity of flora and fauna. There is also the possibility that OC will never reach functional equivalence as most studies did not follow marshes for a sufficient duration of time to showed equivalence.

Most studies on the eastern coast of the United States found a trend of increasing SOC/SOM over time (Craft, 2001; Havens et al., 2002; Morgan and Short, 2002; Craft et al., 2003). A study of different-aged New England salt marshes found that SOM increased steadily from 2% at a 1-year-old site to 15% at a 15-year-old site (Morgan and Short 2002). Studies from the western coast did not find strong directional trends of SOM over time. In Tacoma, Washington SOM stayed between 2-4% over 5 years (Simenstad and Thom 1996) and in San Diego, California only a slight increase of 3% was found over 11 years (Zedler and Calloway, 1999). These differences in trajectories may be more a case of land use than geography. Both of the west coast studies took place in large urban areas, whereas the east coast studies took place in a variety of locales, none as developed as Tacoma and San Diego.

Only two studies documented tidal marshes that reached functional equivalence with their natural references in terms of SOC. The tidal marshes were 25 (Craft et al., 1999; Craft et al., 2003) and 42 years old (Craft, 2001). Both these marshes are located in the southeastern U. S. These marshes achieved functional equivalence possibly because they, or their references, differed from most of the marshes

studied. The 42-year-old marsh differed because it was a restored marsh and not a marsh constructed from dredge spoil. Instead, it had been disturbed by a dike that prevented tidal inundation but was removed after only 8 years (Craft 2001). The 25-year-old marsh differed because its reference marsh was a relatively new natural salt marsh, which contrasted to other reference marshes that are greater than 2,500 years old (Craft et al., 1999). Because the reference was relatively young, its soils resembled spoil (90% sand) more than a Histosol (>10% OM). They were mineral Entisols with a high bulk density and low OC content (<1.4%). Reference marshes can determine whether or not a constructed marsh reaches functional equivalence because their mean attribute values represent functional equivalence "finish lines."

Factors Affecting Functional Equivalence

The reference marsh used affects the functional equivalence of the constructed marsh. In the last example (Craft et al., 1999), if the SOC pool of the 25-year-old constructed marsh had been compared to the SOC pool of the 2,500-year-old natural marsh with a high OM content, the authors would have concluded that the constructed marsh had not yet reached functional equivalence. Many studies choose nearby natural marshes as references without regard to their similarities to constructed marshes. Studies in urban areas are particularly limited by reference sites as the restored site is often the only large area of marsh remaining (Simenstad and Thom 1996). Morgan and Short (2002) solved some of the problems associated with reference site choice when they chose reference sites after comparing constructed sites to potential reference sites using a principle components analysis (PCA) based on physical attributes like aspect, slope, and size. They used the PCA to choose two well-matched reference sites for each constructed site. Because reference marsh is a major factor in whether a constructed marsh reaches functional equivalence, it should not be chosen arbitrarily.

While between-system factors affect whether a constructed wetland reaches functional equivalence, so do within-system factors like elevation, depth in the soil profile, and variation in sedimentation rates. Even when a constructed marsh as a whole is far from reaching functional equivalence in terms of SOC, some parts of it may be closer than others. Several studies found higher SOC at low elevations in constructed marshes (Lindau and Hossner 1981; Craft et al., 2002). Lower elevations are inundated by tides for longer periods of time, which leads to more highly reducing conditions that can encourage OC storage. In most soils or sediments, OC naturally decreases with depth, which may hinder the ability of lower depths to reach functional equivalence. In one of the only studies to examine OC at different depths in the soil profile, upper depths reached functional equivalence quickly while OC values at lower depths did not increase over 7 years (Havens et al., 2002). Sedimentation of mineral particles dilutes SOC concentrations. Creek banks often have lower OC concentrations than the interior of marshes (Craft et al., 2002) because they experience greater sedimentation of mineral particles (e.g. Temmerman et al., 2003). Simenstad and Thom (1996) cited sedimentation as a reason why SOM in a restored marsh did not increase with time.

Storing Carbon versus Sinking Carbon

Even though most constructed marshes do not yet store the same amount of C as their natural counterparts, they may still be acting as C sinks. A few studies examined OC accumulation rates as well as SOC pools and found that OC accumulation rates in constructed marshes are as high as or higher than rates in constructed marshes (Cammen 1975; Craft et al., 2002; Craft 2001). The mean OC accumulation rate of 8 different-aged constructed wetlands in North Carolina was 42 g C m^{-2} yr^{-1} compared to 43 g C m^{-2} yr^{-1} in the reference wetlands, even though the OC pools (g C m^{-2}) in the constructed wetlands were significantly lower (Craft et al., 2003). Additionally, some young marshes have high sedimentation rates (Morgan and Short 2002). Sedimentation may encourage OC accumulation while reducing SOC concentrations resulting in a reciprocal relationship as was demonstrated in Bay of Fundy marshes (Connor et al., 2001). High sedimentation rates may have prevented SOC pools from increasing in the Tacoma and San Diego constructed marshes while encouraging OC accumulation, which unfortunately was not measured in those studies.

New Directions

The extensive studies on coastal marsh functional trajectories have been broad in scope and therefore unable to examine OC dynamics in constructed marshes with sufficient detail. SOC is a conglomeration of pools that include a labile pool, a slowly oxidized pool, a very slowly oxidized pool, and a recalcitrant pool. The pool matters, as the one containing the most OC affects the overall sequestration abilities of a wetland. A wetland with most of its OC in the recalcitrant pool is going to sequester C longer than a wetland with most of its OC in a labile pool like microbial biomass, which has frequent turnover. A study of macro organic matter (MOM), precursor of SOM, in constructed marshes showed that younger marshes had more labile MOM than older marshes indicating they were less likely to sequester OC in the long term (Craft et al., 2003).

Sources of SOC may also be important as they influence OC lability, carbon to nitrogen ratios (C:N), and the rate at which OC accumulates. Morgan and Short (2002) hypothesized that the lag time in OC accumulation is because macrophytes must first become established before they contribute to the SOM pool. Others claim that seston, a mixture of plankton and detritus, is the main source of SOM so accumulation should occur whether a site has macrophytes or not (Cammen 1975). Organic matter C:N ratios may also be a significant parameter because the ratios indicate whether accumulation of C is likely. If the C:N ratio is low, the microbes may be starved for C, and therefore more likely to decompose OC and respire CO_2.

Because these studies have been so broad in scope, they also do not take the time to use the best methodology for measuring OC. While loss on ignition (LOI) is the easiest way, the high carbonate content of coastal sediments/soils may interfere with the results (Nieuwenhuize et al., 1994). Furthermore, LOI is a measure of OM so conversion factors, with their associated errors, need to be used to convert an OM value to an OC value. Older studies (e.g. Cammen 1975) used the Walkley-Black chromic acid oxidation method to determine OC, which is only 75-90% efficient at obtaining a true OC value (Nieuwenhuize et al., 1994). While errors in the method are not a significant problem for

comparison studies reviewed here, they are a concern if constructed wetlands are to be used for C emission offsetting. Future work should consider *in situ* acidification techniques and subsequent analysis with an elemental analyzer as the standard method for OC measurements (Nieuwenhuize et al., 1994).

Lastly, research is needed that addresses the permanency of C storage in constructed and restored coastal systems. Disturbances like changes in nutrient loading, invasive species, and hurricanes can affect C storage. A nutrient loading experiment in a North Carolina salt marsh increased microbial respiration and caused a subsequent net loss of SOC over 12 years (Morris and Bradley 1999). Alternatively, the spread of a native, yet invasive, grass species in a natural coastal marsh in France was found to increase SOC storage (Valery et al., 2004). These studies occurred in natural marshes, and studies that examine disturbance effect on SOC in constructed coastal systems are needed because constructed systems may be less resilient than natural systems. In order for constructed coastal systems to become a viable option for C offsets, these effects need to be understood and quantified.

More thorough studies are needed on the C sink capabilities of restored and constructed coastal ecosystems. Thus far, the vast majority of studies have been carried out in salt marshes. Studies are needed in coastal ecosystems like mangrove forests and seagrass beds whose destruction is also routinely mitigated with restoration and construction. If researchers can prove that constructed systems are effective C sinks by demonstrating that they not only follow trajectories of increasing SOC, but also have OC accumulation rates similar to natural ecosystems, then constructing coastal ecosystems may become an accepted way to offset CO_2 emissions. Institutions such as Climate Neutral (www.climateneutral.org) and the Chicago Climate Exchange (www.chicagoclimatex.com) could use coastal ecosystem construction to offset emissions like they now do with certain forestry and agricultural practices.

Sediment Organic Carbon Source Determination

One of the new directions functional trajectory studies could take is examining sources of SOC in constructed coastal systems. Determining SOC sources is important to the study of OC storage in coastal ecosystems as the identity of sources is one of the factors that determine OC lability and accumulation rates. Hedges (1992) stated that understanding the types of OM that accumulate in marine sediments was one of the key questions that needs to be answered in order to better understand global biogeochemical cycles. The source determination question most often studied is whether the SOC is of allochthonous (via sedimentation) or autochthonous origin (Middelburg et al., 1997; Bouillon et al., 2003; Golding et al., 2004). If most SOC is of autochthonous origin, then contributions to SOC from the primary producers needs to be teased apart, but this detailed question is harder to answer and rarely studied (Bouillon et al., 2003). There are many possible SOC sources in seagrass beds, mangroves, and salt marshes. Coastal ecosystems can have allochthonous OC inputs of planktonic origin from the open sea or of terrestrial plant and anthropogenic origin. These systems also have numerous potential autochthonous OC inputs. Within a seagrass bed OC can come from different species of seagrass, epiphytes, macroalgae, or benthic algae. Seagrass beds can also receive OC inputs from adjacent mangroves (Kennedy et al., 2004; Lin et al., 1991). The complexities of seagrass beds,

mangroves, and salt marshes make OC source determination difficult. However, making sense of complex OC sources and their role in C accumulation and storage is important for the conservation of coastal ecosystems in the face of increased nutrient loading and sea level rise and the maintenance of their C sink capabilities.

Many methods have been used to determine SOC sources; however, no single method has offered a definitive answer among and within system types. Some methods were developed for two end-member systems—systems in which there are only two distinct sources of OC such as allochthonous terrestrial plant matter and autochthonous marine plankton. Such simple systems may be encountered in estuaries that lack submerged aquatic vegetation (Golding et al., 2004). Sometimes researchers simply group the sources of OC into two end-member groups. For example, in salt marshes the SOC inputs from *Spartina* can be distinguished from the inputs from all other sources because they differ in their $\delta^{13}C$ values (Middelburg et al., 1997). These two end-member models are often useful in estimating the major categories of OC sources (i.e. whether allochthonous or autochthonous), but they cannot fully partition the individual SOC sources.

The variety of methods used to determine OC inputs range widely in terms of time and equipment involved. Methods can be as simple as comparing C:N ratios of possible sources with sediment C:N ratios or as complicated as searching for a biomarker and then isolating and concentrating that specific compound for isotopic analysis. The most widely used method involves stable isotopes—either comparing bulk composition of $\delta^{13}C$ in possible sources and sediments or comparing composition of $\delta^{13}C$ in lipids found in possible sources and sediments. Lipids and other biomarkers can also be used singly to determine sources. Other methods, which include petrographic analysis and nuclear magnetic resonance spectroscopy (NMR), involve comparing relative amounts of different OC structures in the soil.

Stable Isotopes

In salt marshes, mangrove forests, and seagrass beds many researchers have tried to determine SOC sources by matching the $\delta^{13}C$ isotopic signatures of the bulk sediment to the $\delta^{13}C$ isotopic signatures of the sources via strait comparison of the numbers, mixing models, or diagrams (Table 2-5). In order for stable isotopes to elucidate OC sources, the sources need to have consistently distinct isotopic signatures (Papadimitriou et al., 2005). The various primary producers in coastal systems develop distinct isotopic signatures through their discrimination against heavy isotopes during carbon uptake and fixation. Discrimination against the heavy isotope is highest when the inorganic C exceeds supply. Generally, C_3 plants are lighter ($\delta^{13}C$ = -35 to -20 ‰) than C_4 plants ($\delta^{13}C$ = -15 to -9 ‰) in their $\delta^{13}C$ signatures due to the strong isotopic discrimination of the carboxylase Rubisco, an enzyme that is not found in the C fixation pathway of C_4 plants (Hemminga and Mateo 1996; Hemminga and Duarte 2000). Luckily for C source determination in coastal systems, the principal primary producers of seagrass beds, mangrove forests, and salt marshes all have isotopic signatures distinct from the signatures of the less abundant primary producers within these systems. Seagrass are relatively heavy isotopically with average $\delta^{13}C$ values of -10 to -11 ‰ (Hemminga and Mateo 1996). Mangroves, a C_3 plant, have isotopic signatures close to that of many terrestrial primary producers with $\delta^{13}C$ values around -28 ‰

(Jennerjahn and Ittekkot 2002; Kennedy et al., 2004). *Spartina* species that dominate salt marshes are C_4 plants with $\delta^{13}C$ values around -12 to -13 ‰ (Haines 1976; Middelburg et al., 1997). The isotopic signatures of other primary producers such as plankton and epiphytes generally fall below that of seagrass and *Spartina* and above that of mangroves (Kennedy et al., 2004; Papadimitriou et al., 2005), but this is not always the case.

In order for the bulk stable isotope method to be accurate, the $\delta^{13}C$ signature of the sources must not change during decomposition, or if they do change, the magnitude of the change needs to be small when compared to inter-source differences (Papadimitriou et al., 2005). Changes during decomposition are often small, like the 0.7 ‰ difference found between fresh and senescent mangrove leaves in Brazil (Jennerjahn and Ittekkot 2002), but are variable in direction and magnitude depending on the plant (Dai et al., 2005).

A study of OC inputs into seagrass (*Posidonia oceanica*) sediments of 22 sites in the northwestern Mediterranean by Papadimitriou et al., (2005) is a good example of the potential of bulk isotopic studies and their inherent weaknesses. They measured $\delta^{13}C$ and ^{15}N isotopic signatures of the top 2cm of fine fraction (>63um) sediments and of potential sources—seston (assumed to represent phytoplankton), above- and below-ground seagrass tissues, and epiphytes. $\delta^{13}C$ values of the sediments ranged from -15.8 ‰ to -21.5 ‰ and average $\delta^{13}C$ values of seston, epiphytes, below-ground seagrass tissues, and above-ground tissues were -22.1 ‰, -17.8 ‰, -12.1 ‰, and -12.6 ‰, respectively. No systematic differences in the ^{15}N values of the potential sources were found; most likely because discrimination against different N isotopes is not due to physiology of primary producers and because N is often a limiting nutrient. At all sites, SOC was more depleted isotopically than seagrass tissues but less depleted than seston. Using a mixing equation based on one developed by Dauby (1989), they were able to find a range of fractional contribution values of each source.

$$^{13}C_{sediment} = f_{seston}\delta^{13}C_{seston} + f_{epeiphytes}\delta^{13}C_{epiphytes} + f_{seagrass}\delta^{13}C_{seagrass}$$

In equation 2-1, f_i is the unknown proportion of the OC from source *i* in the SOC pool and $\delta^{13}C$ is the isotopic signature of source *i*. This equation is used to find the range of *f* values for each source needed to satisfy the equation and equal the sediment $\delta^{13}C$ value. With this model, they were able to determine which sites had seston as the major contributor to SOC and which sites had seagrass as the major contributor to SOC. If this were simply a two end member system involving seagrass and seston, the elucidation of sources to the sediments using this model would have been straightforward. But these sites also included epiphytes, and their intermediate $\delta^{13}C$ signature made it impossible for the model to assign them reasonable contribution ranges (often the ranges included a 0% contribution). Thus the relative contribution of epiphytes to SOC could not be determined by bulk isotopic methods alone.

A similar method was employed by Kennedy et al., (2004) when they examined SOC sources in seagrass beds, mangroves, and mixed seagrass/mangrove systems in the South China Sea. They measured $\delta^{13}C$ values of sediments, particles in sediment traps, and potential sources (seagrass leaves, mangrove leaves, epiphytes, and seston). They found consistent and distinct differences in the $\delta^{13}C$ values of potential sources. The use of the mixing equation gave broad estimates of source contribution, which

suggested that seagrass and mangroves contributed to the SOC in their respective systems, but that seston and epiphytes were probably the dominant sources. As with the study by Papadimitriou et al., (2005), the presence of intermediate signatures (epiphytes and seston) between the two end members (seagrass and mangroves) made determination of contributions from sources with intermediate $\delta^{13}C$ values difficult.

In salt marshes, similar problems are encountered. While the importance of the main primary producer's contribution can be easily elucidated, the contributions of other sources with less distinct $\delta^{13}C$ signatures cannot be. In one of the first studies that used C isotopes to examine SOC sources, *Spartina* had the distinctive enriched $\delta^{13}C$ values (-12.3 to -13.6 ‰) of C_4 plants, but all other vascular plants including species as disparate as *Juncus roemerianus* and *Salicornia virginica* had signatures between -22.8 and -26 ‰ because they were C_3 plants (Haines, 1976). Benthic diatoms in this study were plagued with the same intermediately-valued problem as the previously-discussed epiphytes with $\delta^{13}C$ values between -16.2 and -17.9$^0/_{00}$. Through comparing the primary producer and sediment values, Haines concluded sediment $\delta^{13}C$ values generally reflected values of plants growing in the sediments. Sediments beneath C_4 plants were slightly more depleted in $\delta^{13}C$ than the C_4 plants, and sediments beneath the C_3 plants were slightly more enriched in $\delta^{13}C$ than the C_3 plants. The sediments' differences from *in situ* vegetation may have been due to C_3 and C_4 plant detritus mixing or input from benthic diatoms. Inputs to SOC from individual species of C_3 plant were unknown because their signatures were not distinct.

Middelburg et al., (1997) avoided problems associated with intermediate and indistinct values by using $\delta^{13}C$ isotopes for the sole purpose of determining the amount of *Spartina*-derived SOC in salt marshes in Massachusetts (Great Marsh) and the Netherlands (Waarde Marsh). In Great Marsh, high marsh SOC $\delta^{13}C$ value (-13.4 to -14.5 ‰) was similar to the *Spartina* value (-12.5 ‰), but low marsh SOC $\delta^{13}C$ value (-21 to -19.5 ‰) was not. In Waarde Marsh SOC was 9-12 ‰ less than the *Spartina* value (-12.7 ‰). They hypothesized that depletions of SOC values in Waarde marsh and the low marsh of Great Marsh were due to the input of allochthonous OM such as marine plankton and non-local macrophyte, but since they did not measure these sources they could not definitively identify which contributed to the depletion. They concluded Great Marsh was a peaty marsh where C accumulation was due to *Spartina* inputs and Waarde marsh was a mineral marsh where accumulation was due to sedimentation.

Problems with intermediate values were encountered by all previously discussed studies that tried to comprehensively measure $\delta^{13}C$ values of all major sources. These studies often used either a variation of the mixing equation developed by Dauby (1989) or a simple comparison of sources and sediment isotope values. However, other ways to calculate source contributions may partially eliminate problems with intermediate values. Gonneea et al., (2004) used a ternary mixing diagram to elucidate relative source contributions of seagrass, mangroves, and seston to SOC. With a ternary mixing diagram, all sources were end members as they formed a triangle on a graph of C:N ratios plotted against $\delta^{13}C$ values. Sediment C:N and $\delta^{13}C$ values were also plotted on this diagram, which had a 10% tolerance interval to account for natural variability in source values. Sediment samples that fell in the middle of the triangle were assumed to be a mixture of all three sources, samples that fell along a line connecting two end-members were considered a mixture of those two sources, samples that fell near one end

member were assumed to have OC predominantly from that source, and samples that fell outside the triangle were assumed to have OC contributions from additional sources. This method is limited to systems with three main sources. Conclusions of research, like Papadimitriou et al.,'s (2005) study of seagrass, seston, and epiphytes, could have benefited from this method if had they measured C:N ratios.

Comparisons of actual values to values from a predicted model can sometimes help elucidate sources better than a mixing equation based on the actual data. These models are based on biomass of potential sources (Chmura et al., 1987), primary productivity (Bull et al., 1999), or %SOC (Middelburg et al., 1997). The problem with these models is that they assume sources contribute to SOM in the same relative proportions as their biomass/productivity. This assumption may not be correct because sources differ in their degrees of lability, in their litterfall, and in the amount of their biomass that is exported out of the system. However, models are good approximations, especially in more peaty coastal wetlands where sediments have high OC and sedimentation of allochthonous OC inputs is minimal.

For the above methods of calculating SOM sources from isotope values, parameters other than the $\delta^{13}C$ values, such as biomass or C:N ratios, are needed. These other parameters also support conclusions based on isotopic values alone. Many studies combine C:N ratios with isotopic measurements (Middelburg et al., 1997; Bouillon et al., 2003; Soto-Jimenez et al., 2003; Thimdee et al., 2003; Gonneea et al., 2004). Correlations between C:N ratios or %SOC and $\delta^{13}C$ values are used to assist in determining SOC sources. A mild relationship ($R^2 = 0.26$) was found to exist between $\delta^{13}C$ values and C:N ratios in a Mexican salt marsh where less negative $\delta^{13}C$ values corresponded to lower C:N ratios (Soto-Jimenez et al., 2003). In the Mexican marsh lower C:N ratios were thought to be indicative of marine producers, specifically plankton. A brief review of sediment $\delta^{13}C$ and C:N values from mangrove literature also showed an inverse relationship between the two variables (Bouillon et al., 2003). Similar trends were found when comparing sediment $\delta^{13}C$ and %SOC values in mangroves (Bouillon et al., 2003) and salt marshes (Middelburg et al., 1997). More depleted $\delta^{13}C$ values corresponded with higher %SOC in mangroves and more enriched $\delta^{13}C$ values corresponded with higher %SOC in salt marshes. Generally, the higher the sediment %OC values, the closer the sediment $\delta^{13}C$ values are to the $\delta^{13}C$ values of the dominant vegetation (Bouillon et al., 2003).

There are other complications with the use of stable isotopes for OC source determination. Problems not already discussed include inherent variation of isotopic signatures within different tissues of a single individual (Papadimitriou et al., 2005) and within a single species (Hemminga and Mateo 1996) across sites (Kennedy et al., 2004), seasons, and years (Anderson and Fourqurean, 2003; Fourqurean et al., 2005). These variations are most pronounced in seagrass (Thimdee et al., 2003). Such variation may be due to changes in relative uses of dissolved CO_2 and HCO_3^- (sources of inorganic C in water) (Lin et al., 1991), and changes in irradiance, photosynthesis rates, and temperature (Hemminga and Mateo 1996). Kennedy et al., (2004) found that isotopic signatures of sources such as seagrass ($\delta^{13}C$ = -5.8 to -13.3 ‰) and seston ($\delta^{13}C$ = -9.6 to -22.9 ‰) varied greatly among 15 different sites in the South China Sea. The order trend of potential sources' $\delta^{13}C$ signatures (seagrass > epiphyte > seston > mangrove) remained constant, however. Variation by location means that conclusions based on measurement of SOC $\delta^{13}C$ values without measuring potential sources may not be valid. Soto-Jimenez et al., (2003) inappropriately used isotopes when they only measured sediments signatures in a Mexican marsh.

Average $\delta^{13}C$ value of sediment was -20.4 ‰ and they assumed, based on previously published $\delta^{13}C$ signatures of sources in *temperate* estuaries, that dominant SOC sources were plankton and macrophytes. While $\delta^{13}C$ values of putative sources at each site need to be measured at least once per site, Fourqurean et al., (2005) proposed further determination of source $\delta^{13}C$ values seasonally and annually.

Lipid Biomarker Compounds

The use of specific organic compounds, called biomarkers, to identify SOC sources in coastal ecosystems is becoming more common. These compounds are generally lipids including sterols, fatty acids, and hydrocarbons. The ways these organic compounds are used vary because the compounds vary in their specificity—some can identify groups of organisms such as vascular plants or algae while others may be specific to one genera or species (Canuel et al., 1997). Less specific biomarkers can be used in conjunction with stable isotopes to further differentiate sources from general groups (i.e. vascular plants into C_3 and C_4 groups). Many studies used biomarkers in concert with bulk stable isotopes (Hernandez et al., 2001; Wang et al., 2003) or measured the stable isotopic composition of biomarker compounds in sources and sediment (Canuel et al., 1997; Bull et al., 1999; Hernandez et al., 2001; Mead et al., 2005).

This method uses a gas chromatography (GC) to determine lipids after a complex and lipid-type specific extraction process. The different lipids are separated by the GC column due to their different retention time within the column. Different lipids can be identified by comparing their relative retention times on the resulting gas chromatogram with relative retention times on a gas chromatogram of a known standard. Relative amounts of each lipid can also be determined by calculating areas underneath each peak on the chromatogram—larger areas correspond to a larger amount of that lipid in the sample. Isotopic signatures of these lipids can be determined when the GC is connected to a mass spectrometer in a method known as isotope ratio-monitoring gas chromatography-mass spectroscopy (irm-GCMS) (Canuel et al., 1997).

Canuel et al., (1997) examined the usefulness of isotopic signatures of specific lipid compounds to identify SOC sources in coastal ecosystems. The study examined isotopic signatures of bulk organic matter, total lipid extracts, and a whole suite of lipid compounds in three vascular plants, *S. alterniflora, J. roemerianus*, and *Zostera marina*, suspended particulate matter (SPM), and sediment in North Carolina. Vascular plants had similar molecular compositions of sterols and fatty acids but differed in hydrocarbon compositions, specifically in ranges and maxima of *n*-alkanes and the presence of monosaturated alkenes. SPM had a different lipid composition than vascular plants; the majority (>50%) of SPM's hydrocarbons were C_{25} highly branched isoprenoids (HBI) alkenes. Among different vascular plant lipids there was a variety of isotopic signatures with an average depletion of 3-5 ‰ in lipid $\delta^{13}C$ values relative to bulk values. Lipids in *Z. mostera*, *S. alterniflora*, and *J. roemerianus* followed the same trend in $\delta^{13}C$ values as bulk tissues with mean $\delta^{13}C$ values (in ‰) of -14.8 to -18.9, -18.4 to -22.6, and -29.0 to -33.8 for lipids and of -10.0, -12.6, and -26.0 for bulk tissues, respectively. Differences between bulk and lipid signatures demonstrated another reason caution should be used in analyzing bulk isotopic studies because compounds preserved in SOC may not have the same signature as bulk plant matter

(Canuel et al., 1997). $\delta^{13}C$ values of lipids in sediments were different than those for the same lipids in vascular plants, but similar to SPM lipids. Sediments had a higher diversity of lipids than vascular plants, small amounts of major vascular plant biomarkers like C_{21} and C_{29} (maxima observed in the *Z. mostera* and *S. alterniflora* tissues), and a lot of the major SPM biomarker, C_{25} HBI. The study concluded vascular plants were only minor contributors to SOC.

The North Carolina study showed how the molecular distribution (diversity of types and dominant types) of lipids and isotopic signatures of lipids can be used to determine SOC sources. The method was not without problems, however. Use of compound classes that may not be the "most" diagnostic for vascular plants may have skewed results. Furthermore, this technique is biased toward extractable, not bound, lipids in sediments (Canuel et al., 1997). It is important to note that only the top 0.5 cm of sediment was analyzed in this study. Thus, depths where macrophyte roots may contribute to SOC through exudates or senescent tissue were ignored.

Not all studies examine a wide range of lipids. It is common to examine only one lipid class such as *n*-alkanols (Bull et al., 1999) or n-alkanes (Wang et al., 2003). Studying the $\delta^{13}C$ values of one type of lipid biomarker can help solve the intermediate-value problem that muddles analysis of SOC sources in bulk isotope studies. The use of compounds that are specific to vascular plants (n-alkanes) or to plankton and algae (HBI alkenes; Canuel et al., 1997) for isotope studies can help clarify their contributions to SOC (i.e.: whether a sediment bulk $\delta^{13}C$ value of -18 ‰ is due to an even mix of C_4 and C_3 plants, only plankton, or a mixture of all three). The n-alkanol homologue, C_{32}, was chosen for a study addressing contributions of *S. alterniflora* and *Puccinellia maritima* to salt marsh SOC in the United Kingdom (Bull et al., 1999). By only examining $\delta^{13}C$ values of an n-alkanol, which plankton cannot produce, plankton's confounding intermediate $\delta^{13}C$ value was removed as a factor from the isotopic mixing equation. Using a two-member mixing model based on $\delta^{13}C$ values of the C_{32} n-homologue, contributions of *S. alterniflora* to SOC were calculated. *S. alterniflora* contributed about 100% of primary biomass to sediments directly beneath *S. alterniflora* stands and about 50% to sediments beneath *P. maritima* stands. To fully understand all sources to SOC, this method should be expanded to include group-specific biomarkers for both vascular plants and plankton. Otherwise when analyzing only $\delta^{13}C$ values of a biomarker for one plant group, contributions of plants outside that group remain unknown.

Mead et al., (2005) took the examination of group-specific series of homologues a step farther when they used an n-alkane-based proxy, P*aq*, along with compound-specific stable isotopes to elucidate sources along a gradient of freshwater marsh to estuarine mangrove forests to marine seagrass beds in the Florida Everglades. P*aq* is calculated from abundances of different *n*-alkane homologues.

$$Paq = (C_{23} + C_{25}) / (C_{23} + C_{25} + C_{29} + C_{31}) \qquad (2\text{-}2)$$

In equation 2-2, C_x is the amount of the C_x *n*-alkane. Submerged and floating macrophytes like seagrass contained more abundant mid chain *n*-alkanes and therefore had a higher P*aq* than emergent macrophytes and terrestrial plants like mangroves. This method was able to resolve sources to a greater extent than studies using only isotopes or only biomarkers in estuaries because sources with similar P*aq* values were differentiated using n-alkane $\delta^{13}C$ values and vice versa. Generally, as the gradient went

from freshwater marsh to seagrass beds there was a trend of increasing sediment $\delta^{13}C$ values and increasing P*aq* values. These trends were further connected to contributions of individual sources through a PCA based on compound specific $\delta^{13}C$ and P*aq*.

An example using a species-specific biomarker is the study of different homologues of the *n*-alkane-2-ones lipid series to elucidate SOC sources in the Harney River estuary and the adjacent Florida shelf (Hernandez et al., 2001; Mead et al., 2005). Lipids in this series generally have odd-numbered C chains ranging from 19 to 33 C's in length. There has been some debate about whether n-alkane-2-ones arise is sediments directly from plant detritus or whether they arise from microbial oxidation of alkanes. However, Hernandez et al., (2001) were able to find significant amounts of n-alkane-2-ones in tissues of seagrass and mangroves. In seagrass, the most common (82% to 88% of the ketone fraction) n-alkane-2-one was the C_{25} homologue, and in mangroves the most common n-alkane-2-ones were the C_{27}-C_{31} homologues. Gas chromatograms of sediments in the lower estuary and the shoreward section of the Florida shelf showed a predominance of seagrass-derived C_{25} homologues, implying that seagrass was a major SOC source there. In upper estuarine sediments, there was a predominance of higher molecular weight homologues implying mangroves were the major SOC source. Isotopic measurements of bulk SOC and n-alkane-2-ones confirmed these conclusions about primary SOC sources because sediment $\delta^{13}C$ values became more enriched (i.e. more like seagrass-derived SOC) as the samples went from the upper estuary to the Florida shelf. By using biomarkers specific to vascular plants, however, contributions to SOC from algae and plankton were unknown.

As with the use of bulk stable isotope measurements, there are caveats with the use of lipid biomarkers. First, this method has not been as extensively studied as the use of bulk isotopes. Inherent variation of molecular distributions and compound specific isotope signatures within different tissues of an individual plant, within plants of the same species, across geographical areas, and across seasons has yet to be documented (Canuel et al., 1997). Also, the more specific biomarkers may not be applicable to all species of the same plant type. The temperate seagrass, *Z. marina*, did not have the predominant C_{25} n-alkane-2-one homologue that sub-tropical seagrass species had (Hernandez et al., 2001). Not all major ecosystem components will have appropriate species-specific biomarkers, so a combination of species-specific and group-specific biomarkers may have to employed (Mead et al., 2005). Biomarkers confirm the presence of a certain source in SOC, but they do not necessarily yield relative contributions of sources because not all sources are represented in each lipid type. Just because the isotopic mixing equation indicates that 50% of a biomarker is from *S. alterniflora* and the other 50% is from *P. maritima* does not mean each species contributes to 50% of the bulk SOC. Furthermore, when examining isotopes of group-specific lipids, one must make sure that the lipids being examined have similar abundances in each species. Otherwise, what seems like a greater abundance of one species in SOC might actually be due to a greater abundance of that biomarker in tissues of that species relative to other species. In cases where biomarkers are not likely to be at the same concentration among species, relative abundances of biomarkers within each plant should be included in isotopic mixing equations (Bull et al., 1999).

Petrographic Analysis

Petrographic analysis of sediments is a lesser-used method to determine SOC sources. Petrographic analysis microscopically examines organic matter for recognizable organic components such as macrophytic tissues, differentiated based on their level of decomposition, and algae. Marchand et al., (2003) examined SOC sources in mangrove forests of various ages using six different categories of plant tissues: Translucent ligno-cellulosic debris (TLC), which exhibited preserved cell wall structures, degraded ligno-cellulosic debris (DLC), which exhibited decaying cell walls, gelified particles (GP), which were orange brown gel-like particles produced by cellulose degradation, reddish amorphous organic matter (RAOM), in which the cellulose is completely degraded, oxidized opaceous ligno-cellulosic debris (OLC), which were dark and structureless refractory land-derived OM, and grayish amorphous organic matter (GAOM), which were the remains of algae and phytoplankton. This study looked at relative proportions of these various components to understand whether SOC sources to mangrove forests were autochthonous algae, mangroves, or allochthonous riverine detritus. Combining proportions of these OM components with C:N ratios, they found that sediments of younger mangrove forests, with their low C:N ratios and high proportion of GAOM, were dominated by algal-derived OM and that more mature forests, with their higher C:N ratios and ligno-cellulosic debris, were dominated by mangrove-derived OM. They also found that the upper sediments of the younger forests and the deeper sediments of the older forests had a lot of OLC, indicating a trapping of allochthonous OM from the river. With this method, known proportions of various OM components can be measured directly, in contrast to isotopic methods. However, OM components such as TLC and GP cannot be directly attributed to any one species of primary producer, but rather to broad classes of producers. This study did not take seagrass into consideration, which may have similar-looking partially decomposed ligno-cellulosic tissues as mangroves, making it hard to differentiate between those two sources using this method.

Nuclear Magnetic Resonance Spectroscopy

Nuclear magnetic resonance spectroscopy (NMR), specifically ^{13}C-NMR, is another method that can be used to identify SOC sources. However, no known studies document this method in seagrass, mangrove, or salt marsh sediments, and therefore an estuarine study is used to illustrate this method. In NMR spectroscopy, a sediment sample is subjected to a magnetic field which causes the nuclei in the ^{13}C atoms to process as a gyroscope does (Stevenson 1994). A second alternating magnetic field is then added, and when the frequency of that second magnetic field matches the frequency of the nuclei's precession, the nuclei of the atom then resonate causing a voltage change that is amplified and recorded. A spectrum is produced from this resonance signal. The nuclei resonate at different frequencies depending on their chemical environment. Each sample resonates at several frequencies, and from the different resonance signals, spectra with several distinct peaks are produced. Spectra are plotted using the chemical shift—the difference between resonance frequencies of samples and the resonance frequency of a standard, tetramethylsilane (TMS) solution. This chemical shift calculation is analogous to the calculation of δ^{13}C values based on how much the samples' values differ from the PDB standard. Each organic structure, such as an aromatic ring or a carboxyl group, has a different resonance subsequent chemical shift; therefore this method allows scientists to assign categories of OM

to specific chemical shifts (Golding et al., 2004). Using this method, types and relative amounts of OM structures in sediments can be elucidated.

Golding et al., (2004) used ^{13}C-NMR to study whether SOM was terrestrial- or marine-derived in upper (fluvial) and lower (marine) sections of Australian estuaries. They studied four groups of organic carbon structures—carbonyls, aromatics, *O*-alkyls, and alkyls. They associated both *O*-alkyl C and aromatic C with terrestrial plant sources because they assumed *O*-alkyl C was from cellulosic carbohydrates and aromatic C was from lignin and tannins of vascular plants. The presence of alkyl C indicated marine origins because they associated it with planktonic material. They cautioned, however, that alkyl C may also be present due to microbial decomposition of terrestrial OM. The authors concluded that upper portions of estuaries had higher proportions of *O*-alkyl C and aromatic C, and therefore higher amounts of terrestrially-derived SOC, than lower portions of estuaries.

NMR has similar problems as petrographic analysis because structures being studied cannot be directly assigned to specific primary producers; the relationship between the producer and the structure must be inferred, and one structure type can come from several producers. This technique may be best suited to situations where sources are grouped into a couple of components such as a study of seagrass/mangrove-derived SOC and planktonic SOC. Despite problems associated with SOC source determination using ^{13}C-NMR, this tool may help scientists better elucidate roles of plankton and algae, whose proportions in SOC are difficult to determine via stable isotopes because of their variable and intermediate δ^{13}C values. This method also helps scientists understand the OC structures, not just the OC sources in coastal sediments.

Conclusion

This review sought to cover SOC topics relevant to this thesis research in three types of coastal ecosystems—salt marshes, mangrove forests, and seagrass beds. The original research in this thesis covers OC pools and sources in a constructed mangrove and seagrass system. This review covered salt marshes in order to add both depth and breadth because C accumulation and constructed ecosystem development are currently better understood in salt marshes than in mangrove forests and seagrass beds. This review discussed C accumulation rates of salt marshes, mangrove forests, and seagrass beds, functional trajectories of OC attributes in restored and constructed salt marshes, and SOC source determination methods in salt marshes, mangrove forests, and seagrass beds. The C accumulation section showed that these coastal ecosystems are globally significant C sinks. The functional trajectory section showed how OC functions in constructed salt marshes and emphasized the need for further and more in-depth studies of OC in constructed coastal ecosystems. The SOC source determination section showed pros and cons of different SOC determination methods, including bulk stable isotopes, which are utilized in the original thesis research.

Table 6. Global area of mangrove forests, salt marshes, and seagrass beds.

System	Global area (km^2)	Data source[a]
Mangrove forests	200,000	1
	218,000	2
	240,000	3
Salt marshes	300,000	2
	400,000	4
Seagrass beds	300,000	5
	600,000	6

[a]1, Jennerjahn and Ittekkot 2002; 2, Twilley et al., 1992; 3, Mitsch and Gosselink 2000; 4, Duarte and Cebrián 1996; 5, Suzuki et al., 2003; 6, Hemminga and Duarte 2000.

Table 7. Global rates of carbon accumulation in coastal ecosystem sediments.

System	Average type	Global areal rate ($g\ C\ m^{-2}\ yr^{-1}$)	Global rate ($Tg\ C\ yr^{-1}$)	Data source[a]
Mangrove forests	Mean[1]	92	20	1
	Mean	100	22	2
	Estimate[2]	200	44	3
	Mode[1]	115	25	4
Salt marshes	Mean	50-5,000	17.5-1750	5
	Mean	100	35	1
	Mean	175	61	2
	Mode	115	25	4
Intertidal[3]	Mean	210	120	6
Seagrass beds	Estimate[4]	1.2	0.54	7
	Mean	133	60	2
	Estimate	270	122	8, 9
	Mode	36.5	16.5	4
Open ocean	Mean	0.22	170	2
Terrestrial systems[5]				
Tundra		0.2-2.4		10
Temperate forest		0.7-10		10

Tropical rainforest	2.3	10
Temperate grassland	2.2	10
Temperate desert	0.8	10

[a]1, Twilley et al., 1992; 2, Duarte and Cebrián 1996; 3, Jennerjahn and Ittekkot 2002; 4, Cebrián 2002; 5, Rabenhorst 1995; 6, Chmura et al., 2003; 7, Suzuki et al., 2003; 8, Duarte and Chiscano 1999; 9, Hemminga and Duarte 2000; 10, Schlesinger 1990.

[1]Both the mean and mode numbers were derived from compiling numbers from published studies. [2]The estimates were either scaled up from a single study or derived from a rough "back-of-the-envelope" calculation. [3]Number includes contribution of both mangrove forests and salt marshes. [4]Estimate is of amount being exported and subsequently buried in the open ocean sediments, not *in situ* accumulation. [5]These numbers represent long term accumulation rates measured since the end of the last ice age.

Table 8. Rates of carbon accumulation in coastal ecosystem sediments and the methods used to calculate the time component of the rates.

Location	Site	C accumulation ($g\ C\ m^{-2}\ yr^{-1}$)	Time scale	Method	Data source[a]
Mangrove forests					
Herbert River Estuary, Australia		180	Century	^{210}Pb profiles	1
Jiulonglijang Estuary, China	High intertidal	168	Century	^{210}Pb profiles	2
	Mid intertidal	204	Century	^{210}Pb profiles	2
	Low intertidal	841	Century	^{210}Pb profiles	2
Florida Keys, Florida	Rhizophora mangle	159[1]	30 Year	^{137}Cs profiles	3
	Avicennia germinans	105[1]	30 Year	^{137}Cs profiles	3
Rookery Bay, Florida	Fringe	228[1]	Annual	Feldspar marker	4
	Basin	328[1]	Annual	Feldspar marker	4
	Exposed island	291[1]	Annual	Feldspar marker	4
	Sheltered island	191[1]	Annual	Feldspar marker	4
Matang Forest Preserve, Malaysia		150	8,000 Year	Estimate	5
	5-yr-old stand	101	Century	^{210}Pb profiles	6
	18-yr-old stand	110	Century	^{210}Pb profiles	6
	85-yr-old stand	127	Century	^{210}Pb profiles	6

table cont.

Celestun Lagoon, Mexico		55-70	Century	^{210}Pb profiles	7
Chelem Lagoon, Mexico		67-104	Century	^{210}Pb profiles	7
Terminos Lagoon, Mexico		33	Century	^{210}Pb profiles	7
Sawi Bay, Thailand		184-281	Decadal	^{137}Cs and ^{210}Pb profiles	8
Brackish Marshes					
Cameron Parish, Louisiana	Natural waterway	700[1]	Annual	Feldspar marker	9
	Restricted canal	35[1]	Annual	Feldspar marker	9
	Restricted natural waterway	30[1]	Annual	Feldspar marker	9
Fina La Terre, Louisiana	Unmanaged	75[1]	Annual	Feldspar marker	10
	Managed	10[1]	Annual	Feldspar marker	10
Rockefeller Refuge, Louisiana	Unmanaged	335[1]	Annual	Feldspar marker	10

table cont.

Location	Site	C accumulation (g C m^{-2} yr^{-1})	Time scale	Method	Data source [a]
Salt marshes					
Upper Bay of Fundy,	Low marsh	39	30 Year	^{137}Cs profiles	11
	High marsh	194	30 Year	^{137}Cs profiles	11
Outer Bay of Fundy, Canada	Low marsh	76	30 Year	^{137}Cs profiles	11
	High marsh	188	30 Year	^{137}Cs profiles	11
St Marks NWR, Florida	Low marsh	117	12 Year	^{14}C bomb uptake[2]	12
	Mid marsh	101	12 Year	^{14}C bomb uptake[2]	12
	High marsh	65	12 Year	^{14}C bomb uptake[2]	12
	Low marsh	25	400-600 Year	^{14}C profiles[2]	12
	Mid marsh	22	400-600 Year	^{14}C profiles[2]	12
	High marsh	20	400-600 Year	^{14}C profiles[2]	12
Lafourche Parish, Louisiana	Continuous canal	300[1]	Annual	Feldspar marker	10

	Discontinuous canal	200[1]	Annual	Feldspar marker	10
	Natural waterway	650[1]	Annual	Feldspar marker	10
Cedar Creek, Maryland		89	150 Year	^{210}Pb profiles[2]	13
		18.5	Millennia	^{14}C profiles[2]	13
Hell Hook, Maryland		78	150 Year	^{210}Pb profiles[2]	13
		39.8	Millennia	^{14}C profiles[2]	13
Barnstable, Massachusetts		96	NA	Modeled	14
Biloxi Bay, Mississippi		180	Decadal	^{137}Cs profiles	3
Waarde Marsh, Netherlands		105	NA	Modeled	14
Consultant, North Carolina	3-yr-old constructed	39	3 Year	ΔOC / Time[2]	15
	Natural reference	35-51	Decadal	^{137}Cs and ^{210}Pb profiles	15
Drum Inlet, North Carolina	Bare spoil	80	16 Month	ΔOC / Time[3]	16
	Spoil planted with Spartina	87	16 Month	ΔOC / Time[3]	16
	Fertilized spoil	96.8	16 Month	ΔOC / Time[3]	16
Dill's Creek,	13-yr-old constructed	62	13 Year	ΔOC / Time[3]	15

table cont.

table cont.

Location	Site	C accumulation (g C m^{-2} yr^{-1})	Time Scale	Method	Data Source [a]
"DOT," North Carolina	1 yr-old constructed	99	1 Year	ΔOC / Time[3]	15
	Natural reference	30-36	Decadal	^{137}Cs and ^{210}Pb profiles	15
Jacob's Creek, North Carolina	Irregularly-flooded streamside	146	Decadal	^{137}Cs profiles	17
	Irregularly-flooded backmarsh	107	Decadal	^{137}Cs profiles	17
"Marine Lab," North Carolina	26-yr-old constructed	34	26 Year	ΔOC / Time[3]	15
	Natural reference	15	Decadal	^{210}Pb profiles	15
Oregon Inlet, North Carolina	Regularly-flooded streamside	58.9	Decadal	^{137}Cs profiles	17
	Regularly-flooded backmarsh	21.3	Decadal	^{137}Cs profiles	17
Pine Knoll Shores, North Carolina	21-yr-old constructed	125	11 Year	ΔOC / Time[4]	18
	Natural reference	115	11 Year	ΔOC / Time[4]	18
"Port," North Carolina	8-yr-old constructed	27	8 Year	ΔOC / Time[3]	15
	Natural reference	28-32	Decadal	^{137}Cs and ^{210}Pb profiles	15

Snow's cut, North Carolina	25-yr-old constructed	99	11 Year	ΔOC / Time[4]	18
	Natural reference	159	11 Year	ΔOC / Time[4]	18
Swansboro, North Carolina	11-yr-old constructed	18	11 Year	ΔOC / Time[3]	15
	Natural reference	105-115	Decadal	^{137}Cs and ^{210}Pb profiles	15
Aransas NWR, Texas		167[1]	Decadal	^{137}Cs profiles	3
San Bernard NWR, Texas		207[1]	Decadal	^{137}Cs profiles	3
United States Average		83	NA	Compiled	19
Seagrass Beds					
Aburatsubo Bay, Japan		1.2-1.5[5]	NA	Modeled	20
Celestun Lagoon, Mexico		40	Century	^{210}Pb profiles	7
Terminos Lagoon, Mexico		53-65	Century	^{210}Pb profiles	7
Cala Culip, Spain		19-191	600 Year	Shipwreck	21
Fanals Point, Spain		182	Annual	Sediment trap	22

table cont.

[a]1, Brunskill et al., 2002; 2, Alongi et al., 2005; 3, Callaway et al., 1997; 4, Cahoon and Lynch 1997; 5, Ong 1993; 6, Alongi et al., 2004; 7, Gonneea et al., 2004; 8, Alongi et al., 2001; 9, Cahoon and Turner 1989; 10, Cahoon 1994; 11, Connor et al., 2001; 12, Choi and Wang 2004; 13, Hussein et al., 2004; 14, Middelburg et al., 1997; 15, Craft et al., 2003; 16, Cammen 1975; 17, Craft et al., 1993; 18, Craft et al., 1999; 19, Hopkinson 1988; 20, Suzuki et al., 2003; 21, Romero et al., 1994; 22, Gacia et al., 2002.

[1]This author reported organic matter accumulation rates, so rates were divided by 2 in order to obtain these numbers. [2]Modeled sediment profiles instead of measuring them directly. [3]Calculated by subtracting the OC in 0-30 cm from the OC in top 10 cm, divided by the age of the site [4]Calculated by subtracting the OC at time 0 from the OC at time 1, divided by time 1-time 0 [5]Denotes carbon buried after exportation to the open ocean not in situ.

Table 9. Studies comparing organic carbon in restored and constructed coastal marshes to OC in natural reference marshes.

Location	Site	Age (years)	Constructed OC (units)	Reference OC (units)	Depth sampled (cm)	Method[a]	Source[b]
Tacoma, Washington[1]	Gog-Le-Hi-Te, Site 1	1	3.5 %	3.3 – 8.7 %	0-2	1	1
		2	3.0				
		3	4.0				
		6	3.5				
	Gog-Le-Hi-Te, Site 2	1	4.0				
		2	4.5				
		3	5.5				
		6	9.0				
	Gog-Le-Hi-Te, Site 3	1	2.5				
		2	2.0				
		3	2.2				
		6	1.2				
	Gog-Le-Hi-Te, Site 4	1	2.0				
		2	2.0				
		3	3.0				
Maine, New	Great Bay Estuary	1	2.0 %	mean = 23 %	0-5	1	2
		2	1.5				
		3	3.0				
		6	2.5				
		14	16				
Core Banks, North	Sound-side marsh,	0	77.3 g OC m^{-2}	362.7 g OC m^{-}	0-13	2	3
		1.3	184.3				
	Sound-side marsh,	0	77.3				
		1.3	193.3				
	Sound-side marsh,	0	77.3				
		1.3	206.4				
San Diego, California[1]	San Diego Bay	2	3.5 %	7.5 – 11 %	Not reported	1	4
		4	5.5				
		8	7.5				
		11	7/0				

Location	Site	Age (years)	Constructed OC (units)	Reference OC (units)	Depth sampled (cm)	Method[a]	Source[b]
Georgia	Sappelo Island	42	1264 g C m^{-2}	1372 g C m^{-2}	0-10	1	5
North Carolina	Pamlico River Estuary	5	886 kmol C ha^{-1}	10270 kmol C ha^{-1}	0-30	3	6
		15	1866				
Virginia	Gloucester Point	5	95 g C m^{-2}	129 – 163 g C m^{-2}	0-2	1	7
		12	120				
		5	50	146 – 174	14-16		
		12	53				
North Carolina[2]	"DOT"	1	400 g C m^{-2}	3800 g C m^{-2}	0-30	1	8
	Consultant	3	600	4600			
	Port	8	900	2000			
	Swansboro	11	1000	4600			
	Dill's Creek	13	1800	4900			
	Pine Knoll	24	1200	1000			
	Marine Lab	26	2900	5100			
	Snow's Cut	28	2900	10000			

[a]1, loss-on-ignition; 2, Walkley-Black oxidation; 3, CHN analyzer. [b]1, Simenstad and Thom 1996; 2, Morgan and Short 2002; 3, Cammen 1975; 4, Zedler and Calloway 1999; 5, Craft 2001; 6, Craft et al., 2002; 7, Havens et al., 2002; 8, Craft et al., 2003

[1]Signifies study measured organic matter (OM) only, not organic carbon (OC). [2]Signifies study did not measure the same wetland overtime but instead used a space-for-time substitution.

Table 10. Stable Isotope values and dominant source conclusions from carbon source determination studies in coastal ecosystems.

Location	Potential sources	Source $\delta^{13}C$	Site description	Sediment $\delta^{13}C$	Main sources [a]	How main sources determined	Data source [b]
Mangrove forest							
Eastern Brazil	Seston	-21 to -22[1]	Mangroves	-26.9	4	Comparison	1
	Spartina	-26.8[2]	Riverine	-23.8	4		
	Mangroves	-27	Shelf	-21.3	2		
			Slope	-20.5	2		
Gazi Bay, Kenya	Mangroves	-28.25	*Rhizophora mucronata*	-25.3	4	Comparison	2
Gazi Bay, Kenya	Mangroves	-24.12	*Ceriops tagal*	-22.7	4	Comparison	2
Southeast Asia	Seston	-20.5 to -23[3]	Coringa Wildlife Sanctuary, India	-29.4 to -20.6[3]	2	Compared to curve of 2 source mixing model	3
	Mangroves	-27 to -29[3]	Galle, Sri Lanka		4		
			Pampala, Sri Lanka		4		
Mangroves and salt marsh							
Chiricahueto,	NR		Marsh	-20.4	2	Compared to	4

Mexico						literature values	
Salt Marsh							
Florida	NR		*Spartina alterniflora*	-16.9		Comparison	5
			Juncus roemerianus	-23.9			
Sapelo Island, Georgia	Diatoms	-17.0	Bare creekbank	-18.9	NR	Comparison	6
	S. alterniflora	-12.9	Tall *Spartina*	-16.0	NR		
	S. virginica	-26.0	Short *Spartina*	-17.9	5		
	D. spicata	-13.1	*S. virginica* high marsh	-21.6	NR		
	S. virginicus	-13.3	Sand flat	-22.6	NR		
	J. roemerianus	-22.8	Mixed vegetation	-19.3	NR		
	B. frutescens	-26.0					
Barataria Bay, Louisiana	S. Alterniflora	-12.1 to -13.6	Marsh	-16.2	5	Compared to predicted values based on producer biomass	7

Location	Potential sources	Source $\delta^{13}C$	Site description	Sediment $\delta^{13}C$	Main sources [a]	How main sources determined	Data source [b]
Plum Island,	S. Alterniflora	-13.3	Mid marsh	-18.9[4]	both	Comparison and	8
Massachusetts	T. latifolia	-25.3	Upper marsh	-22.81[4]	both	distributions of long	
			Mudflat	-19.39[4]	Both	chain n-Alkanes	
Waarde Marsh,	Spartina	-12.7[2]	Marsh	-22 to	6	Compared to curve of	9
Netherlands	Allochthonous OM	-25.5		-24.6		2 source mixing model	
Cape Lookout Bight,	Seston	-18.4	Fall	-17.8	2	Comparison with lipid	10
North Carolina	Seagrass	-10.0	Spring	-20.3	2	distributions and lipid	
	Spartina	-12.6				$\delta^{13}C$	
	J. roemerianus	-26.0					
Dorset,	Spartina anglica	-12.1	*S. anglica*	-17.6	5	Mixing model using	11
United Kingdom	Puccinellia maritima	-26.9	*P. maritima*	-21.4	5 (50%)	compound specific	
			Mudflat	-20.4	5 (40%)	$\delta^{13}C$	

Barnstable, Massachusetts	Spartina Allochthonous OM	-12.5[1] -25.5[1]	High marsh Low marsh	-13.4 to -14.5 -21 to -19.5	5 6	Compared to curve of 2 source mixing model	9
Seagrass beds							
Gazi Bay, Kenya	Seagrass Mangroves Sediment Traps	-19.7 -26.7[5] -23.3	Closest to mangroves	-22.9	4,1	Comparison	12
Gazi Bay, Kenya	Seagrass Mangroves POM	-18.3 -26.7[5] -22.5	Closer to mangroves	-20.6	4,1	Comparison	2
Gazi Bay, Kenya	Seagrass Mangroves POM	-15.8 -26.7[5] -19.2	Farther from mangroves	-18.5	1	Comparison	2

Gazi Bay, Kenya	Seagrass	-10.70	Farthest from mangroves	-15.14	1	Comparison	2
	Mangroves	-26.7[5]					
	POM	-13.7					

Location	Potential sources	Source $\delta^{13}C$	Site description	Sediment $\delta^{13}C$	Main sources [a]	How main sources determined	Data source [b]
Chale Lagoon, Kenya	Seagrass	-10.72		-14.8	1	Comparison	12
	Mangroves	-26.7[5]					
Silaqui, Philipines	Seston	-16.4		-10.3	3	Percent contribution ranges from mixing equation	12
	Seagrass	-5.8					
	Epiphytes	-9.6					
Pislatan, Philipines	Seston	-16.5		-14.9	2	Percent contribution ranges from mixing equation	12
	Seagrass	-7.5					
	Epiphytes	-10.5					
Spain	Seston	-22.1[3]	Iberian Coast	-15.8 to -21.6[3]	2,1 (3?) 1,2 (3?)	Percent contribution ranges from mixing equation	13
	Seagrass	-12.4[2,3]					
	Epiphyte	-17.8[3]	Balearic Islands	-15.8 to			

				-21.6[3]			table cont.
Fanals Point, Spain	Seston	-24.7		-20.07	2	Percent contribution ranges	14
	Seagrass	-12.2				from mixing equation and	
	Epiphyte	-17				microscopic examinations	
	POM	-21.5					
Can Rhan Lagoon,	Seston	-19.6		-18.6	NR	Percent contribution ranges	12
Vietnam	Seagrass	-8.6				from mixing equation	
Dam Ghia Bay,	Seston	-17.7		-15.8	2	Percent contribution ranges	12
Vietnam	Seagrass	-6.0				from mixing equation	
	Epiphyte	-8.6					
Mi Giang II, Vietnam	Seston	-12.1		-13.2	NR	Percent contribution ranges	12
	Seagrass	-7.6				from mixing equation	
Seagrass and mangroves							
Celestun, Mexico	Seston	-22.1	Fringing mangrove	-24	4,2	Ternary mixing diagram	15

Seagrass	-16.1[2]	Lagoon center	-20	1,2	of $\delta^{13}C$ and N:C	table cont.
Mangrove	-28.6[2]					

table cont.

Location	Potential sources	Source $\delta^{13}C$	Site description	Sediment $\delta^{13}C$	Main sources [a]	How main sources determined	Data source [b]
Chelem, Mexico	Seston	-22.1	Fringe mangrove	-23.1 to	1,2	Ternary mixing diagram	15
	Seagrass	-15.4[2]		-26.1[6]	4,2	of $\delta^{13}C$ and N:C	
	Mangrove	-27.1[2]	Seagrass bed	-17.2 to			
				-22.4[6]			
Terminos, Mexico	Seston	-25.3	Fringe mangrove	-26	4,2	Ternary mixing diagram	15
	Seagrass	-11.9	Seagrass bed	-16	1,2	of $\delta^{13}C$ and N:C	
	Mangrove	-28.6[2]					
Santa Barbara, Philipines	Seston	-19.0	Seagrass bed	-22.7	4	Percent contribution ranges from mixing equation	12
	Seagrass	-10.9					
	Epiphyte	-12.9					
	Mangrove	-28.6					
Buenavista, Philipines	Seston	-17.7	Seagrass bed	-15.7	1	Percent contribution ranges	12
	Seagrass	-11.7					

	Epiphyte	-13.1				from mixing equation	
	Mangrove	-28.1					
Umalagan, Philipines	Seston	-27.6	Seagrass bed	-26.6	2 or 4	Percent contribution ranges	12
	Seagrass	-12.3				from mixing equation	
	Epiphyte	-22.9					
	Mangrove	-28.4					
Khung Krabaen Bay, Thailand	Seston	-20.6[2]	Canals	-26.5	4	Comparison	16
	Seagrass	-10.5	Mangroves	-26.3	4		
	Macroalgae	-15.6[5]	Inner bay	-15.1	1,7		
	Mangrove	-28.8[2,5]	Mouth of bay	-19.2	2		
	Shrimp feed	-22.5	Offshore	-17.5	2		
Ghia Luan, Vietnam	Seston	-21.6	Seagrass	-24.6	4	Percent contribution ranges	12
	Seagrass	-13.3				from mixing equation	
	Mangrove	-27.9					

[a]The numbers in the main sources column signify the following: 1, seagrass; 2, seston; 3, epiphytes; 4, mangroves; 5, *Spartina*; 6, other; 7, macroalgae.
[b]1, Jennerjahn and Ittekkot 2002; 2, Hemminga et al., 1994; 3, Bouillon et al., 2003; 4, Soto-Jimenez et al., 2003; 5, Johnson and Calder 1973; 6, Haines 1976; 7, Chmura et al., 1987 ; 8, Wang et al., 2003; 9, Middelburg et al., 1997; 10, Canuel et al., 1997; 11, Bull et al., 1999; 12, Kennedy et al., 2004; 13, Papadimitriou et al., 2005; 14, Gacia et al., 2002; 15, Gonneea et al., 2004; 16, Thimdee et al., 2003. [1]The values were not measured in the study and were taken from published values in the literature. [2]Averaged values of leaf, root, rhizome and litter tissue or across sites to obtain one stable isotope value. [3]Average or range of entire study because the authors did not provide the specific values for each site. [4]Averaged values of e ach 2 cm section in the top 10 cm of sediment. [5]Average of several species. [6]Range taken from a graph. NR = not reported

SEDIMENT ORGANIC CARBON STORAGE IN A CONSTRUCTED MANGROVE AND SEAGRASS SYSTEM

INTRODUCTION

Coastal ecosystems such as salt marshes, mangrove forests, and seagrass beds are being degraded and lost worldwide as a result of the eutrophication, sedimentation, and destruction that accompany coastal development for human habitation, agriculture, and aquaculture (Valiela et al., 2001; Kennish 2002; Zedler 2004). In the United States development and infilling are the main causes of coastal ecosystem loss (Dahl, 1990). In the last two decades, humans have caused the loss of 18% of the known worldwide area of seagrass beds, and in the last five decades, have caused the loss of about 35% of the world's mangrove forests (Valiela et al., 2001; Alongi 2002). In the United States, about 50% of salt marshes have been lost historically (Kennish 2001) and 25% of mangrove forests have been lost since the 1950's (Bridgham et al., 2006). United States seagrass beds had a relatively constant area between 1986 and 1997 in, what is to our knowledge, the only nationwide seagrass inventory (Dahl 1990). Smaller scale studies, however, have demonstrated local declines in the extent of seagrass (Zieman et al., 1999; Short et al., 2006). When coastal systems are lost, we lose not only wildlife habitat, storm surge protection, and economically-important fish and shellfish nurseries, but also biogeochemical functions like phosphorus retention, denitrification, and carbon (C) sequestration (Alongi 2002; Duarte 2002; Zedler and Kercher 2005).

The United States has a policy of no net wetland loss that includes coastal wetlands as part of the Clean Water Act (Zedler 2004; Zedler and Kercher 2005). Florida policy applies this no-net-loss principle to seagrass beds as well (Florida Administrative Code, Chapter 18-21). Destruction of mangrove and seagrass ecosystems in Florida requires compensatory mitigation via restoration of an existing ecosystem or construction of a new ecosystem. Mitigation can result in the replacement of fully functioning ecosystems with ineffective surrogates that do not provide the same functional value (Zedler 2004). Success of most mitigation projects is judged on the survival of macrophytes, not on proper functioning of the ecosystem. With the majority of ecosystem functions are not assessed, the true success of mitigation projects is usually unknown.

One major function of coastal ecosystems is C sequestration. The value of this ecosystem function is increasing with mounting concern about climate change. Anthropogenic release of greenhouse gases like carbon dioxide (CO_2) and methane (CH_4) through fossil fuel burning and deforestation, and livestock production, respectively, is the major cause of global climate change (IPCC 2001). Coastal ecosystems dominated by macrophytes including salt marshes, seagrass beds, and mangrove forests are high productive habitats that act as sinks for CO_2 and therefore mitigate climate change. Worldwide, salt marshes and mangroves store at least 44.6 Pg C in their sediments (Chmura et al., 2003). Seagrass beds, which make up only 0.15% of the global marine area, account for 15% of the global marine organic C (OC) storage (Hemminga and Duarte 2000). Global rates of C sequestration in vegetated marine sediments are estimated between 111 and 216 Tg C y^{-1} (Duarte et al., 2005). Based on the low estimate, globally mangroves bury 23.6 Tg C y^{-1}, salt marshes bury 60.4 Tg C y^{-1}, and seagrass bury 27.4 Tg C y^{-1} (Duarte et al., 2005). In the United States, salt marshes store 400 Tg C and sequester 4.4 Tg C y^{-1},

and mangroves store 61 Tg C and sequester 0.5 Tg C y^{-1} (Bridgham et al., 2006); the C stored and sequestered by seagrass systems is unknown. Coastal ecosystems also export C to the oceans where another portion is buried (Duarte et al., 2005).

The capacity of coastal ecosystems to sequester C, like freshwater wetlands, is greater than the capacity of uplands. These "wetlands" are a natural C sink, while upland systems eventually reach an equilibrium where amount of C fixed equals the amount respired annually, if disturbances like fire do not cause a loss of C first (Rabenhorst 1995). Constant accumulation of C in wet ecosystems is due to their anaerobic sediments where alternate electron acceptors, which are not as efficient as oxygen, must be utilized to decompose C. The capacity of coastal ecosystems to sequester C is also greater than that of freshwater wetlands (Bridgham et al., 2006). Bridgham et al., (2006) found that estuarine wetlands sequestered C 10 times faster on an aerial basis than other wetlands. These high rates are due to estuarine wetlands' high sedimentation rates, high percent soil C, and burial due to sea level rise (Connor et al., 2001; Bridgham et al., 2006). Coastal ecosystems have another advantage over freshwater wetlands. They have lower rates of methanogenesis, so the C they store is not being converted to CH_4, a more potent greenhouse gas than CO_2. In the United States, freshwater mineral wetlands emit 2.4 Tg CH_4 y^{-1} while salt marshes and mangroves emit only 0.027 and 0.004 Tg CH_4 y^{-1}, respectively (Bridgham et al., 2006).

When these coastal ecosystems are impacted, a portion of the biosphere's C storage and sequestration capacity is lost, which may exacerbate climate change by causing more CO_2 to be in the atmosphere than would be if these systems were intact. Loss of vegetated coastal ecosystems has caused at least a 25% decrease in their global C sequestration capacity (Duarte et al., 2005). Bridgham et al., (2006) estimated that losses of salt marshes and mangroves in the conterminous United States have caused a net flux of 402 Tg C y^{-1} into the atmosphere.

The upside is that restoration and construction of coastal systems may help mitigate the effects of climate change by increasing C sequestration. For example, if all dyked salt marshes in Canada were restored, an additional 2.4 to 3.6 x 10^{11} g C y^{-1} would be sequestered, which would contribute 5% to Canada's CO_2 emissions reduction identified in the Kyoto Protocol (Connor et al., 2001). It is therefore important to know if restoration and construction of coastal systems returns the C accumulation and storage capacity of these C sinks. Such research can indicate whether mitigation is effective and if coastal wetland restoration can become a policy tool for reducing CO_2 emissions as was suggested by Connor et al., (2001). Studies that focus on functional trajectories of OC in restored/constructed systems and compare OC between restored/constructed and natural systems help answer these questions.

Functional trajectories are used to track the progress of constructed systems over time and to compare constructed and reference systems (Simenstad and Thom 1996; Zedler and Callaway 1999; Morgan and Short 2002). Functional trajectory studies examine many "ecological attributes" that act as indicators of more complex ecosystem functions (Simenstad and Thom 1996; Craft et al., 2003). Attributes reach functional equivalence when they have a value similar to the reference. Functions can follow linear, asymptotic, and sigmoidal trajectories (Kentula et al., 1992) or no trajectory at all (Zedler and Calloway

1999). Craft et al., (2003) proposed that different attributes follow one of three trajectories depending on whether they are part of hydrologic, biological, or "soil" development processes. OC pool formation is a soil development process, and soil development processes generally follow the longest trajectory before reaching functional equivalence (Craft et al., 2003). There have been many studies documenting functional trajectories of sediment OC (SOC) or organic matter (OM) in restored and constructed tidal marshes (Simenstad and Thom 1996; Craft 2001; Havens et al., 2002; Morgan and Short 2002; Craft et al., 2003) but, to our knowledge, only one in seagrass beds (Evans and Short 2005) and only a comparison study in mangrove forests (McKee and Faulkner 2000).

Given the limited scope of these studies, many questions remain unexplored. First, the majority of studies on restored coastal systems have been performed in temperate salt and brackish marshes. Second, these studies only measured SOC or sediment OM as one of a suite of variables and did not deeply examine various SOC pools or characteristics. Third, these studies only examined long term trends and not short term changes that may occur immediately following construction of an ecosystem.

Whether constructed mangrove and seagrass ecosystems provide the same ecological services as their natural counterparts with respect to the C sink, and if the restoration of this service follows a functional trajectory is currently unknown. In this study, OC storage in a constructed seagrass and mangrove system in the Indian River Lagoon, FL was examined and its OC storage functioning was compared with the functioning of adjacent mature systems. Specific objectives were to: 1) determine whether extractable OC, microbial biomass C, total OC pools, and OC lability follow a short term trajectory in sediments of a constructed mangrove forest and seagrass bed and 2) evaluate whether the constructed system has reached functional equivalence by comparing SOC between constructed and natural systems. We hypothesized that, in the short term, SOC storage would increase in the constructed system but would not reach the level of SOC storage in natural systems.

METHODS

Study Site

SL-15 (Figure 60) is a mitigation site located in the Indian River Lagoon (IRL) adjacent to Fort Pierce, Florida. It is one of many spoil islands created in the IRL during the construction of the Atlantic Intracoastal Waterway that sit several meters above sea level. These islands are populated by many exotics, such as Australian Pine (*Casuvina casuarina*) and Brazilian Pepper (*Schinus terebinthifolius*), in their interiors and by native red, black, and white mangroves (*Rhizophora mangle*, *Avicennia germinans*, and *Laguncularia racemosa*) around their margins. To mitigate destruction of a nearby mangrove forest and seagrass bed, seagrass and mangrove systems were created on SL-15. These systems were created by burning and removing interior vegetation and removing dredge spoil to create several different elevations. The seagrass bed, which remains submerged during low tide, is at the lowest elevation, the mangrove forest, which is exposed at low tide, is at the middle elevation, and a maritime forest occurs above sea level at the highest elevation. The mangrove fringe of SL-15 was left intact except for a few flushing channels. Between the constructed seagrass and mangrove systems a thin *Spartina alterniflora*

buffer was planted. The mangrove forest was planted with *R. mangle*, and maritime forests were planted with *Coccoloba uvifera*, *Borrichia frutescens*, *Rapanea guinensis*, *Conocarpus erectus*, and *Distichlis spicata*, but seagrass were left to colonize naturally. Natural systems near SL-15 include its original mangrove forest fringe, surrounding seagrass beds, and mangrove fringes of adjacent spoil islands, which are at least 40 years old.

Sediment Sampling

Four, 2 m x 2 m plots were established in the mangrove forest and in the seagrass bed on SL-15 (Figure 60). Three, 7 cm in diameter sediment cores from each of these plots were retrieved in November 2005, January (mangrove only), February (seagrass only), May, July, and November 2006. Cores were taken from different areas of the plots each time to ensure an area was not re-sampled. For references, three randomly-selected plots were established in natural mangrove forests and seagrass beds within 1 km of SL-15. These plots were sampled in July and November 2006 using the same procedure as for SL-15 plots. Sediment cores were sectioned in the field and stored in plastic bags on ice for transport and then in a 4ºC refrigerator. SL-15 cores were initially divided into 0-5 cm and 5-10 cm sediment depths. In subsequent samplings, material had accumulated on top of the seagrass bed, which was collected and analyzed separately from the original sediment depths as an accreted layer. Surface layers—floc from seagrass systems, algal mats from the SL-15 mangrove system, and litter layers from the reference mangrove system—were collected from each core and were composited for each plot. Differences in color and texture were used to separate accreted and surface layers from original depths except for floc, which was the fraction of the accreted layer that poured off (Figure 61). Average heights of accreted and surface layers were measured for bulk density calculations. One core per plot was retrieved in September 2006 and brought intact to the laboratory for pH and Eh (redox potential) measurements.

Laboratory Analyses

Sectioned sediments and surface layers were weighed to determine bulk density. Rocks, roots, and detritus were removed from the sample before homogenization, and the volume and weight of large rocks were taken into account when calculating bulk density. After homogenization of each sample, a subsample was weighed to determine moisture content and the remaining sample was split into two parts. One part (wet sample) was stored in airtight containers at 4ºC and the other was freeze-dried for 48 hours. Moisture content was determined after subsamples were dried at 105ºC for 24 hours.

Intact cores from September 2006 were incubated upright in tanks filled with 25 ppt saltwater made with Instant Ocean (Marineland Labs, Moorpark, CA). Platinum electrodes were inserted into each core at 2.5 cm, at 7.5 cm, at 12.5 cm (reference seagrass only), and halfway through the accreted layer (SL-15 seagrass only). Platinum electrodes stabilized for 24 hours, and then Eh was measured using an Accumet AP71 handheld meter and an Accumet calomel reference electrode. Eh values were corrected relative to a standard hydrogen electrode. Cores were then sectioned into 0-5 cm, 5-10 cm, and 10-15 cm or accreted depths as previously described. pH was measured on 5 g of each depth using a Fisher Accumet AR50 pH meter.

Total OC (TOC) and total nitrogen (TN) were measured on freeze-dried sediment and surface layer samples. Freeze-dried samples were composited by plot and sieved through a 1 mm mesh screen to remove large shell pieces and carbonate rock, which were weighed so their mass could be accounted for in calculations. Samples were then ball-milled to a fine powder in stainless steel canisters. Inorganic C (IC) in was removed from samples via vapor acidification (Hedges and Stern 1984; Harris et al., 2001). Samples were weighed out into 9 x 5 mm or 10 x 10 mm silver capsules (Thermo Scientific, Waltham MA and CE Elantech, Lakewood, NJ), moistened with deionized water, and placed in an airtight container with a beaker of concentrated HCl (12 *M*) for 24 hours before being dried at 60ºC for 24 hours. Samples were then rolled and analyzed for OC on an elemental analyzer (ECS 4010, Costech Analytical Technologies, Valencia, CA). Peach leaves (NIST 1547) were used for calibration standards, and sucrose and an internal soil standard were used for quality control. Tests were run on sand samples with various carbonate percentages and total weights to assess the efficacy of vapor acidification and to determine the maximum sample mass that still ensured complete removal of IC. Furthermore, concurrent measures of ^{13}C were used to confirm complete removal of IC, and if incomplete IC removal was suspected, samples were rerun at a lower total mass. Unacidified samples were run separately in tin capsules (Costech) on a Flash EA 1112 series elemental analyzer (Thermo Scientific, Waltham, MA) for TN. Acetinilide was used for calibration standards, and peach leaves (NIST 1547) and an internal soil standard were used for quality control.

Extractable organic C (ExOC) and microbial biomass C (MBC) were measured using a modified fumigation-extraction procedure (Vance et al., 1987; Joergensen and Mueller, 1995). Approximately 5 g of moist sample was weighed out in duplicate for sediment, algal mat, and litter samples and 10 g was weighed out for floc samples. One set of samples was immediately extracted with 25 mL of 0.5 M K_2SO_4 for an hour and then filtered through a Whatman 42 filter. The second set was fumigated in an ethanol free-chloroform atmosphere for 24 hours before being extracted as above. Extracts were diluted, acidified, and run for OC on a Shimadzu TOC-5050A (Shimadzu North America, Columbia, MD). OC in non-fumigated samples was ExOC. The difference between OC in fumigated and non-fumigated samples, multiplied by a correction factor of 2.22 (Wu et al., 1990; Joergensen and Mueller, 1995), was MBC.

Sediment oxygen demand (SOD; APHA 1992), normalized to TOC, was used as a measure of OC lability. SOD was measured by mixing 10 mg of wet sample with about 300 mL of oxidized, salt water in dark biological oxygen demand (BOD) bottles. The salt water was created by dissolving Instant Ocean Sea Salt (Marineland Labs) into deionized water until the solution reached 25 ppt. Dissolved oxygen (DO) content of the water was measured initially and after 24 hours by a Fisher Accumet AR40 DO meter. Measurements were taken after the water and sample in each BOD bottle were thoroughly mixed on stir plates for 30 minutes. While abiotic and chemotrophic reactions can cause decreases in DO, these reactions most likely did not cause a significant O_2 reduction during this experiment because samples were already exposed to O_2 during processing. Furthermore, NH_4^+ levels in the samples were low (unpublished data) and pH did not change during incubation, which would have indicated oxidation of sulfide in the samples,. The majority of O_2 depletion was therefore assumed to be due to biological, heterotrophic oxidation of OC.

OC accumulation rates (g OC m^{-2} y^{-1}) in SL-15 were calculated using equation 3-1 (Cammen 1975; Craft et al., 1999).

$$OC_{accumulation} = \frac{OC_f - OC_i + OC_a}{A_{system}} \quad (3\text{-}1)$$

In equation 3-1, OC_f is the final amount of TOC (g OC m^{-2}) in the top 0-10 cm, OC_i is the initial amount of TOC in the top 0-10 cm, OC_a is the amount of TOC in the accreting layer, and A_{system} is the age of the system in years. Without dating sediments using ^{137}Cs, ^{210}Pb, or ^{14}C profiles, OC accumulation rates in reference systems could not be calculated.

Statistical Analyses

Repeated measures analysis of variance (ANOVAs) were run to investigate if parameters in SL-15 sediments and surface layers followed a functional trajectory over time. ANOVAs were run with a spatial power covariance structure to account for the unequal spacing between time points. Subjects were SL-15 plots and the repeated factor was month. The 0-5 and 5-10 cm depths were run together in each system in ANOVAs with depth as a main effect. Floc, algal mat, and accreted layers were run separately in ANOVAs. Replicate cores had to be averaged for each plot and month so the data fit the structure required for repeated measures analysis. A parameter followed a trajectory if its repeated measures ANOVA had a significant time effect and it demonstrated an increasing or decreasing (in the case of bulk density) trend over time. Analyses were run using the mixed procedure in SAS Version 8 (SAS Institute, Cary, NC).

Comparisons between SL-15 and reference sites were analyzed using one factorial ANOVA each for the mangrove and seagrass sediments and one factorial ANOVA each for the mangrove and seagrass surface layers. Sediment ANOVAs consisted of three fixed factors—site, month, and depth. Surface layer ANOVAs consisted of site and month factors. All two way interactions were tested. Plot data were pooled into two site treatments, SL-15 and reference. July and November 2006 were the months. For seagrass sediment analysis, SL-15 and reference depths were assigned to 3 categories in order to make comparisons: SL-15 accreted and reference 0-5 cm were depth 1, SL-15 5-10 cm and reference 0-5 cm were depth 2, and SL-15 5-10 cm and reference 10-15 cm were depth 3. Factorial ANOVAs only compare the same depths across different sites and not different depths across different sites (*i.e.*: it compares SL-15 mangrove 0-5 cm to reference 0-5 cm but not SL-15 mangrove 0-5 cm to reference 5-10 cm), so one-way ANOVAs were also run when site*depth interactions of the factorial ANOVAs did not reveal all interesting trends. Data were averaged by each site and depth over July and November samplings for these one-way ANOVAs. Factorial and one-way ANOVAs were run on JMP Version 6 (SAS Institute, Cary, NC).

For all analyses most data were transformed to meet the normality requirement (see Appendix A for details). Post hoc multiple comparisons were carried out on significant effects using the Tukey test. Significance was decided using an alpha level of 0.05.

RESULTS

Sediment Characteristics

SL-15 sediments (0-5 cm and 5-10 cm) had higher bulk densities than reference sediments (Table 1; site effect, $p<0.0001$, Table 2) as did the SL-15 mangrove algal mat. The seagrass accreted layer had a bulk density similar to the 0-5 cm depth of the seagrass reference. In seagrass sediments, bulk densities were greatest in the lowest depths, but in mangrove sediments were greater in 0-5 cm depths (Table 1; depth effect, $p<0.026$, Table 2). SL-15 seagrass sediments had orders of magnitude more shell fragments than reference sediments, while SL-15 and reference mangrove sediments had similar amounts of shell fragments (Table 1). pH in SL-15 seagrass and reference sediments and in SL-15 mangrove sediments ranged from 8.0 – 8.3. Reference mangrove sediments had a pH of 7.5 (Table 1). Redox potentials in the upper sediment depths were similar between SL-15 and reference sites, but were more negative in the lower depths of the reference sediments (Table 1).

Trajectory of Constructed System

Parameters measured in SL-15 sediments did not follow a trajectory over time, except for mangrove sediment bulk density, which significantly decreased with time (month effect, $p<0.0001$, Table 3, Figure. 62). OC parameter values seemed to shift randomly when there were significant monthly changes as for MBC in all sediments, and ExOC and TOC in seagrass 0-10 cm sediments (month effect, $p<0.021$, Table 3, Figure 63). OC parameters followed a pattern in seagrass sediments in which low values occurred in February and July while high values occurred in May and November (Figure 63). TN and C:N also changed without direction when they did change significantly (month effect, $p<0.041$, Table 3). There were no significant changes in lability for either mangrove or seagrass sediments. Significant differences between depths were few. In mangrove sediments, 0-5 cm depths had greater bulk density and TN, and in all sediments, 0-5 cm depths had greater lability (depth effect, $p<0.031$, Table 3).

SL-15 surface layers (mangrove algal mat and seagrass floc) followed a trajectory of significantly increasing MBC ($p<0.0051$, Table 3, Figure 64). Extractable OC, TOC, and TN significantly changed with time in floc, with TOC and TN generally increasing ($p<0.050$, Table 3, Figure 64). C:N significantly changed without a trend in floc ($p<0.043$, Table 3). Lability of OC in the mangrove algal mat significantly increased with time, while lability in seagrass floc significantly decreased with time ($p<0.052$, Table 2).

Constructed and Reference Comparisons

TOC was significantly higher in reference than in SL-15 mangrove and seagrass systems on both a concentration and storage basis, except in seagrass floc where TOC was similar between sites (site effect, $p<0.0005$, Table 2; Table 4). TOC differences between sites were greatest in mangrove sediments (Figure 65). On a concentration basis in seagrass sediments, sites had similar TOC in depth one, but had different TOC in depths two and three (site x depth interaction, $p=0.018$, Table 2; Figure 65b). On a storage basis, all layers had lower TOC in SL-15 so there was not a significant interaction, but a Tukey revealed layers one and three had similar TOC across sites (Figure 65b; one-way ANOVA, df=5,

p<0.0001). In seagrass sediments, TOC was greatest in depth one (depth effect, p>0.013, Table 2; Table 4).

TN was significantly higher in reference than in SL-15 mangrove and seagrass systems (site effect, p<0.0001, Table 2; Table 4), except in seagrass floc where month affected which site had higher TN (site x month interaction, p=0.041, Table 2; Table 4). C:N was significantly higher in the sediments and surface layers of mangrove references but was similar in the sediments and surface layers of seagrass sites (site effect, p<0.0097, Table 2; Table 4).

In mangrove sediments, ExOC was significantly higher in references but, in seagrass sediments, was significantly higher in SL-15 (site effect, p>0.0013, Table 2, Table 5). ExOC (storage basis) of the 0-5 depth in SL-15's mangrove system was similar to reference depths while SL-15's 5-10 depth had significantly lower ExOC (site x depth interaction, p=0.058, Table 2; Figure 66a). In the seagrass systems, ExOC (concentration basis) was similar in depths two and three across sites while depth one in SL-15 had greater ExOC than depth one in the reference (site x depth interaction, p<0.0001, Table 2; Figure 66b). On a storage basis, however, ExOC of depths two and three in SL-15 were higher than the references, but depth one had similar ExOC across sites (site x depth interaction, p=0.0017, Table 2; Figure 66b). Upper depths had significantly more ExOC in both mangrove and seagrass sediments (depth effect, p<0.0038, Table 2; Table 5). Surface layer ExOC did not significantly differ except for seagrass floc where ExOC was greater on a concentration basis in SL-15 (site effect, p=0.020, Table 2; Table 5).

MBC was significantly higher in reference sites for mangrove and seagrass sediments on a concentration and storage basis (site effect, p<0.0001, Table 2; Tables 5; Figure 67). In mangrove sediments, SL-15 0-5 cm depths had similar MBC to reference 5-10 cm depths on a storage basis (Figure 67a; one-way ANOVA, df=3, p<0.0001). On a concentration basis, depths two and three of SL-15 seagrass sediments had significantly lower MBC than those depths in reference sediments, while depth one MBC was similar across sites (Figure 67b; one-way ANOVA, df=5, p<0.0001). On a storage basis, depths two and three had similar MBC across sites, but depth one had significantly lower MBC in SL-15 (site x depth interaction, p<0.0001, Table 2; Figure 67b). MBC was significantly greater in November than in July for both mangrove and seagrass sediments (month effect, p<0.0009 Table 2; Table 5). MBC was significantly greater in upper depths of both mangrove and seagrass sediments (depth effect, p<0.0066, Table 2; Table 5 and 6). Surface layers had similar MBC to respective references (Table 2; Table 5).

SL-15 systems had significantly greater OC lability than reference systems in all sediments and surface layers except for floc (site effect, p<0.013, Table 2; Table 6). Only depth one in seagrass sediments was similar across sites (site x depth interaction, p<0.0001, Table 2; Table 6). In mangrove sediments, the 0-5 cm depth had significantly greater lability than the 5-10 cm depth while in seagrass sediments, depth two had the greatest lability (depth effect, p<0.0027, Table 2; Table 6). In mangrove surface layers, lability of the SL-15 algal mat increased while lability of reference litter decreased from July to November (site x month interaction, p<0.0001, Table 2; Table 6).

Organic Carbon Accumulation Rates

OC accumulation rates in SL-15 sediments were between 168 to 231 g OC m^{-2} y^{-1} in the seagrass sediments, but were between -119 to -148 g OC m^{-2} y^{-1} in the mangrove sediments. When algal mat accumulations were added to mangrove sediments accumulations, rates ranged from 29 to 236 g OC m^{-2} y^{-1}. Floc OC accumulations were not added to seagrass sediments due to the transient nature of floc, which is easily swept away by currents.

DISCUSSION

Sediment Characteristics

SL-15 and reference sediments are physically different from one another because SL-15 sediments' parent material is dredge spoil, as is apparent from their high amount of shell fragments (Table 1). Furthermore SL-15 sediments were compacted during construction. SL-15's accreted layer differs from other SL-15 sediments because it is a layer of post-construction deposition and was not compacted by equipment. In comparisons of constructed and reference salt marshes and mangrove forests, bulk density was almost always greater in constructed sites (Craft et al., 1999; McKee and Faulkner 2000; Craft et al., 2002). Redox potentials of all sites were negative implying anaerobic conditions and a slow rate of decomposition. Redox potentials in this study are generally more negative than those found in other mangrove (Otero et al., 2006) and seagrass sediments (Terrados et al., 1999), and sediment pH in this study are generally higher than in other mangrove sediments (McKee and Faulkner 2000; Otero et al., 2006) but similar to other seagrass sediments (Burdige and Zimmerman 2002; Daby 2003).

C:N ratios only differed between mangrove constructed and reference sediments (Table 4). Lower C:N ratios in the mangrove SL-15 sediments are due to their very low TOC. The rest of the C:N ratios are the same between SL-15 and reference sites due to similar proportions of C and N despite SL-15 having lower amounts of C and N overall. In this study, C:N ratios could therefore not be used as the ultimate metric of restoration success as was suggested for salt marshes by Craft (2001).

Trajectory of Constructed Site

In SL-15 sediments, only mangrove bulk density followed a functional trajectory in which it decreased within 2 months of construction completion but remained higher than the reference values (Figure 62). This initial decrease may have occurred as these sediments decompressed, aided by water movement into interstitial spaces, once compaction-causing construction ceased and tides could access the site. The seagrass section of SL-15 was completed a month before the rest of SL-15. Seagrass sediments therefore decompressed earlier and may have experienced a similar bulk density decrease before sampling began.

OC parameters in SL-15 sediments did not follow a trajectory, although OC pools in SL-15 seagrass sediments seemed to follow a pattern (Figure 63). External, seasonal factors, not ecosystem development, were likely the force driving these patterns. A review of physical and chemical water column data in IRL from November 2005 to November 2006 revealed potential correlations that could

explain the pattern (SFWMD 2007; station IRL 36). Lows in OC parameters corresponded with lows in salinity, highs in total Kjeldahl nitrogen, and the lowest (February) and highest (July) water temperatures of the year. Nitrogen probably did not cause these trends because N levels in the IRL are not high enough to be toxic to bacteria, but temperature or salinity may have. If the overlying water affected OC parameters in seagrass sediments, it explains why mangrove sediments, which are only in contact with water at high tide, experienced the pattern to a much lesser extent.

In both SL-15 surface layers, TOC and MBC followed a trajectory where they increased over time (Figure. 64). As a surface layer, seagrass floc is more likely to respond to water column changes than sediments. Floc TOC and MBC, however, followed a different pattern than seagrass sediments and IRL salinity and temperature. Floc OC pool increases match increases in IRL total suspended solids from February to November 2006 (SFWMD 2007; station IRL 36). Since the floc is mostly water (95%), it is likely that its solids are correlated to water column solids, which include OC substrate and microbes. Algal mat MBC increases are likely due to the algal mat's maturation as it became larger and denser throughout the year (personal observation).

Overall, during the first year following construction, with the exception of the mangrove algal mat, OC changes in SL-15 are due to seasonality and water quality. These seasonality-caused changes are large and may obscure any changes due to increasing functions. High interannual variability that mask directional changes has been observed in a restored California salt marsh (Zedler and Callaway 1999). SL-15 changes were greatest in ExOC and MBC, pools with fast turnover rates. One year may not be ample time to observe changes in more stable OC pools like TOC.

Constructed and Reference Equivalence

Organic carbon pools

A lack of trajectories does not preclude OC on SL-15 from being functionally equivalent to reference OC. Examining depths separately, 0-5 cm SL-15 mangrove sediments approached functional equivalence on a storage basis for ExOC and MBC (Figure 66). Most depths of SL-15 seagrass sediments were at or exceeded functional equivalence for all OC pools on a storage basis (Figure 66 and 67). The reason for this equivalence was bulk density. Because bulk density of SL-15 0-10 cm sediments is greater than reference sediments, when OC concentrations are multiplied by bulk density in order to be reported on a storage basis, the resulting parameters in SL-15 are often greater than or equal to the resulting parameters in reference sediments. Accreted layers were an exception because their bulk densities were the same as the references' and their heights were usually less than the references' 5 cm.

TOC equivalence did not occur on a concentration or a storage basis in the mangrove sediments but occurred for accreted and 0-5 cm depths in seagrass sediments. Accreted layers reached equivalence because the material accumulating from the water column is likely the same material being trapped by seagrass in reference sediments. It is odd, at first, that 5-10 cm depths reached equivalence before 0-5 cm depths because inputs of OC to SL-15 sediments were most likely coming the water column and benthic vegetation, which in the first year did not include deeply rooting plants. The 5-10 cm depth, however, was not completely dredge spoil. It contained mangrove clay from pre-construction mangrove

areas and a buried “A horizon” from the seagrass bed that occupied the site before spoil island creation (Fischler, 2006). These other sediments were exposed and mixed with dredge spoil during construction and had more OC than dredge spoil due to their origins in vegetated systems.

In the surface layers, the seagrass floc reached or exceeded equivalence in terms of all OC pools. SL-15 floc may have exceeded reference values due to its position inside the mangrove fringe of SL-15. In the subtidal portion of SL-15 there were areas of slower tidal flow that caused settling of water column material (Fischler, 2006), which would include OC. The algal mat reached equivalence in ExOC and MBC but not TOC. Lower TOC in the algal mat than in the litter layer is because the litter layer consisted of higher plant material like mangrove leaves and seagrass that contain more recalcitrant C than algae (Kristensen 1994). Surface layers are first to receive inputs that contribute directly and indirectly to OC pools, such as of light, water column nutrients, and detritus. Therefore, it is not surprising that most of their OC parameters would reach equivalence within the first year. Upper depths reached OC functional equivalence quickly while lower depths failed to increase over 7 years in a constructed Virginia salt marsh (Havens et al., 2002).

The majority of studies that test functional trajectories of TOC or organic matter (OM) in restored and created salt marshes do not see OC reach functional equivalence. In studies that ranged from one- to 42-year-old marshes, only two reached equivalence with their natural wetland references in terms of SOC (Simenstad and Thom 1996, Zedler and Calloway, 1999; Craft 2001; Havens et al., 2002, Morgan and Short 2002, Craft et al., 2003). They were 25 (Craft et al., 2003) and 42 (Craft 2001) years old. These authors concluded that it takes a long time for restored salt marshes to develop SOC pool equivalence and acknowledged that such equivalence may never be reached.

Predictions from salt marsh studies may be valuable for understanding trajectories of constructed mangrove forests because both are intertidal systems that take a long time to reach equivalence. Sediment OM in a 6-year and 14-year-old mangrove forests in Southwest Florida remained at 18 to 32% of reference forest values (McKee and Faulkner 2000). SL-15 mangrove sediment TOC was well below that of references. The lack of a TOC trajectory for mangrove sediments contrasts to findings in salt marsh studies and indicate that not reaching equivalence is a possibility. In all salt marsh studies except one (Simenstad and Thom 1996) a trajectory of increasing OC/OM was documented (Zedler and Calloway, 1999; Craft 2001; Craft et al., 2002; Havens et al., 2002; Morgan and Short 2002; Craft et al., 2003). Even a young constructed salt marsh in North Carolina increased its sediment TOC by over 100% in 1.3 years (Cammen 1975).

Predictions from salt marshes studies do not work for constructed and restored seagrass beds. OM content of restored sediments was higher than one reference and lower than another throughout the first 8 years in the only other known study of seagrass functional trajectories (on the New Hampshire coast, Evans and Short 2005). In SL-15 seagrass sediments, TOC was functionally equivalent in 2 out of 3 depths within a year.

There are several reasons why OC in seagrass sediments reach functional equivalence before OC in mangrove forests and in salt marshes. The first reason is elevation. In several studies of restored and

constructed salt marshes, soil development was correlated to marsh elevation so that OC/OM was higher at lower elevations (Lindau and Hossner 1981; Moy and Levin 1991; Craft et al., 2002). OC equivalence occurs faster at lower elevations because they are inundated for longer periods of time (always in the case of seagrass beds), which can create more highly reducing conditions that slow OM decomposition. More contact with water also means more contact with, and accumulation of, the dissolved organic carbon (DOC), particulate organic carbon (POC), and nutrients that water transports. Nutrients and OC stimulate bacterial production in sediments, nutrients stimulate autotrophic production of OC, and POC settles becoming part of sediment OM (Gacia et al., 2002).

The second reason seagrass sediments reach OC functional equivalence first is parent material. In most constructed salt marshes, in the SL-15 mangrove system, and in the Southwest Florida restored mangroves, the parent material was dredge spoil that is practically devoid of OM. As previously discussed, dredge spoil was not the only material found in SL-15 seagrass sediments. There was also OM-rich material originating from old vegetated sediments that were disturbed during construction, in 5-10 cm depths. At time zero OC is therefore greater in seagrass sediments. In the New Hampshire seagrass study, the sediment material was not spoil but a previously vegetated, estuarine "A horizon" that had been devoid of seagrass for 12 years (Evans and Short 2005). Like in the 5-10 cm depth of the SL-15 seagrass sediments, it is likely OC was present before restoration began.

The third reason seagrass sediments reach equivalence before mangrove and salt marsh sediments is the different OC amounts among the three coastal systems. OC content varies greatly, even among nearby reference sites (Craft et al., 1999), but generally seagrass sediments have the lowest OC and mangrove sediments the highest. Reported range in seagrass %OC is 0.15 to 1.3 (Evans and Short, 2005; Vichkovitten and Holmer 2005). Reported range in salt marsh %OC is 1.7 to 13.5 (Moy and Levin, 1991; Simenstad and Thom, 1996; Zedler and Calloway, 1999; Morgan and Short, 2002). Reported range in mangrove %OC is 2.3 to 37 (McKee and Faulkner 2000; Alongi et al., 2001; Jennerjohn and Ittekkot 2002; Alongi et al., 2004; Bouillon et al., 2004; Otero et al., 2006). The functional equivalence "bar" is therefore lowest for seagrass sediments, which was true in this study where reference sediments' mean %OC was 1.4 in mangroves and only 0.74 in seagrass. Lower than reported %OC values in this study's reference mangrove sediments are likely due to their position around spoil islands—mangrove reference sites, just as SL-15, began development in dredge spoil.

No known functional trajectory studies have measured OC pools with short turnover times like ExOC and MBC. These OC pools were the only pools to approach equivalence in mangrove sediments. Because these pools are more active (Buyanovsky et al 1994; Rochette and Gregorich 1998), they are likely to develop faster in sediments. Constructed and reference sediments in this study had MBC that was about equal to greater than MBC in a North Sea tidal flat, a Brazilian mangrove forest, and an arctic salt marsh (Joergensen and Mueller, 1995; Otero et al., 2006; Buckeridge and Jefferies 2007). Those other studies are the only known to measure MBC via fumigation extraction in a marine environment. MBC measured by fumigation-extraction has been found to correlate well with MBC measured by phospholipids fatty acid (PLFA) analysis but not by DNA analysis or substrate-induced respiration (Bailey et al., 2002; Leckie et al., 2004). A relationship between fumigated and extracted C and total PLFA concentrations has been developed by Bailey et al., (2002).

$$CFE_{flush} = 2.4(PLFA_{total}) + 46.2 \quad (3\text{-}2)$$

In equation 3-2, CFE is the uncorrected flush of OC (μg C g^{-1} soil) resulting from fumigation and $PLFA_{total}$ (nmol g^{-1} soil) is the total amount PLFA extracted from the soil. Multiplying the results by the 2.22 CFE to MBC correction factor, MBC from this study was compared to MBC in studies that used the PLFA method. Converted measurements of PLFA yielded MBC values that were the same order of magnitude as our constructed system sediments—MBC was 193 to 715 mg C kg^{-1} in a European seagrass bed (Boschker et al., 2000), 289 to 769 mg C kg^{-1} in a California salt marsh (Cordova-Kreylos et al., 2006), and 182 mg C kg^{-1} in an Australian seagrass bed (Moriarty et al., 1985). Note that this conversion equation came from sandy soils, not marine sediments, so values are not exact but are estimates for comparison purposes.

Organic carbon lability

The magnitude of OC pools is not the only factor that affects C storage, so further data exploration is needed to assess whether SL-15 stores sediment C as well as other seagrass beds and mangroves forests. SL-15 sediments must not only have OC pools equal to or greater than references to function as a significant C store, they must also have their OC stored in long term pools, where it can be sequestered away from the atmospheric C pool for decades, centuries, and even millennia. Relative amounts of the OC pool are important because the pool containing the most OC affects the overall storage abilities of a system. A system with most of its OC in non-reactive, recalcitrant pools is going to store C longer than a system with most of its OC in active pools like microbial biomass (Buyanovsky et al., 1994).

The constructed system generally stored more OC in short-term pools than references. In all sediments except constructed mangrove sediments, ExOC made up less than 1% of the TOC pool (Figure 68), but the percentage of the TOC pool made up by MBC was greater in constructed than in reference sediments. In SL-15 mangrove sediments, 53 to 63% of their TOC was MBC, while in reference sediments 11 to 15% of TOC was MBC (Figure 68). This trend was the same in mangrove surface layers. In SL-15 seagrass 0-10 cm sediments, 24 to 38% of their TOC was MBC, while in references 17 to 20% of TOC was MBC (Figure 68). SL-15 accreted layers and reference 0-5 cm depths had similar percentages that ranged from 19 to 27% (Figure 68). Sediments in this study had more TOC stored as MBC than in other coastal systems, which generally had less than 10% of their MBC as TOC (Boschker et al., 2000; Bouillon et al., 2004; Cordova-Kreylos et al., 2006). OC limitation is a possible reason for high microbial biomass. The low C:N ratios of constructed and reference sediments suggested a C limitation. When microbes are C limited they tend to sequester C in their cells instead of respiring C for energy (Anderson 2003). This mechanism is supported by another study with high MBC percentages (23 to 50% of TOC), as its sediments also had low TOC (<1.0%) (Joergensen and Mueller 1995).

Constructed sediments do not store OC as well as reference sediments because the lability of SL-15 OC was greater than references at all depths except for seagrass floc and accreted layers. Lability is a proxy for the decomposability of OC—the greater the lability, the faster OC is decomposed releasing C back to the atmosphere. It is therefore unlikely that labile OC would be stored in sediments for long periods of

time. One study of macro organic matter (MOM), a precursor of sediment OM, in constructed marshes showed that younger marshes had more labile MOM than older marshes indicating they were less likely to sequester OC in the long term (Craft et al., 2003)

Organic carbon accumulation

Rates of OC accumulation are another factor that determines how well constructed systems function as OC stores. Pool sizes measure how much C systems are keeping from the atmosphere, lability indicates how long C is likely sequestered, and accumulation rates measure how much C is being actively taken from the atmosphere (via plant production). Salt marsh studies found equal and even greater OC accumulation rates in constructed marshes (Cammen 1975; Craft et al., 1999; Craft et al., 2003). In this study, OC accumulation rates in constructed seagrass sediments were similar to those in other studies, but rates of constructed mangrove sediments were much lower than other studies unless the algal mat was included (Table 7, Figure 68). Negative rates in mangrove sediments were due to a decrease bulk density throughout the year while TOC concentrations remained constant, but if the algal mat becomes more permanent (i.e. buried) its OC will more than compensate for negative rates. Positive rates in seagrass sediments were driven by the accreting layer. It is unknown whether the accreted layer in seagrass sediments of SL-15 will continue to accumulate material at the same rates as in the first year. Continued accumulation depends on how much the accreted layer formation was due to a physical response to an uneven benthic surface after construction and how much was due to macroalgae and seagrass trapping particles from the water column.

CONCLUSION

Mangrove sediments are farther from being equivalent C stores than seagrass sediments. Mangrove sediments have only begun to reach equivalence in active pools (ExOC and MBC) and contain a relatively small amount of TOC, while seagrass sediments have equivalent TOC at most depths (Figure 68). The difference between constructed and reference OC lability is also much greater in mangrove than in seagrass sediments, and OC accumulation rates in mangrove sediments are negative (if the algal mat is excluded). However, if constructed mangrove sediments do begin to follow a functional trajectory, their potential OC storage is greater than constructed seagrass sediments because mangrove reference sediments have larger TOC pools, less OC stored as MBC (Figure 68), and lower OC lability than seagrass reference sediments. Overall, due to potential OC limitations, low TOC values for their ecosystem type, and nitrogen eutrophication (Morris and Bradley 1999; Sigua and Tweedale 2003) IRL coastal ecosystems are probably not as effective at storing C as their counterparts elsewhere.

The C storage capabilities of coastal ecosystems make them a great contender for use as C offsets. One year is not enough time to discern whether these systems will become significant C stores. More studies should investigate constructed coastal ecosystems as potential C sinks by measuring functional trajectories, OC lability, and OC accumulation rates. If constructed systems are similar to natural systems, then constructing coastal ecosystems may become an accepted way to offset CO_2 emissions, which would encourage more restoration.

Table 11. Mean (± SE) bulk density, % shell pieces, pH, and Eh (redox potential) of the sediments according to depth and site. The bulk density and % shell data were averaged over the July and November 2006 sampling periods (n=24 for SL-15 and n=18 for references). The pH and Eh data were measured in September 2006 (n=3).

System	Depth	Bulk density ($g\ cm^{-3}$)		Shells >1mm (%)		pH		Eh (mV)	
		SL-15	Reference	SL-15	Reference	SL-15	Reference	SL-15	Reference
Mangrove	Algal mat/ Litter	1.03 (0.2)	0.41 (0.1)	0	0	NA	NA	NA	NA
	0-5 cm	1.62 (0.03)	0.95 (0.04)	24 (2)	21 (5)	8.3 (0.04)	7.6 (0.07)	-71 (90)	-130 (40)
	5-10 cm	1.48 (0.03)	0.93 (0.05)	30 (2)	14 (2)	8.2 (0.03)	7.5 (0.03)	-5.7 (100)	-160 (8)
Seagrass	Floc	0.32 (0.04)	0.54 (0.09)	0	0	NA	NA	NA	NA
	Accreted	0.91 (0.05)		6.0 (1)		8.2 (0.04)		-98 (50)	
	0-5 cm	1.51 (0.04)	0.89 (0.04)	20 (3)	0.33 (0.1)	8.3 (0.04)	8.0 (0.18)	-230 (4)	-150 (30)
	5-10 cm	1.48 (0.04)	1.03 (0.03)	19 (5)	0.49 (0.2)	8.2 (0.03)	8.3 (0.07)	-180 (60)	-240 (8)
	10-15 cm		1.20 (0.02)		0.48 (0.1)		8.3 (0.14)		-320 (40)

Table 12. Results of factorial ANOVAs comparing SL-15 and references. Sediments and surface layers of the mangrove and seagrass systems were each run individually. BD=Bulk Density, ExOC=Extractable organic carbon, MBC=Microbial biomass carbon, TOC=Total organic carbon, and TN=Total nitrogen. Concentration (conc.) parameters are reported in mg kg-1 dry soil, and storage parameters are reported in g m-2.

ANOVA	Effect	BD	TOC		TN	C:N	ExOC		MBC		Lability
			(conc.)	(storage)	(conc.)		(conc.)	(storage)	(conc.)	(storage)	
Sediment											
Mangrove	Site	***	***	***	***	**	***	***	**	***	*
	Month	NS	NS	NS	NS	NS	NS	NS	***	***	NS
	Depth	*	NS	NS	NS	NS	**	**	**	***	**
	Site*Month	NS	NS	NS	NS	NS	NS	NS	*	*	NS
	Site*Depth	NS	NS	NS	*	NS	*	NS	NS	NS	NS
	Month*Depth	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
Seagrass	Site	***	**	**	***	NS	***	***	***	***	***
	Month	NS	NS	NS	**	NS	***	***	***	***	NS
	Depth	***	***	***	***	NS	***	***	***	***	***
	Site*Month	NS	NS	NS	NS	NS	NS	NS	NS	**	NS
	Site*Depth	***	*	NS	*	NS	***	**	NS	***	***
	Month*Depth	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS
Surface											

table cont.

Mangrove algae/litter	Site	*	***	***	**	**	NS	NS	NS	NS	**
	Month	NS	NS	*	NS	NS	NS	NS	NS	NS	NS
	Site*Month	NS	NS	NS	NS	*	NS	NS	NS	NS	*
Seagrass floc	Site	*	NS	NS	NS	NS	*	NS	NS	NS	NS
	Month	NS	NS	NS	NS	*	NS	NS	**	NS	NS
	Site*Month	NS	NS	NS	*	**	NS	NS	NS	NS	NS

For significance NS=not significant, * p = or <0.05, **p < 0.01, ***p < 0.0001. Please see Appendix A for a table listing how these data were transformed prior to running the factorial ANOVA.

Table 13. Results of the repeated measures ANOVAs for SL-15 mangrove and seagrass sediments (0-5 cm, 5-10 cm, and seagrass accreted) and surface layers (algal mat and floc).

ANOVA	Effect	BD ($g\ cm^{-3}$)	ExOC ($mg\ kg^{-1}$)	MBC ($mg\ kg^{-1}$)	TOC (%)	TN (%)	C:N (molar ratio)	Lability ($mg\ O_2\ g^{-1}OC\ hr^{-1}$)
Mangrove								
0-10	Month	***	NS	***	NS	*	*	NS
	Depth	**	NS	NS	NS	**	NS	*
Algal mat	Month	NS	NS	**	NS	*	NS	*
Seagrass								
0-10	Month	NS	***	***	**	NS	NS	NS
	Depth	NS	NS	NS	NS	NS	NS	*
Accreted	Month	NS	NS	*	NS	*	NS	NS
Floc	Month	*	*	***	*	*	*	*

For significance NS=not significant, * $p < 0.05$, ** $p < 0.01$, *** $p < 0.0001$. Please see Appendix A for a table listing how these data were transformed prior to running the repeated measures ANOVA.

Table 14. Mean (± SE) organic carbon concentrations (%) and storage (g m^{-2}), nitrogen concentrations, and carbon to nitrogen molar ratios of SL-15 (n=4) and reference (n=3) mangrove and seagrass sediments according to depth and month. TOC=total organic carbon and TN=total nitrogen.

Month and system	Depth	TOC (%)		TOC (g m^{-2})		TN (%)		C:N (molar ratio)	
		SL-15	Reference	SL-15	Reference	SL-15	Reference	SL-15	Reference
July mangrove	Algal mat/ litter	2.49 (0.7)	11.9 (3)	170 (20)	670 (50)	0.26 (0.07)	1.1 (0.3)	8.2 (.22)	9.3 (0.29)
	0-5 cm	0.13 (0.04)	1.3 (0.1)	110 (30)	610 (50)	0.018 (0.005)	0.096 (0.02)	5.0 (0.99)	9.6 (2.5)
	5-10 cm	0.11 (0.03)	1.4 (0.4)	77 (20)	620 (90)	0.010 (0.003)	0.11 (0.03)	8.0 (1.7)	9.2 (1.3)
Nov. mangrove	Algal mat/ litter	3.27 (0.7)	18.0 (3)	310 (60)	1300 (400)	0.37 (0.1)	0.79 (0.1)	8.1 (0.92)	21 (5.4)
	0-5 cm	0.17 (0.02)	1.3 (0.3)	140 (10)	600 (90)	0.024 (0.004)	0.12 (0.02)	5.5 (0.33)	7.0 (1.1)
	5-10 cm	0.14 (0.03)	1.7 (0.5)	110 (30)	760 (200)	0.013 (0.001)	0.14 (0.03)	6.2 (0.77)	8.8 (0.9)
July seagrass	Floc	1.8 (0.4)	2.7 (1)	60 (20)	84 (40)	0.20 (0.06)	0.39 (0.1)	8.2 (0.95)	5.7 (0.55)

table cont.

	Accreted	0.9 (0.01)		260 (50)		0.096 (0.01)		7.6 (1.1)	
	0-5 cm	2260 (0.07)	0.91 (0.6)	170 (40)	440 (20)	0.023 (0.008)	0.12 (0.01)	6.9 (0.27)	6.7 (0.16)
	5-10 cm	0.27 (0.1)	0.65 (0.8)	200 (80)	330 (20)	0.027 (0.01)	0.084 (0.01)	7.0 (0.98)	6.6 (0.19)
	10-15 cm		0.63 (1.0)		370 (40)		0.077 (0.01)		7.0 (0.21)
Nov. seagrass	Floc	5.0 (1)	3.7 (0.3)	130 (30)	96 (10)	0.62 (0.1)	0.30 (0.04)	6.8 (0.08)	10.8 (0.65)
	Accreted	1.0 (0.1)		340 (40)		0.13 (0.02)		6.1 (0.38)	
	0-5 cm	0.25 (0.05)	0.97 (0.1)	180 (30)	390 (30)	0.027 (0.006)	0.15 (0.02)	6.7 (0.76)	5.4 (0.22)
	5-10 cm	0.41 (0.1))	0.65 (0.6)	280 (60)	340 (10)	0.041 (0.01)	0.10 (0.01)	7.3 (1.1)	5.6 (0.35)
	10-15 cm		0.62 (0.3)		370 (1)		0.089 (0.009)		6.1 (0.36)

Table 15. Mean (± SE) concentration (mg kg^{-1}) and storage (g m^{-2}) of two relatively labile types of organic carbon in SL-15 (n=12) and reference (n=9) mangrove and seagrass sediments according to depth and month. ExOC=extractable organic carbon and MBC=microbial biomass carbon.

Month and system	Depth	ExOC (mg kg^{-1} dry soil)		MBC (mg kg^{-1} dry soil)		ExOC (g m^{-2})		MBC (g m^{-2})	
		SL-15	Reference	SL-15	SL-15	Reference	Reference	SL-15	Reference
July mangrove	Algal Mat/ litter	830 (200)	1800 (1000)	8500 (2000)	14000 (5000)	6.7 (2.0)	8.8 (3)	57 (5)	75 (9)
	0-5 cm	47 (4)	130 (30)	740 (20)	1900 (100)	3.8 (0.3)	6.3 (1)	60 (2)	87 (4)
	5-10 cm	38 (3)	86 (7)	690 (10)	1600 (100)	2.8 (0.3)	3.9 (0.5)	51 (2)	69 (4)
Nov. mangrove	Algal Mat/ Litter	600 (100)	750 (200)	12000 (3000)	7200 (1000)	5.7 (1.0)	4.7 (2)	110 (30)	49 (20)
	0-5 cm	52 (7)	76 (5)	900 (30)	1800 (100)	4.2 (0.5)	3.5 (0.2)	75 (4)	83 (3)
	5-10 cm	32 (3)	80 (4)	820 (20)	1800 (200)	2.4 (0.2)	3.8 (0.2)	61 (2)	80 (3)
July seagrass	Floc	100 (9)	89 (10)	18000 (100)	16000 (500)	0.33 (0.08)	0.26 (0.02)	55 (10)	48 (3)
	Accreted	77 (6)		1700 (100)		2.1 (0.1)		47 (4)	
	0-5 cm	29 (2)	53 (4)	840 (40)	2100 (100)	2.2 (0.1)	2.4 (0.1)	64 (2)	100 (4)

table cont.

	5-10 cm	24 (1)	25 (1)	800 (40)	1340 (80)	1.8 (0.1)	1.4 (0.1)	59 (2)	67 (3)
	10-15 cm		24 (3)		1100 (70)		1.2 (0.04)		67 (3)
Nov. seagrass	Floc	130 (11)	91 (6)	24000 (1100)	23000 (3000)	0.37 (0.03)	0.24 (0.0)	65 (4)	60 (9)
	Accreted	100 (6)		2300 (100)		3.4 (0.3)		75 (5)	
	0-5 cm	31 (2)	62 (5)	990 (30)	2600 (200)	2.3 (0.1)	2.5 (0.1)	74 (3)	100 (2)
	5-10 cm	36 (2)	39 (5)	1000 (40)	1400 (100)	2.6 (0.2)	2.0 (0.3)	73 (3)	71 (5)
	10-15 cm		28 (1)		1300 (40)		1.7 (0.07)		74 (2)

Table 16. Mean (± SE) organic carbon lability of organic carbon in SL-15 (n=4) and reference (n=3) sites according to depth and month.

System and month	Depth	Lability ($mg\ O_2\ g^{-1}OC\ hr^{-1}$)	
		SL-15	Reference
July mangrove	Algal mat/ litter	1120 (300)	760 (290)
	0-5 cm	1480 (580)	520 (170)
	5-10 cm	360 (120)	332 (130)
Nov. mangrove	Algal mat/ litter	2230 (540)	320 (100)
	0-5 cm	1210 (250)	355 (21)
	5-10 cm	572 (350)	198 (48)
July seagrass	Floc	631 (73)	782 (320)
	Accreted	387 (45)	
	0-5 cm	1170 (120)	527 (34)
	5-10 cm	856 (120)	677 (48)
	10-15 cm		469 (29)
Nov. seagrass	Floc	333 (79)	421 (120)
	Accreted	555 (52)	
	0-5 cm	1280 (110)	492 (25)
	5-10 cm	800 (69)	625 (38)
	10-15 cm		626 (40)

Table 17. Organic carbon accumulation rates in mangrove and seagrass systems in this and other studies.

System	Rate ($g\ OC\ m^{-2}\ y^{-1}$)	Location and remarks	Source[a]
Seagrass	195	Florida, USA	This study
	40-65	Mexico	1
	19-191	Spain	2
	182	Spain	3
Mangrove	-189	Florida, USA; sediment of 1-year-old planted system	This study
	120	Florida, USA; above system with algal mat included	This study
	180	Australia	4
	168-841	China	5
	105-159	Florida, USA	6
	191-328[1]	Florida, USA	7
	101-127	Malaysia	8
	33-104	Mexico	1
	184-281	Thailand	9

[a] 1, Gonneea et al., 2004; 2, Romero et al., 1994; 3, Gacia et al 2002; 4, Brunskill et al., 2002; 5, Alongi et al., 2005; 6, Callaway et al., 1997; 7, Cahoon and Lynch 1997; 8, Alongi et al., 2004; 9, Alongi et al., 2001

[1]This author reported organic matter accumulation rates, so rates were divided by 2 to obtain these numbers.

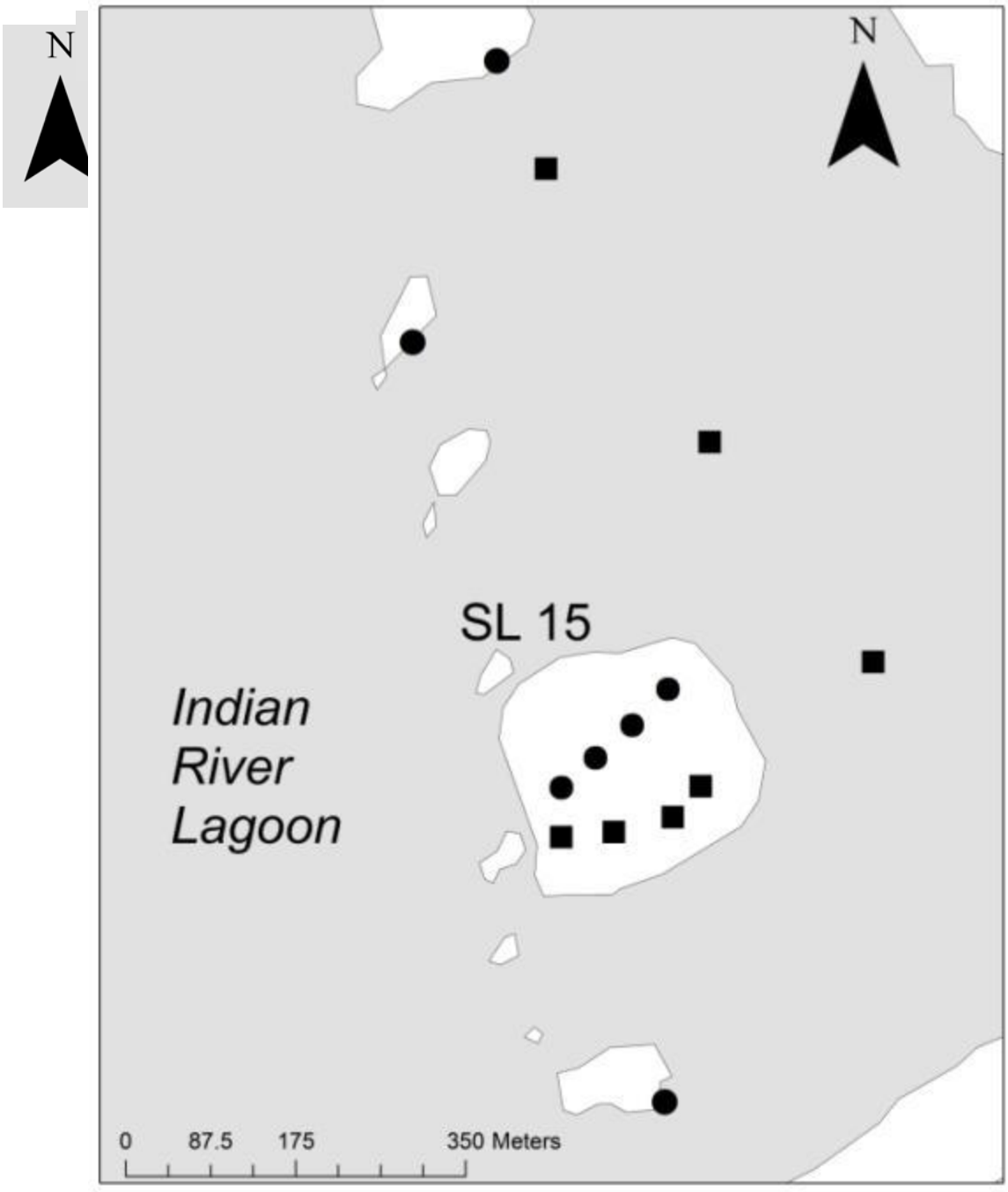


Figure 62. The study area in the Indian River Lagoon, next to Fort Pierce, Florida (inset). SL-15 is the large island in the center. Circles are mangrove system plots and squares are seagrass system plots. Symbols outside of SL-15 are the reference sites, which have one plot each.

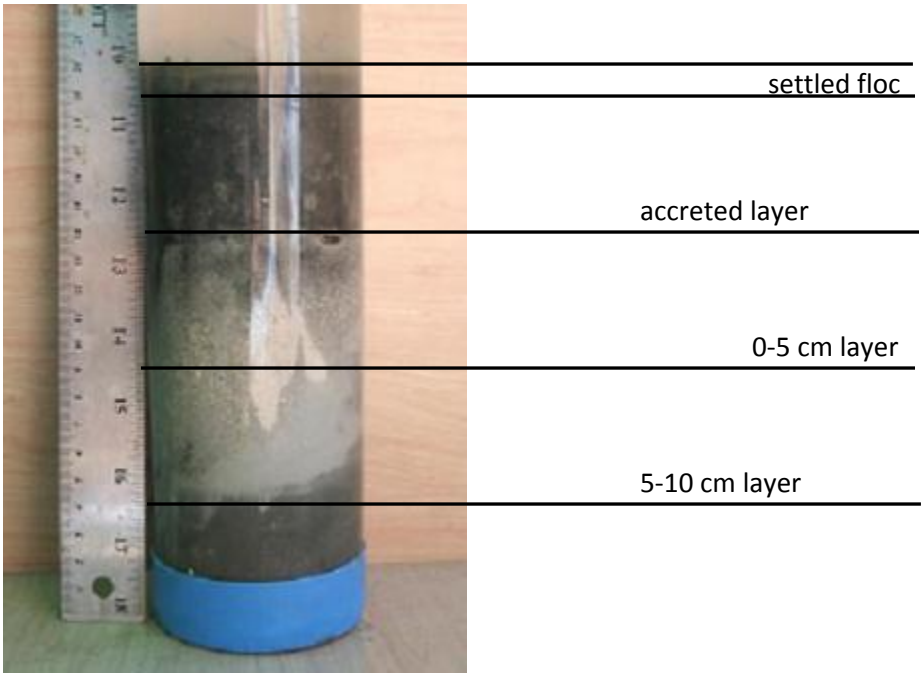


Figure 63. Core from SL-15 seagrass system illustrating the surface layer (floc) and different sediment depths (accreted layer, 0-5 cm, 5-10 cm). Note the difference in color between the accreted layer and 0-5 cm depth.

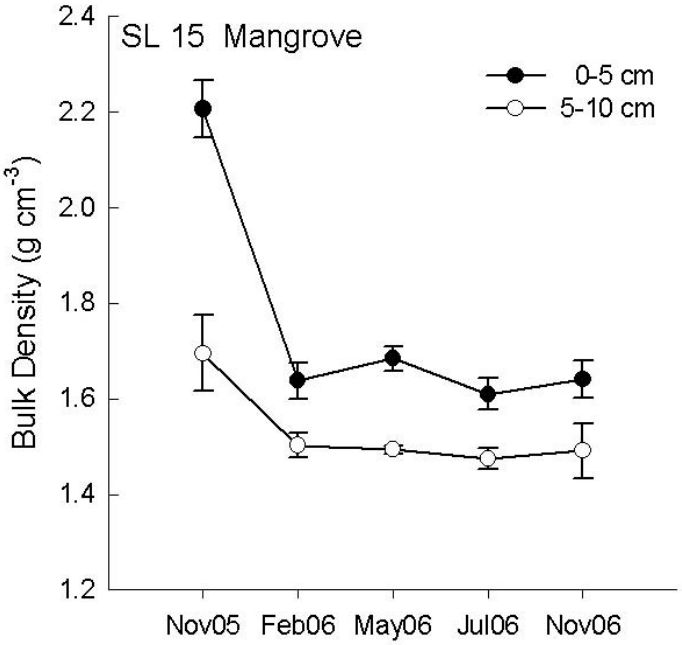


Figure 64. The functional trajectory the bulk density of SL-15 mangrove sediments followed over the first year after construction. The symbols are the mean values for each sampling date (n=12) and error bars are ± SE.

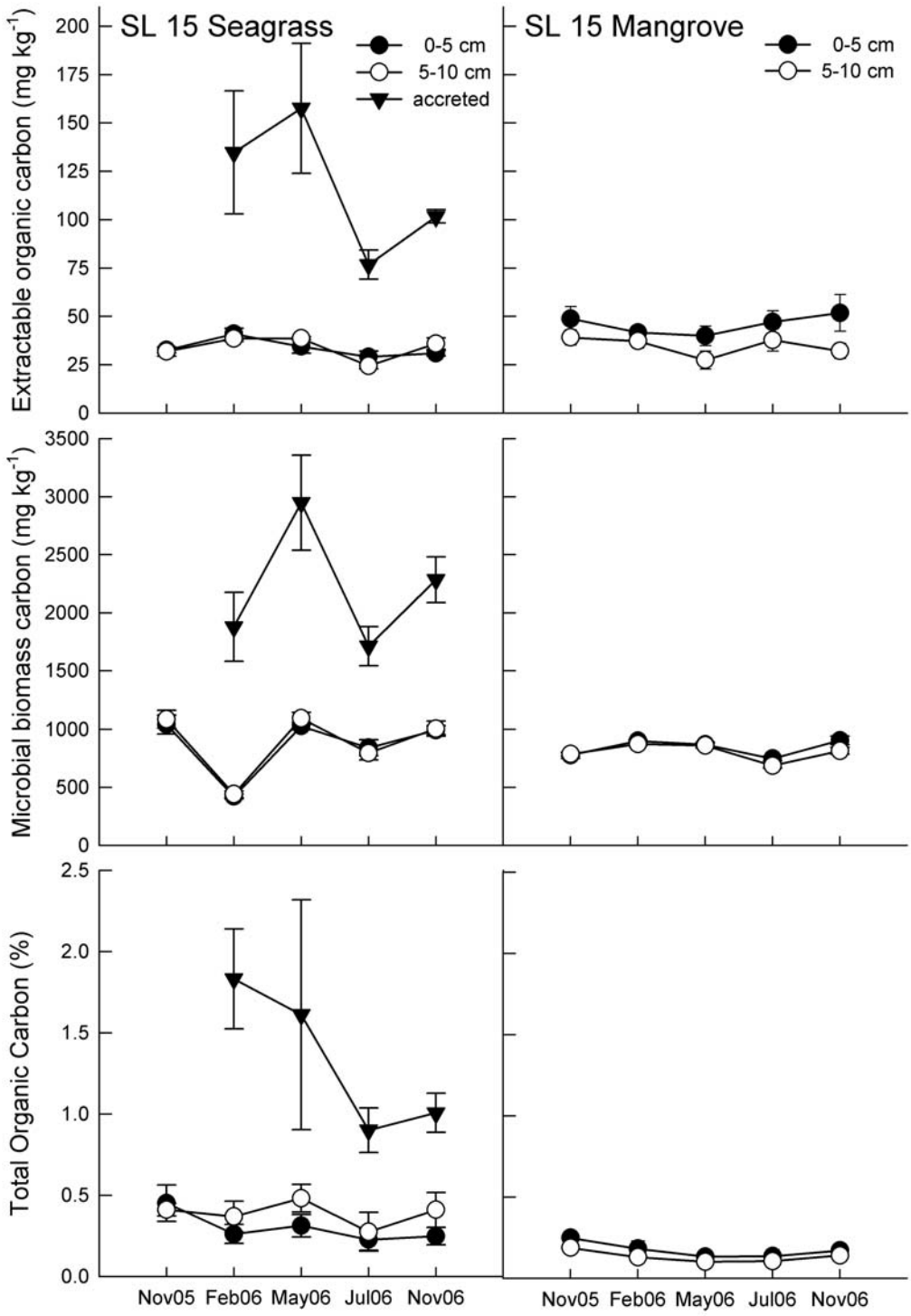


Figure 65. The changes in organic carbon parameters over the first year after construction in SL-15 seagrass and mangrove sediments. The symbols are the mean values for each sampling date (n=12 for ExOC and MBC and n=4 for TOC) and error bars are ± SE.

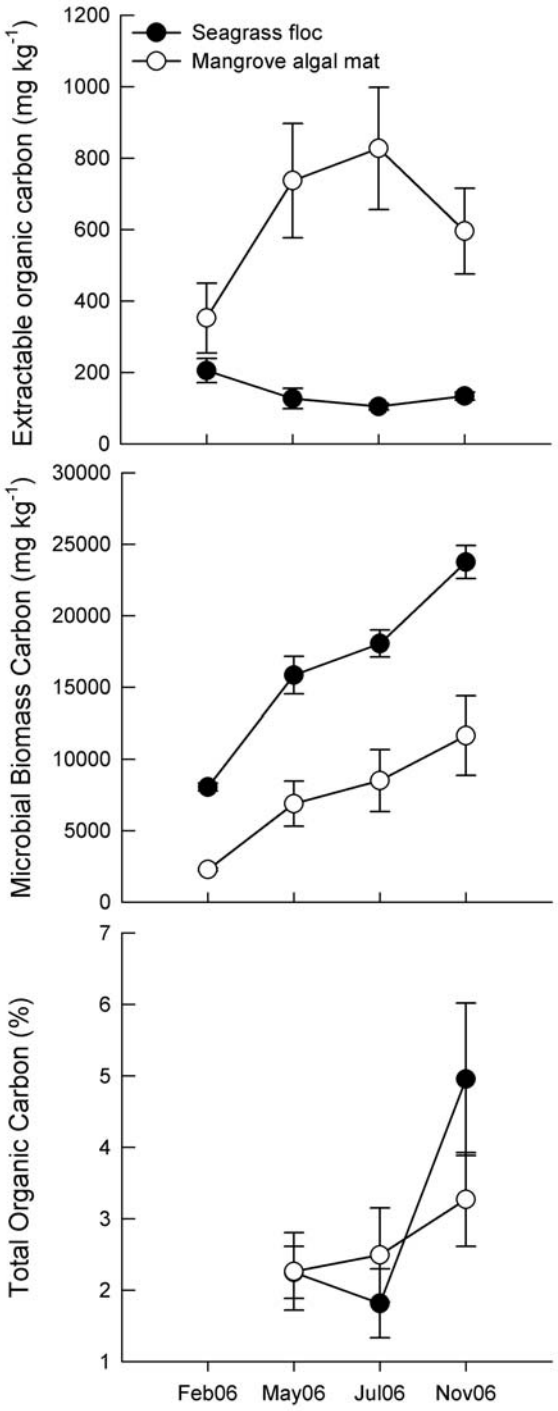


Figure 66. The changes in organic carbon parameters over the first year after construction in SL-15 seagrass and mangrove surface layers. The symbols are the mean values for each sampling date (n=4) and error bars are ± SE.

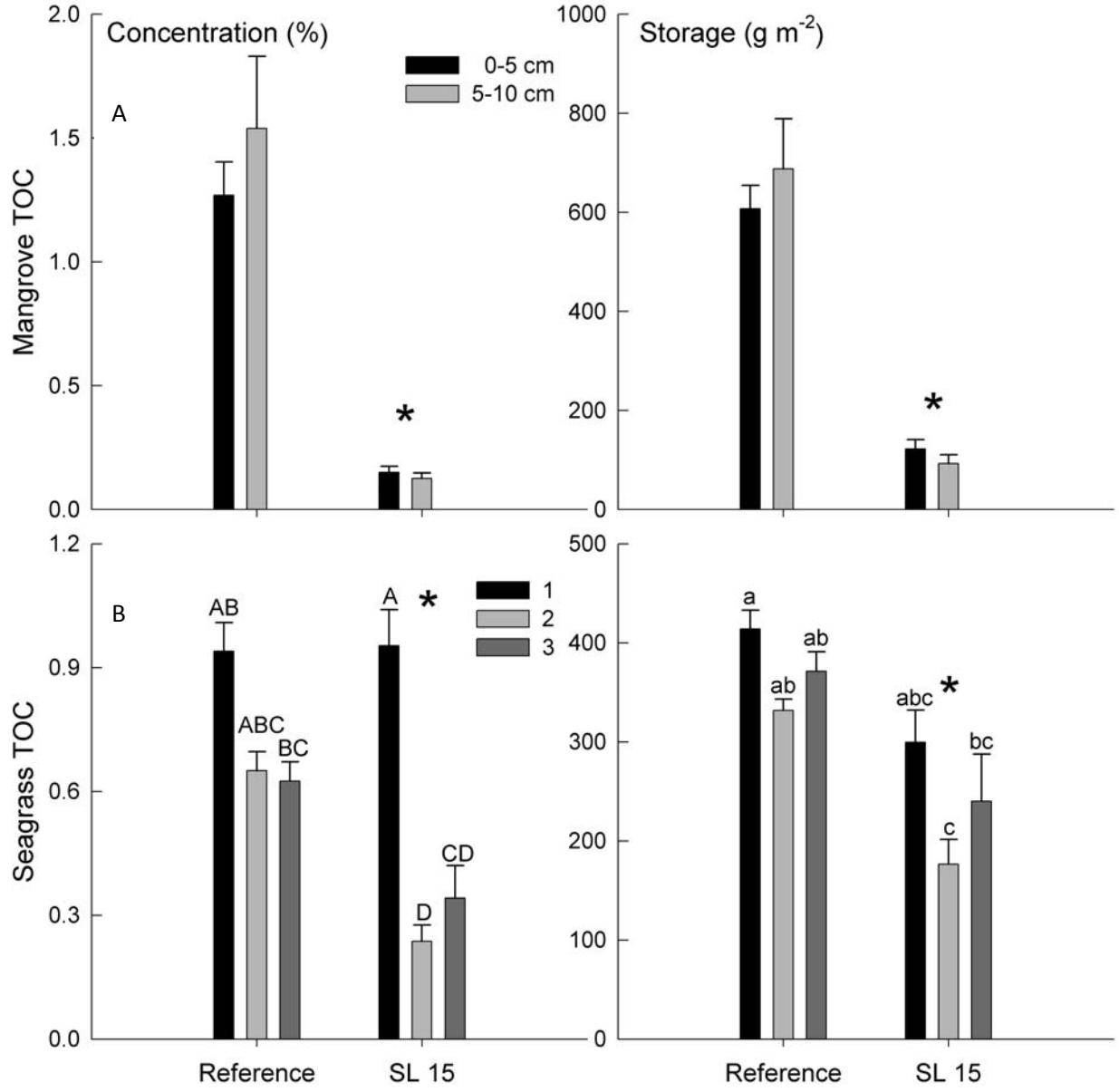


Figure 67. Comparisons between total organic carbon (TOC) in reference and SL-15 mangrove (top) and seagrass (bottom) sediments. The bars are mean TOC averaged over month (July and November 2006) for each depth of sediment (n=4 for SL-15 and n=3 for reference). Error bars are ± SE. Depths in the seagrass systems are as follows: 1= SL-15 accreted and reference 0-5, 2= SL-15 0-5 and reference 5-10, 3= SL-15 5-10 and reference 10-15. An asterisk indicates a significant site effect (Table 3-5). Capital letters are results of a Tukey test performed after a significant site x depth interaction, and lowercase letters are results of a Tukey performed after an insignificant site x depth interaction, but a significant one-way ANOVA. Bars that share letters are not significantly different.

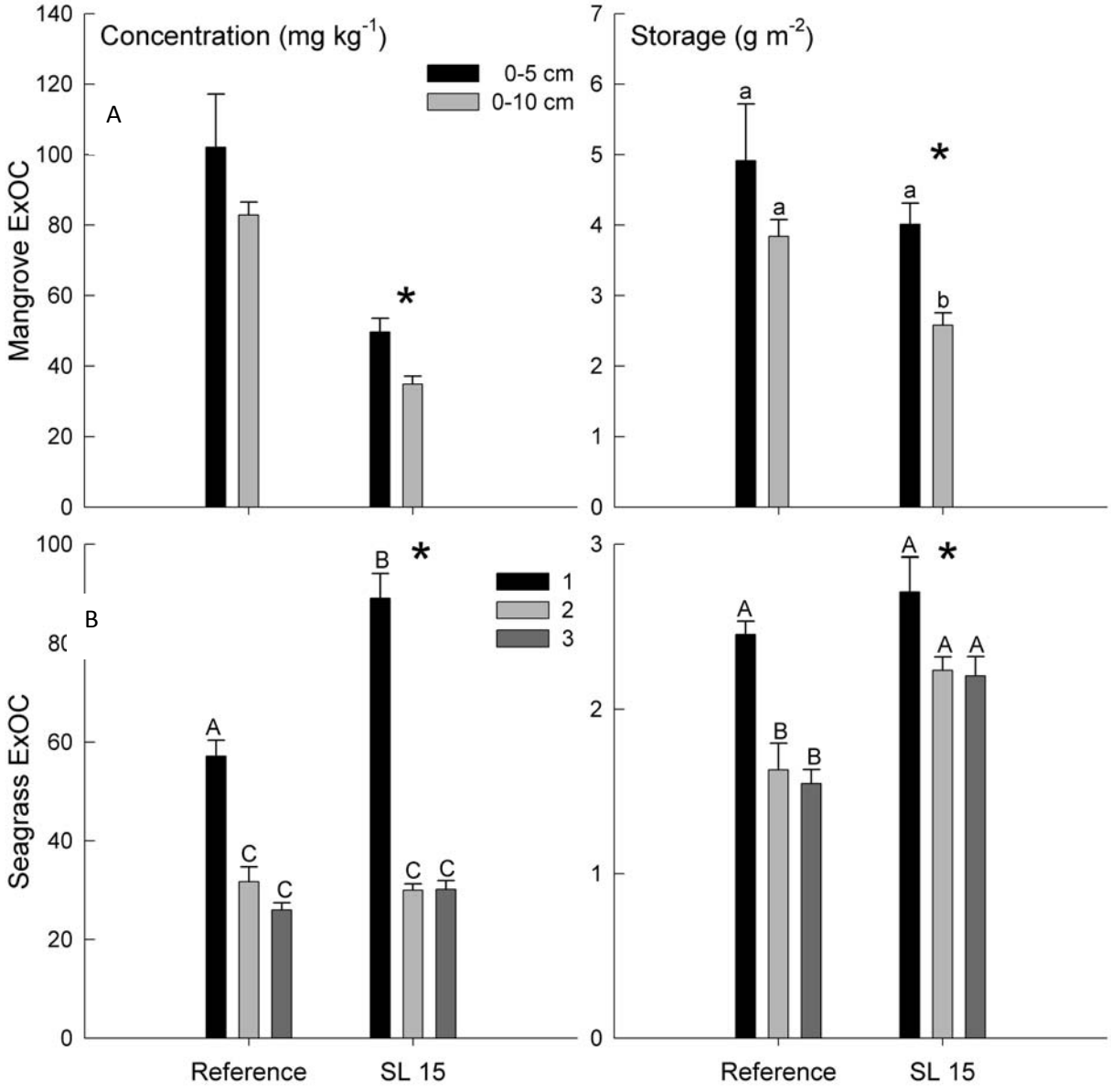


Figure 68. Comparisons between extractable organic carbon (ExOC) in reference and SL-15 mangrove (top) and seagrass (bottom) sediments. The bars are mean ExOC averaged over month (July and November 2006) for each depth of sediment (n=12 for SL-15 and n=9 for reference). Error bars are ± SE. Depths in the seagrass systems are as follows: 1= SL-15 accreted and reference 0-5, 2= SL-15 0-5 and reference 5-10, 3= SL-15 5-10 and reference 10-15. An asterisk indicates a significant site effect (Table 3-5). Capital letters are results of a Tukey test performed after a significant site x depth interaction, and lowercase letters are results of a Tukey performed after an insignificant site x depth interaction, but a significant one way ANOVA. Bars that share letters are not significantly different.

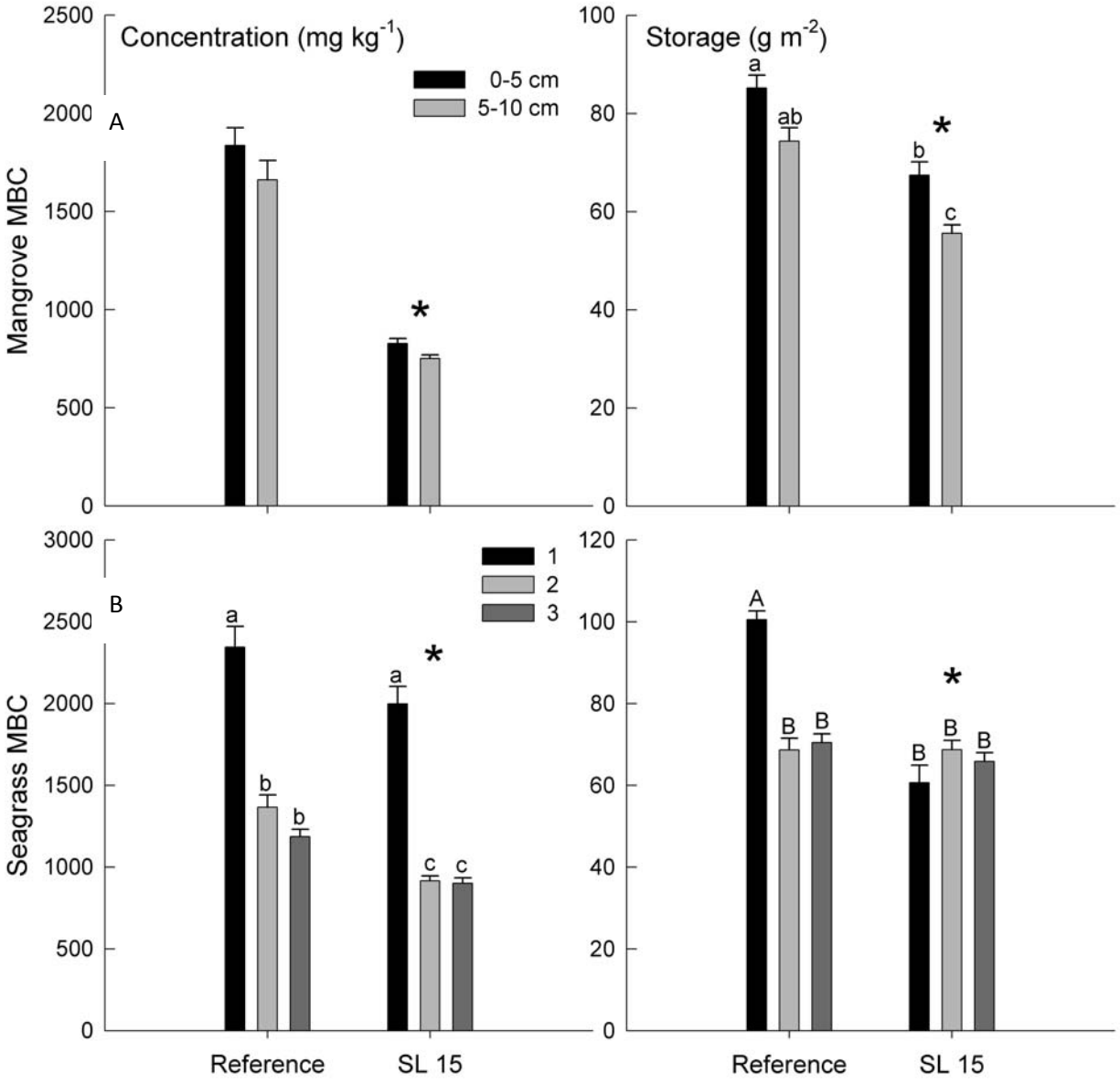


Figure 69.Comparisons between microbial biomass carbon (MBC) in reference and SL-15 mangrove (top) and seagrass (bottom) sediments. The bars are mean MBC averaged over month (July and November 2006) for each depth of sediment (n=12 for SL-15 and n=9 for reference). Error bars are ± SE. Depths in the seagrass systems are as follows: 1= SL-15 accreted and reference 0-5, 2= SL-15 0-5 and reference 5-10, 3= SL-15 5-10 and reference 10-15. An asterisk indicates a significant site effect (Table 3-5). Capital letters are results of a Tukey test performed after a significant site x depth interaction, and lowercase letters are results of a Tukey performed after an insignificant site x depth interaction, but a significant one way ANOVA. Bars that share letters are not significantly different.

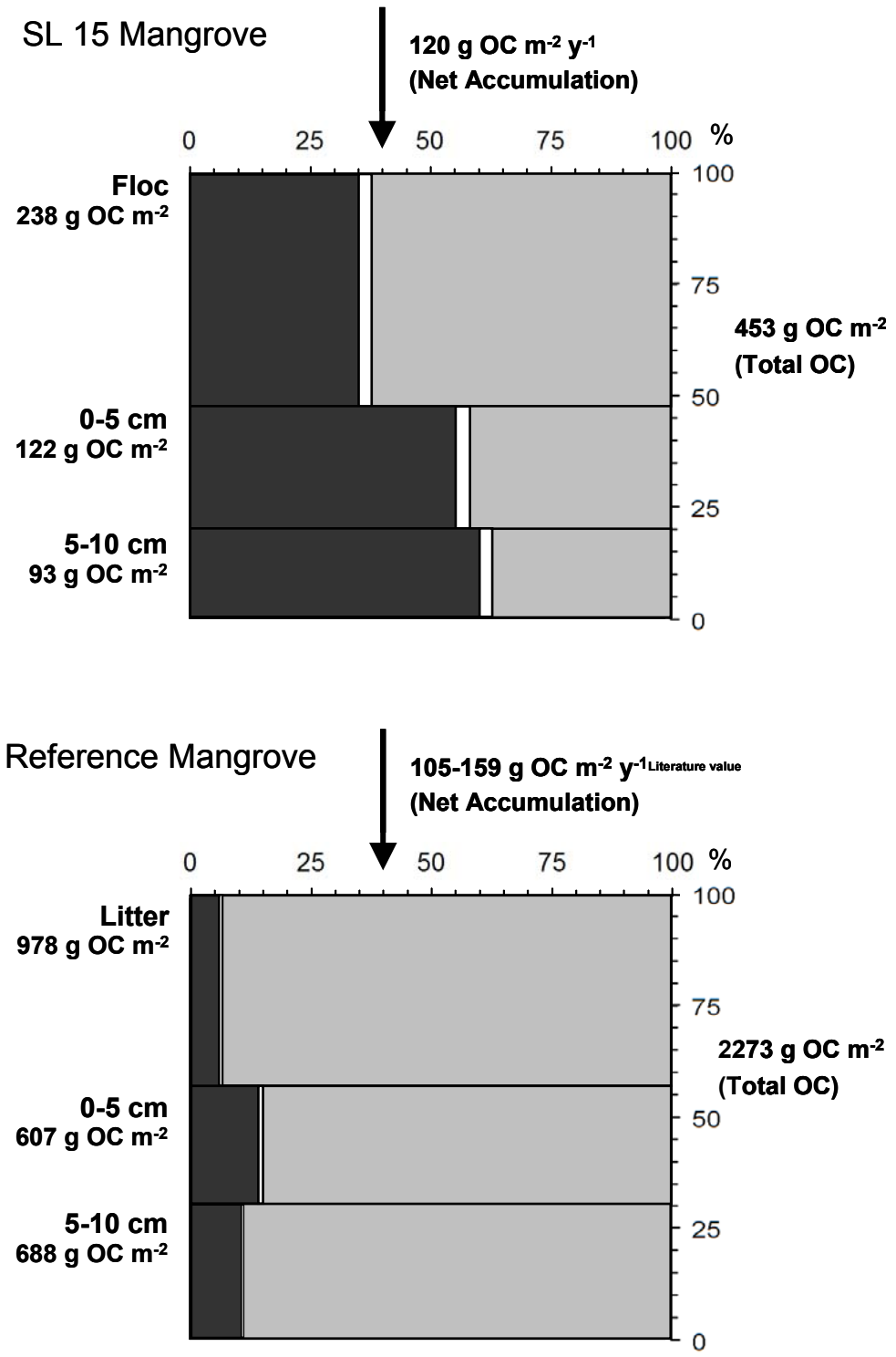
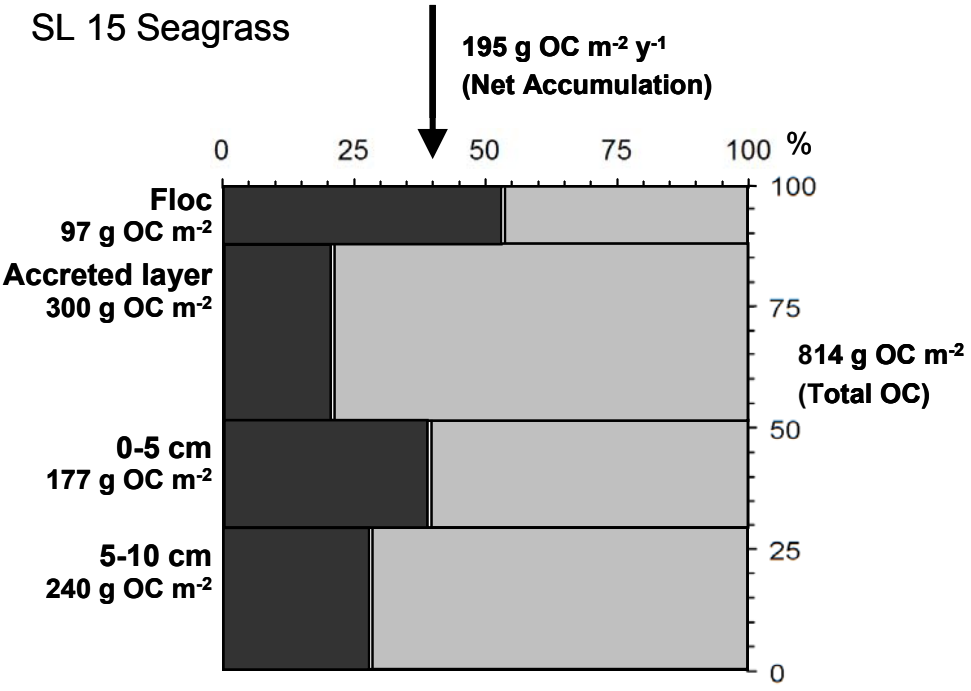


Figure 70. Organic carbon (OC) pools in SL-15 and reference mangrove and seagrass sediments. Beside each box is the total amount of OC in the depths analyzed. OC accumulation rates were calculated in this study for SL-15 sediments (includes algal mat for SL-15 mangrove) but are literature values for reference sediments (Callaway et al., 1997 for mangrove and Gonnoeea et al., 2004 for seagrass). Boxes showed the percentage distribution of the total OC in each depth and OC pool—MBC (dark grey), ExOC (white), and other (light grey).



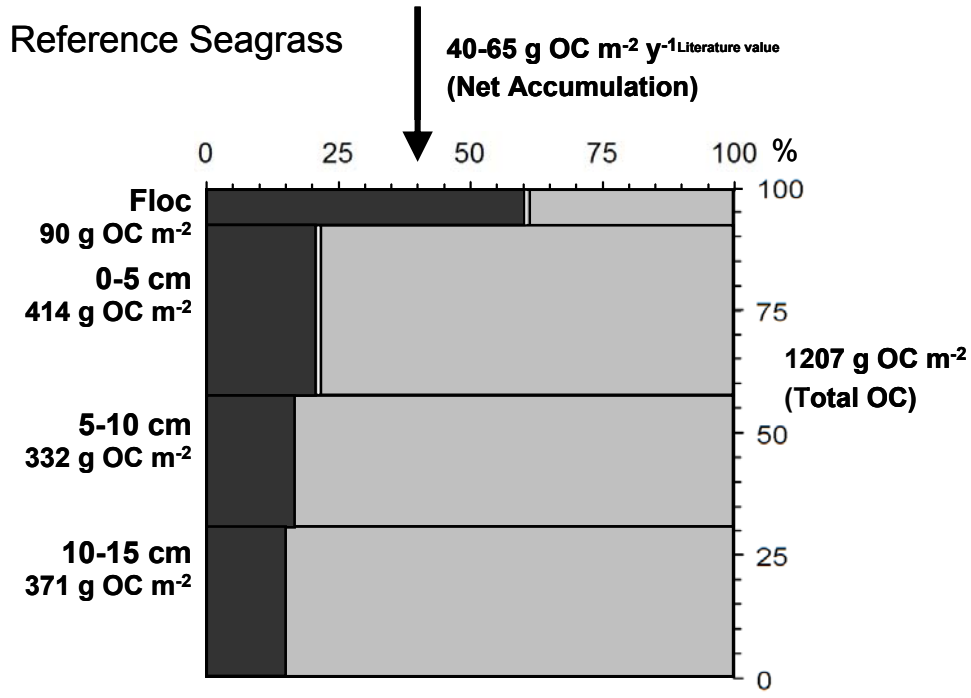


Figure 70 cont.

SOURCES OF SEDIMENT ORGANIC CARBON IN A CONSTRUCTED MANGROVE AND SEAGRASS SYSTEM

INTRODUCTION

Sediments can accumulate organic carbon (OC) from *in situ* vegetation, drift macroalgae, plankton, and water column terrestrial- and marine-derived detritus. Understanding sources of OC in soils and sediments is important to our understanding of local and global C cycles (Hedges, 1992). The source of OC influences the quality and stability of OC in sediments. OC sources, like temperature and oxygen availability, affect decomposition rates (Chapin et al., 2002), which in turn affect OC sequestration. Certain ecosystems, like macrophyte-dominated coastal systems, accumulate and store large amounts of OC in their sediments. These salt marshes, mangrove forests, and seagrass beds are sinks for CO_2 and therefore mitigate climate change by keeping C out of the atmosphere. Worldwide, salt marshes and mangroves store at least 44.6 Pg C in their sediments (Chmura et al., 2003), equivalent to 2% of the global soil C pool (Lal et al., 1995). Seagrass beds, which make up only 0.15% of global marine area, account for 15% of the global marine OC storage (Hemminga and Duarte 2000). Determining the vegetation that are the main OC sources to coastal sediments helps researchers predict how changing environmental conditions may affect the future of these significant C stores.

Coastal ecosystems are experiencing great losses worldwide (Valiela et al., 2001; Alongi 2002). The loss of vegetated coastal ecosystems has caused at least a 25% decrease in their global C sequestration capacity (Duarte et al., 2005). Constructing coastal ecosystems may restore a portion of the lost C sink (Connor et al., 2001). Knowing OC sources of constructed coastal systems can indicate whether these constructed systems can become effective at storing OC. For example, a constructed mangrove system whose principle sedimentary OC (SOC) source is relatively labile macroalgae will not store as much C for as long amount of time as a well-established mangrove system whose main OC sources are the more recalcitrant leaves and roots of mangroves.

There are a myriad of methods researchers utilize to determine OC sources. The most widely used method measures bulk stable isotopes (usually ^{13}C and ^{15}N) in possible sources and sediments. Bulk analyses measure isotopic signatures of entire OC pools in sediments or of whole plant parts. Sources are then determined by a simple comparison of source and sediment isotopic signatures (Haines 1976; Hemminga et al., 1994; Jennerjahn and Ittekkot 2002; Thimdee et al., 2003) or by mixing models (Dauby 1989; Kennedy et al., 2004; Papadimitriou et al., 2005; Zhou et al., 2006;). Other parameters are used with isotopic signatures to determine sources using ternary diagrams of N:C ratios plotted against $\delta^{13}C$ (Gonnoeea et al., 2004; Miserocchi et al., 2007) or more complex mixing models using $\delta^{13}C$ and biomass or %OC as parameters (Chmura et al., 1987; Middelburg et al., 1997; Bouillon et al., 2003;). Sources must have consistently distinct stable isotopic signatures for this method to be useful (Papadimitriou et al., 2005). Lipids are also used as biomarkers to determine OC sources (Wang et al., 2003). The lipids, generally sterols, fatty acids, or hydrocarbons, vary in specificity as some can identify groups of organisms such as vascular plants or algae while others may be specific to one genera or species (Canuel et al., 1997). Finer resolution of sources is possible when the isotopic signatures of lipids are measured in compound specific stable isotope analyses (Canuel et al., 1997; Bull et al., 1999; Hernandez et al.,

2001; Mead et al., 2005). Some lesser-used methods involve comparing relative amounts of certain OC structures in the soil, either visually as in petrographic analysis (Lallier-Verges et al., 1998; Marchand et al., 2003) or chemically as in nuclear magnetic resonance spectroscopy (Golding et al., 2004).

Stable isotopes of bulk compositions have successfully identified the main SOC sources in subtropical and tropical coastal ecosystems dominated by mangroves and seagrass because potential sources in these ecosystems have a wide range of $\delta^{13}C$ (Hemminga et al., 1994; Jennerjahn and Ittekkot 2002; Gonnoeea et al., 2004; Kennedy et al., 2004; Papadimitriou et al., 2005; Smit et al., 2005; Zhou et al., 2006). Mangroves have the most depleted $\delta^{13}C$ because Rubisco carboxylase discriminates against isotopically heavy C during C_3 photosynthesis (Hemminga and Mateo 1996; Hemminga and Duarte 2000). Seagrass have the most enriched $\delta^{13}C$, despite C_3 characteristics, because of diffusional constraints on C uptake in an aquatic environment (Hemminga and Mateo 1996). Isotopic signatures of other potential sources such as plankton and epiphytes generally fall between mangrove and seagrass values (Kennedy et al., 2004; Papadimitriou et al., 2005).

In this study, we determine: 1) significant sources to the SOC in a constructed mangrove and seagrass system, 2) how sources change over time in a constructed system, and 3) how sources differ between the constructed system and nearby mangrove and seagrass reference sediments. We hypothesized that SOC sources in the constructed system will initially be macroalgae or seston, while SOC sources in the reference systems will be vascular plants like mangroves and seagrass.

METHODS

Study Site

SL-15 (Figure 71) is a mitigation site located in the subtropical portion of the Indian River Lagoon (IRL) adjacent to Fort Pierce, Florida. The IRL is a long, shallow, and microtidal water body that lies in both temperate and subtropical climates. SL-15 is one of many spoil islands created in the Indian River Lagoon during the construction of the Atlantic Intracoastal Waterway. These islands sit several meters above sea level and are populated by many exotics, such as Australian Pine (*Casuvina casuvina*) and Brazilian Pepper (*Schinus terebinthifolius*), in their interiors and by native red, black, and white mangroves (*Rhizophora mangle*, *Avicennia germinans*, and *Laguncularia racemosa*) around their edges. To mitigate destruction of a nearby mangrove forest and seagrass bed, seagrass and mangrove systems were created on SL-15. These systems were created by burning and removing interior vegetation and removing dredge spoil down to several different elevations. The seagrass bed, which remains submerged during low tide, is at the lowest elevation, the mangrove forest, which is exposed at low tide, is at the middle elevation, and at the highest elevation, above sea level, is a maritime forest. The mangrove fringe of SL-15 was left intact except for a few flushing channels. In between the constructed seagrass and mangrove systems a thin *Spartina alterniflora* buffer was planted. The mangrove forest was planted with *R. mangle*, and maritime forests were planted with *Coccoloba uvifera*, *Borrichia frutescens*, *Rapanea guinensis*, *Conocarpus erectus*, and *Distichlis spicata*, but seagrass were left to colonize naturally. Natural systems near SL-15 include its original mangrove forest fringe, surrounding seagrass beds, and mangrove fringes of adjacent spoil islands, which are at least 40 years old.

Litter Bags

Plant material from *Syringodium filiforme*, *Thalassia testudinum*, *Halodule wrightii*, *Acanthophora spicifera*, *Sargassum* spp, *A. germinans*, and *R. mangle* were collected in July 2006. Living seagrass fronds were taken from the beds around SL-15, which is similar to the material ripped off by wind and wave events (Moore and Fairweather 2006). Clumps of live macroalgae were taken from the subtidal areas in and around SL-15. Yellow mangrove leaves, the kind about to fall, were taken from trees on the edge of SL-15 and surrounding islands. Plant material was transported back to the laboratory and rinsed. Epiphytes were removed from seagrass fronds and macroalgae. Plant material was then air dried for several weeks before being weighed by species into 2-3 g (only 1 g for *A. spicifera*) allotments and placed intact into 13 cm x 13 cm litter bags of nylon mesh with 0.5 x 0.25 mm holes.

This litter bag study is a "common garden" study where we investigated the relative decomposition rates of the potential sources to SOC, so all litter bags were placed in the same area of the SL-15 mangrove system. On September 8, 2006, litter bags were placed on the sediment surface, pinned down with metal stakes, and overlaid with large wire mesh to prevent them from washing away. Three litter bags from each plant species were randomly collected at 2, 4, 8, 16, and 32 weeks. Sediment and algae were rinsed from the litter bags in the laboratory before the bags were air dried for several weeks. Once dry, the bags were opened, and plant material in each bag was weighed.

Source Sampling

Plants were sampled on SL-15 in January, July, October, and November 2006 and at reference sites in July and November 2006. Sampled plants included all potential sources to SOC found in and around SL-15 and reference sites and fell into 3 main groups: Subtidal, which include seagrass (*S. filiforme*, *T. testudinum*, *H. wrightii*, *Halophila johnsonii*) and macroalgae (epiphytes on seagrass, *Acanthophora spicifera*, *Caulerpa sertulariodes*, *Sargassum* spp., *Ulva* spp., *Chaetomorpha linum*, *Rosenviga intricata*, *Hypnea cervicornis*, *Gracilaria tikvahiae*, and *Enteromorpha* spp.); Intertidal, which included mangroves (*Avicennia germinans*, *Rhizophora mangle*, *Laguncularia racemosa*), *Sueda linearis*, and *Spartina alterniflora*; and Terrestrial (*Schinus terebenthifolius*, *Casuarina equisetifolia*, *Coccoloba uvifera*, and *Triplasis purpurea*). Not all plants were collected at all sampling dates because some plants, particularly species of macroalgae were not present throughout the year. Vascular plant samples were a composite of 3-5 live, healthy leaves or fronds from greater than three individuals collected across the sampling area (i.e. SL-15 or reference sites). Macroalgae samples were composites of different clumps collected from across the sampling area. Epiphyte samples were composites of algal material scraped from seagrass fronds in the laboratory. Roots of seagrass and mangroves were taken from sediment cores for analysis; they were not identified to species. Roots of *A. germinans*, *R. mangle*, and *S. alterniflora* were collected in the field as well. At the laboratory, seagrass fronds were scraped clean, and seagrass, roots, and macroalgae were rinsed. All plants were dried at 60ºC for three days before being initial ground on a Wiley mill (if necessary) and then ground to a fine powder using a ball mill.

Seston was collected in May, September, October, and November 2006 and February 2007. For each seston sample, 500 mL of water was collected from the middle of the water column in the subtidal area

of SL-15. Three samples each were taken on a flood and an ebb tide except in February 2007, where only ebb tide samples were collected. Water samples were kept on ice and transported to the laboratory where they were filtered through pre-combusted Whatman GF/F glass fiber filters. Blanks of 500 mL of deionized water were also filtered for each sampling event. Filters were then freeze-dried for 24 hours.

Sediment Sampling

Four, 2 m x 2 m plots were established in the mangrove forest and in the seagrass bed on SL-15 (Figure 71). Three, 7 cm in diameter sediment cores from each of these plots were retrieved in November 2005, January (mangrove only), February (seagrass only), May, July, and November 2006. Cores were taken from different areas of the plots each time to ensure an area was not re-sampled. For references, three randomly-selected plots were established in natural mangrove forests and seagrass beds within 1 km of SL-15. These plots were sampled in July and November 2006 using the same procedure as for SL-15 plots. Sediment cores were sectioned in the field and stored in plastic bags on ice for transport and then in a 4ºC refrigerator. SL-15 cores were initially divided into 0-5 cm and 5-10 cm sediment depths. In subsequent samplings, material had accumulated on top of the seagrass section, which was collected and analyzed separately from the original sediment depths as an accreted layer. Surface layers—floc from seagrass systems, algal mats from the SL-15 mangrove system, and litter layers from the reference mangrove system—were collected from each core and were composited by plot. Differences in color and texture were used to separate accreted and surface layers from original depths except for floc, which was the fraction of the accreted layer that poured off (Figure 72).

Laboratory Analyses

Rocks, roots, and detritus were removed from each sample prior to homogenization. Samples were then freeze-dried for 48 hours. Freeze-dried sediment samples were composited by plot and sieved through a 1 mm mesh to remove large shell pieces and carbonate rock, which were weighed so their mass could be accounted for in calculations. Sediment and surface layer samples were then ball-milled to a fine powder in stainless steel canisters.

TOC, TN, and $\delta^{13}C$ were measured in sediment, surface layers, seston filters, and plant samples. TOC and TN were used to calculate C:N ratios on a molar basis. Inorganic carbon (IC) was removed from sediment, surface layer, and seston samples via vapor acidification (Hedges and Stern, 1984; Harris et al., 2001; Gonneea et al., 2004). Sediment and surface layer samples were weighed out into 9 x 5 mm or 10 x 10 mm silver capsules (Thermo Scientific, Waltham MA and CE Elantech, Lakewood, NJ), which were arranged in plastic well plates and moistened with deionized water before acidification. Three holes (7 mm in diameter) were cut from each seston filter with a hole punch and arranged in plastic well plates. The filled well plates were then placed in a glass desiccator with a beaker of concentrated HCl (12 *M*) for 24 hours before being dried at 60ºC for another 24 hours. Seston filter samples were then put into 10 x 10 mm silver capsules. Plant samples were weighed into 9 x 5 mm tin capsules (Costech Analytical Technologies, Valencia, CA). All samples were combusted on an elemental analyzer (ECS 4010, Costech) in line with an isotope ratio mass spectrometer (ThermoFinnigan MAT Delta Plus XL, Thermo Scientific,

Waltham MA) for %OC and $\delta^{13}C$. Plants were analyzed for %TN simultaneously. Peach leaves (NIST 1547) were used for EA calibration, with sucrose and an internal soil standard used as check standards. Sucrose and Peach leaves were used as internal standards for mass spectrometry measurements. C isotopes were reported in per mil notation based on deviations from the Pee Dee Dolomite standard. Tests were run on sand samples with various carbonate percentages and total weights to assess the efficacy of the vapor acidification method and determine the maximum sample mass that still ensured complete removal of IC. Furthermore, $\delta^{13}C$ values were used to confirm complete removal of IC because the presence of carbonate greatly raised $\delta^{13}C$ values. If incomplete IC removal was suspected, samples were rerun at a lower total mass. The $\delta^{13}C$ of filter blanks were accounted for in the calculation of seston $\delta^{13}C$. Unacidified sediment, surface layer, and seston samples were run separately in tin capsules (Costech) on an elemental analyzer (Flash EA 1112 Series, Thermo Scientific, Waltham, MA) for TN. Acetinilide was used for calibration standards, while peach leaves (NIST 1547) and an internal soil standard were used for quality control.

Data Analyses

Individual plant $\delta^{13}C$ and C:N were averaged across sites and sampling dates. Values of certain species were also averaged together into plant groups of seagrass, macroalgae, mangroves, or C_3 terrestrial. Differences in $\delta^{13}C$ between sampling date and tide phase (ebb or flood) were tested on seston samples using one-way analyses of variance (ANOVAs) in JMP Version 6. For all sources, C:N ratios are reported, even though N:C ratios are used in graphs, so data can be easily compared across studies. Litter mass loss for each species was modeled using a first-order exponential decay curve.

$$M_t = M_0 * e^{(-kt)} \qquad (4\text{-}1)$$

In equation 4-1, M_0 is the initial litter mass, M_t is the litter mass at time t, and k is the decay constant. The decay constant for each species was estimated using nonlinear models in JMP Version 6 (SAS Institute, Cary, NC).

To investigate whether $\delta^{13}C$, TOC, or C:N changed through time in SL-15 sediments and surface layers, repeated measures ANOVAs were run for both mangrove and seagrass areas. A spatial power covariance structure was used to account for unequal spacing between time points. Subjects were the plots on SL-15, and the repeated factor was time. For the 0-5 and 5-10 cm depths in each system, the ANOVAs were run with depth as a main effect and a time*depth interaction term. The floc, algal mat, and accreted layers were each run separately in ANOVAs where time was the only effect. These analyses were run using the mixed procedure in SAS Version 8 (SAS Institute, Cary, NC).

Comparisons between SL-15 and the reference sites were analyzed using one factorial ANOVA each for the mangrove and seagrass sediments and one factorial ANOVA each for the mangrove and seagrass surface layers (algal mat/litter and floc). Sediment ANOVAs consisted of three fixed factors—site, month, and depth. Surface layer ANOVAs consisted of only the site and month factors. All two way interactions were tested. SL-15 plot and reference site data were pooled into two site treatments, SL-15 and reference. Months used in these analyses were July and November 2006, the sampling dates for

which both SL-15 and reference data were available. For seagrass sediment analysis, SL-15 and reference depths were assigned to 3 categories in order to make comparisons: SL-15 accreted and reference 0-5 cm were depth 1, SL-15 5-10 cm and reference 0-5 cm were depth 2, and SL-15 5-10 cm and reference 10-15 cm were depth 3. Factorial ANOVAs were run on JMP Version 6 (SAS Institute, Cary, NC).

A portion of the above analyses were performed on data transformed to meet the normality requirement (see Appendix A for details). Post hoc multiple comparisons were carried out on significant effects using the Tukey test. Significance was decided using an alpha level of 0.05.

Ternary diagrams (Dittmar et al., 2001; Goni et al., 2003, Gonnoeea et al., 2004) were used to determine the main SOC sources. Because ternary diagrams can only have three end members, field observations and the position of mean sediment $\delta^{13}C$ relative to mean potential source $\delta^{13}C$ on a $\delta^{13}C$ line (Figure 73) were used to choose the three most likely end members for each constructed and reference sediment and for the mangrove litter layer and seagrass floc. N:C of the three end members and sediments were plotted against $\delta^{13}C$. N:C ratios are used instead of C:N ratios because with the larger number in the denominator, they are more statistically robust (Goni et al., 2003). End members' N:C and $\delta^{13}C$ were averaged for all sampling dates and species within that group (e.g.: mangroves), but for plants where multiple parts were measured, only leaf/frond values were used. The three end members create a triangle that is expanded according to the standard deviations of the end members to account for natural variability and analytical error. Sediment samples that fall in the middle of the triangle are assumed to be a mixture of all three sources, samples that fall along a line connecting two end-members are considered a mixture of those two sources, and samples that fall around the vertex of an end member are assumed to have OM from mainly that source. Samples that fall outside of the expanded triangle have OC contributions from additional sources or have undergone changes during diagenesis.

RESULTS

Source Characteristics

$\delta^{13}C$ and C:N varied among plant groups. Generally, the lowest $\delta^{13}C$ and greatest C:N were found in mangrove leaves and roots and C_3 terrestrial plant leaves (Table 1). The greatest $\delta^{13}C$ and a relatively low C:N were found in seagrass fronds. *S. alterniflora* had a low $\delta^{13}C$ and high C:N. Seston had low $\delta^{13}C$ and low C:N. Seston samples had greater $\delta^{13}C$ in fall than in winter (ANOVA, df=4, p=0.0002) but did not differ between ebb and flood tides (ANOVA, df=1, p=0.54). Compared to variation among plant groups, variation of $\delta^{13}C$ within plant groups was usually low with mangrove tissues of all species varying by less than 2.5‰ and seagrass tissues (except *H. johnsonii*) by less than 3.4‰. The exception was the macroalgae group, whose $\delta^{13}C$ varied by 15‰. Macroalgae had high variability with C:N ratios as well. Plant tissue type influenced C:N ratios with greater C:N in roots than in leaves for both mangroves and seagrass.

Plants also differed in their decay rates, even within groups (Table 2). The greatest decay constants, and fastest rates of decay, were for a macroalgae (*A. spicifera*) and a seagrass (*S. filiforme*). The slowest decay rates were for a seagrass (*H. wrightii*) and a mangrove (*R. mangle*).

Sediments and Surface Layers

$\delta^{13}C$ of SL-15 sediments and surface layers, with the exception of the 0-10 cm seagrass sediments, changed significantly over time (month effect, $p<0.034$, Table 3, Figure 74). Mangrove sediments and seagrass accreted layers had $\delta^{13}C$ that increased towards the mean $\delta^{13}C$ of their respective references over the first year after construction (Figure 74). The mangrove algal mat's $\delta^{13}C$ also increased but moved away from reference values (Figure 74). Most of the layers did not have changing %OC or C:N ratios throughout the year. C:N ratios changed significantly without direction in mangrove sediments and seagrass floc (Chapter 3). TOC significantly changed in seagrass 0-10 cm sediments and floc, but only with direction in floc, where it increased over time (Chapter 3). The $\delta^{13}C$, TOC, or C:N values did not differ among sediment depths (Table 4-3, Chapter 3).

SL-15 seagrass sediments had lower $\delta^{13}C$ than reference seagrass sediments (site effect, $p<0.0001$, Tables 4-3 and 4-4), but SL-15 mangrove sediments had $\delta^{13}C$ similar to reference mangrove sediments ($p=0.40$, Tables 4-3 and 4-4). SL-15 floc was more depleted than reference floc in July but more enriched than reference floc in November (month x site interaction, $p<0.0001$, Table 4-3 and 4-4). SL-15 algal mat was more enriched than reference litter in both months, but the difference was greater in November (month x site interaction, $p<0.0001$, Table 4-3 and 4-4). TOC (%) was generally lower in SL-15 sediments than references with the exception of the SL-15 seagrass accreted layer and floc, which had similar TOC to the reference's 0-5 cm depth and floc, respectively (Chapter 3). C:N ratios were similar in seagrass sediments and floc but were lower in SL-15 mangrove sediments and surface layers than in respective mangrove references (Chapter 3).

Source Determination

Putting source (plants and seston) and sediment $\delta^{13}C$ data together indicates potential sources to the various sediments and surface layers (Figure 73). Using observations from the field and Figure 73, the three ternary diagram end members for SL-15 mangrove sediment were seston, algal mat, and terrestrial plants (Figure 75a). Seston, litter, and mangroves were the end members for reference mangrove sediments (Figure 75b). Seston, seagrass, and macroalgae were the end members for SL-15 and reference seagrass sediments and floc (Figure 76 and 77b). Seston, seagrass, and mangroves were the end members for reference mangrove litter (Figure 77a). SL-15 algal mats did not need a diagram because they are their own source as primary producers. Ternary diagrams explained 74% of SL-15 mangrove sediment samples, 92% of reference mangrove sediment samples, 71% of SL-15 seagrass sediment samples, 33% of reference seagrass sediment samples, 100% of reference mangrove litter samples, and 89% of seagrass floc samples (Figure 75 through 77). All of the samples that fell outside the ternary plots, regardless of site or depth, did not fit because their N:C ratios were greater than that of the sources.

The majority of SL-15 mangrove sediment samples fell near the seston end member. Some samples fell in the middle of the triangle and others fell close to the terrestrial end member (Figure 75a). Most reference mangrove sediment samples fell between seston and litter end members (Figure 75b). In terms of $\delta^{13}C$, but not in terms of N:C, most SL-15 and reference seagrass sediment samples were within

the range of macroalgal sources (Figure7 6). SL-15 seagrass sediment samples fell far from the seagrass end member (Figure 76a). SL-15 seagrass 0-10 cm and accreted depths did not differ in their sources. Most reference seagrass samples fell outside the diagram due to high N:C ratios (Figure 76b). Examining only $\delta^{13}C$, reference seagrass sediments were more enriched than macroalgae and seston but more depleted than seagrass (Figure 73). Reference mangrove litter layer samples from July fell between seston and seagrass end members but November samples fell in the middle or at the mangrove vertex (Figure 77a). Reference seagrass floc samples fell between seston and macroalgae end members in July but outside the diagram in November (Figure 77b). SL-15 seagrass floc fell between seston and seagrass regardless of sampling data (Figure 77b.)

DISCUSSION

Source Characteristics

$\delta^{13}C$ of the main potential sources in the studied part of the Indian River Lagoon were within the range of literature from similar estuarine studies (Table 4-5). Our sources' C:N values were also within reported literature values of 30 to 99 for mangrove leaves and roots (Lallier-Verges et al., 1998; Thimdee et al., 2003; Gonnoeea et al., 2004; Muzuka and Shunula 2006), of 15 to 21 for seagrass fronds (Thimdee et al., 2003; Gonnoeea et al., 2004; Machas et al., 2006), of 5.8 to 9.3 for seston (Gonnoeea et al., 2004; Zhou et al., 2006), and of 7 to 30 for macroalgae (Kristensen 1994; Thimdee et al., 2003).

$\delta^{13}C$ of plants vary within different tissues (Vizzini et al., 2003; Papadimitriou et al., 2005;), within a single species (Hemminga and Mateo 1996), across sites (Kennedy et al., 2004), seasons (Vizzini et al., 2003), and years (Anderson and Fourqurean 2003; Fourqurean et al., 2005). Variations are most pronounced in seagrass (Thimdee et al., 2003) and macroalgae. In submerged vegetation variation is due to the relative uses of dissolved CO_2 and bicarbonate, the source of inorganic C in the water, temperature, irradiance, and subsequent photosynthesis rates (Lin et al., 1991; Hemminga and Mateo 1996). Seston $\delta^{13}C$ can also vary temporally, spatially, and between ebb and flood tides (Hemminga et al., 1994). These variations in source $\delta^{13}C$ make it necessary to measure all potential sources' $\delta^{13}C$ for each study area, instead of relying on literature values, and ideally, measure significant sources across tissues, sites, and seasons. $\delta^{13}C$ variations within individual sources and plant groups in this study were generally smaller than differences among main sources, so the variations most likely do not affect our source determinations. Furthermore, where $\delta^{13}C$ did overlap among main sources their C:N ratios set them apart, as with seston and mangroves, or they were not both end members for the same ternary diagram.

There is some concern about whether $\delta^{13}C$ of plant tissues changes during diagenesis because large changes in $\delta^{13}C$ could lead to misleading source determinations. Studies that measured fresh and senescent mangrove leaves and seagrass found small (generally >1‰) differences (Thimdee et al., 2003; Gonnoeea et al., 2004). Decomposition studies found significant but minor (0.55 to 2‰) changes in seagrass, mangrove, and macroalgae $\delta^{13}C$ during diagenesis (Fenton and Ritz 1988; Fourqurean and Schrlau, 2003), but others found no significant changes (Machas et al., 2006). Where $\delta^{13}C$ did change in decomposition studies of multiple species, the initial differences in $\delta^{13}C$ between species were still clear.

Unfortunately, we did not measure changes in $\delta^{13}C$ of our plant tissues during decomposition. Given the small magnitude of changes found in other studies, and the large differences in $\delta^{13}C$ between groups of potential sources, diagenetic changes in $\delta^{13}C$ are unlikely to cause misidentification of the main SOC sources in this study. Changes in C:N during decomposition also occur and can be greater in magnitude than $\delta^{13}C$ changes (Fourqurean and Schrlau, 2003). Studies of mangrove, seagrass, and macroalgal decomposition have found decreases and increases in C:N ratios that were dependent upon species or tissue (Twilley et al., 1986; Bourgues et al., 1996; Fourqurean and Schrlau, 2003); others found no change in C:N ratios (Machas et al., 2006).

Decay constants of seagrass on SL-15 were within literature values, which ranged from 0.002 to 0.12 day^{-1} (Mateo and Romero 1996; Machas et al., 2006; Moore and Fairweather 2006). *T. testudinum* had a greater decay constant and therefore faster decomposition in this study than in Florida bay (Fourqurean and Schrlau, 2003). Mangrove decay constants were also within literature values that ranged from 0.0048 to 0.022 day^{1} (Fourqurean and Schrlau, 2003; Ake-Castillo et al., 2006; Ramos e Silva et al., 2006). *R. mangle*'s decay constant in this study fell on the low end of *R. mangle* reported values. Estimated macroalgae decay constants ranged widely from 1 to 0.014 (Mews et al., 2006). The decay constant of *Sargassum spp.* in our study was at the low end of the range, probably because *Sargassum* has more structural components than most other macroalgae. Surprisingly, differences among decay constants in this study did not fall along plant groups. We expected mangroves to have the lowest decay constants and macroalgae to have the highest with seagrass falling in between (Kristensen 1994; Bourgues et al., 1996; Fourqurean and Schrlau, 2003). However, *S. filiforme* decomposed as fast as the macroalgae and *T. testudinum*'s decomposition was at the rate of *A. germinans*. These results indicate that in terms of decomposition, species identity matters more than the group to which a species belongs. For source determination, these results specify which species of an end member group are more likely to contribute to SOC because the slower a species decomposes, the better chance its OC will be buried in sediments.

Sediments and Surface Layers

Changes in SL-15 SOC $\delta^{13}C$ over the course of a year indicate new SOC sources are adding to the sediment TOC pool or, without a change in TOC, decomposition of old source OC while new source OC accumulates. These changes were greater in upper sections of both mangrove (0-5 cm) and seagrass (accreted layer) sediments because the inputs of new OC reach the top of sediments first. Bioturbation then brings new OC inputs deeper into the profile. Bioturbating organisms were observed in mangrove, but not in seagrass sediments, which may explain why $\delta^{13}C$ of deeper mangrove sediments changed over time but deeper seagrass sediments did not. Surface layers had the greatest $\delta^{13}C$ changes through the year.

All changes were positive so that the new SOC sources to SL-15 after construction must be more enriched than old OC sources. Old OC sources were relatively depleted in $\delta^{13}C$ as they were most likely the terrestrial plants that inhabited SL-15 pre-construction. In mangrove sediments, the new source was most likely the algal mat and in seagrass accreted layers and floc the new sources were macroalgae or seagrass (Figure 73). Enrichment of algal mat $\delta^{13}C$ is due to changing inorganic C sources, as unlike

other sediments and layers, the algal mat is its own producer of OC. As the algal mats grow, so does their influence on the biogeochemistry of their environment. Photosynthesis and respiration within the algal mat changes the pH of the water around it (Kayombo et al., 2002). At night respiration decreases the pH, which can cause $CaCO_3$ in the sediment below the algal mat to dissolve. $CaCO_3$ dissolves into various carbonate species (CO_3^{-2}, HCO_3^{-1}), which inherit the high $\delta^{13}C$ of $CaCO_3$ (≈ 0) (Lin et al., 1991). These carbonate species then may be utilized by algae as inorganic C sources during daytime photosynthesis.

$\delta^{13}C$ in the literature ranges from -29.4‰ to -20.6‰ for mangrove sediments (Bouillon et al., 2003; Thimdee et al., 2003; Gonneea et al., 2004) and from -10.3‰ to -26.6‰ for seagrass sediments (Hemminga et al., 1994; Kennedy et al., 2004; Papadimitriou et al., 2005). Sediment $\delta^{13}C$ in this study are for the most part within literature values. SL-15 and reference mangrove sediments span the range of literature values from -27.5‰ to -19.4‰. SL-15 and reference seagrass sediments are at the lower end of the literature values with $\delta^{13}C$ ranging from -23.2‰ to -19.4‰. Differences in $\delta^{13}C$ among SL-15 and reference sediments and surface layers suggested their SOC sources differ. Observations of the distribution of primary producers around the sites also suggested sources differ, even between SL-15 and reference mangrove sites, whose $\delta^{13}C$ were not significantly different.

Source Determination

The ternary diagram indicated that seston was the dominant source for SL-15 mangrove sediments with some OC being contributed by terrestrial plants and the algal mat (Figure 75a). Terrestrial sources most likely contributed to SOC before and during construction. During construction, we observed terrestrial plant parts that were not fully removed by burning and clearing being mixed into spoil within SL-15's intertidal zone. The algal mat's influence as a source was supported by $\delta^{13}C$ enrichment of mangrove sediments over the first year. Mangroves were not included as a source in the ternary diagrams because SL-15 mangroves were young (>2 years old) and mangrove litter was very sparse. Seston was also a dominant source for reference mangrove sediments according to the ternary diagram, but in this instance it shared this designation with the litter layer (Figure 75b). According to Figure 73, mangroves also contributed to SOC because mean sediment $\delta^{13}C$ was more depleted than mean seston and litter values.

Seston and macroalgae were the dominant OC sources in SL-15 seagrass sediments according to the ternary diagram (Figure 76a). Seagrass, which had colonized most of SL-15 at generally low densities by July 2006 (Fischler, 2006), were not yet important SOC sources. Seston and macroalgae as main SOC sources were further supported by observations—drift macroalgae was frequently found buried in the accreted layer section of cores throughout the study where it seemed to trap particles from the water, driving accretion. High N:C (low C:N) ratios of seagrass reference sediments interfered with determining sources via the ternary diagram (Figure 76b). Samples outside of the diagram can indicate an unknown source of SOC, but that is unlikely as almost all plants encountered were measured and none had high N:C (low C:N) ratios (Table 4-1). According to $\delta^{13}C$ only, seston and seagrass are probably both sources because reference seagrass SOC $\delta^{13}C$ falls in the middle of those end members. The contribution of macroalgae is unknown though due to its intermediate $\delta^{13}C$. Mangroves were not chosen as a potential

source for seagrass sediments as mangrove litter was observed infrequently on seagrass sediments. Therefore mangrove's influence to SOC was believed to be mediated through seston.

Sources to the reference mangrove litter layer change with season as the ternary diagram indicates that seagrass and seston are the dominant sources in July but mangroves are the dominant sources in November (Figure 77a). Conclusions from the ternary diagram match field observations. The litter layer was primarily seagrass wrack in July but was primarily partially-decomposed mangrove leaves in November. Since the litter layer is one of the main sources to reference mangrove SOC, seagrass and mangroves are therefore also sources to mangrove SOC through the litter. Sources to seagrass floc varied seasonally for references but not SL-15. Seston was a dominant floc OC source for all seasons and sites according to the ternary mixing diagram (Figure 77b). Macroalgae was also a dominant source for November reference floc samples.

Ternary diagrams indicated that seston is a dominant source to almost all sediments and surface layers regardless of site. Seston is not the only source, however, because all sediments and surface layers are more enriched in ^{13}C than seston (Figure 73). A review of source determination studies in mangrove and seagrass sediments found that seston was a dominant source at 47% of sites (Chapter 2). OC in sediments of young mangrove forests were dominated by algal and seston sources, just as the constructed sediments were in this study (Marchand et al., 2003; Alongi et al., 2004). In sediments with low %OC, as in this study (Chapter 3), the dominant macrophytes such as mangroves or seagrass seemed less likely to be significant OC sources (Gonnoeea et al., 2004; Kennedy et al., 2004). Middelburg et al., (1997) showed a significant relationship of decreasing $\delta^{13}C$ (more depleted than *in situ* macrophyte $\delta^{13}C$) with decreasing %SOC. These trends may be because when seston settles onto sediments, it does so with inorganic particles, which dilute SOC, lowering the %OC.

Seston comes from a variety of sources as it is made up of phytoplankton, zooplankton, bacteria, and detritus (Figure 78). Its high $\delta^{13}C$ in this study is indicative of a mangrove or terrestrial origin. Its high N:C (low C:N), however, indicates a mixture of phytoplankton, which have C:N ratios from 7.7 to 10.1, and bacterioplankton, which have C:N ratios from 2.6 to 4.3 (Lee and Fuhrman 1987). Other estuarine studies similarly had seston with low $\delta^{13}C$ and low C:N ratios (Hemminga et al., 1994; Cifuentes et al., 1996; Zhou et al., 2006). Cifuentes et al., (1996) demonstrated that bacteria in the water column were likely immobilizing N in the process of decomposing terrestrial-derived organic matter, which could lead to incorporation of that nitrogen into organic matter during humification and a lower C:N ratio.

Low C:N ratios of many sediment samples are concerning because it may have lead us to overstate the importance of seston as a source because it is the only source with equally low C:N ratios. Just as microbial activity likely lowered C:N ratios in seston (Cifuentes et al., 1996), it could lower C:N ratios in sediments. Decreasing sediment C:N ratios during diagenesis has also occurred in other source determination studies (Thimdee et al., 2003; Gonnoeea et al., 2004; Kennedy et al., 2004). Changes due to bacteria are likely because a high percentage of TOC in these sediments is microbial biomass (11 to 63%; Chapter 3). Decreases in source C:N ratios during decomposition may also explain the relatively low C:N ratios of the sediments compared with living source material. Unfortunately, C:N ratios during decomposition were not measured in this study. Results of studies that measured decomposition in

similar systems were equivocal (see source characteristics section). Another reason for low C:N ratios is the eutrophication of the IRL, which has greatly increased the availability of inorganic N sources (Sigua and Tweedale 2003). Due to the influence of factors other than source identity in determining sediment C:N ratios, caution is emphasized in interpreting ternary diagram results.

CONCLUSION

In all sediments, seston was a dominant source and diagenesis of organic matter within sediments lowered sediment C:N ratios (Figure 79). Because the other main sources differed between SL-15 and reference sediments (Figure7 9), their abilities to sequester SOC probably differ too. The litter bag decomposition study suggested which SOC sources are likely to be sequestered in sediments the longest. This information allows us to predict how OC storage will differ in sediments of SL-15 and reference sites. Since seston is an OC source for all sediments, the fact that its decomposition was not measured should not greatly affect these predictions. Because fast-decaying macroalgae OC dominates in SL-15 seagrass sediments, they are unlikely to store OC for as long as reference seagrass sediments. A year after construction, SL-15's seagrass sediments therefore do not store C as well as references. OC in both SL-15 and reference mangrove sediments are a mixture of seston and vascular plants (terrestrial plants in SL-15 and mangrove/seagrass via litter in references). Since terrestrial plants most likely have decay rates similar to mangroves and slower than most seagrass species, it is possible that the length of OC storage in SL-15 and reference mangrove sediments are currently similar. The labile algal mat, however, has the potential of becoming a main source in constructed mangrove sediments because it caused sediment $\delta^{13}C$ enrichment throughout the year, which may ultimately shorten the length of constructed mangrove OC storage.

Table 18. $\delta^{13}C$ (‰) and C:N ratios for all potential sources of organic carbon to mangrove and seagrass sediments in SL-15 and reference sites averaged over various collection times and plant parts (unless otherwise noted). Values in parentheses are ± SE; where no standard error is listed, the value is for a single composite sample.

Location	Species	$\delta^{13}C$ (‰)	C:N
Subtidal	Seagrass	-11.54 (0.84)	
	Leaves	-10.95 (1.3)	14 (0.52)
	Roots	-12.42 (0.78)	31 (2.5)
	Syringodium filiforme	-9.23 (0.86)	14 (0.5)
	Thalassia testudinum	-9.83 (1.2)	13 (0.9)
	Halodule wrightii	-12.62	11
	Halophila johnsonii	-20.07	16
	Epiphytes	-16.20 (1.9)	11 (0.6)
	Macroalgae	-21.00 (0.98)	18 (1.6)
	Acanthophora spicifera	-17.11 (0.78)	12 (0.3)
	Caulerpa sertulariodes	-18.36	14
	Sargassum spp.	-17.48 (0.29)	28 (0.9)
	Daysa baillouviana	-32.06	15
	Ulva spp.	-20.64	14
	Chaetomorpha linum	-25.29	25
	SL subtidal macroalgae	-21.75 (0.86)	16 (2)
	Rosenviga intricata	-20.94 (1.1)	16 (0.8)
	Hypnea cervicornis	-19.69 (1.2)	21 (0.1)
	Gracilaria tikvahiae	-22.50 (1.4)	11 (0.4)

table cont.

	Enteromorpha spp.	-25.24	19	
	Seston	-26.29 (0.47)	6.5 (0.2)	
	May 2006	-27.30 (0.44)	7 (0.5)	
	September 2006	-25.78 (2.2)	6 (0.5)	
	October 2006	-24.23 (0.21)	6 (0.2)	
	November 2006	-24.58 (1.3)	6.5 (0.7)	
	February 2007	-30.10 (0.84)	7.5 (1)	
Intertidal	Spartina alterniflora	-12.94 (0.30)		
	Leaves	-13.37 (0.27)	27 (2)	
	Roots	-12.30 (0.04)	42 (11)	
	Sueda linearis	-29.22	14	
	Mangrove	-26.95 (0.24)		
	Leaves	-27.27 (0.32)	27 (1.4)	
	Roots	-26.18 (0.20)	58 (3.7)	
	Avicennia germinans	-27.46 (0.53)		
	Leaves	-27.77 (0.49)	22 (1.7)	
	Roots	-25.26	55	table cont.
	Rhizophora mangle	-27.17		

		(0.36)	
	Leaves	-27.32 (0.47)	31 (1.7)
	Roots	-26.34 (0.30)	55 (9)
	Laguncularia racemosa	-26.31 (0.84)	26 (3.0)
Terrestrial	C_3 terrestrial	-27.54 (0.44)	33 (4)
	Schinus terebenthifolius (leaves)	-28.30	31
	Casuarina equisetifolia (needles)	-26.33	34
	Coccoloba uvifera (leaves)	-27.77 (0.32)	32 (11)
	Borrichia frutescens	-27.86 (0.60)	21 (0.9)
	Distichlis spicata	-13.67	27 (2.6)

table cont.

Table 19. Decay constants (± SE) and turnover times calculated from a nonlinear regression (exponential decay) of litter bag experiment data.

Species	k (day^{-1})	Turnover time (days)
Halodule wrightii	0.0049 (0.0006)	203
Thalassia testudinum	0.0099 (0.001)	101
Syringodium filiforme	0.046 (0.006)	22
Acanthophora spicifera	0.070 (0.006)	14
Sargassum spp.	0.019 (0.001)	53
Avicennia germinans	0.0093 (0.0004)	109
Rhizophora mangle	0.0047 (0.0004)	213

Table 20. Results of ANOVAs comparing $\delta^{13}C$ values in SL-15 and reference mangrove and seagrass sediments and surface layers (right) and of repeated measures ANOVAs of $\delta^{13}C$ values in SL-15 sediments and surface layers (left).

ANOVA	Effect	$\delta^{13}C$
Repeated measures		
Mangrove 0-10	Month	*
	Depth	NS
	Month*Depth	NS
Mangrove algal mat	Month	**
Seagrass 0-10	Month	NS
	Depth	NS
	Month*Depth	NS
Seagrass accreted	Month	*
Seagrass floc	Month	*

ANOVA	Effect	$\delta^{13}C$
Sediment comparison		
Mangrove	Site	NS
	Month	NS
	Depth	NS
	Site*Month	NS
	Site*Depth	NS
	Month*Depth	NS
Seagrass	Site	***
	Month	NS
	Depth	NS
	Site*Month	NS
	Site*Depth	NS
	Month*Depth	NS
Surface Comparison		
Mangrove algal mat/litter	Site	***
	Month	NS
	Site*Month	***
Seagrass floc	Site	**
	Month	***
	Site*Month	***

For significance NS=not significant, * $p < 0.05$, ** $p < 0.01$, *** $p < 0.0001$. Please see Appendix A for a table listing how these data were transformed prior to running the 3-way ANOVA.

Table 21. Mean $\delta^{13}C$ and C:N (± SE) for sediments and surface layers of SL-15 and reference mangrove and seagrass systems.

System and month	Depth	$\delta^{13}C$		C:N (molar)	
		SL-15	Reference	SL-15	Reference
July mangrove	Algal Mat/ Litter	-15.72 (0.46)	-18.21 (0.63)	8.2 (0.2)	9.3 (0.3)
	0-5 cm	-23.57 (1.03)	-24.12 (0.91)	5.0 (1)	9.6 (3)
	5-10 cm	-23.70 (0.50)	-22.17 (1.40)	8.0 (2)	9.2 (1)
Nov. mangrove	Algal Mat/ Litter	-11.96 (0.47)	-24.43 (1.56)	8.1 (0.9)	21 (5)
	0-5 cm	-21.96 (0.18)	-23.42 (0.70)	5.5 (0.3)	7.0 (1.1)
	5-10 cm	-24.11 (0.46)	-24.10 (0.31)	6.2 (0.8)	8.8 (0.9)
July seagrass	Floc	-21.05 (0.14)	-18.93 (0.27)	8.2 (1)	5.7 (0.6)
	Accreted	-21.00 (0.44)		7.6 (1)	
	0-5 cm	-21.03 (0.35)	-18.97 (0.29)	6.9 (0.3)	6.7 (0.2)
	5-10 cm	-20.93 (0.60)	-19.11 (0.20)	7.0 (1)	6.6 (0.2)
	10-15 cm		-18.79 (0.33)		1.0 (0.2)
Nov. seagrass	Floc	-19.50	-24.46	6.8	10.8 (0.6)

table cont.

	(0.15)	(0.56)	(0.1)	
Accreted	-20.06 (0.20)		6.1 (0.4)	
0-5 cm	-21.55 (0.41)	-17.87 (0.21)	6.7 (0.8)	5.4 (0.2)
5-10 cm	-20.80 (0.48)	-18.80 (0.46)	7.3 (1)	5.6 (0.4)
10-15 cm		-19.10 (0.48)		6.1 (0.4)

Table 22. Mean $\delta^{13}C$ or $\delta^{13}C$ ranges of means for sources in this study and in the literature. Means are averaged across and ranges are across plant parts, species, and sites for this study and where applicable in the literature. The sources listed here are the main SOC sources that were used in this study's ternary diagrams.

Source	$\delta^{13}C$ (‰) This study	$\delta^{13}C$ (‰) Other studies	Data source[a]
Seagrass	Mean: -11.54	-10.0	1
	Range: -20.07 to -9.23	-19.7 to -10.7	2
		-13.3 to -5.8	3
		-12.4	4
		-12.2	5
		-16.1 to -11.9	6
		-10.5	7
		-23 to -3, -10 (mode)	8
		-10.4 to -7.2	9
		-14.6 to -8.8	10
		-12.7 to -11.4	11
Mangrove	Mean: -26.95	-27.0	12
	Range: -27.77 to -25.26	-28.3 to -24.1	2
		-29.0 to -27.0	13
		-28.4 to -27.9	3
		-28.8	7
		-28.2	14
		-27.9	15
		-30.1 to -28.3	16
		-29.7 to -25.9	17
Macroalgae	Mean: -21.00	-31.7 to -16.6	11
	Range: -32.06 to -17.11	-21.5 to -15.0	18
		-26.0 to -20.9	15
		-15.61	7
Seston	Mean: -26.29	-22.0 to -21.0	12
	Range: -30.10 to -24.23	-23.0 to -20.5	13
		-18.4	1
		-23.3 to -13.7[b]	2
		-27.6 to -12.1	3
		-22.1	4

table cont.

		-24.7	5
		-25.32 to -22.06[b]	6
		-20.6	7
		-22.6[b]	19
		-28.1 to -20.8[b]	20
		-26.4[b]	21
Terrestrial (C_3)	Mean: -27.54	-28 to -25	22
	Range: -28.30 to -26.33	-30 to -25	23
		-26	24

[a]1, Canuel et al., 1997; 2, Hemminga et al., 1994; 3, Kennedy et al., 2004; 4, Papadimitriou et al., 2005; 5, Gacia et al., 2002; 6, Gonnoeea et al., 2004; 7, Thimdee et al., 2003; 8, Hemminga and Mateo 1996; 9, Anderson and Fourqurean 2003; 10, Vizzini et al., 2003; 11, Smit et al., 2005; 12, Jennerjahn and Ittekkot 2002; 13, Bouillon et al., 2003; 14, Bouillon et al., 2004; 15, Bouillon et al., 2002; 16, Lallier-Verges et al., 1998; 17, Muzuka and Shunula 2006; 18, Fenton and Ritz 1988; 19, Zhou et al., 2006; 20, Dittmar et al., 2001; 21, Cifuentes et al., 1996; 22, Miserocchi et al., 2007; 23, Kang et al., 2007; 24

[b]Called particulate organic matter (POM) or suspended particulate matter (SPM) by the authors

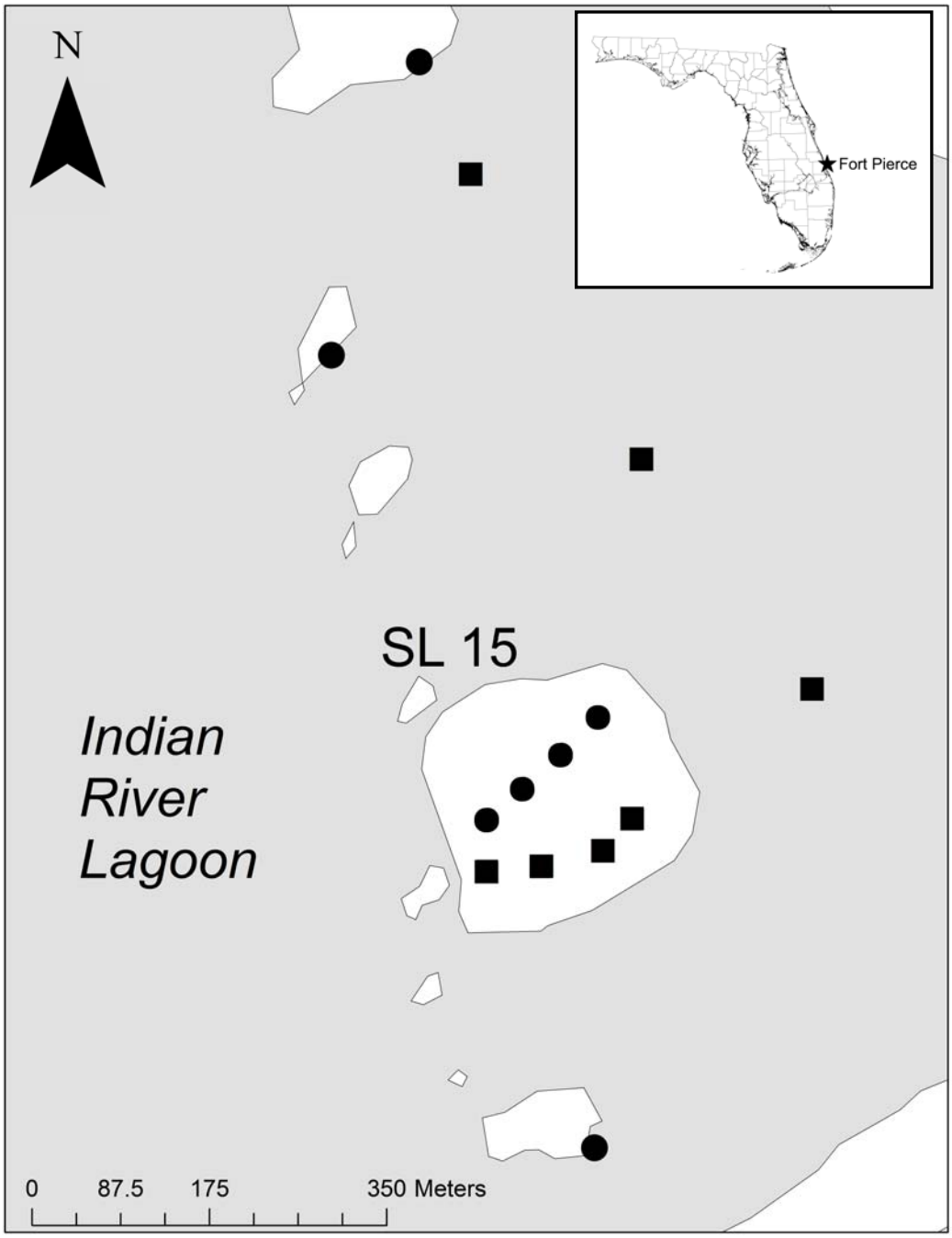


Figure 71. The study area in the Indian River Lagoon, next to Fort Pierce, Florida (inset). SL-15 is the large island in the center. Circles are mangrove system plots and squares are seagrass system plots. Symbols outside of SL-15 are the reference sites, which have one plot each.

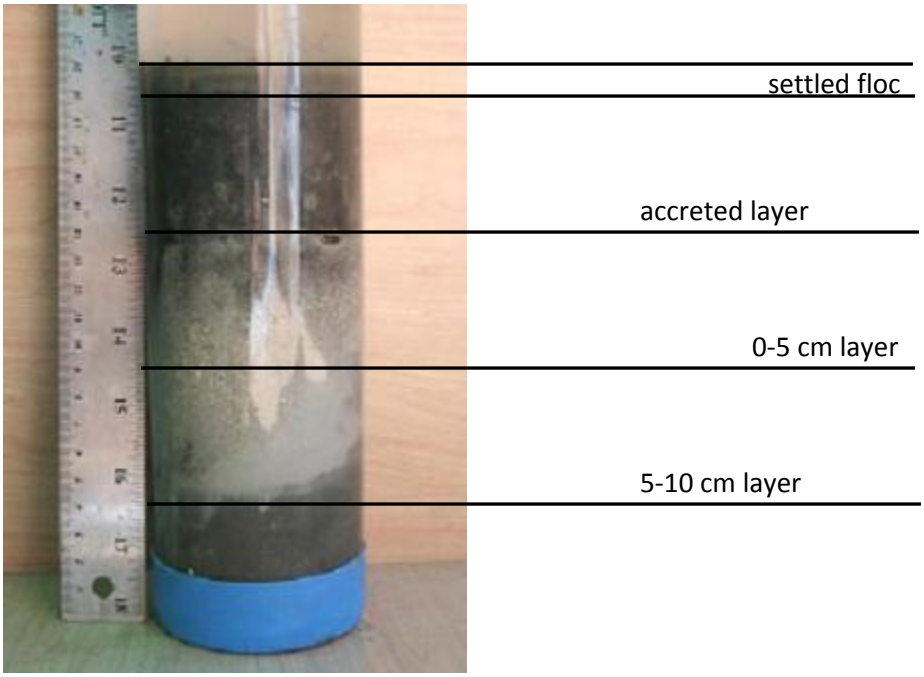


Figure 72. Core from SL-15 seagrass system illustrating the surface layer (floc) and different sediment depths (accreted layer, 0-5 cm, and 5-10 cm). Note the difference in color between the accreted layer and 0-5 cm depth.

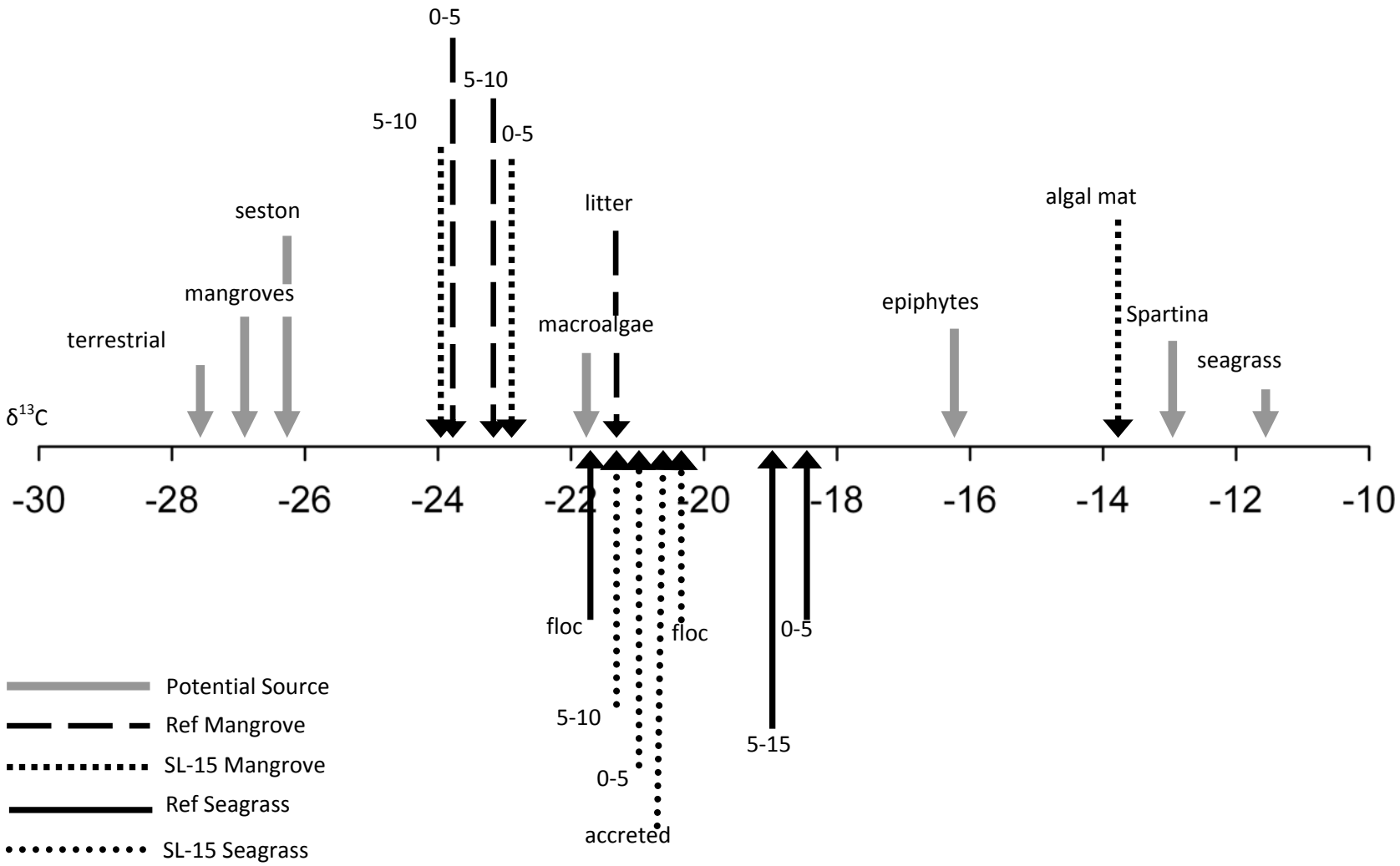


Figure 73. $\delta^{13}C$ averaged over July and November 2006 for SL-15 and reference sediments and surface layers compared to mean $\delta^{13}C$ of potential sources.

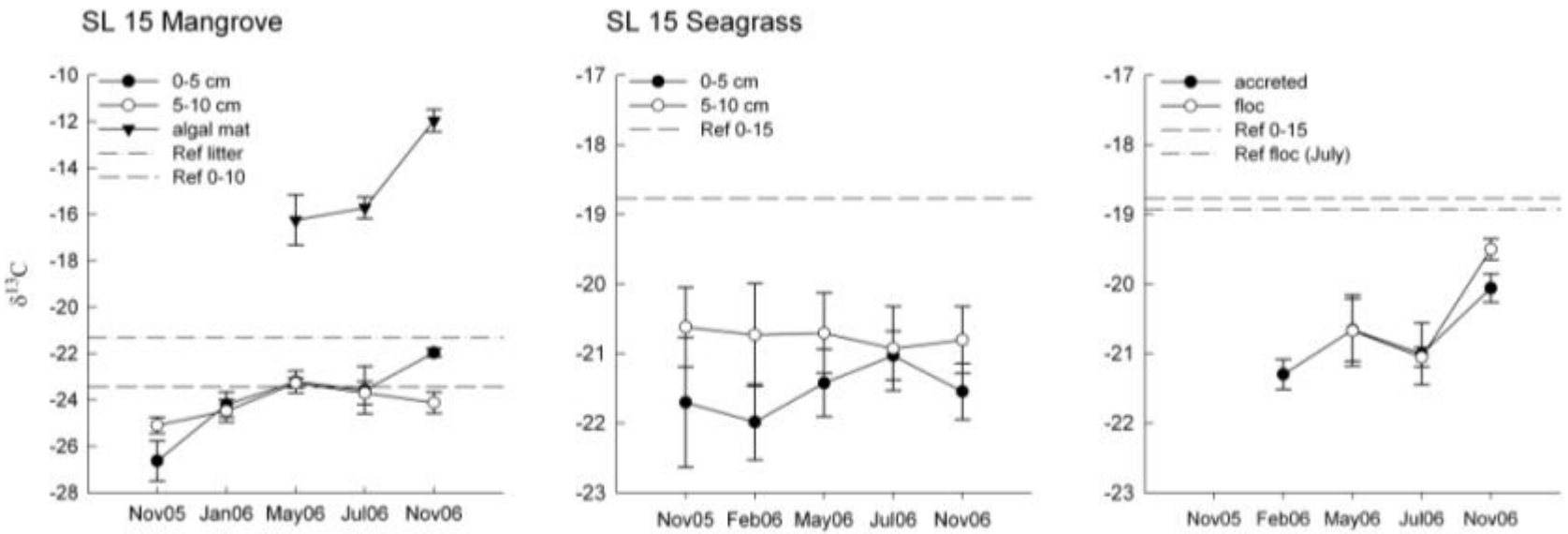


Figure 74. Mean $\delta^{13}C$ of SL-15 sediments and surface layers over the first year after construction. Error bars are ±SE. Reference lines are $\delta^{13}C$ averaged over depth (for sediments) and month (except for reference floc) for the respective reference systems.

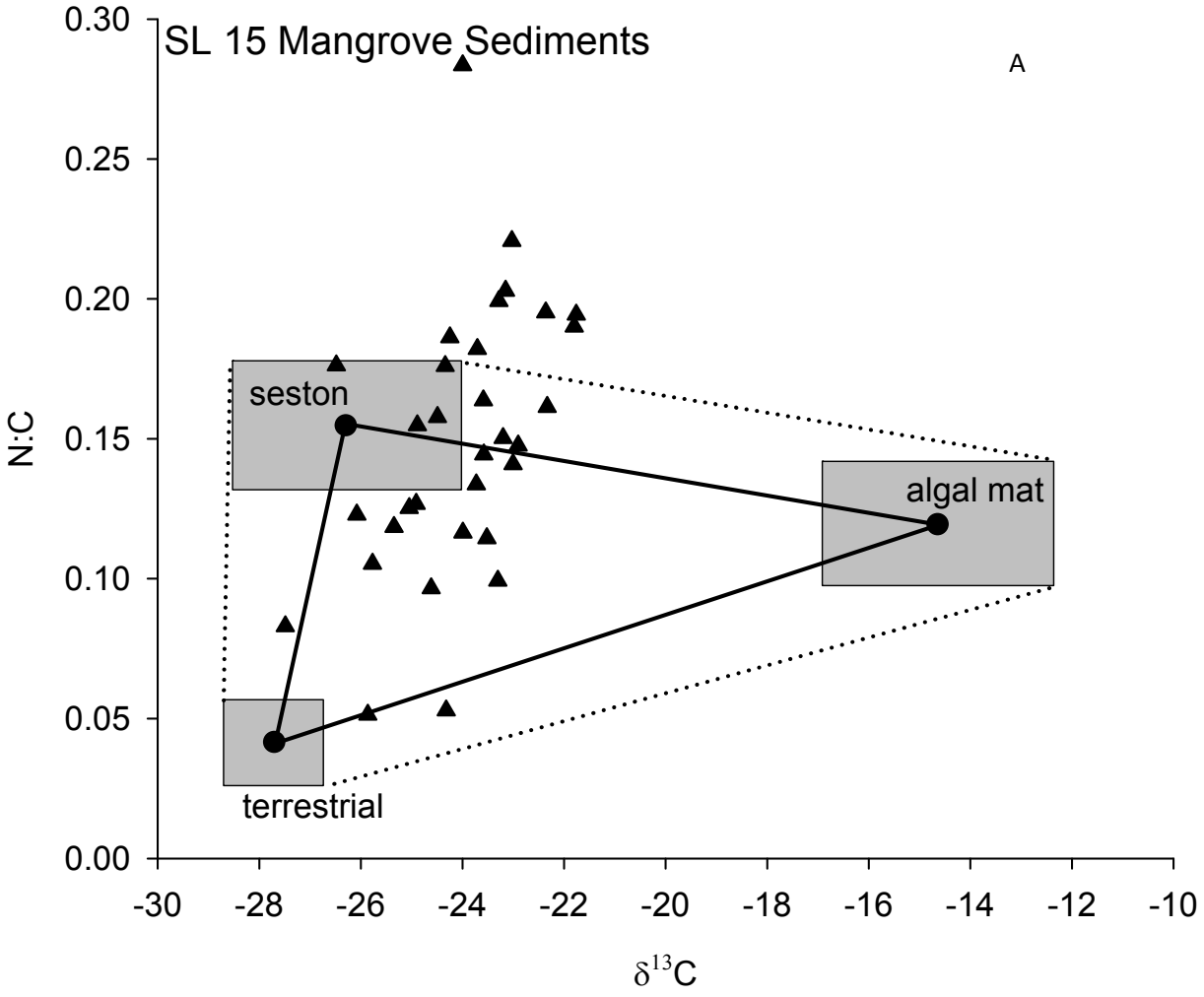


Figure 75. N:C vs. δ13C in ternary mixing diagrams of three potential OC sources and mangrove sediments. Circles are the mean end member values and boxes are ± standard deviation of N:C and δ13C. Triangles are mangrove sediment values for SL-15 (A) and the reference (B).

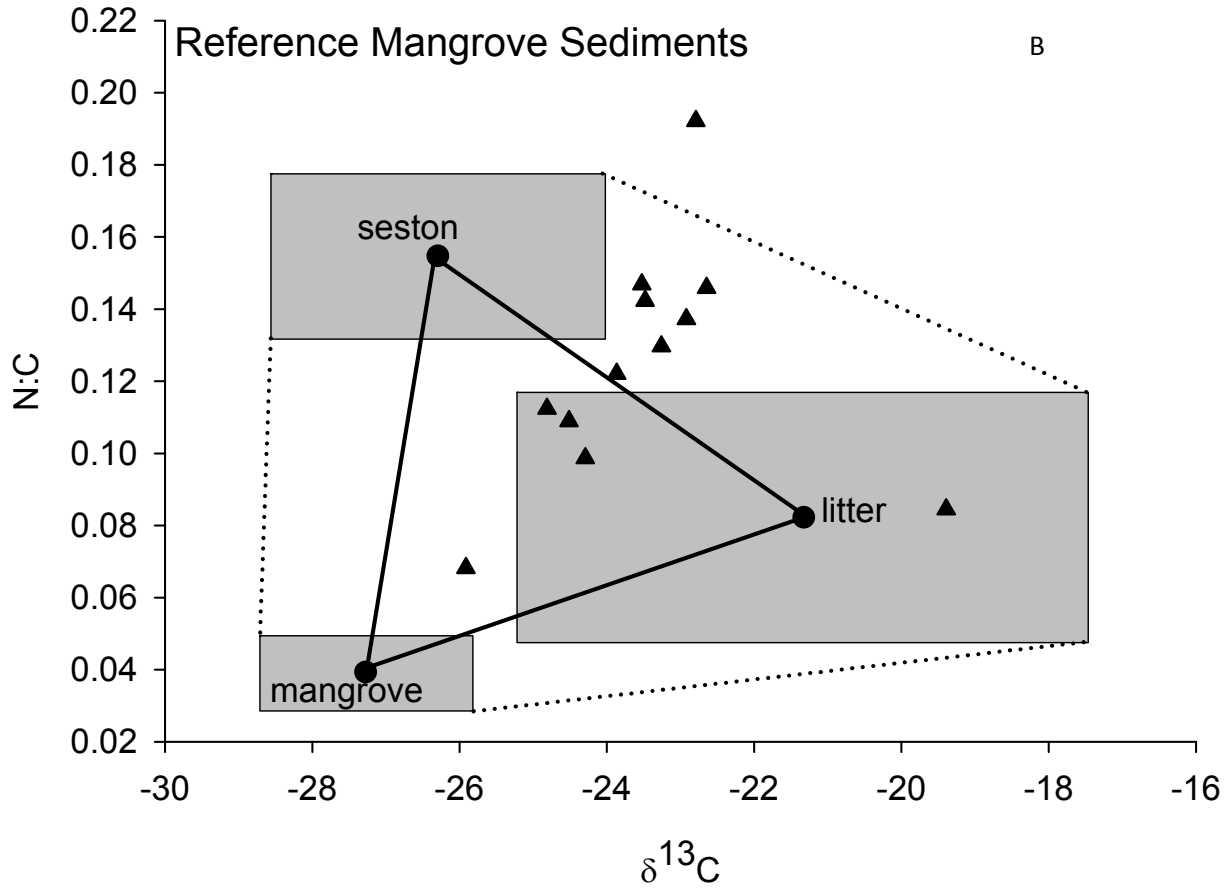


Figure 75 cont.

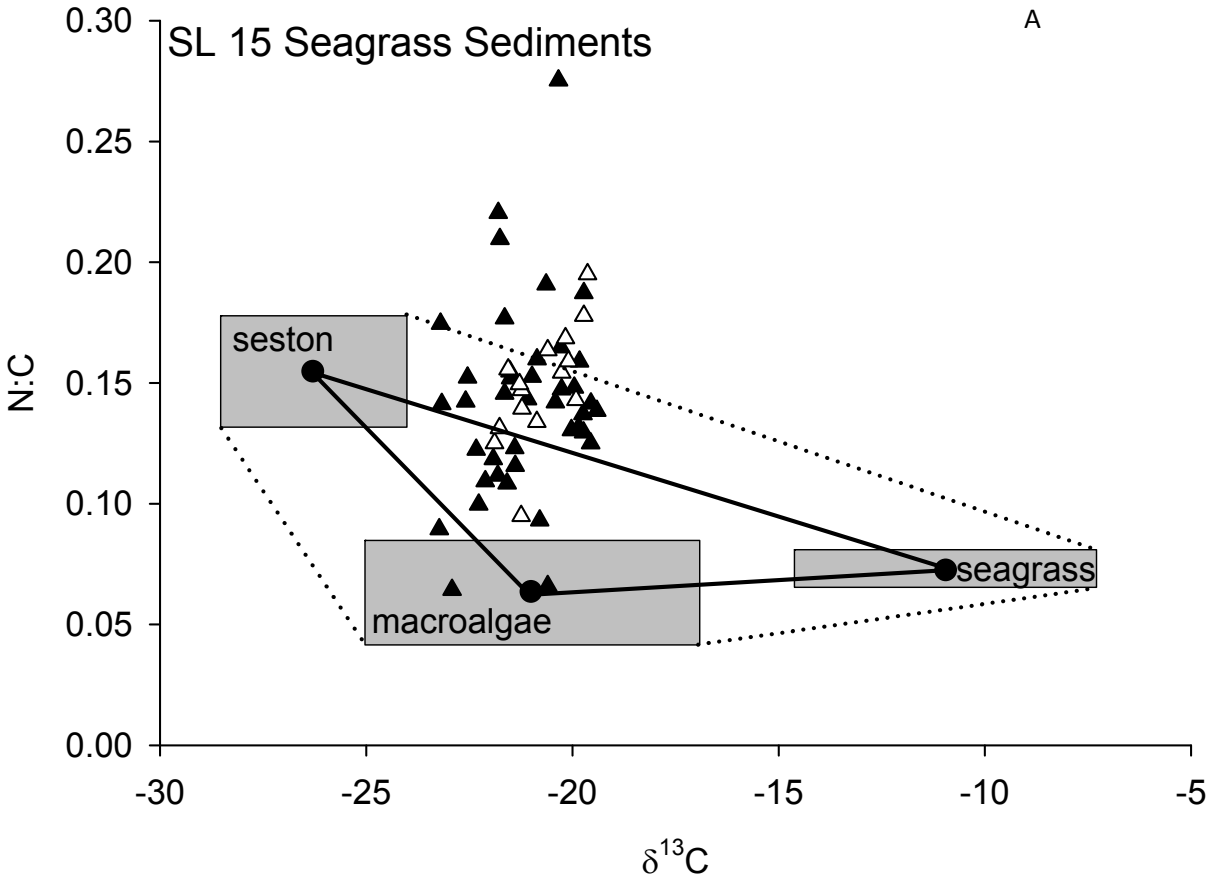


Figure 76. N:C vs. δ13C in ternary mixing diagrams of three potential OC sources and seagrass sediments. Circles are the mean end member values and boxes are ± standard deviation of N:C and δ13C. Filled triangles are 0-10 cm sediment values for SL-15 (A) and 0-15 cm sediment values for reference (B). Open triangles are accreted layer values for SL-15 (A).

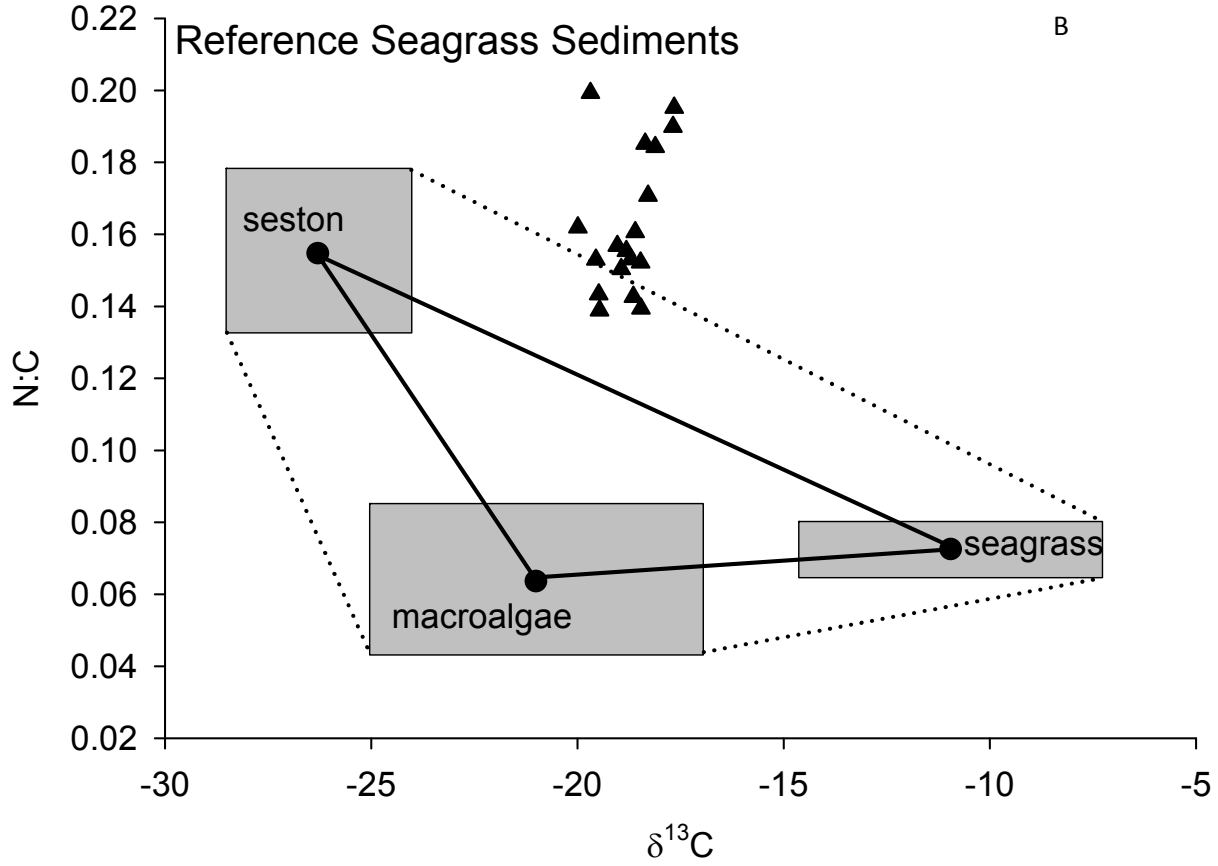


Figure 76 cont.

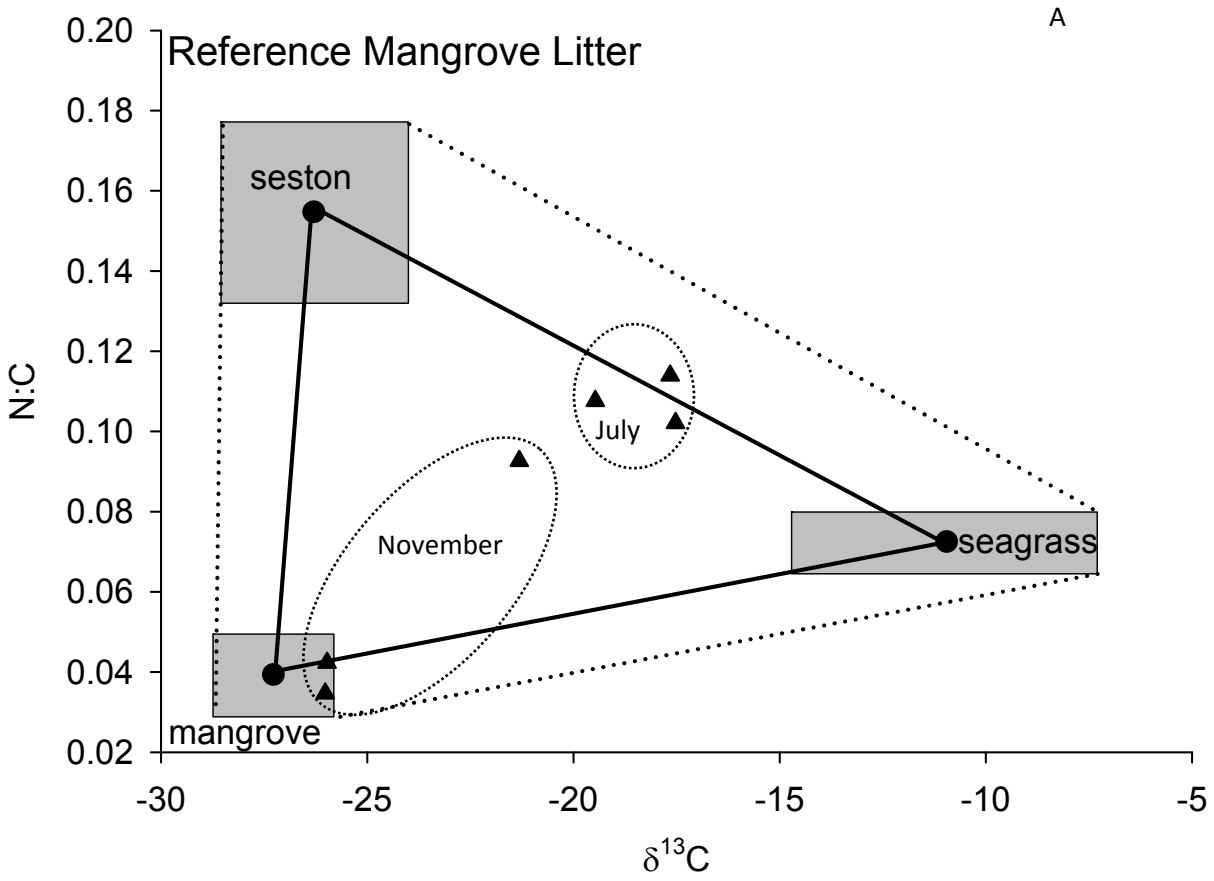


Figure 77. N:C vs. δ13C in ternary mixing diagrams of three potential OC sources and surface layers. Circles are the mean end member values and boxes are ± standard deviation of N:C and δ13C. Filled triangles are reference litter layer values (A) and SL-15 floc values (B). Open triangles are reference floc values (B).

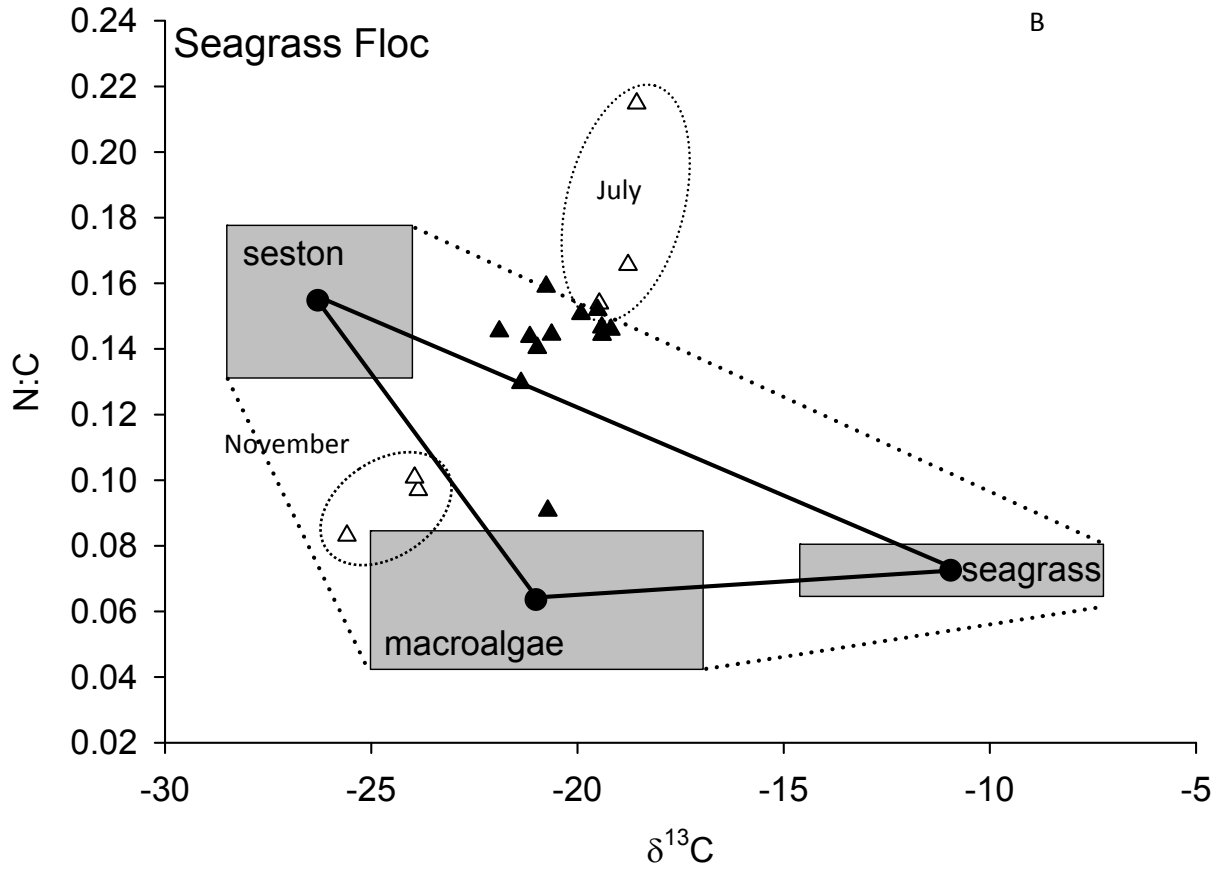


Figure 77 cont.

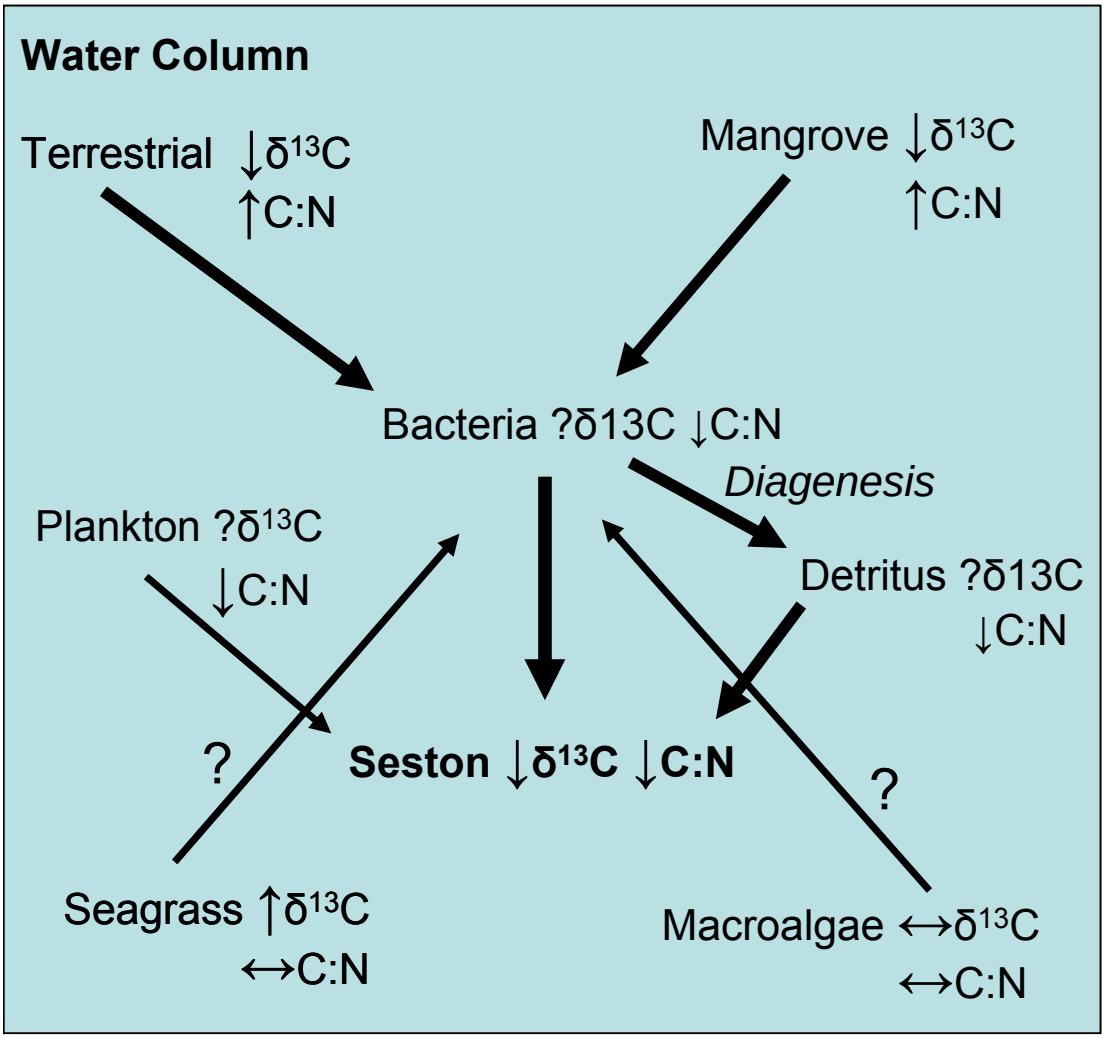
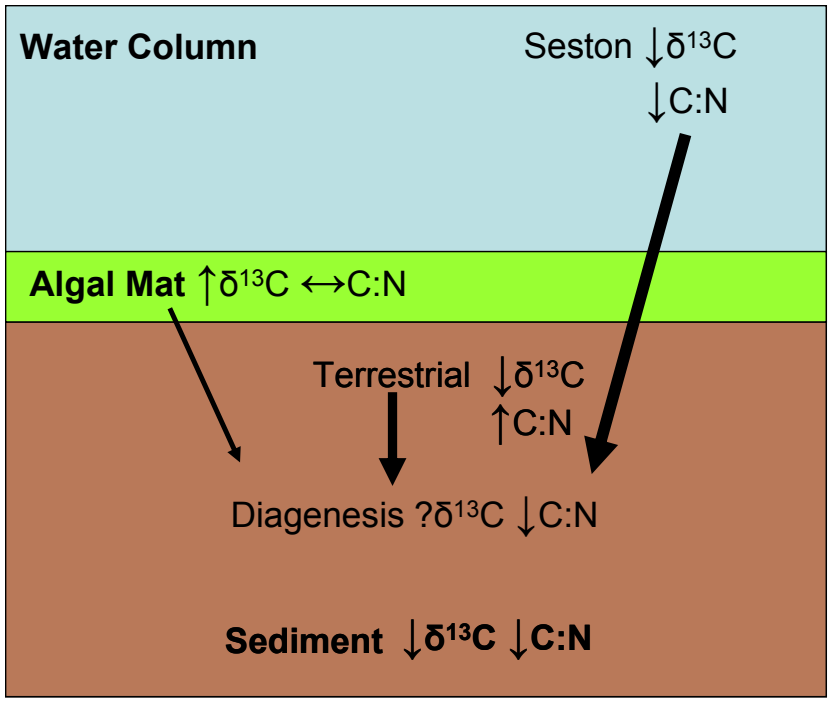


Figure 78. A theoretical diagram of organic carbon sources that may constitute seston and how they affect seston $\delta^{13}C$ and C:N. Arrow sizes indicate the possible relative contributions of each source. ↓ indicates depleted $\delta^{13}C$ and low C:N, ↑ indicates enriched $\delta^{13}C$ and high C:N, and ↔ indicates mid-range $\delta^{13}C$ and C:N.

SL-15 Mangrove



Ref Mangrove

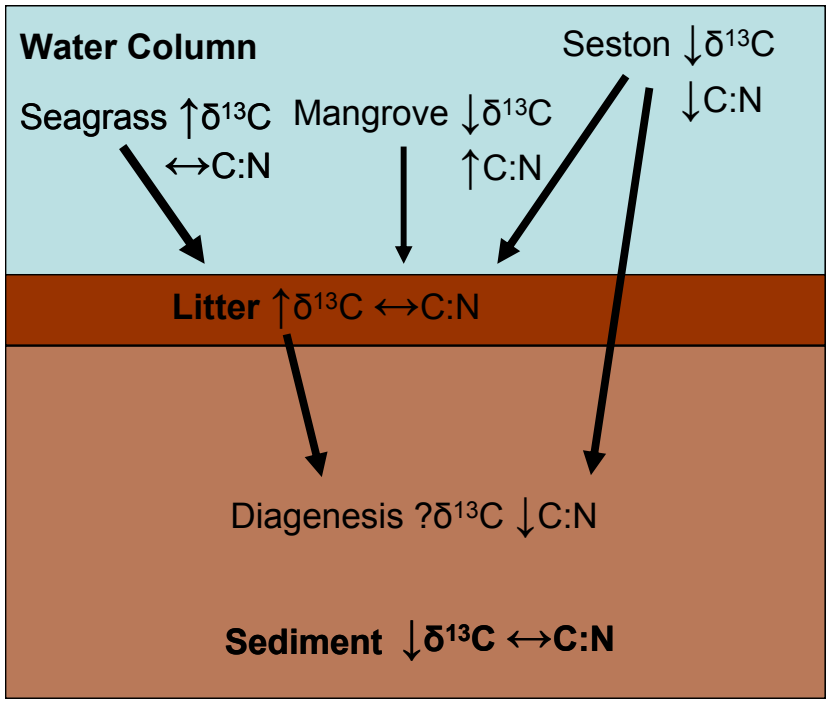
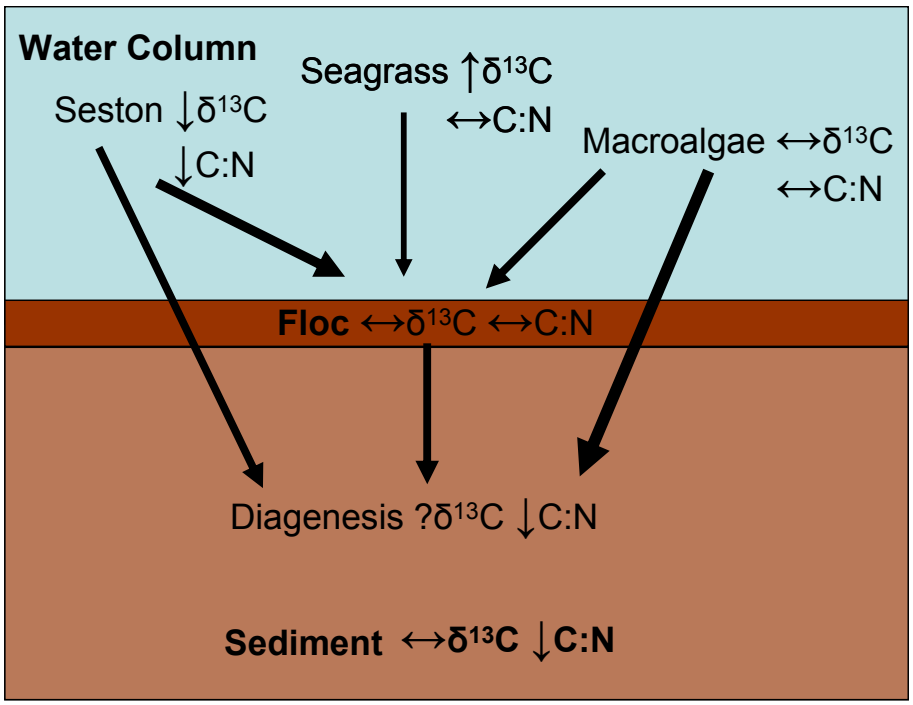


Figure 79. Main sources and how they affect surface layer and sediment $\delta^{13}C$ and C:N. Arrow sizes indicate the relative contributions of each source. ↓ indicates depleted $\delta^{13}C$ and low C:N, ↑ indicates enriched $\delta^{13}C$ and high C:N, and ↔ indicates mid-range $\delta^{13}C$ and C:N.

SL-15 Seagrass



Ref Seagrass

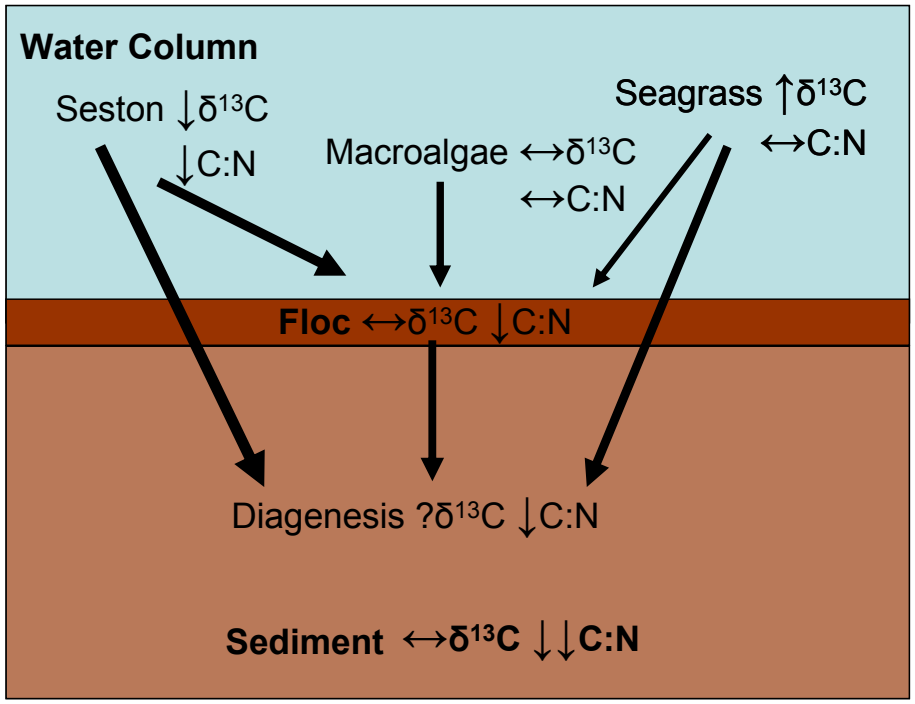


Figure 79 cont

SYNTHESIS

Coastal ecosystems including salt marshes, seagrass beds, and mangrove forests are more effective carbon (C) sinks than terrestrial systems and freshwater wetlands (Chapter 2). These ecosystems store large amounts of OC and actively accumulate OC at high rates; about 44.6 Pg C is stored and about 120 Tg C y^{-1} accumulates in salt marsh and mangrove sediments globally (Jennerjahn and Ittekkot 2002; Chmura et al., 2003). Many coastal ecosystems have been degraded or lost due to anthropogenic disturbances (Valiela et al., 2001; Kennish 2002; Zedler 2004). Destruction of coastal ecosystems increases atmospheric CO_2 concentrations because their organic C (OC) stores are often mineralized as a result and their future OC sequestration capacity is lost (Duarte et al., 2005; Bridgham et al., 2006). Generally in the United States, the destruction of coastal ecosystems must be mitigated by restoring or creating coastal ecosystems elsewhere. Whether mitigation of seagrass and mangrove systems restores the C sink capacity is currently not well-studied.

In the present study, functional trajectories of sediment OC (SOC) parameters in a constructed mangrove and seagrass system in the Indian River Lagoon, Florida were measured. Sediment OC (SOC) parameters in constructed systems were also compared to mature reference systems to indicate if constructed sediments had reached functional equivalence in terms of OC storage. The objectives of this study were: 1) to determine short term trajectories of SOC pools in a constructed mangrove forest and seagrass bed; 2) to compare SOC pools in the constructed system with those in reference systems; 3) to compare the lability of SOC in the constructed and reference systems; 4) to determine and compare significant sources to the total SOC pool in the constructed and reference systems. The hypotheses were: 1) in the short term, storage in the three OC pools studied would increase in the constructed systems, but would not reach the level of storage in the references' OC pools; 2) OC lability would be greater in sediments of constructed systems than in reference sediments; 3) SOC sources in constructed systems would be macroalgae or plankton, while SOC sources in reference systems would be vascular plants, like mangroves and seagrass. Key findings addressing each objective and the validity of the hypotheses are presented below.

Objective One: Short-Term Trajectories of Sediment Organic Carbon Pools

Contrary to the hypothesis, functional trajectories were not followed by OC parameters in the constructed site sediments. Instead of steady increases, SOC parameters either remained unchanged or increased and decreased throughout the year, driven by seasonal changes in the water column. The only sediment functional trajectory was followed by the mangrove system's bulk density, which decreased throughout the year but remained above reference levels. Functional trajectories were somewhat followed by surface layers as both microbial biomass C (MBC) and total OC (TOC) increased. Due to their proximity to OC inputs, it is logical that OC should increase in the surface layers before they increase in sediments. However, whether increases in surface layer OC were due to a recovering function or an annual pattern could not be discerned. For example, the increase in floc MBC and TOC followed the same trend as total suspended solids, a water quality parameter. Overall, one year was not sufficient time to map OC functional trajectories in the constructed mangrove and seagrass system. The

lack of a functional trajectory did not preclude the OC parameters from being functionally equivalent to reference values.

Objective Two: Comparisons of Sediment Organic Carbon Pools

The hypothesis that SOC pools would be smaller in constructed systems was by and large correct for mangrove sediments but not for seagrass sediments (Figure 80 and 81). Floc and accreted layers of constructed seagrass sediments reached or exceeded functional equivalence for all three OC pools—TOC, Extractable OC (ExOC), and MBC. The 0-10 cm depths, also reached equivalence for ExOC pools. On a storage (areal) basis, equivalence was also reached by mangrove 0-5 cm depths for ExOC and MBC and seagrass 0-10 cm depths for MBC. This equivalence was only on a storage basis because it was driven by greater bulk densities in the constructed sediments. Seagrass sediments reached SOC pool equivalence more than mangrove sediments due to their constant inundation, parent material, and lower equivalence goal (reference seagrass sediments had less TOC and ExOC than reference mangrove sediments).

SOC pool sizes were not the only factors that indicated if constructed systems had attained functionally equivalent OC storage—information about OC accumulation rates and OC lability was also needed. OC accumulation rates in the constructed mangrove and seagrass systems were similar to literature values if accumulation in surface layers was considered. It was unknown if the constructed systems could sustain these accumulation rates over the long term or if the rates reflected an immediate response in SOC after construction. Larger proportions of the TOC pool were MBC in constructed 0-10 cm sediments indicating greater SOC lability and therefore less SOC storage in constructed systems.

Objective Three: Comparisons of Sediment Organic Carbon Lability

Generally, constructed systems SOC was more labile than reference system SOC for both mangrove and seagrass sediments (Figure 80 and 81). Lability was only similar between constructed and reference systems in the upper portion of seagrass sediments and in seagrass floc. These results confirm hypothesis two. Greater lability of OC in the constructed system indicates that the constructed system does not function as well as reference systems in terms of OC storage. Even when SOC pool sizes are similar to references', as in the 5-10 cm depth of constructed seagrass sediments, greater OC lability indicates that OC storage is not functionally equivalent. The more labile OC is, the more likely it will be mineralized by microbes and respired to CO_2 instead of being stored in sediments long term. Differences in OC lability were partially due to differing C limitations and to differences in SOC sources between constructed and reference systems.

Objective Four: Comparisons of Sediment Organic Carbon Sources

Sources to the SOC pool differed between constructed and reference systems, but not to the extent that was hypothesized (Figure 80 and 81). Ternary diagrams suggested that seston from the water column was a main SOC source for all systems—constructed and natural. The true importance of seston, however, was unclear because the low sediment C:N ratios that led to the conclusion that seston was a main source, can also result from diagenetic transformations. In mangrove sediments, both systems

had lignin-containing higher plants as other main sources—terrestrial plants in the constructed system and mangroves/seagrass via litter in the reference systems. The effect of sources on OC storage in mangrove sediments was therefore similar; but there was an indication that the labile algal mat was becoming an increasingly important source in constructive sediments, which would shorten OC storage times. Sources in reference seagrass sediments were unclear. It was apparent though, that a greater amount of SOC was derived from macroalgae in the constructed system than in the reference system. Litter bag studies demonstrated that macroalgae generally have the fastest decomposition rates of all aquatic macrophytes, indicating that the macroalgae-derived OC would not be stored in constructed sediments for long amounts of time.

Conclusion

Overall, neither mangrove nor seagrass sediments of the constructed system are functionally equivalent to their respective references in regards to OC storage (Figure 80 and 81). Recovery indices indicate how close various parameters are to equivalence with references.

$$RI = \log\left(X_{constructed} / X_{reference}\right) \quad (5\text{-}1)$$

In Equation 5-1, RI is the recovery index, $X_{constructed}$ is the value of the parameter in the constructed system, and $X_{reference}$ is the value of the parameter in the reference system. RI's equal to zero indicate equivalence, less than zero indicate that equivalence has not been reached, and greater than zero indicate equivalence has been surpassed. The constructed seagrass system is closer to equivalence than the constructed mangrove system (Figure 82). Upper depths of constructed seagrass sediments had similar SOC pools and lability to upper depths of reference seagrass sediments, causing the seagrass sediments to be closer to equivalence. As the mangroves and seagrass within the constructed systems mature, it is likely that their SOC will become less algae-derived, leading to lower OC lability and better OC storage. Dominance of seston as a source in all systems means that a switch in less significant sources may take time to register in the sediments. Ultimately, if the OC pools and lability in reference systems are any indication (Figure 80 and 81) and if functional trajectories are followed in the future, the constructed mangrove system will become more effective at OC storage than the constructed seagrass system.

This study adds to the body of research on functional trajectories. It is one of two known studies on seagrass trajectories and the only known study of mangrove trajectories. More importantly, it is the first known study to examine the trajectory of OC storage in depth. If constructed and restored coastal ecosystems store and accumulate OC as well as their established counterparts, corporations and governments could construct coastal systems in exchange for C credits. This action can replace lost systems and restore many of the ecologically important functions these systems provide. Conclusions based on this study are limited because it only followed constructed systems during the first year of recovery. The constructed mangrove and seagrass system in this study accumulated SOC at rates similar to rates in mature systems over the first year. If these rates are sustained and more OC is stored in long-term, recalcitrant pools, then the constructed systems will be effective C sinks. Longer term studies are needed to fully assess the effectiveness of constructing coastal ecosystems for OC storage.

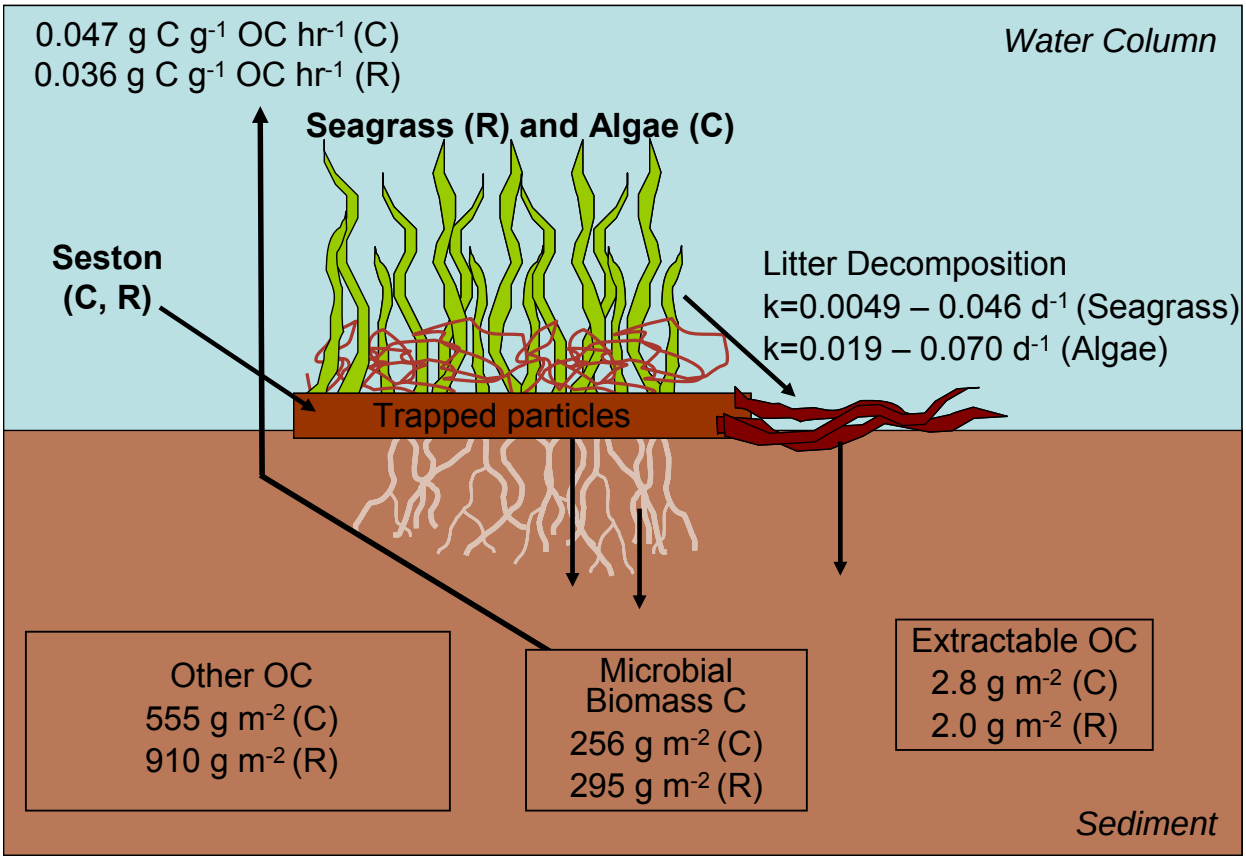


Figure 80. A modified seagrass bed carbon cycle showing values from this study in constructed (C) and reference (R) systems. Organic carbon (OC) pools are the sum of sediment and surface layer means (July and November data). Rates of microbial carbon respiration are the mean of all depths (sediment and surface layers) in July and November, adjusted from an O_2 uptake rate to a carbon release rate by an assumed 1 O_2 to 6 C molar ratio. Bolded words are the main contributors to sediment OC pools.

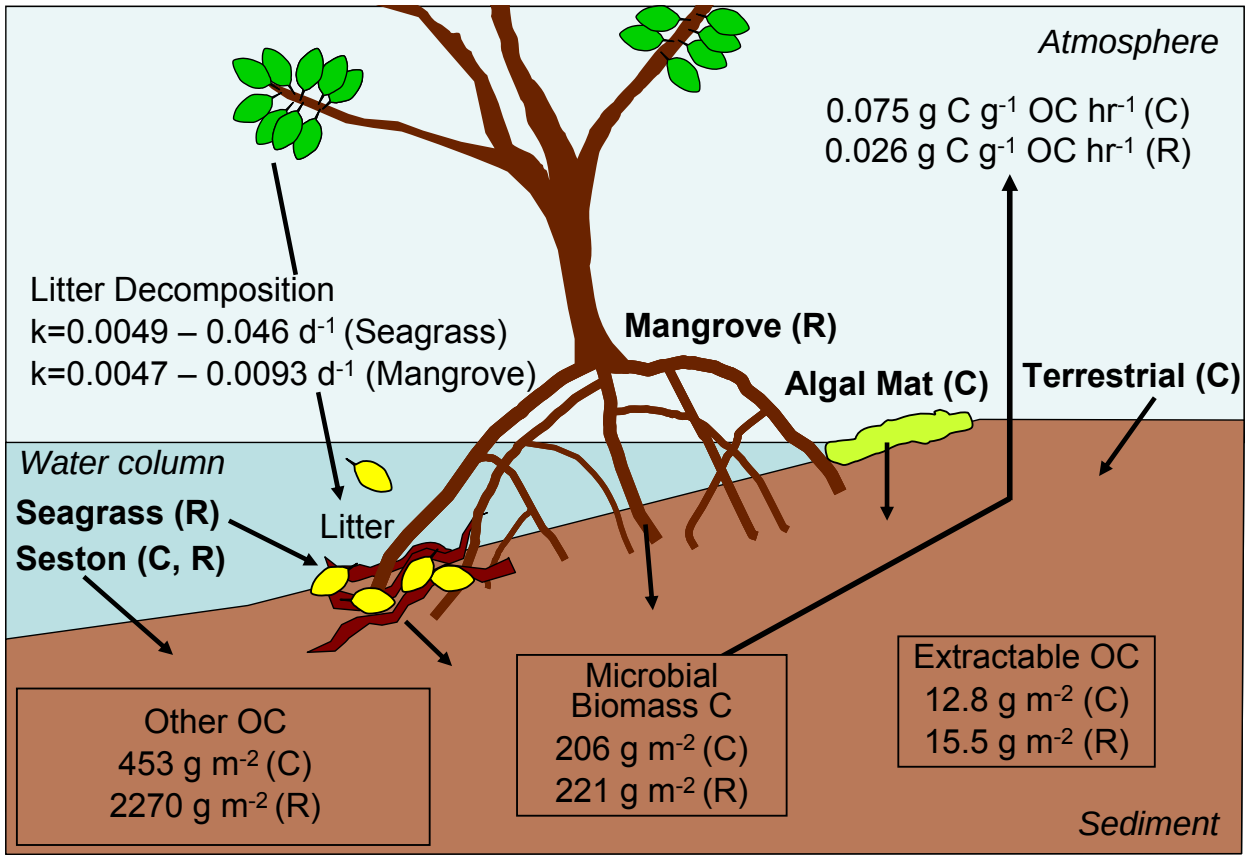


Figure 81. A modified mangrove forest carbon cycle showing values from this study in constructed (C) and reference (R) systems. Organic carbon (OC) pools are the sum of sediment and surface layer means (July and November data). Rates of microbial carbon respiration are the mean of all depths (sediment and surface layers) in July and November, adjusted from an O_2 uptake rate to a carbon release rate by an assumed 1 O_2 to 6 C molar ratio. Bolded words are the main contributors to sediment OC pools.

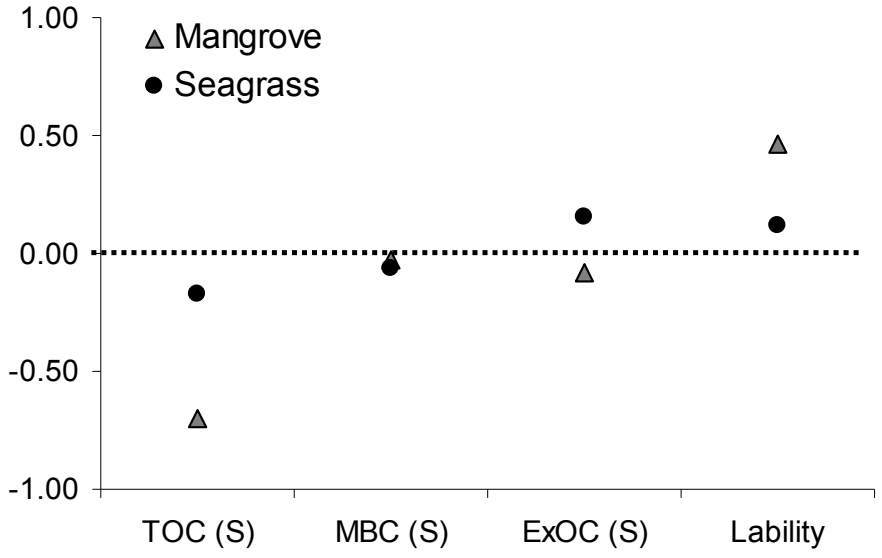


Figure 82. Recovery indices of three organic carbon (OC) pools and OC lability parameters for constructed mangrove and seagrass systems. For each system, OC pools are summed for all sediment depths and surface layers and lability is the average of all depths and surface layers. TOC=total organic carbon, MBC=microbial biomass carbon, ExOC=extractable organic carbon, and (S) indicates that these OC pool parameters were calculated on a storage basis.

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CHAPTER 7: PARTICLE SIZE DISTRIBUTION ANALYSIS OF MARINE SOILS

INTRODUCTION

The purpose of this chapter is to review current methods for determining particle size distribution (PSD). Current methods could be improved to give more useful determinations of PSD. Also, plans for an experiment for improving these methods will be presented at the end of this chapter.

Particle size distribution analysis (PSDA) is a measurement of the size distribution of soil's individual particles, sand, silt and clay (Table 23), which can be used to understand soil genesis, to classify soil or to define texture (Soil Survey Staff, 1993). It is an important soil property that is used at many levels of soil classification and interpretation. With the recent expansion of soil science into marine environments, the USDA is challenged with determining PSD for soils that can be physically and chemically different from terrestrial soils.

USDA PSDA METHODS

The USDA classification of soil texture is based on the proportion of sand 2.0-0.05 mm, silt 0.05-0.002 mm and clay < 0.002 mm particles. Geologists and sedimentologists have been on the forefront of analyzing marine and submerged sediments. Demas was one of the first to take a soil science approach to study the sediments of shallow coastal systems (Demas et al, 1996). A soil science approach to sediments would necessitate the classification of these subaqueous soils into a unified taxonomic system with the Natural Resource Conservation Service's (NRCS) terrestrial soil taxonomy (Soil Survey Staff, 1999). Thomas Reinsch of the NRCS' National Soil Survey Center explained that PSDA data will be collected for subaqueous soils in regards to existing policies and methods with the terrestrial soils (E-mail communications with Dr. Thomas Reinsch). PSDA is a major criterion used to describe these soils and their characteristics.

For now, NRCS PSDA of subaqueous soils will be consistent with the terrestrial standards of the established methods. The current procedure accepted by the NRCS for PSDA is the pipette method (Gee and Bauder, 1986). The USDA uses this method because it is reproducible on many different types of soils (Soil Survey Staff, 1999). This procedure has been relatively unchanged in variations used since 1922 (Muller et al., 2009). In contrast geologists and sedimentologists have begun to rely almost exclusively on the instrumental methods which are more time and cost effective as well as more reproducible (Mudroch et al.,1997; Molinaroli et al., 2000; Muller et al., 2009).

GEOLOGIC PSDA METHODS

The two classes of methods of determining the particle size distribution (PSD) of a given sample consist of classical and instrumental, both of which rely on physical segregation of particles followed by quantification by mass (Mudroch et al., 1997). Examples of classical methods are sieving and pipette methods. As previously mentioned, instrumental methods tend to be faster and more reproducible

(Welch et al., 1979; Mudroch et al., 1997). Examples of instrumental methods are optical determination of particles, electrical sensing zone or electroresistance particle counting (Coulter Counter), X-ray sedimentation (Sedigraph) and laser diffraction (Mudroch et al., 1997; Molinaroli et al., 2000; Goossens, 2008).

Table 23: USDA Particle Size Separates. The experiment will focus on the clay fraction (<0.002 mm). (USDA NRCS Soil Survey Laboratory Methods Manual, 2004)

USDA Particle Size Separates	Size (mm)
Clay, total	<0.002
Silt, total	0.002-0.05
Silt, fine	0.002-0.02
Silt, coarse	0.02-0.05
Sand, total	0.05-2.00
Very fine sand	0.05-0.10
Fine sand	0.10-0.25
Medium sand	0.25-0.50
Coarse sand	0.50-1.00
Very coarse sand	1.00-2.00

Optical determination or direct measurement of particles is one of the oldest methods, where each particle is directly measured with calipers or through magnified digitized photos. This process is time consuming and requires a great deal of particles counted to reach desirable confidence intervals. Electroresistance particle counters were originally designed to count blood cells, but have been used in earth sciences enough to be established in the American Society of Testing Materials. They measure particles on the basis of electrical resistance. When particles pass between electrodes, the machine records the resistance, which is proportional to particle size (Mudroch et al., 1997; Molinaroli et al., 2000). They are popular because they can analyze samples rather quickly, approximately 10-100 seconds per sample and can be used for small quantities of sample; drawbacks are the measuring tube tends to clog and there is no way to know if more than one particle is passing through the electrodes (Mudroch et al., 1997). X-ray sedimentation and laser diffraction particle size instruments are based on sedimentation rates and Stoke's Law, as is the pipette method (Mudroch et al., 1997; Molinaroli et al., 2000). X-ray sedimentation measures the density of the suspension with a cumulative curve of the percentage mass of the silt and clay size fraction versus the logarithm of equivalent diameter (Buchan et al., 1993). This technique is quick and reproducible, but is limited to the fine particles, generally <63 μm, and may be influenced by the different densities of particles (Buchan et al., 1993; Muller, 2009). Laser diffraction spectroscopy uses the same basic principle as the X-ray, but instead uses the intensity of the light scattered by the particle to determine its size (Mudroch et al., 1997, McCave et al., 2006; Taubner et al., 2009). All of these methods have distinct advantages and disadvantages and no one procedure

gives exact results due to the nature and definition of soil particle size (Mudroch et al., 1997; Molinaroli et al., 2000; Goossens, 2008).

ORGANIC MATTER REMOVAL AS A PRE-TREATMENT

With the pipette method, samples are usually pre-treated to avoid interferences, by flocculation and aggregates (Welch et al., 1979; Mudroch et al., 1997). Standard pretreatment and dispersion procedure outlined in the Soil Survey Laboratory Methods Manual is to remove organic matter, carbonates, iron, silica, and to disperse soil aggregates (Soil Survey Staff, 2004). These pre-treatment methods were designed for the use in terrestrial soils and may not be suitable for some of the subaqueous soils. For instance one of the pre-treatment procedures outlined in the Soil Survey Laboratory Methods Manual is to remove carbonates (Soil Survey Staff, 2004). This is not appropriate in soils where the majority of the parent material is composed of carbonaceous material, such as many of our South Florida Subaqueous Soils. The importance of the appropriate pre-treatment is as important as the technique for measuring PSD (Vassma, 2008).

To determine PSD of soil, the organic matter is removed as to not interfere with the mineral components of the sample. There are some associated challenges with the removal of organic matter depending on the type of pre-treatment and on the sample type. For instance exfoliation of mica, dissolution of manganese dioxide, dissolution of carbonates, dissolution of iron and aluminum and the formation of artifacts such as calcium, aluminum and ferric oxalate which can bind particles together (Gee and Bauder, 1986, Mikhail and Briner, 1978, Anderson, 1961). The most common pre-treatment methods for the removal of organic matter is oxidation. Four frequently used methods of oxidizing organic matter are achieved using hydrogen peroxide (H_2O_2), sodium hypochlorite (NaOCl), disodium peroxdisulfate ($Na_2S_2O_8$), and by combustion or loss on ignition (LOI); each of which have associated problems if to be used for PSDA. Mikutta et al., (2005) reviewed the use and results of these reactants with the mineral and organic constituents of soils, and many procedures have been documented depending on the nature of the soil as well as the determination to be run on the sample.

Of the four treatment types removal of organic matter by LOI and $Na_2S_2O_8$ are not typically used for textural analysis. LOI removes the organic matter through combustion, is quick and effective, but also partially removes carbonates, and damages and aggregates small particles (Vassma, 2008). Disodium peroxdisulfate can take as little as 16 hours for removal of 93% of the organic content, but has been reported that after a 2 day reaction time organic carbon was not sufficiently removed (Mikutta et al., 2005). The use of disodium peroxdisulfate has little to no effect on the structural components of Ca and Mn containing minerals, which eliminates the concern of forming oxalates, though is not practical for PSDA because of the large amounts of reactant needed and the variable reaction time (Mikutta et al., 2005).

The use of H_2O_2 to remove organic matter is the NRCS' standard procedure for the removal of organic matter before running PSDA (Gee and Bauder, 1986). The reaction of H_2O_2 can be highly variable depending on many different factors; for instance the structure of the organic compound, pH, and concentration of the solution (Mikutta et al., 2005). At a pH of 9-10 the reaction only removes 5-20% of

organic carbon, but 50-90% at a pH of 6 and 7.5 (Mikutta et al., 2005). It takes a considerable amount of time sometimes several weeks for H_2O_2 to oxidize the organic matter. H_2O_2 also reacts with Ca forming oxalates, leaving residual carbon which can complex with the surface of other minerals to form silt sized particles (Anderson, 1961, Mikutta et al., 2005). Hydrogen peroxide has also been shown to exfoliate and weather mica, vermiculite, and biotite through the destruction of Mn oxides (Mikutta et al., 2005).

NaOCl is more efficient in the destruction of organic matter and does not form oxalates as readily as with H_2O_2. This procedure using NaOCl was proposed by Anderson (1961) for mineralogical analysis. The sample is heated for 15 minutes with NaOCl then centrifuged at 2000 rpm for a total of three treatments (Anderson, 1961). It is limited to 15 minute intervals because of the relatively fast decomposition of NaOCl at high temperatures (Anderson, 1961; Mikutta et al., 2005). Sodium hypochlorite at a pH of 8-9.5 has been used for mineralogical analysis because of the relatively fast reaction times and less affected by the presence of carbonates (Mikutta et al., 2005; Mikhail and Briner, 1978). It does not dissolve Mn oxides and Fe and Al as readily as with H_2O_2 which eliminates the concern of forming silt sized particles in the sample (Mikutta et al., 2005). NaOCl also acts as a dispersing agent which eliminates the use and extra step of adding $(NaPO_3)_6$ typically added when using H_2O_2 as an oxidant (Omueti, 1980).

There has been considerable work on the abundance of analysis techniques and the methods for pretreatments on a diversity of minerals and physically different soils and sediments. There are also recommendations on what the best and most efficient procedure for the removal of organic matter for PSDA. It seems that sodium hypochlorite is the most efficient and least detrimental option for the removal of organic matter in subaqueous soils for PSDA. However there are concerns that have arisen with the use of NaOCl.

FUTURE RESEARCH

PROBLEM STATEMENT

In preliminary studies performed in the University of Florida Soil and Water Science Department Environmental Pedology Laboratory, most samples required additional spinning which may mechanically form clay sized particles. The procedure also requires the supernatant to be decanted after each treatment. It was found that the supernatant is rarely clear after the first two treatments which could imply the suspension of clay and silt sized particles in the supernatant.

In the preliminary studies the majority of the concerns arose from the Key Largo, FL subaqueous soils. These soils mainly consist of calcium carbonate parent materials. The fine-grained soils are mainly calcium carbonate in the form of aragonite and calcite. In the removal of OM through NaOCl and centrifuging there is concern of forming clay sized particles through physical abrasion from coarser fragments of shell and coral. When the fine-grained soils (silt + clay) are spun the supernatant is rarely clear after centrifuging at 2000 rpms for 5 minutes as recommended (Anderson, 1961). The first step is finding the correct spin speed and time to consistently get a clear supernatant on these soils. With

increased speeds and time intervals in conjunction with the presence of coarse shell and coral fragments physical destruction and abrasion may form smaller sized particles skewing the results of PSDA, showing incorrect clay and silt fractions numbers.

Aragonite and calcite are the two crystallographic forms of calcium carbonate. Aragonite, a polymorph of calcite, has an orthorhombic structure while calcite is trigonal. Unlike calcite, aragonite's carbonate ions lie in two planes pointing in opposite directions where as calcite, the carbonate ions lie on a single plane pointing in the same direction. Aragonite is considered unstable at normal surface temperatures and pressures and will spontaneously convert to calcite at 400 degrees C. Aragonite will still preferentially form if conditions are right, such as the magnesium and salt content and turbidity of the crystallizing fluid. Most bivalves, corals and many sea creatures secrete aragonite for their shells laying them in several layers of aragonite. Most of Florida's subaqueous soils are dominated by quartz, though there are calcium carbonate rich soils in South Florida that are made up of the aragonite form of calcium carbonate (Figure 83 and 84).

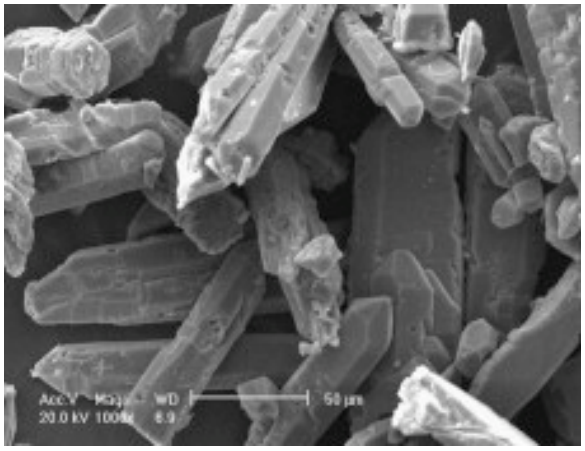

Figure 83. SEM photograph of aragonite Crystals (unknown source).

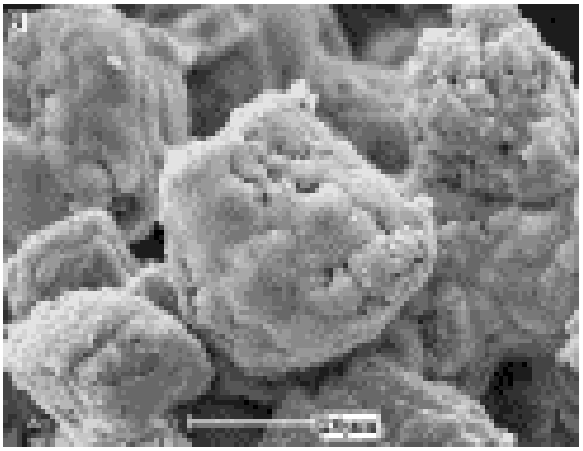

Figure 84. SEM photograph of calcite crystals (unknown source)

OBJECTIVES AND HYPOTHESES

Objective 1: To find the correct spin velocity and duration to obtain a clear supernatant for two South Florida subaqueous soil types; calcium carbonate rich and quartz dominated soils for the analysis of PSD.

Objective 2: Compare the percent clay of two naturally occurring subaqueous soil types. I will determine if spin time and speed mechanically produce clay in subaqueous soils of quartz versus carbonaceous parent material.

Hypothesis 1

RPM and duration of speed will have to be increased in order to get a clear supernatant free of suspended particles.

Rationale: South Florida subaqueous soils tend to have significant amounts of fine textured (silt + clay) particles easily dispersed into the water column. At higher velocities the particles will flocculate more effectively leaving a clear supernatant and minimal suspension of soil material.

Hypothesis 2

The carbonaceous soils will have an increase in percent clay after treatment, while the quartz soils will not be affected.

Rationale: Aragonite crystals preferentially form in the carbonaceous South Florida subaqueous soils. Aragonite is of orthorhombic structure forming acicular needles making aragonite less stable and more susceptible to comminution. Quartz is hard and characteristically spheroidal making it more physically stable mineral, therefore not being as affected.

METHODS

Use a modified version of the procedure for moist soil samples, as well as the Pipette method from the NRCS Soil Survey Laboratory Methods Manual. The modifications are the use of NaOCl instead of Hydrogen Peroxide to avoid the creation of oxalates.

Both soil types will be centrifuged at different velocities and time durations until a clear supernatant can be consistently produced. The spinning will begin at the current procedure recommendation of 2000 rpms for 5 minutes, then increased in time in five minute intervals to a maximum of 15 minutes and increase rpms in 1000 rpm intervals to a maximum of 10,000 rpms with each time duration or until we can consistently get a clear supernatant indicating there are no particles in suspension.

Once the spin time and velocity have been found for our samples, run the procedure for PSDA, including treatment with sodium hypochlorite, and shaking with sodiumhexametaphosphate. Compare percent silt and clay, as well as the silt to clay ratios of the subaqueous soils with differing parent materials, calcium carbonate and quartz. Analyze a total of 30 treatments silt and clay fractions (Table 24)

Table 24. Outline of treatments

	5 minutes			10 minutes			15 minutes		
2000 rpm	Q	Q	Q	Q	Q	Q	Q	Q	Q
	C	C	C	C	C	C	C	C	C
3000 rpm	Q	Q	Q	Q	Q	Q	Q	Q	Q
	C	C	C	C	C	C	C	C	C
4000 rpm	Q	Q	Q	Q	Q	Q	Q	Q	Q
	C	C	C	C	C	C	C	C	C
5000 rpm	Q	Q	Q	Q	Q	Q	Q	Q	Q
	C	C	C	C	C	C	C	C	C
6000 rpm	Q	Q	Q	Q	Q	Q	Q	Q	Q
	C	C	C	C	C	C	C	C	C
7000 rpm	Q	Q	Q	Q	Q	Q	Q	Q	Q
	C	C	C	C	C	C	C	C	C
8000 rpm	Q	Q	Q	Q	Q	Q	Q	Q	Q
	C	C	C	C	C	C	C	C	C
9000 rpm	Q	Q	Q	Q	Q	Q	Q	Q	Q
	C	C	C	C	C	C	C	C	C
10000 rpm	Q	Q	Q	Q	Q	Q	Q	Q	Q
	C	C	C	C	C	C	C	C	C
Control (No Treatments)	Q	Q	Q	Q	Q	Q	Q	Q	Q
	C	C	C	C	C	C	C	C	C

Q	Quartz Soil
C	Calcium Carbonate Soil

A control, where no pretreatments or spinning will included; one of the calcium carbonate soil and one of the quartz soil done in triplicates. Samples of quartz parent material at each RPM speed and time interval, included in triplicates. Samples of calcium carbonate parent material at each rpm speed and time interval, also in triplicates. All samples except for controls will be treated with sodiumhexametaphosphate and NaOCl.

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CHAPTER 8: CONCLUSIONS

Assessing the success of coastal restoration and mitigation sites is challenging because spatial patterns can escape traditional transect-based and random site monitoring. The findings presented in these studies reveals the site complexity that can exist and the utility of a spatial monitoring approach for quantifying those spatial patterns.

In general, the existing soil and vegetation at Lake Surprise was not impacted negatively. Most vegetative and soil parameters measured indicated no change. Seagrass density, namely *Thalassia*, did steadily increase over the monitoring period. This is could possibly be attributed to increased exchange with the outside Florida Bay. This exchange was facilitated by the removal of the US 1 causeway. Therefore it is arguable that the causeway removal has improve the seagrass community in Lake Surprise. Given that the shoot density increase occurred in *Thalassia*, the climax species, it is also arguable that these changes indicate a more permanent improvement.

In contrast, the vegetative change at SL-15 was rapid in some areas. Where Spartina was planted, growth was explosive during the first two years. Following that growth, natural recruitment of mangroves set the stage for mangrove density, height, and canopy volumes that exceeded areas where mangroves were planted. Since this occurred within the initial five year permit monitoring period, it is possible that natural recruitment could be substituted for planting.

In the seagrass recruitment area, natural colonization of the bottom has not occurred to any large extent. Seagrass are slow growing so it is possible that in 10 or 20 years the seagrass coverage will be greater. Anecdotal observations of turbidity on windy days and water levels on low tides suggested that the flushing channels may not be wide and deep enough to allow consistent seagrass growth.

Soil formation at SL-15 is slow and steady. Organic matter (OM) is accumulating, but the trajectory is uncertain. Initial results suggested that sedimentation in the seagrass recruitment area has occurred. The soils of the mangrove planter are still largely dominated by carbonates. Areas of natural recruitment have pockets of finer material that is higher in OM, which most likely is an elevated source of nutrients for the mangroves. This could explain the greater mangrove growth observed in these areas. While the added nutrients in the seagrass recruitment area should facilitate seagrass growth, this has not occurred.

The seagrass transplant study suggested that soil phosphorus is the most important property to consider in areas of new recruitment. We cannot currently recommend soil phosphorus amendments as that would require an experiment. However, we hypothesize that burial of nutrients would greatly facilitate new seagrass growth.

The literature review and proposed experiment on determining particle size would benefit efforts to assess baseline soil conditions in future seagrass restoration areas.

The activities at Lake Surprise suggested that causeway removal can have a positive effect on seagrass growth, growth of naturally recruited vegetation can exceed that of planted vegetation, and that spatial monitoring can provide a superior view of restoration areas compared to transect or random monitoring.

APPENDICIES

APPENDIX A: SUPPLEMENTAL FIGURES AND TABLES FOR CHAPTER 3

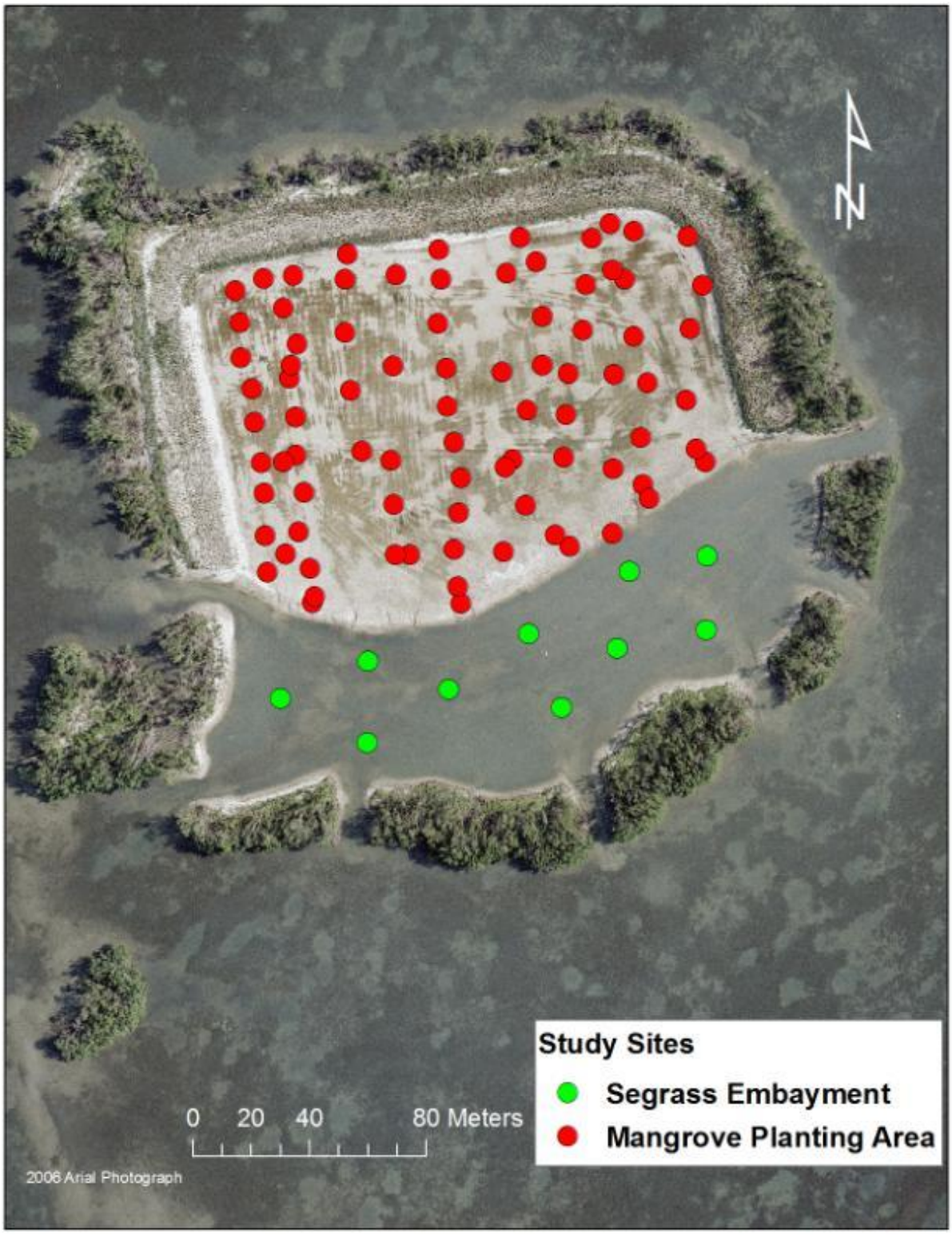

Figure 85. Study sites within the mangrove planting area and seagrass embayment.

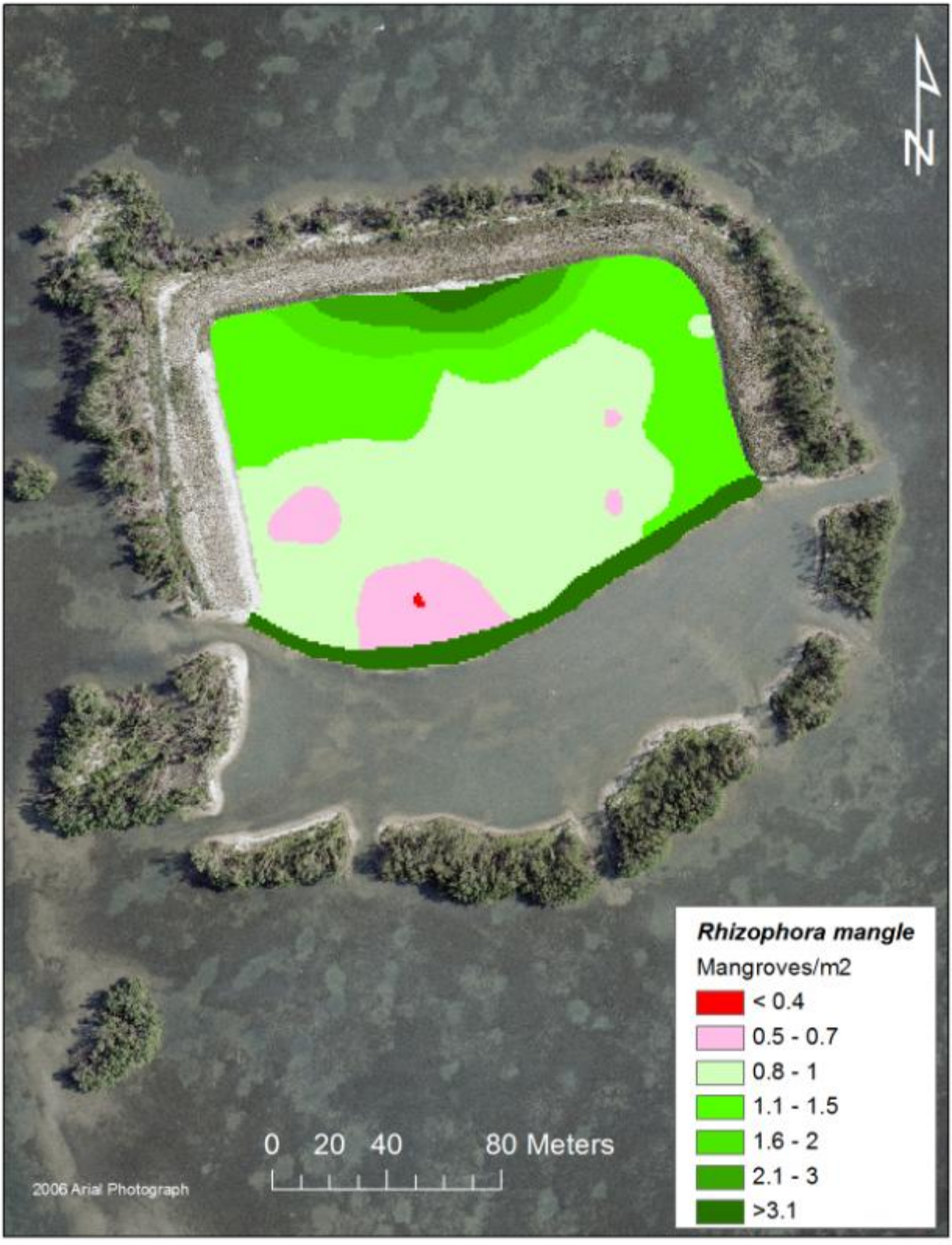

Figure 86. *R. mangle* density in the mangrove planter for summer 2009.

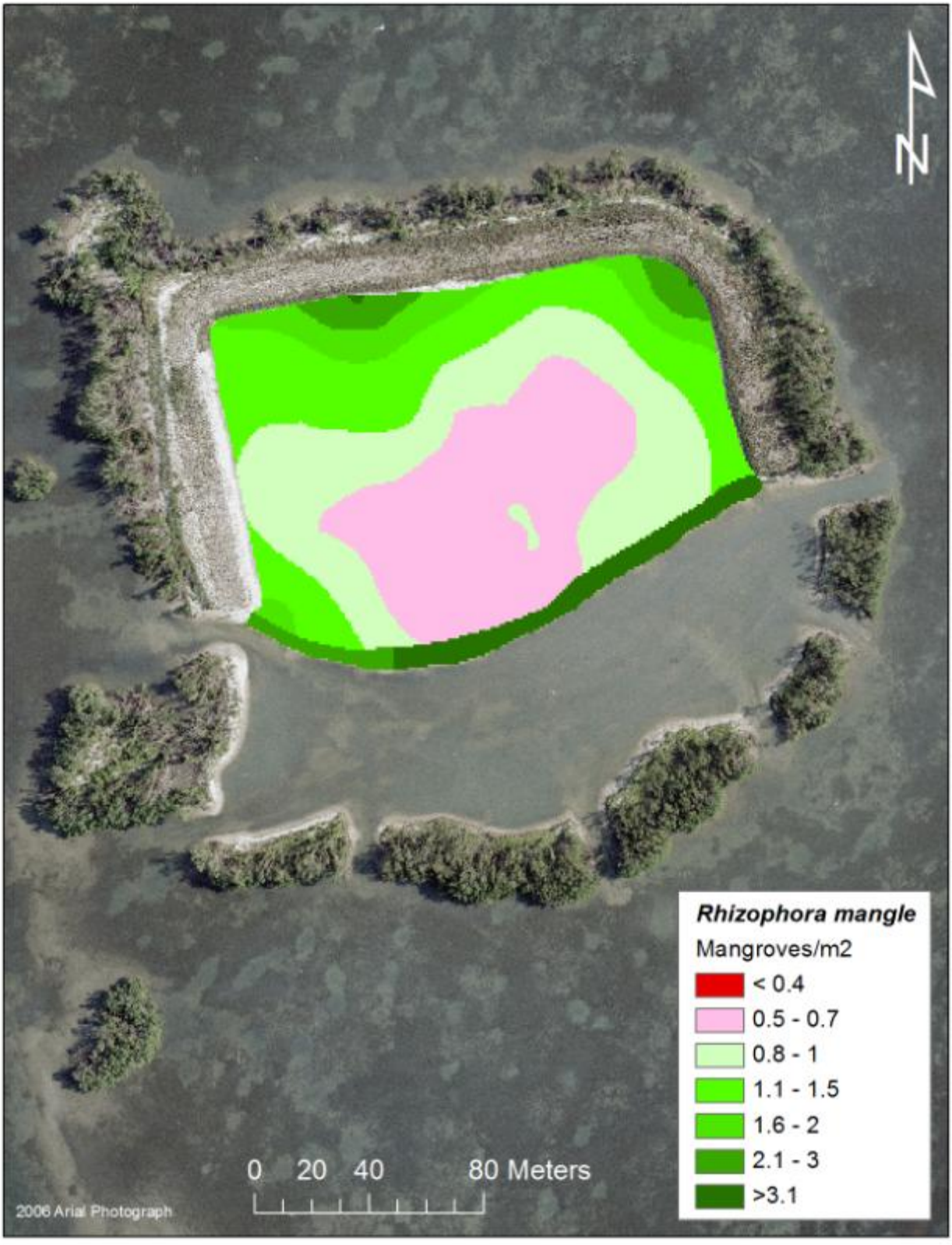

Figure 87. *R. mangle* density in the mangrove planter for winter 2009.

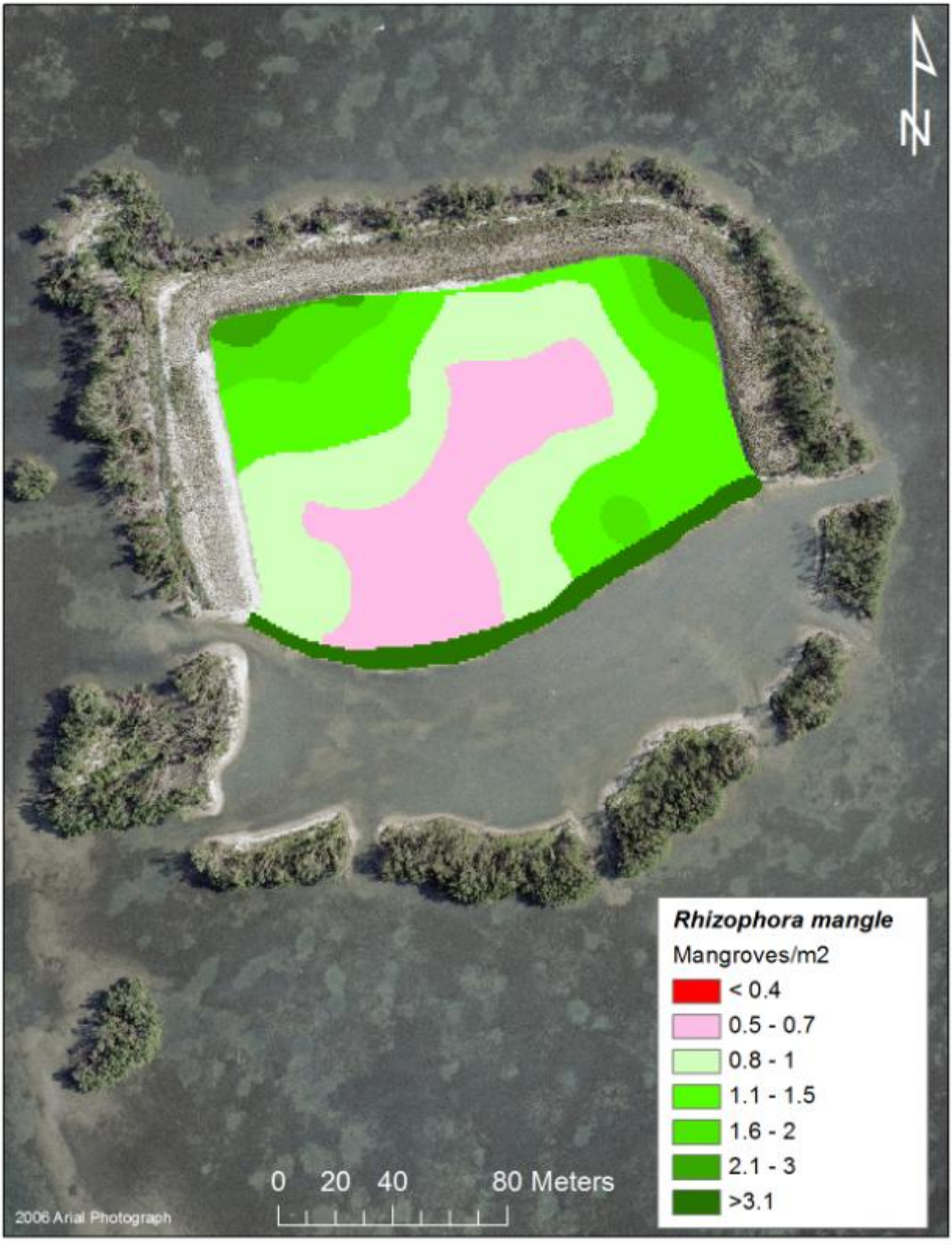

Figure 88. *R. mangle* density in the mangrove planter for summer 2010.

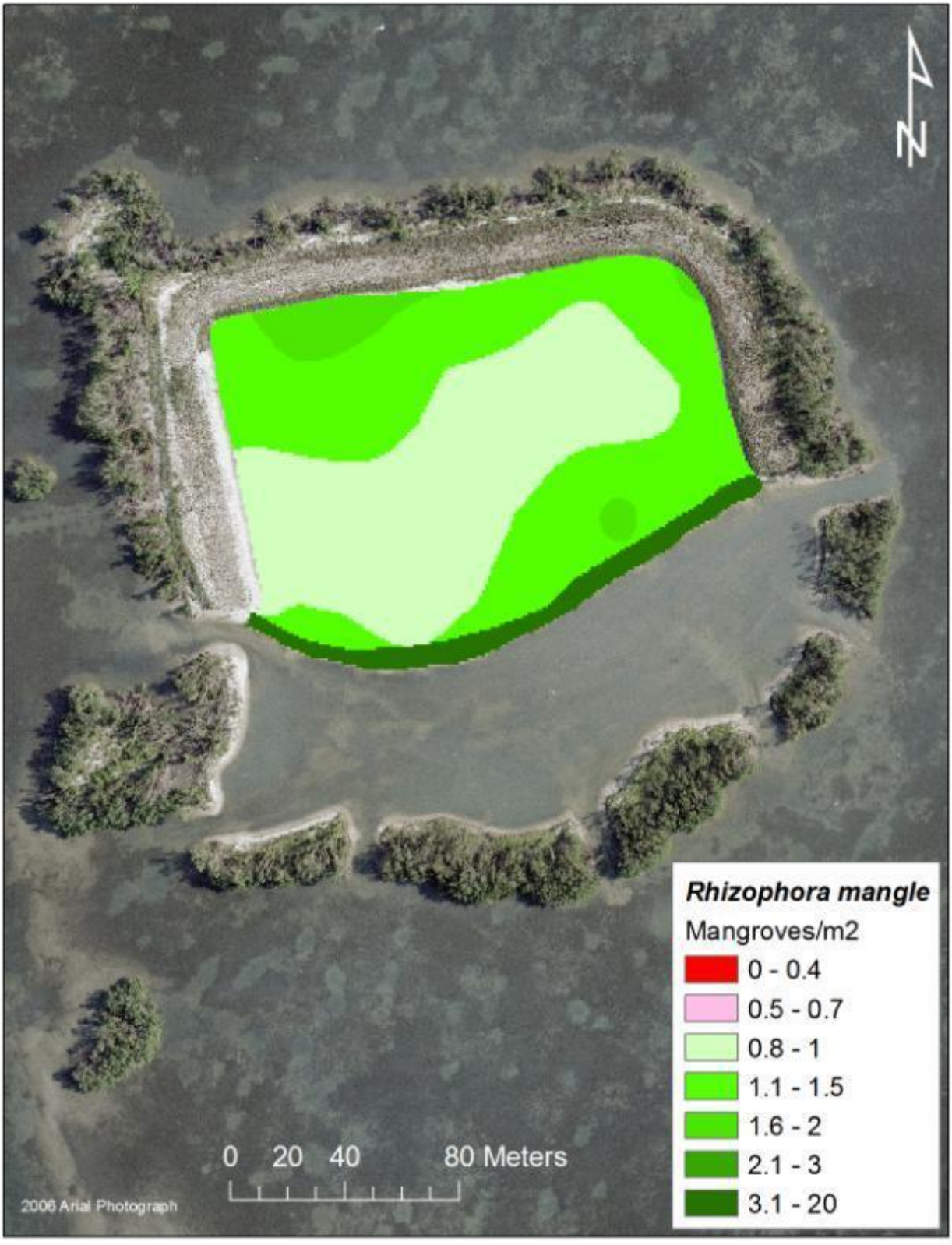

Figure 89. *R. mangle* density in the mangrove planter for winter 2010.

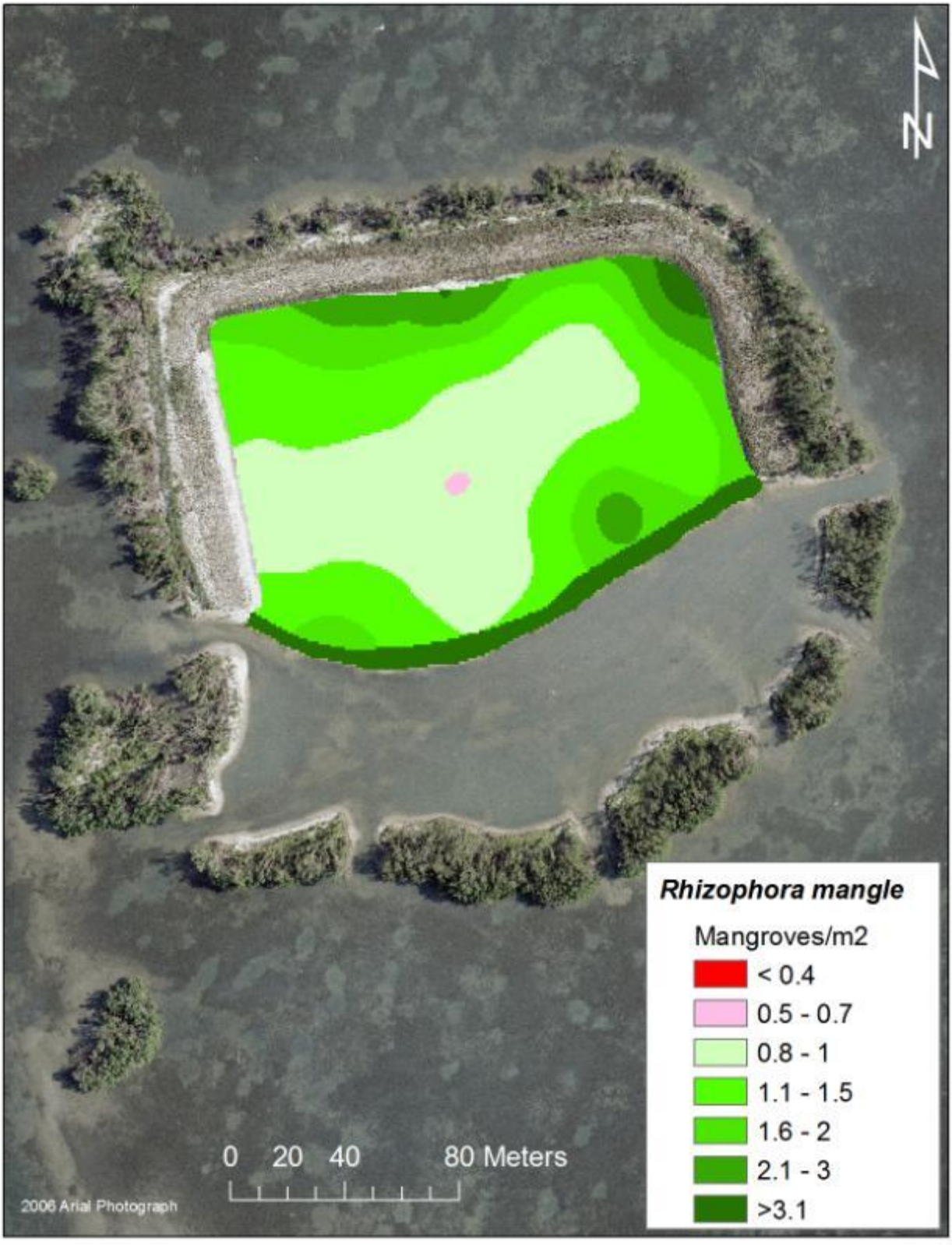

Figure 90. *R. mangle* density in the mangrove planter for summer 2011.

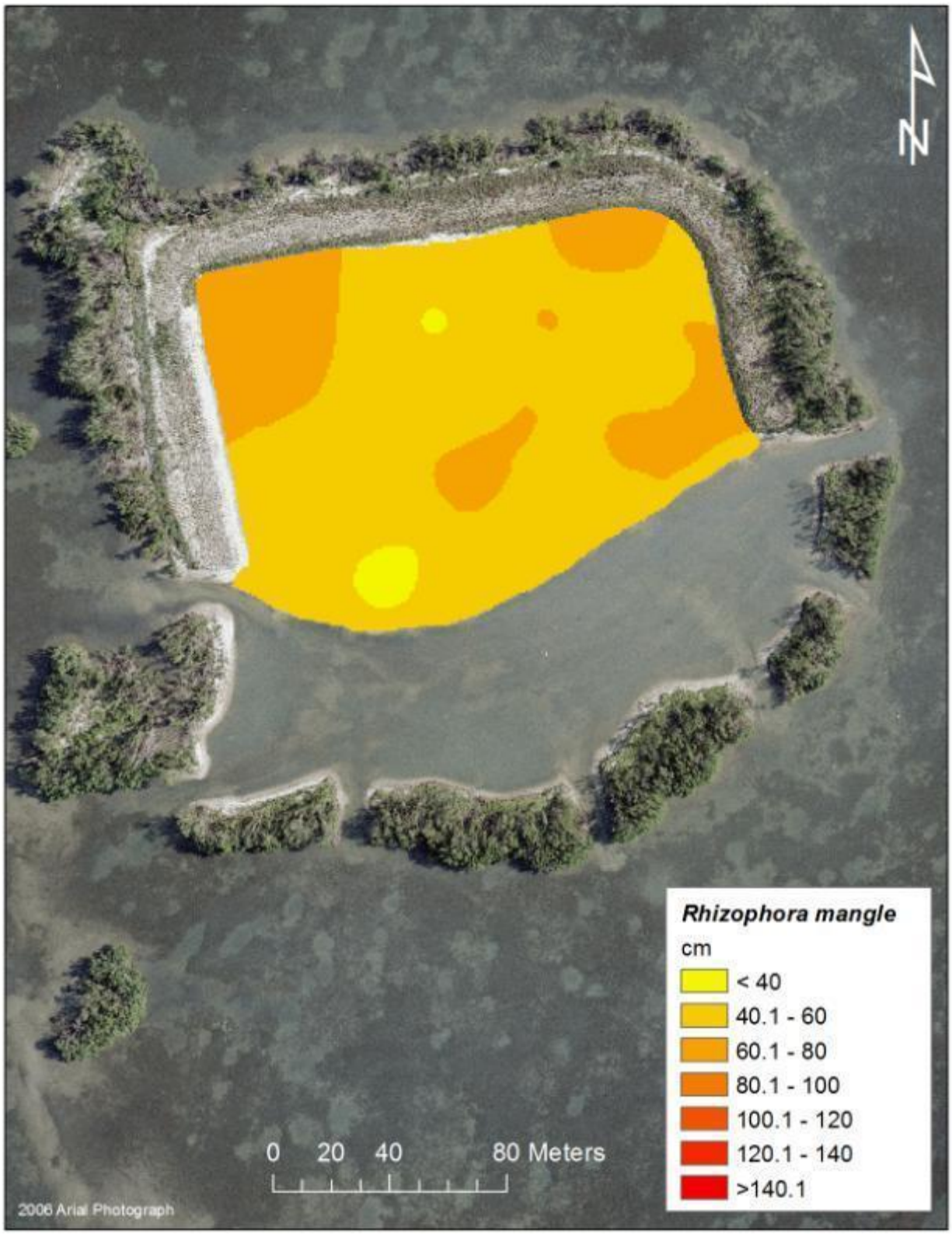

Figure 91. *R. mangle* average mangrove height in summer 2009.

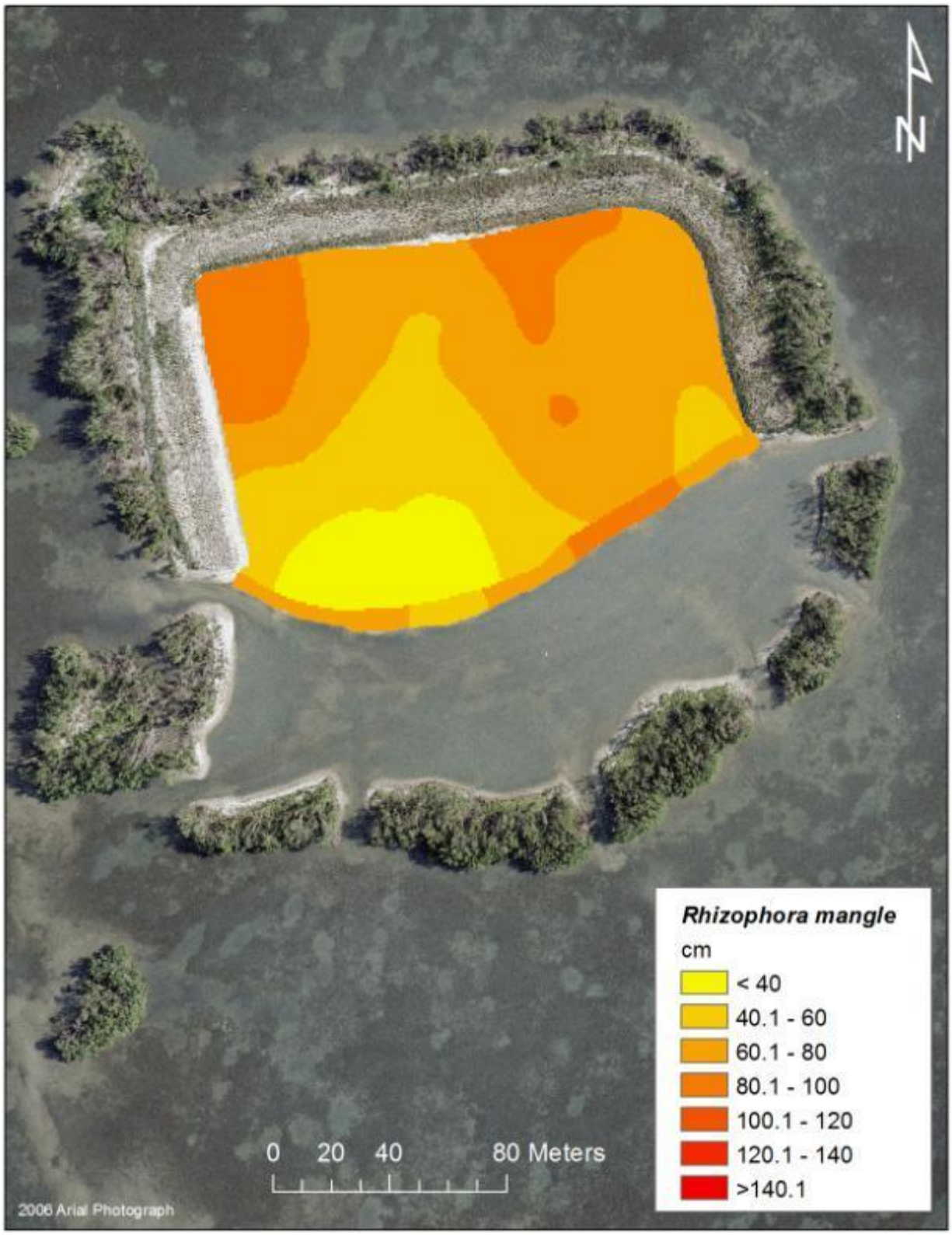

Figure 92. *R. mangle* average mangrove height in winter 2009.

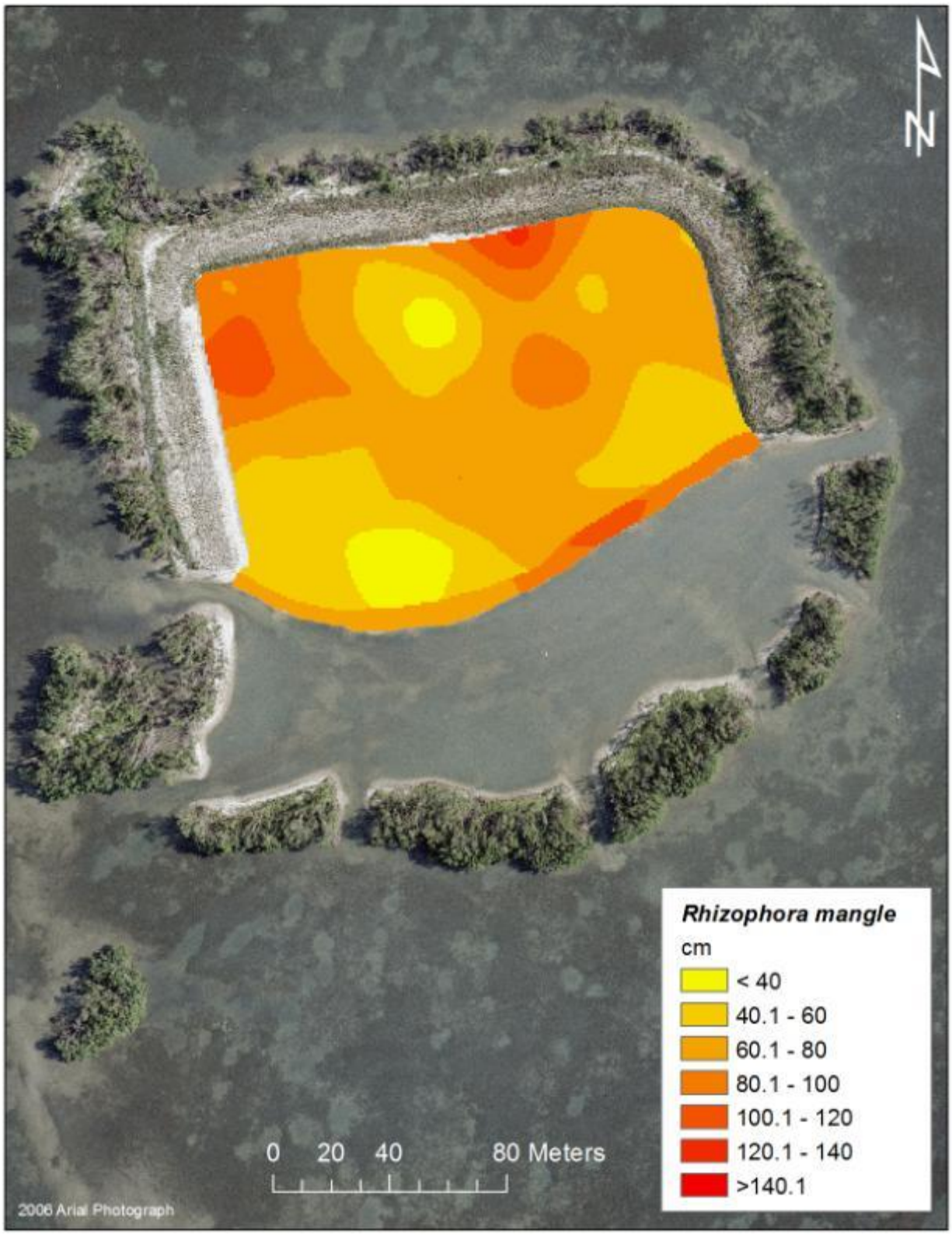

Figure 93. *R. mangle* average mangrove height in summer 2010.

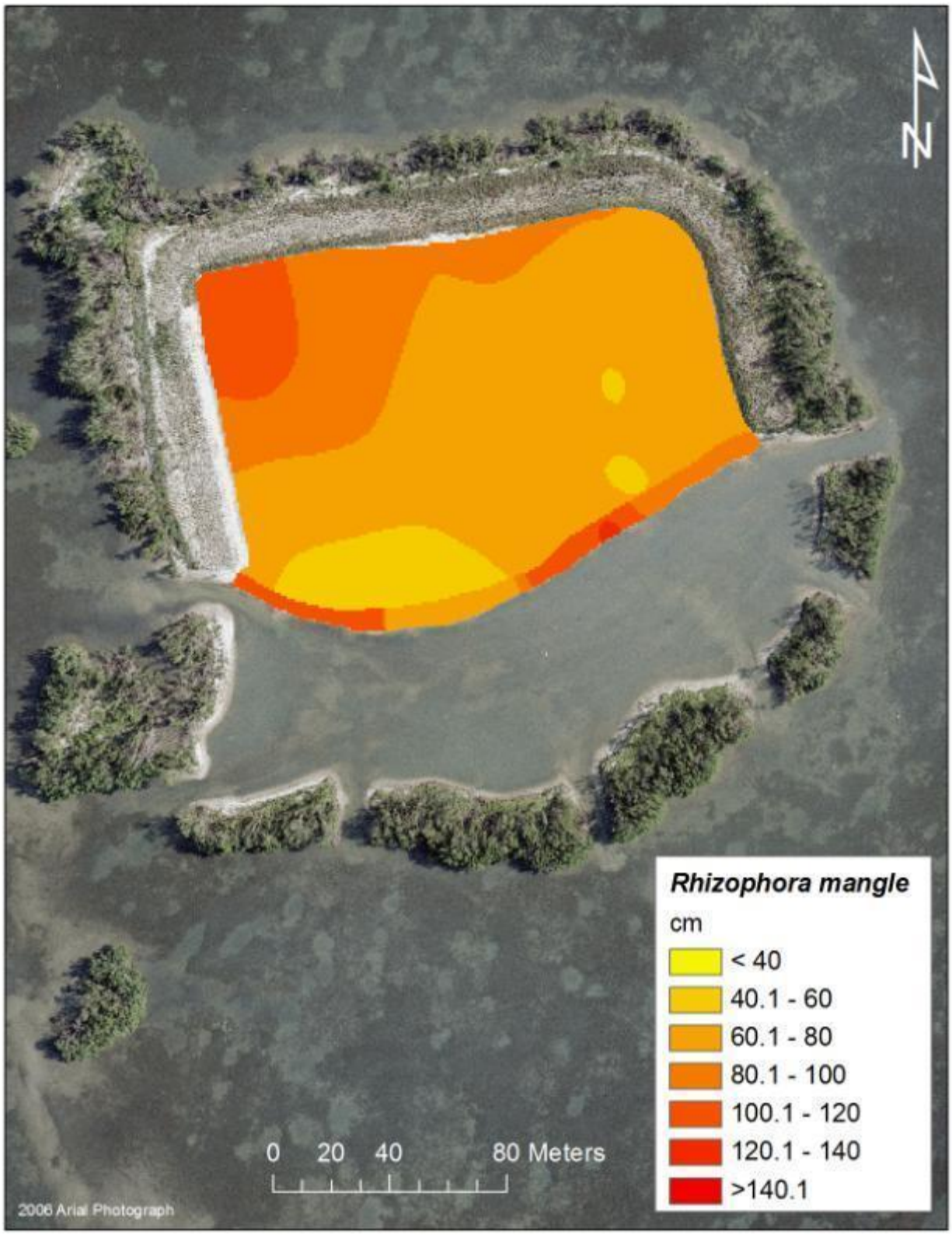

Figure 94. *R. mangle* average mangrove height in winter 2010.

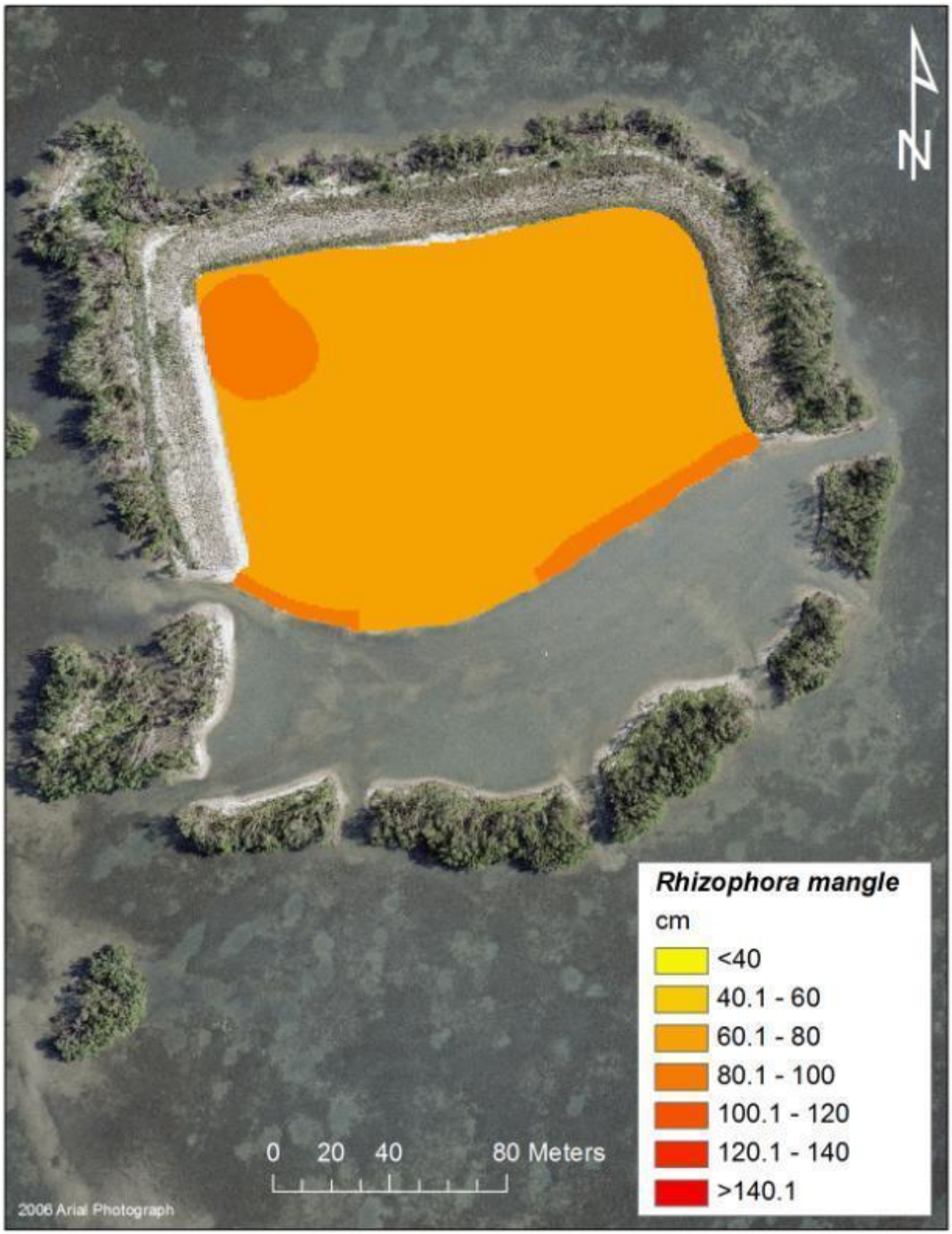

Figure 95. *R. mangle* average mangrove height in summer 2011.

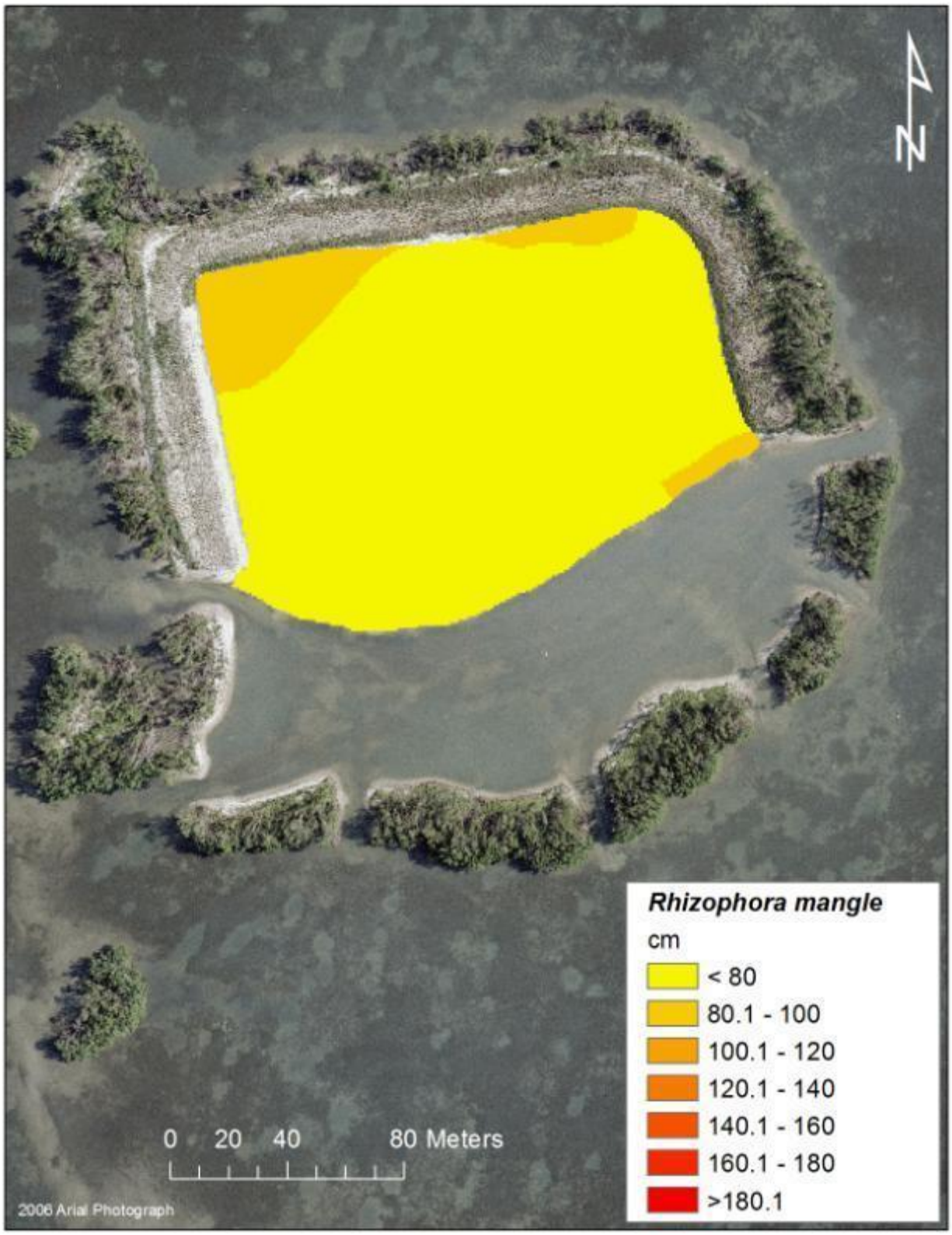

Figure 96. *R. mangle* maximum mangrove height in summer 2009.

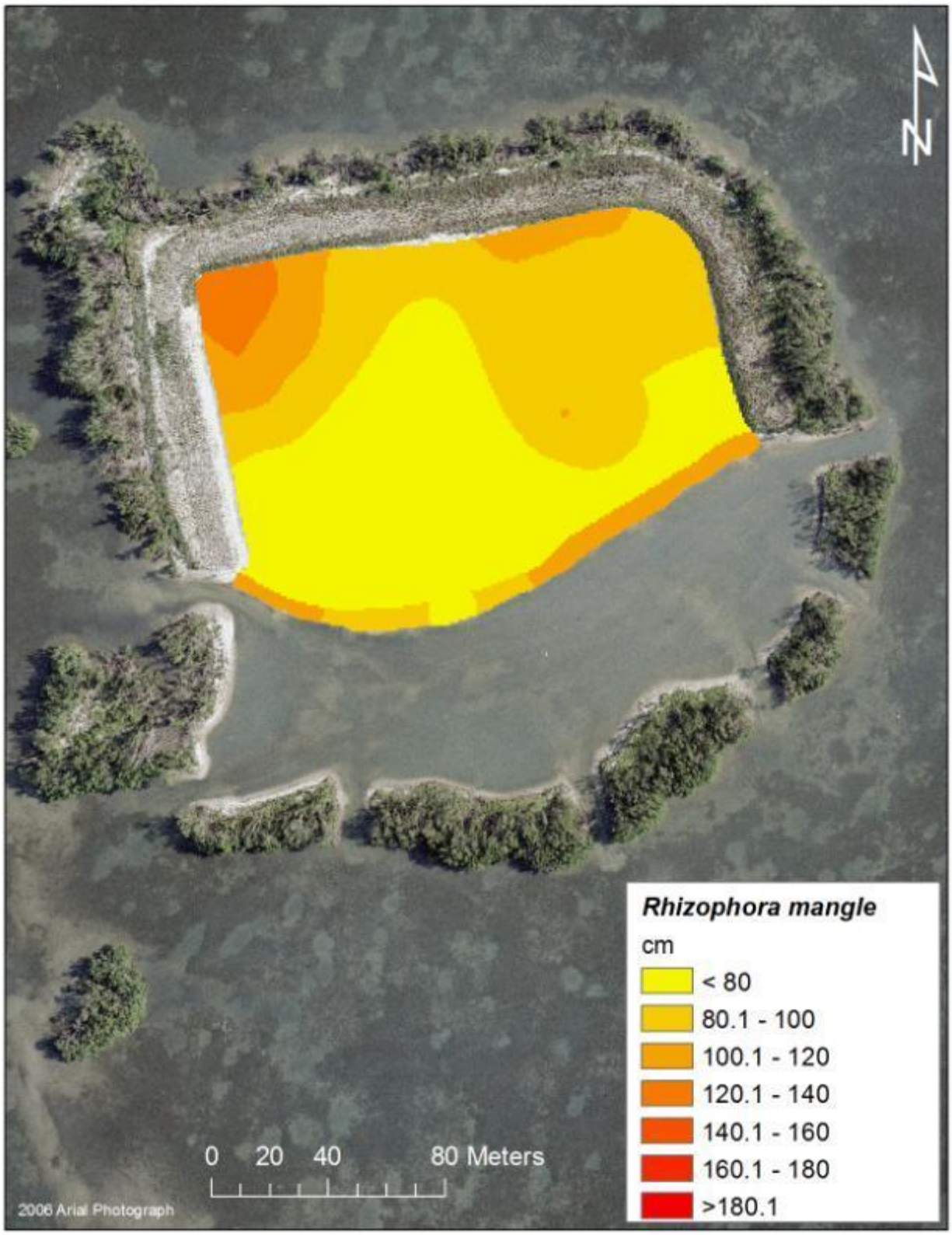

Figure 97. *R. mangle* maximum mangrove height in winter 2009.

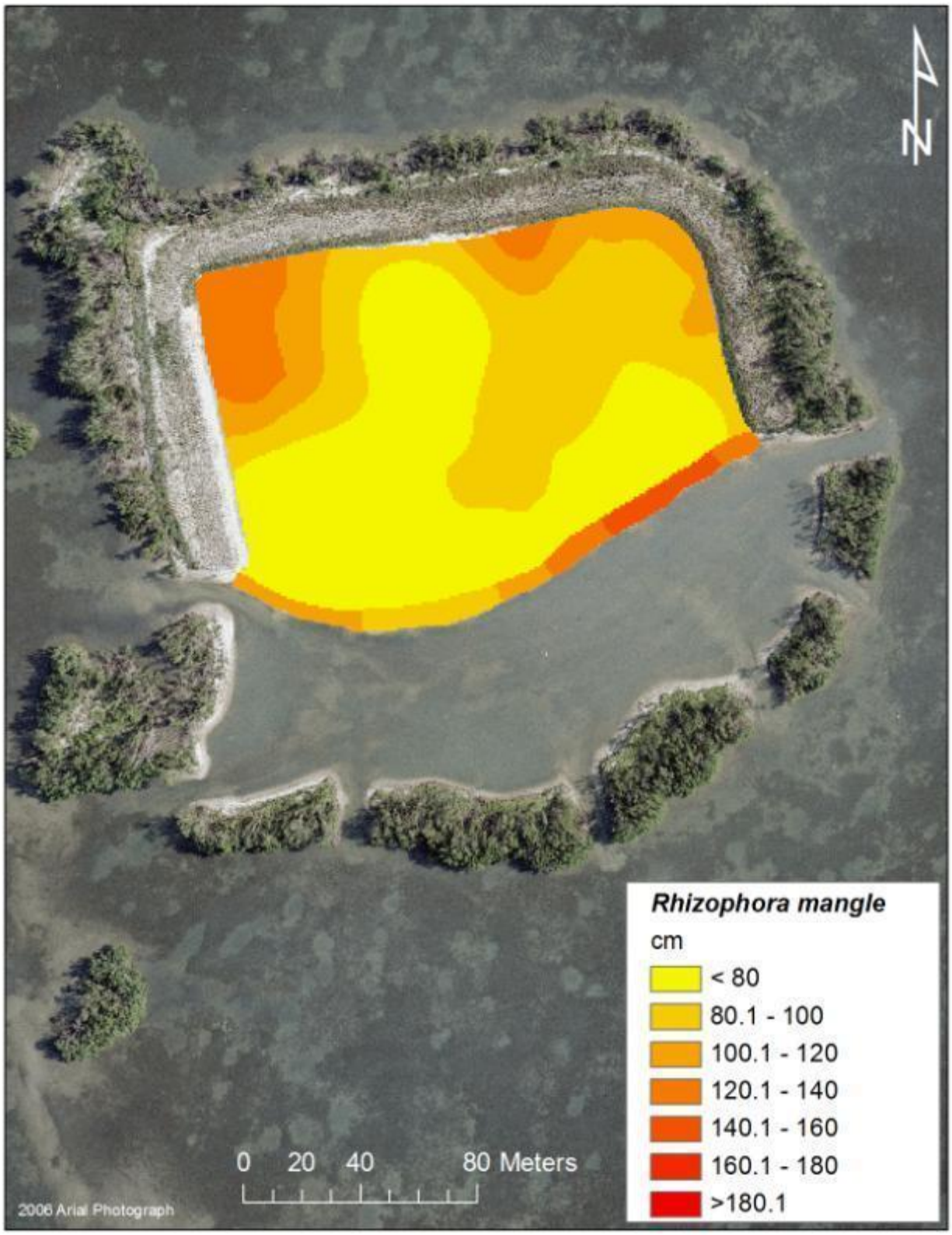

Figure 98. *R. mangle* maximum mangrove height in summer 2010.

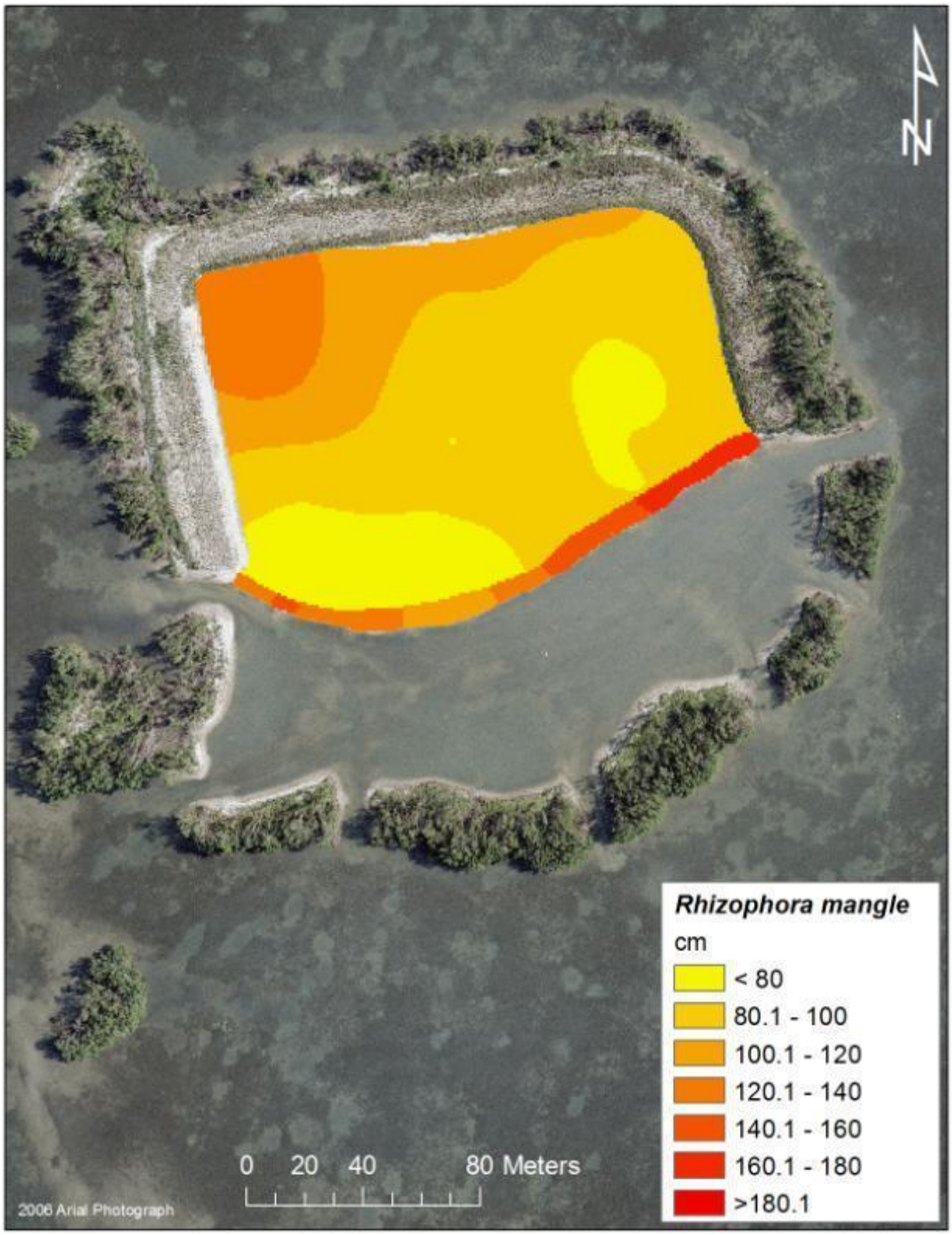

Figure 99. *R. mangle* maximum mangrove height in winter 2010.

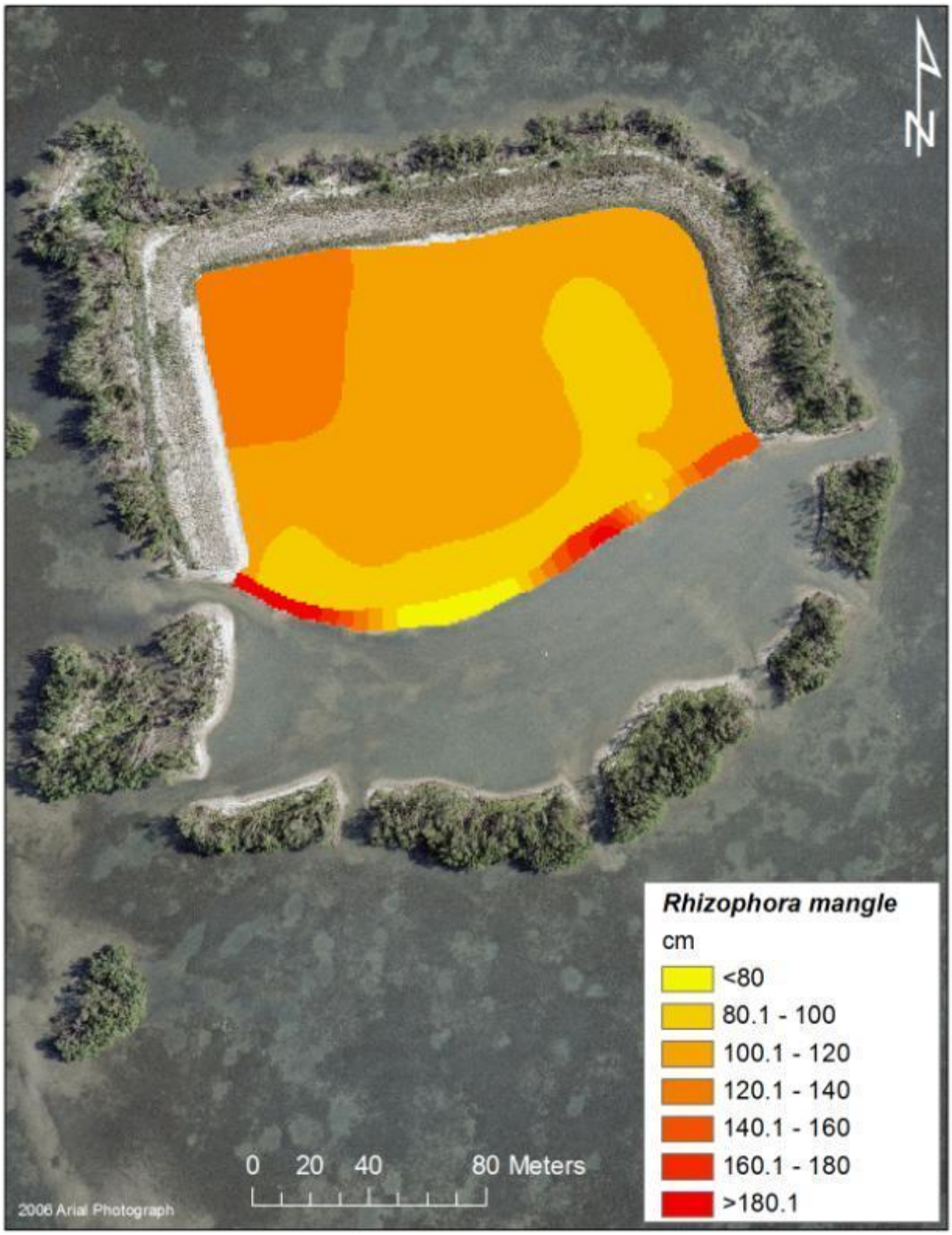

Figure 100. *R. mangle* maximum mangrove height in summer 2011.

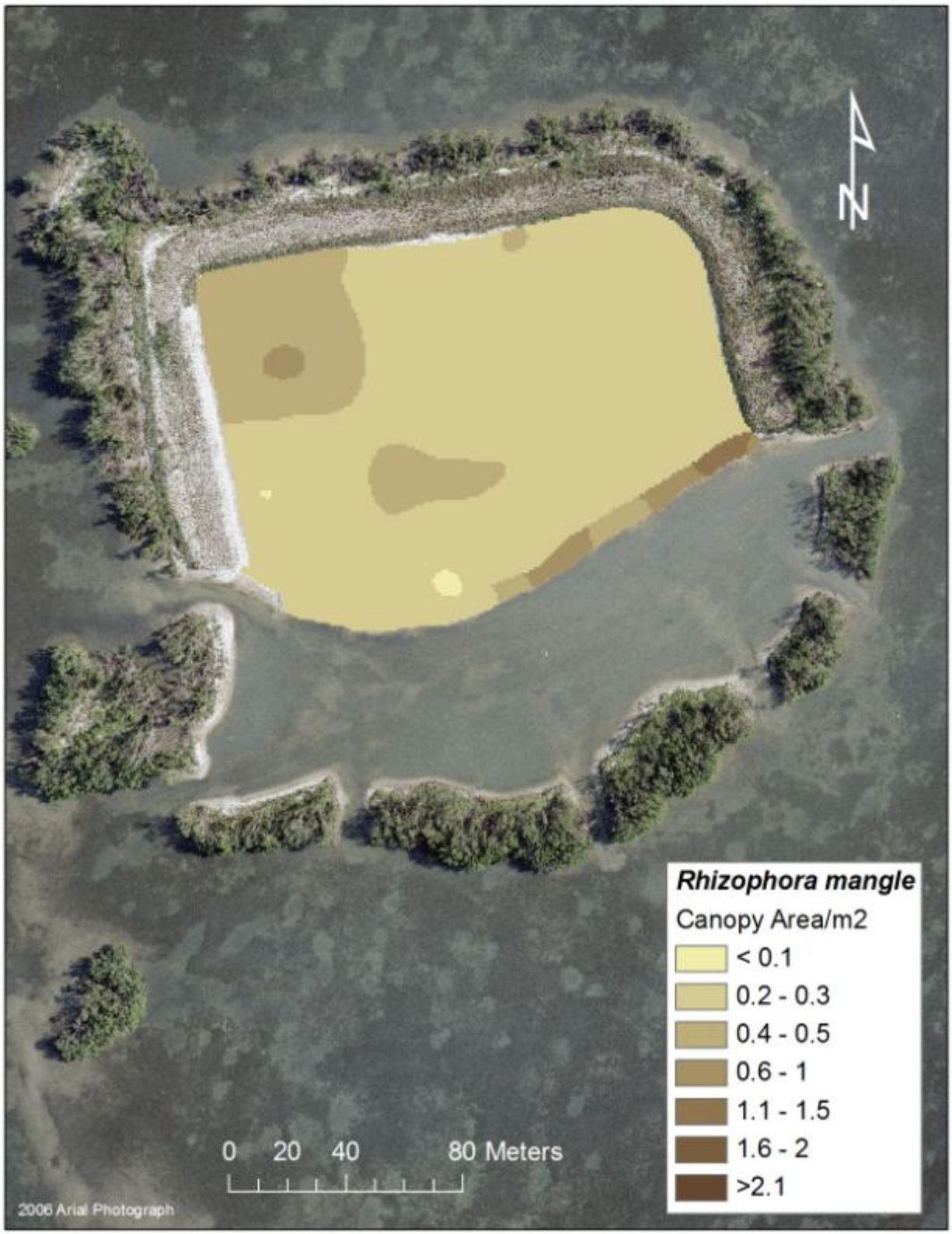

Figure 101. *R. mangle* calculated canopy area/m2 for summer 2011.

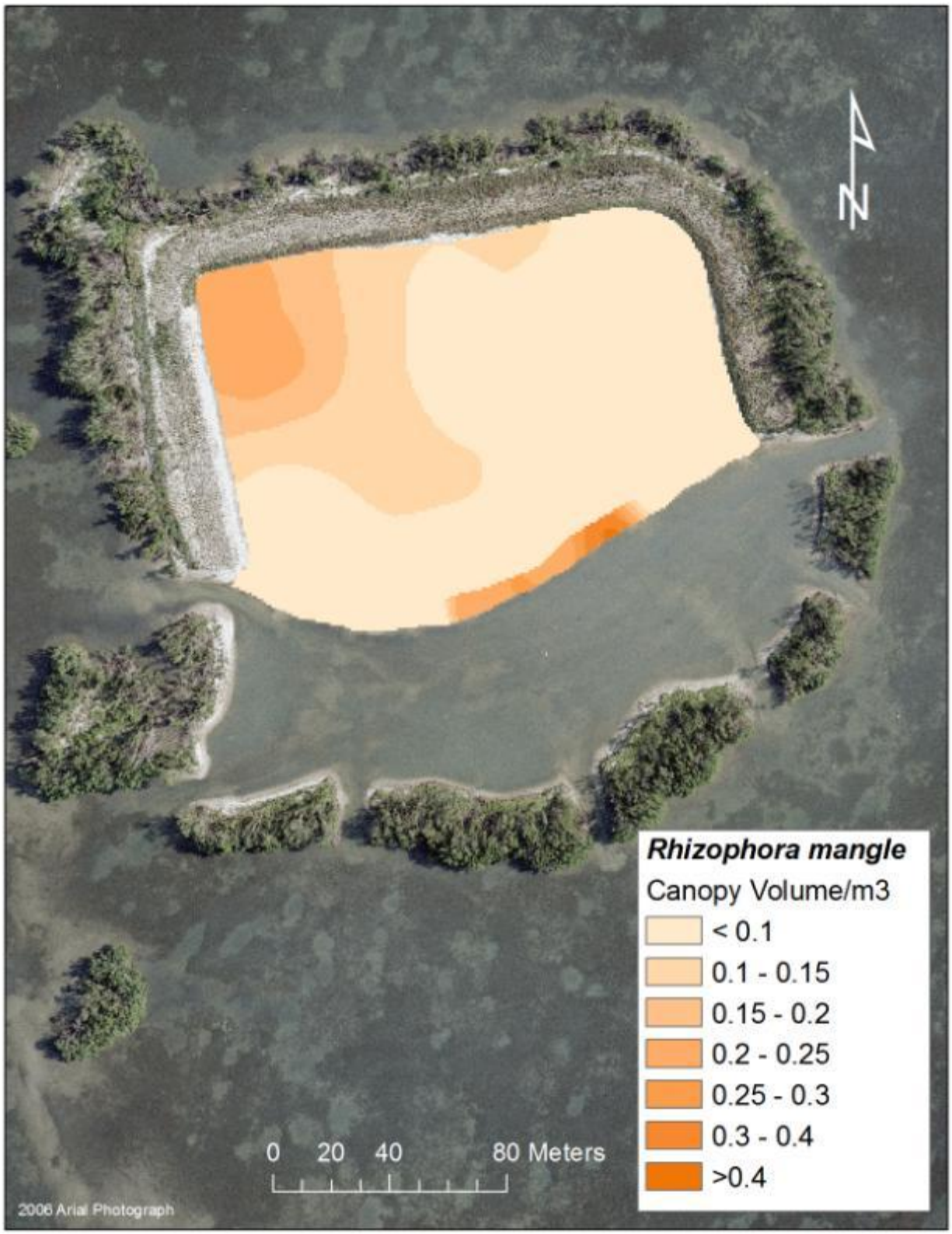

Figure 102. *R. mangle* calculated volume area/m3 for summer 2011.

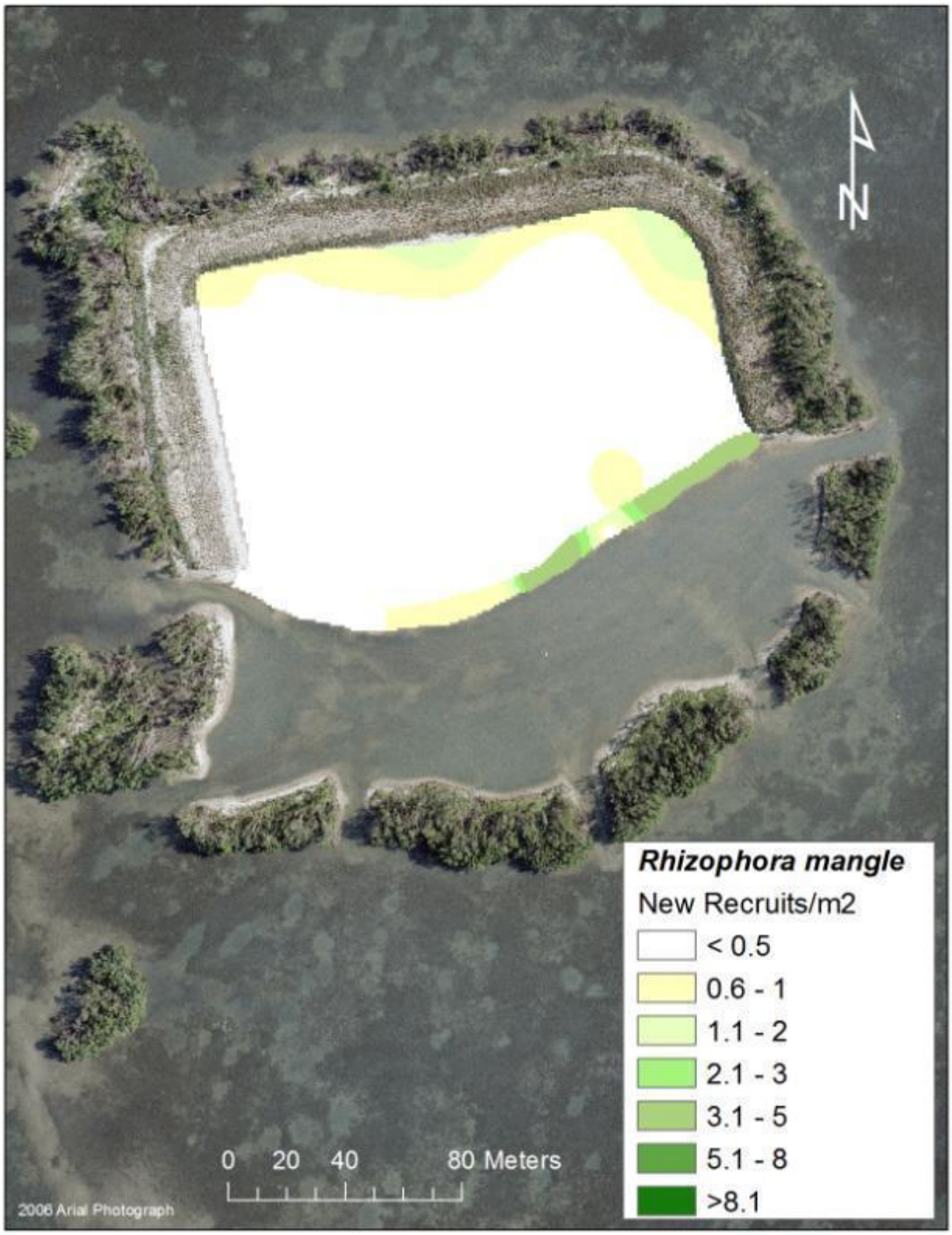

Figure 103. *R. mangle* juvenile new recruits in summer 2011.

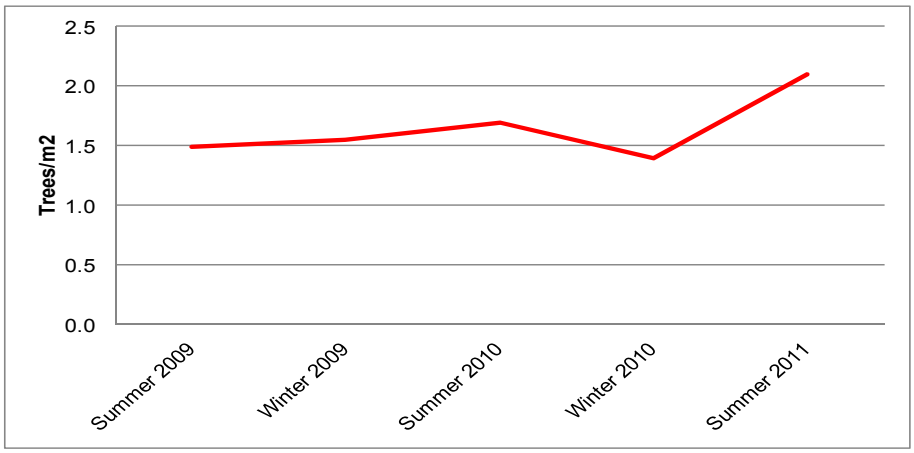

Figure 104. Average *R. mangle* density (trees/m2) from summer 2009 to summer2011.

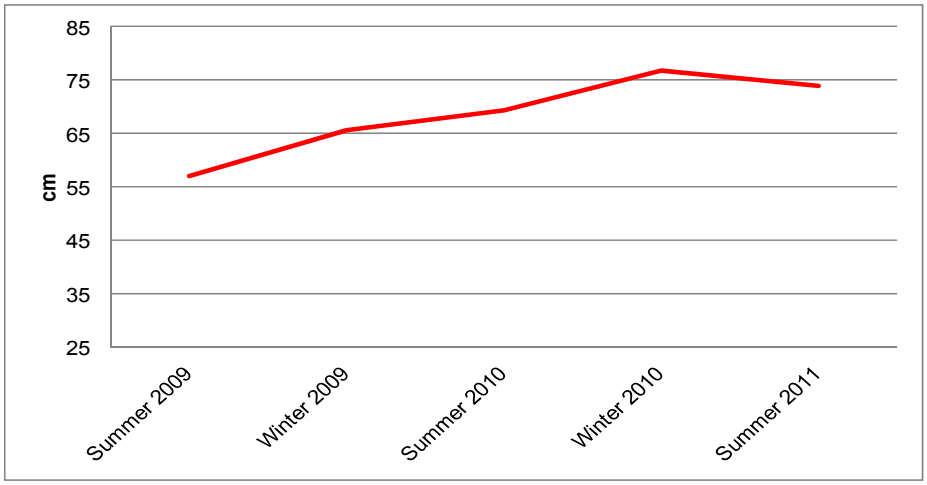

Figure 105. Average *R. mangle* height from summer 2009 to summer2011.

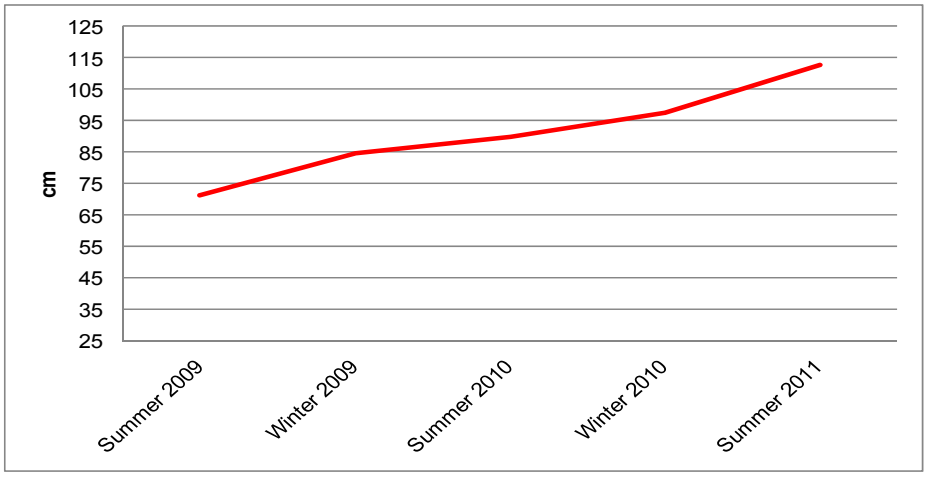

Figure 106. Average *R. mangle* maximum height from summer 2009 to summer2011.

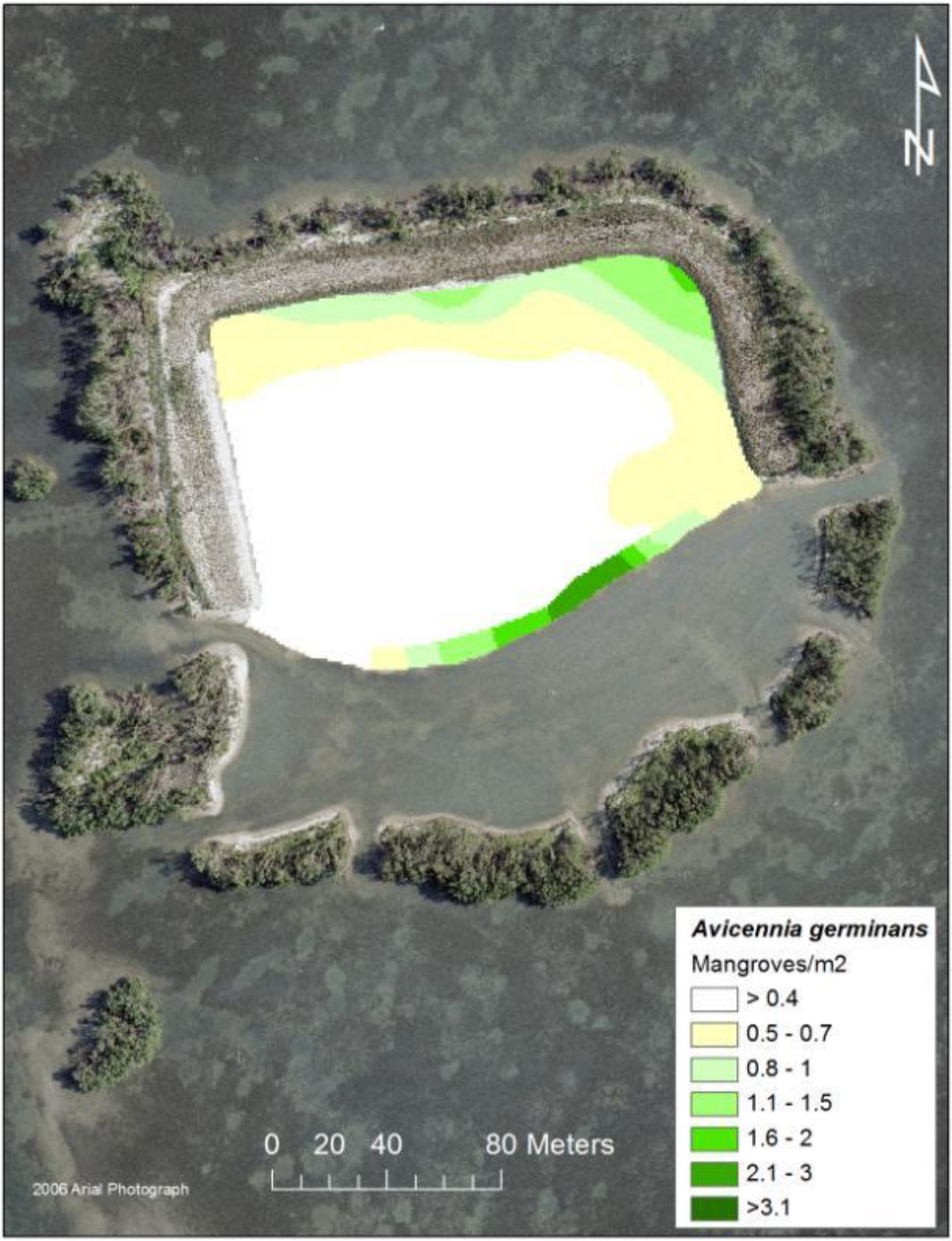

Figure 107. *A. germinans* density in the mangrove planter for summer 2009.

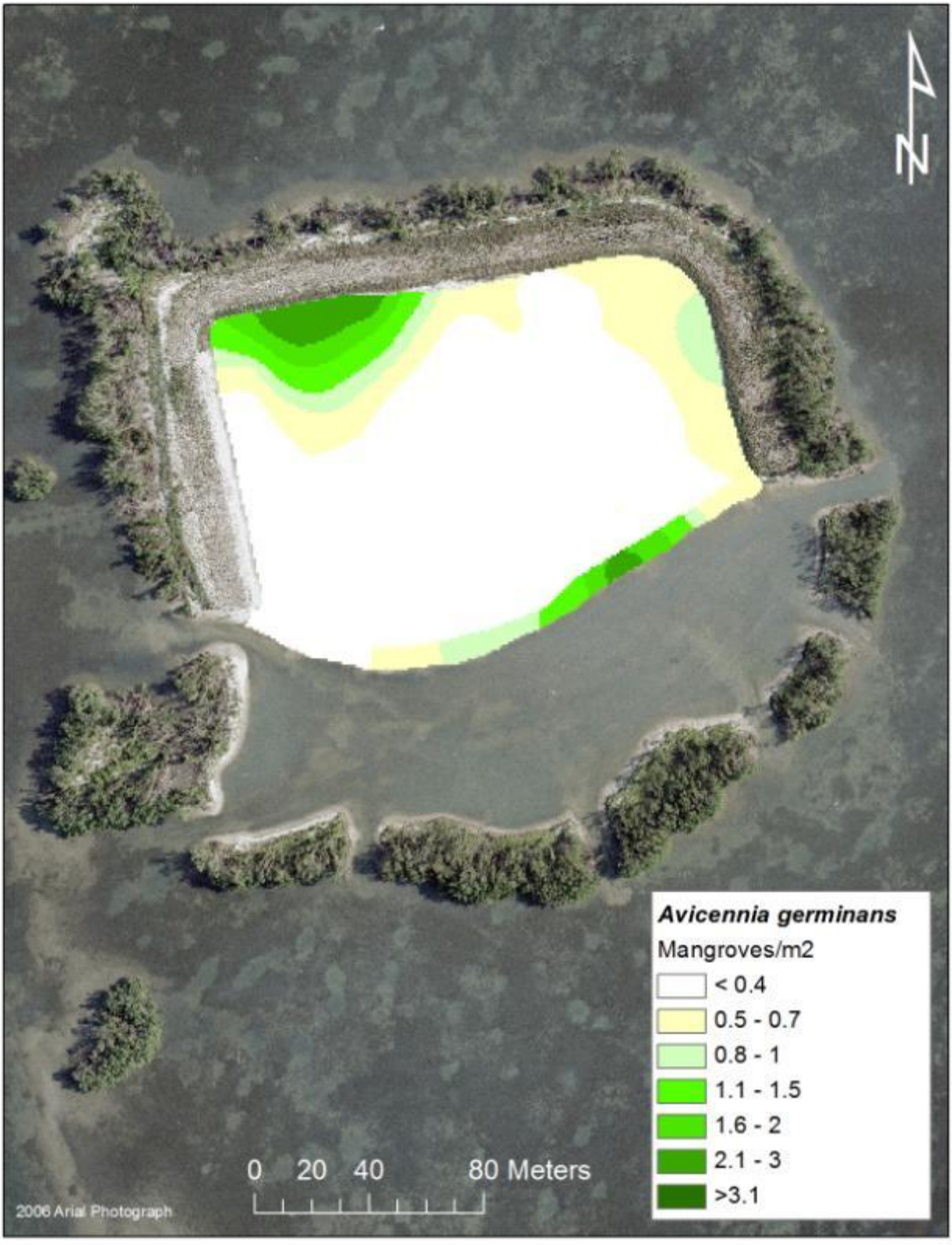

Figure 108. *A. germinans* density in the mangrove planter for winter 2009.

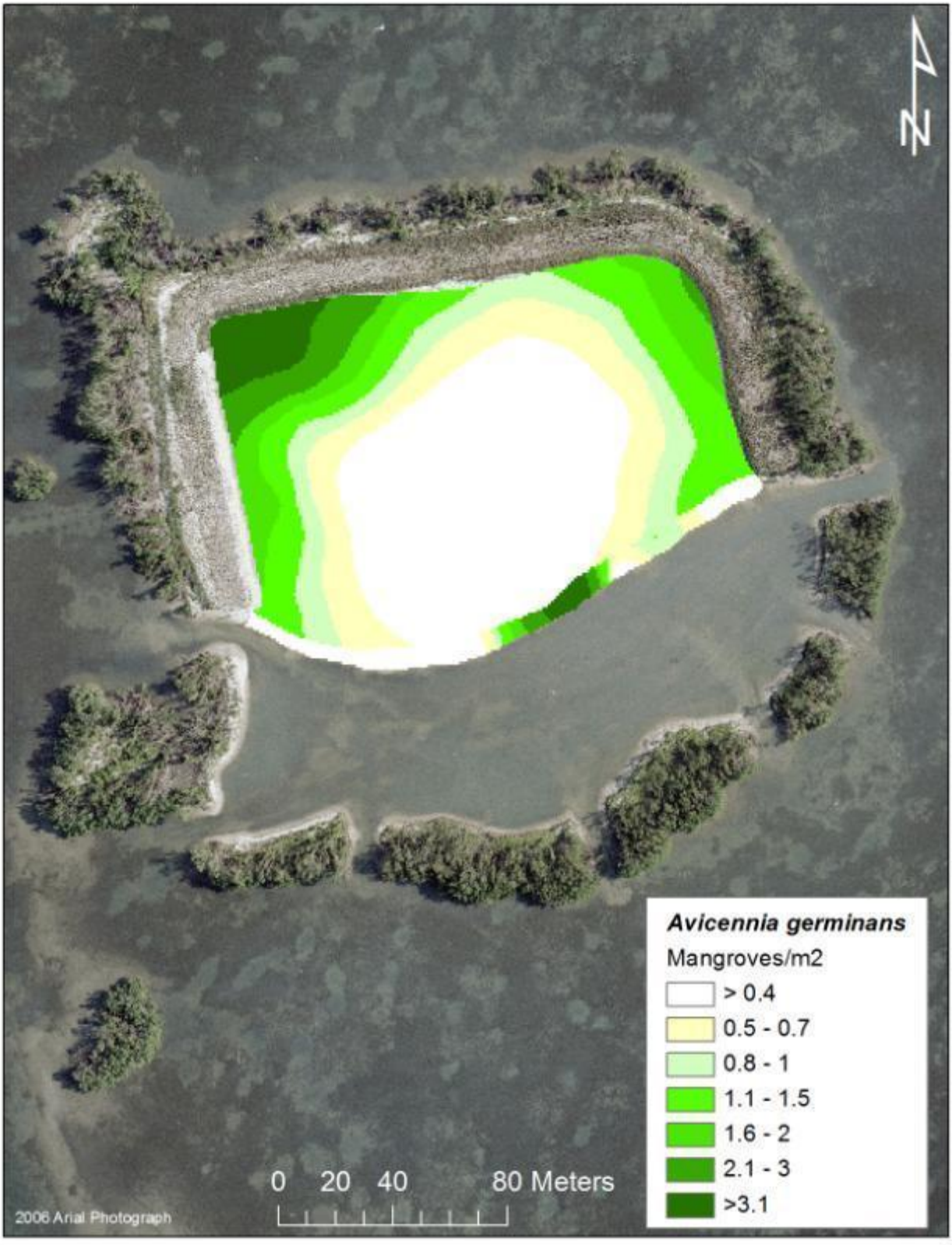

Figure 109. *A. germinans* density in the mangrove planter for summer 2010.

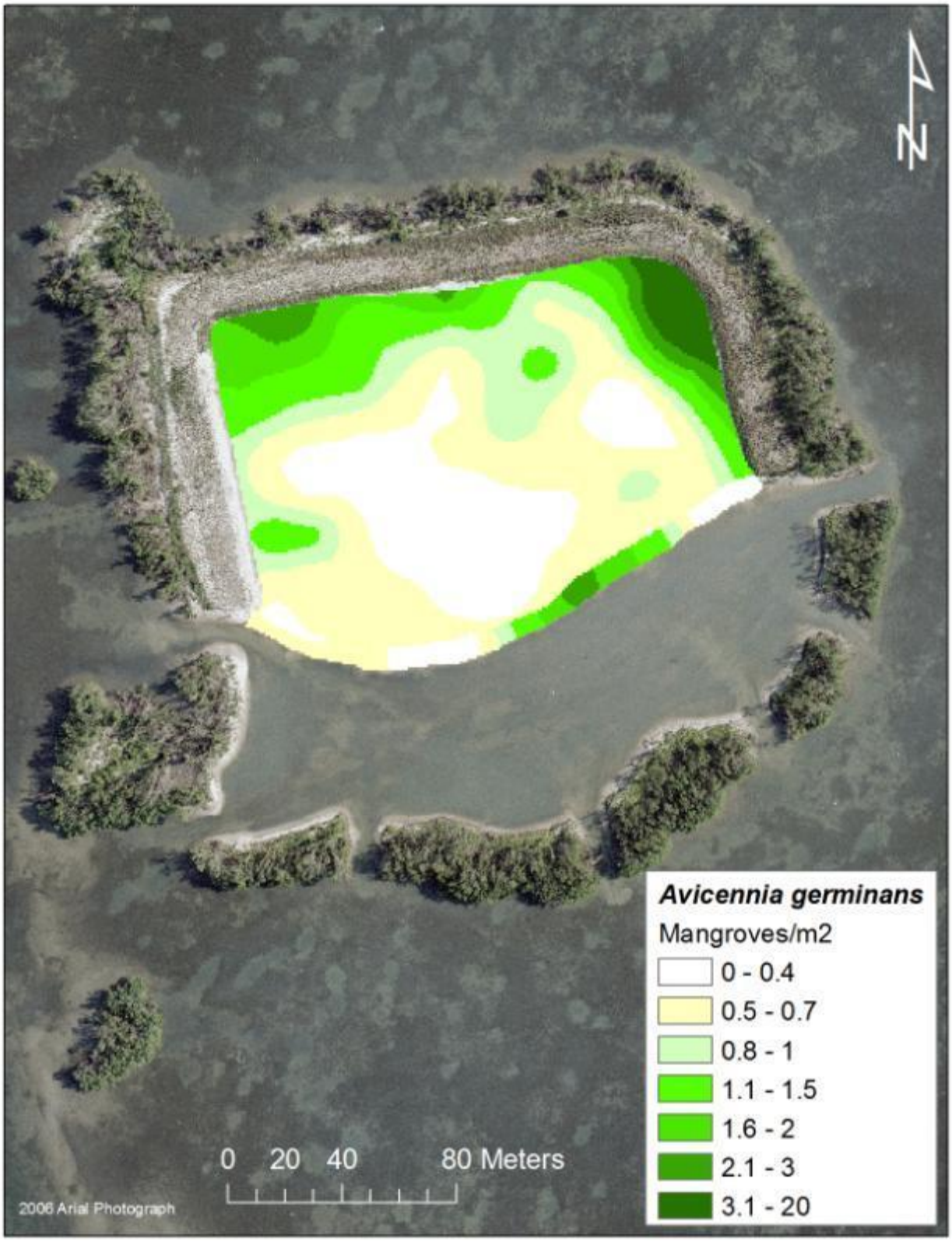

Figure 110. *A. germinans* density in the mangrove planter for winter 2010.

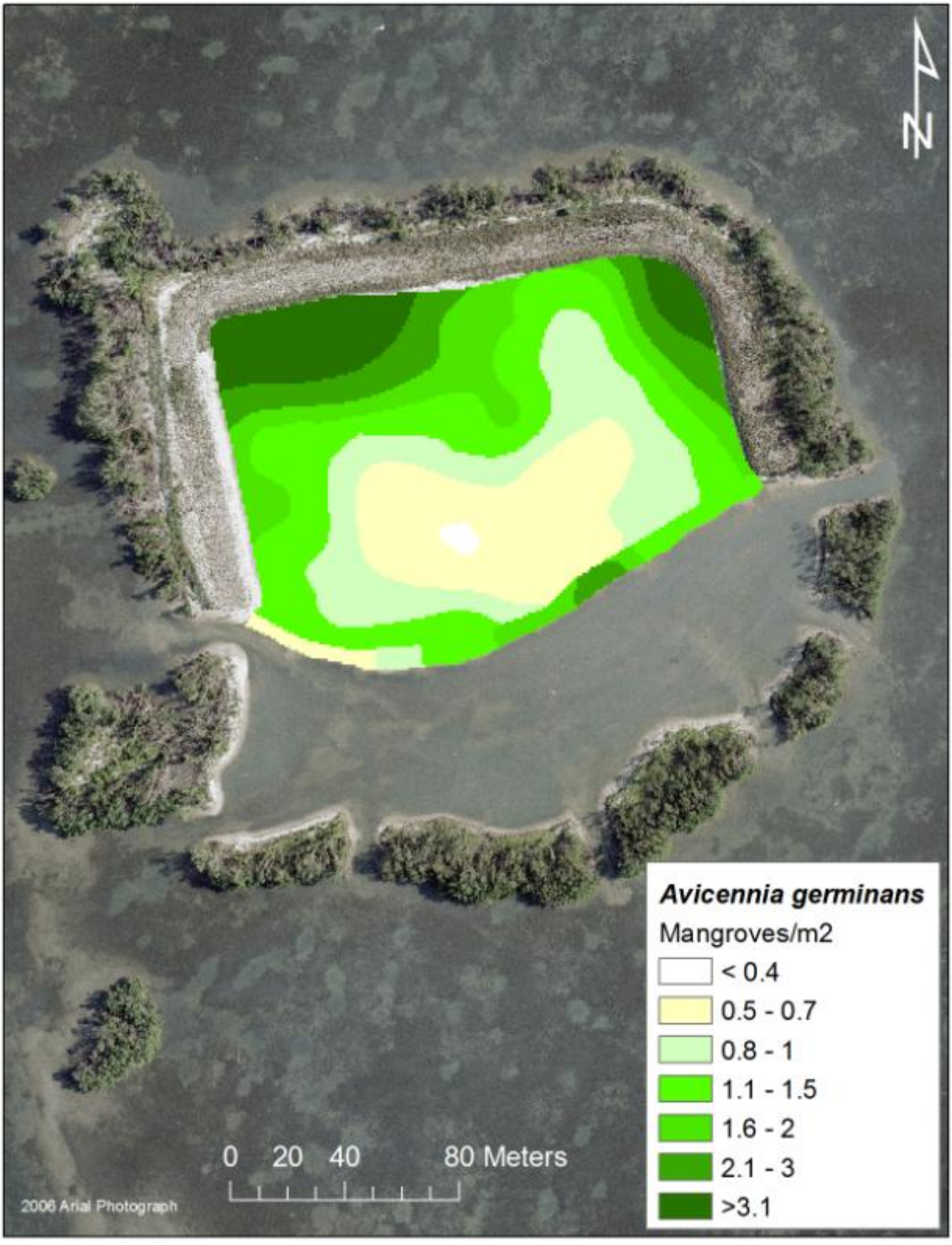

Figure 111. *A. germinans* density in the mangrove planter for summer 2011.

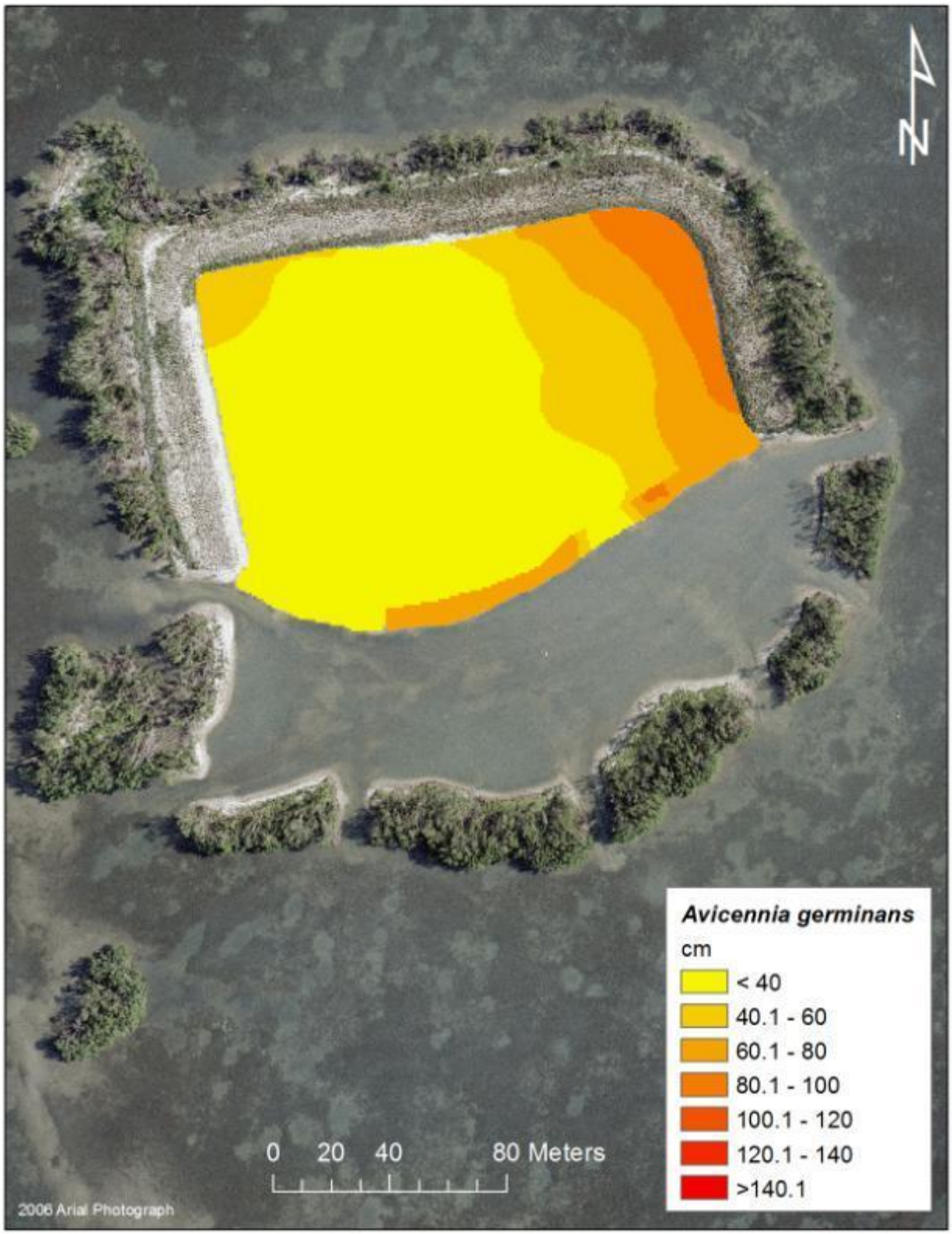

Figure 112. *A. germinans* average height for summer 2009.

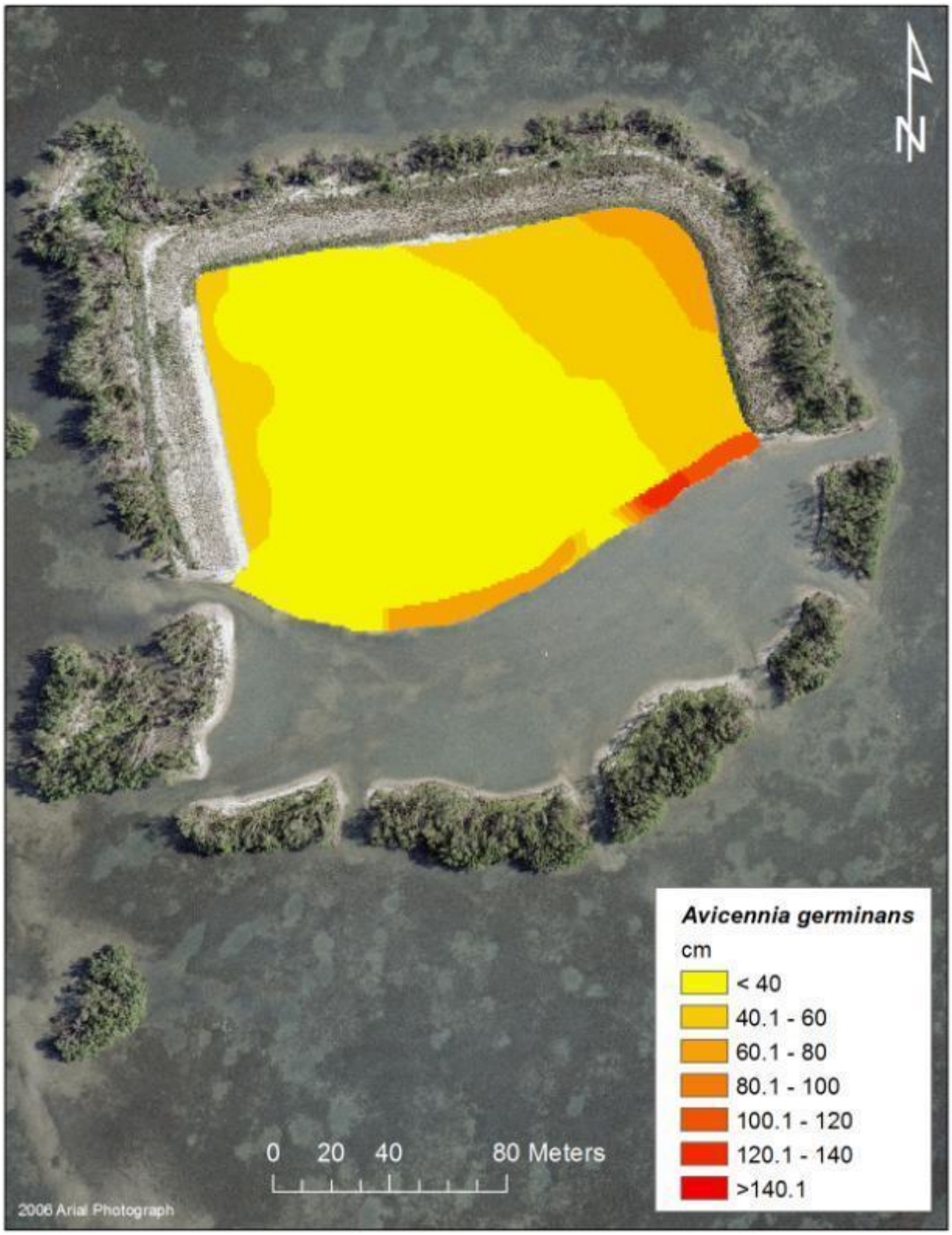

Figure 113. *A. germinans* average height for winter 2009.

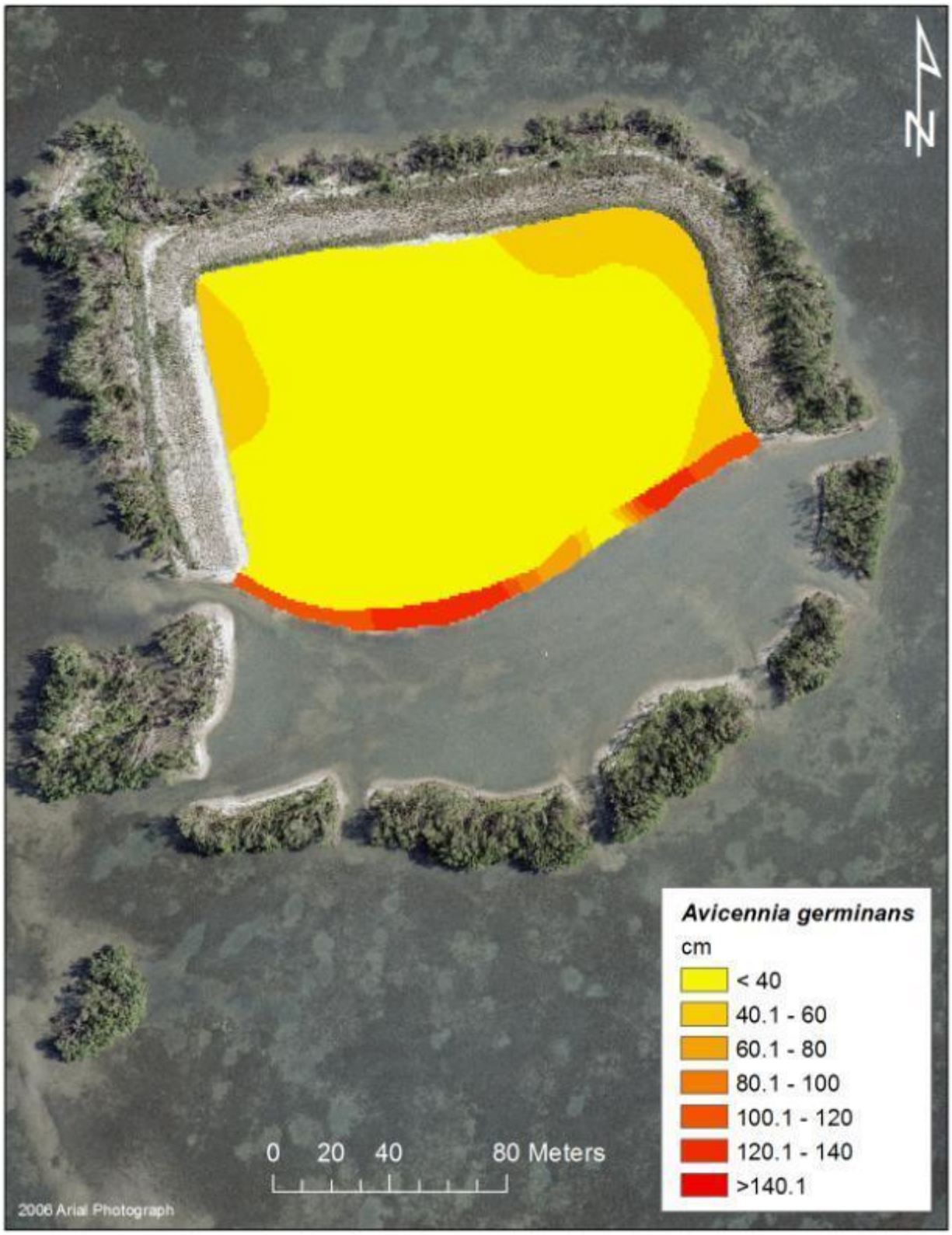

Figure 114. *A. germinans* average height for summer 2010.

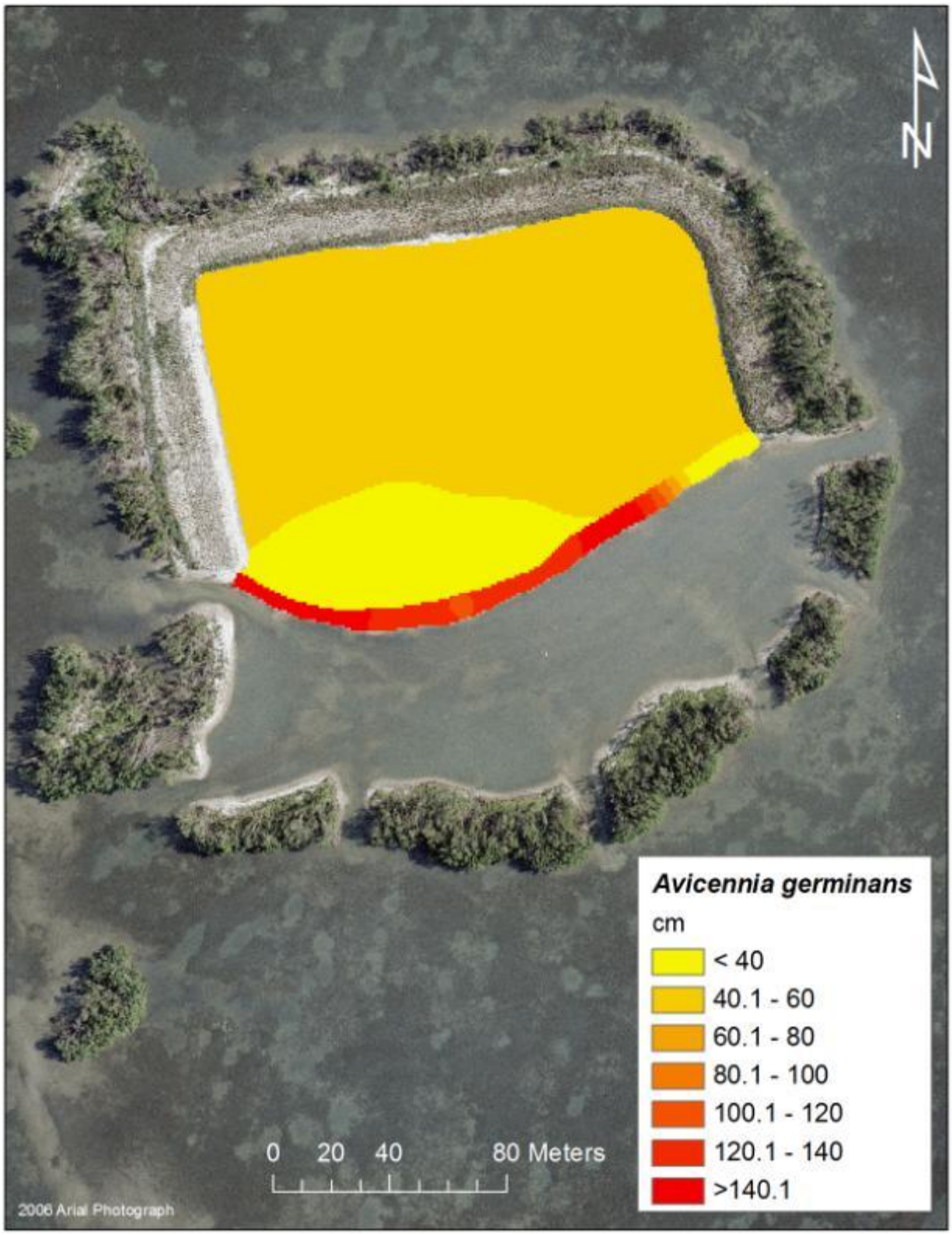

Figure 115. *A. germinans* average height for winter 2010.

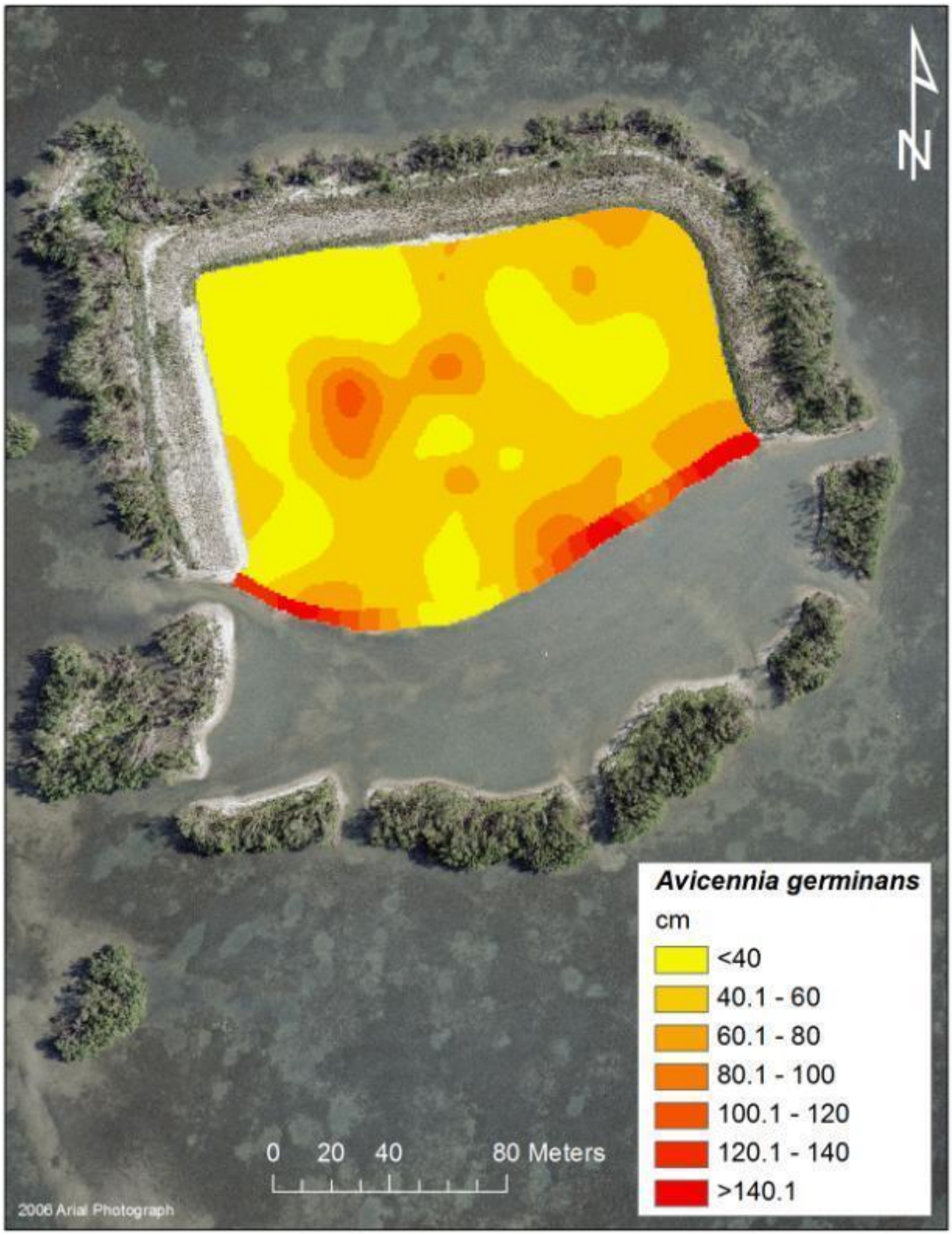

Figure 116. *A. germinans* average height for summer 2011.

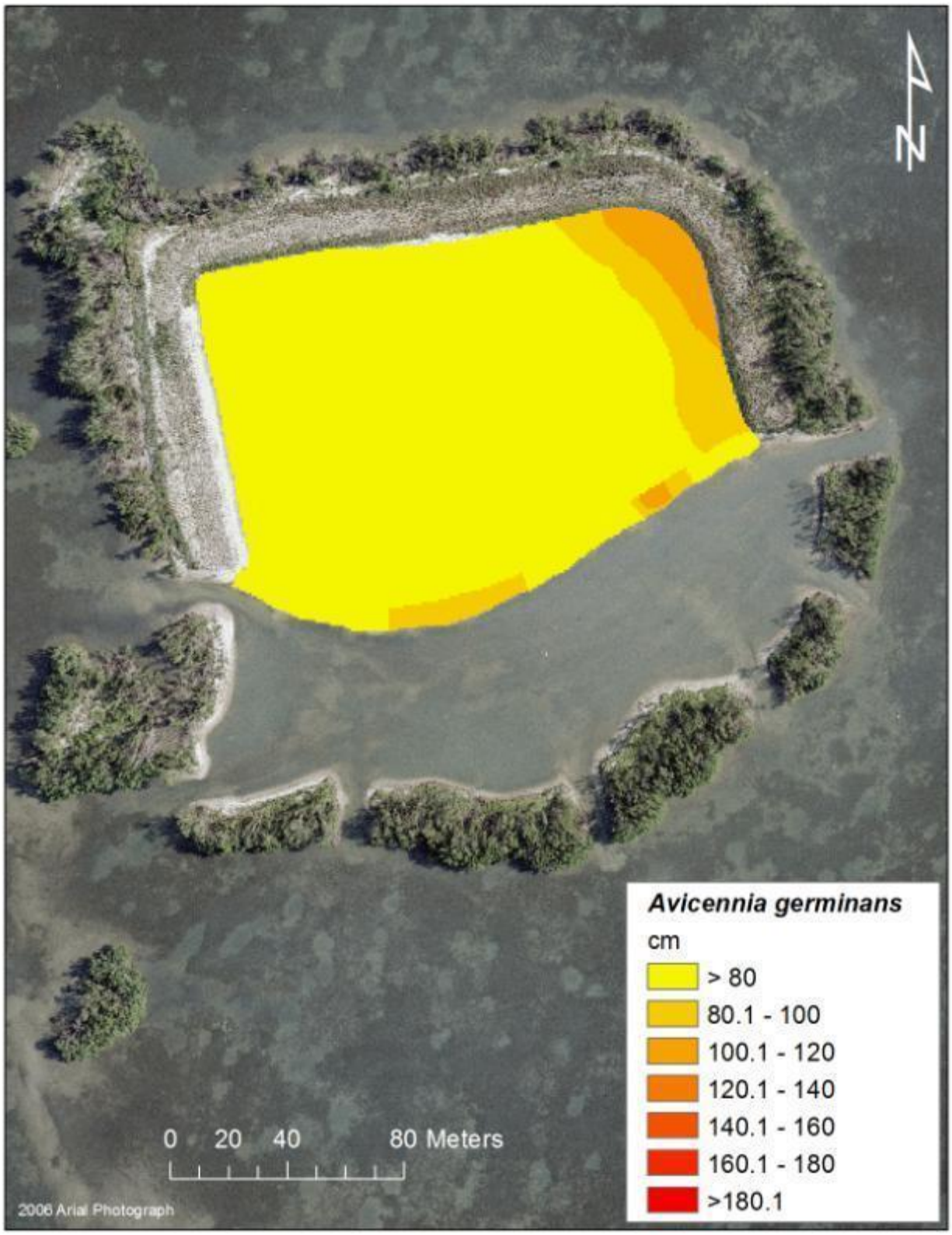

Figure 117. *A. germinans* maximum height for summer 2009.

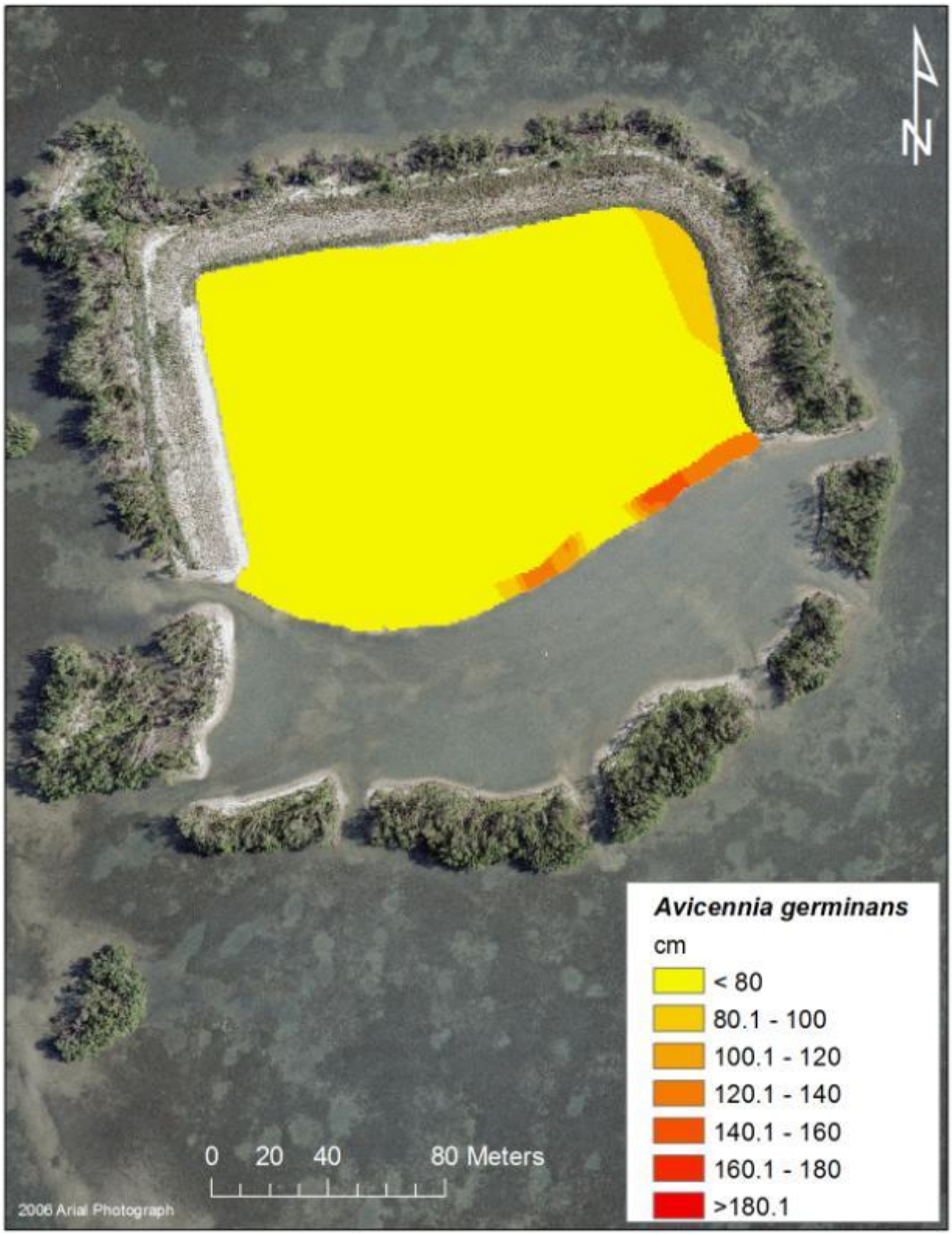

Figure 118. *A. germinans* maximum height for winter 2009.

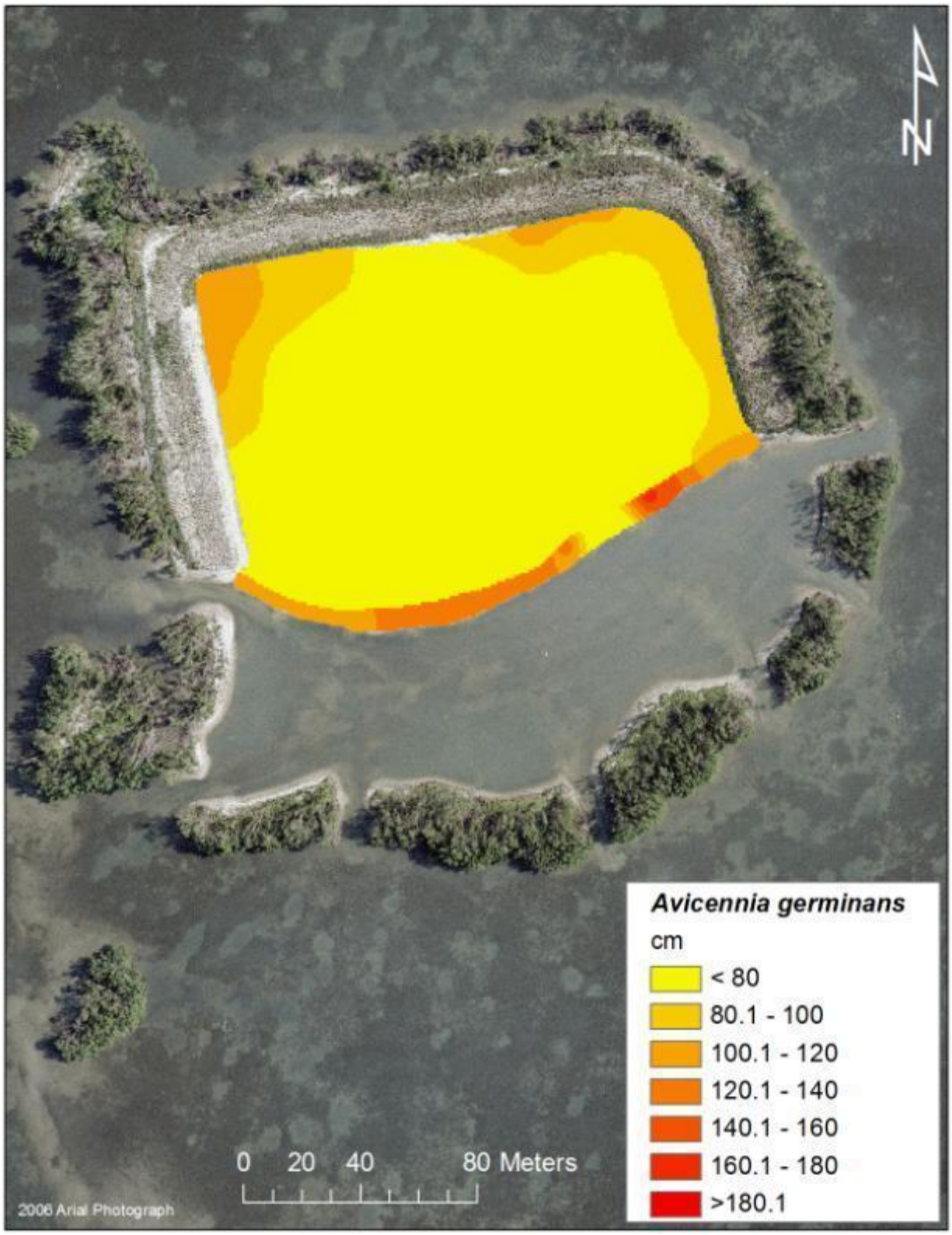

Figure 119. *A. germinans* maximum height for summer 2010.

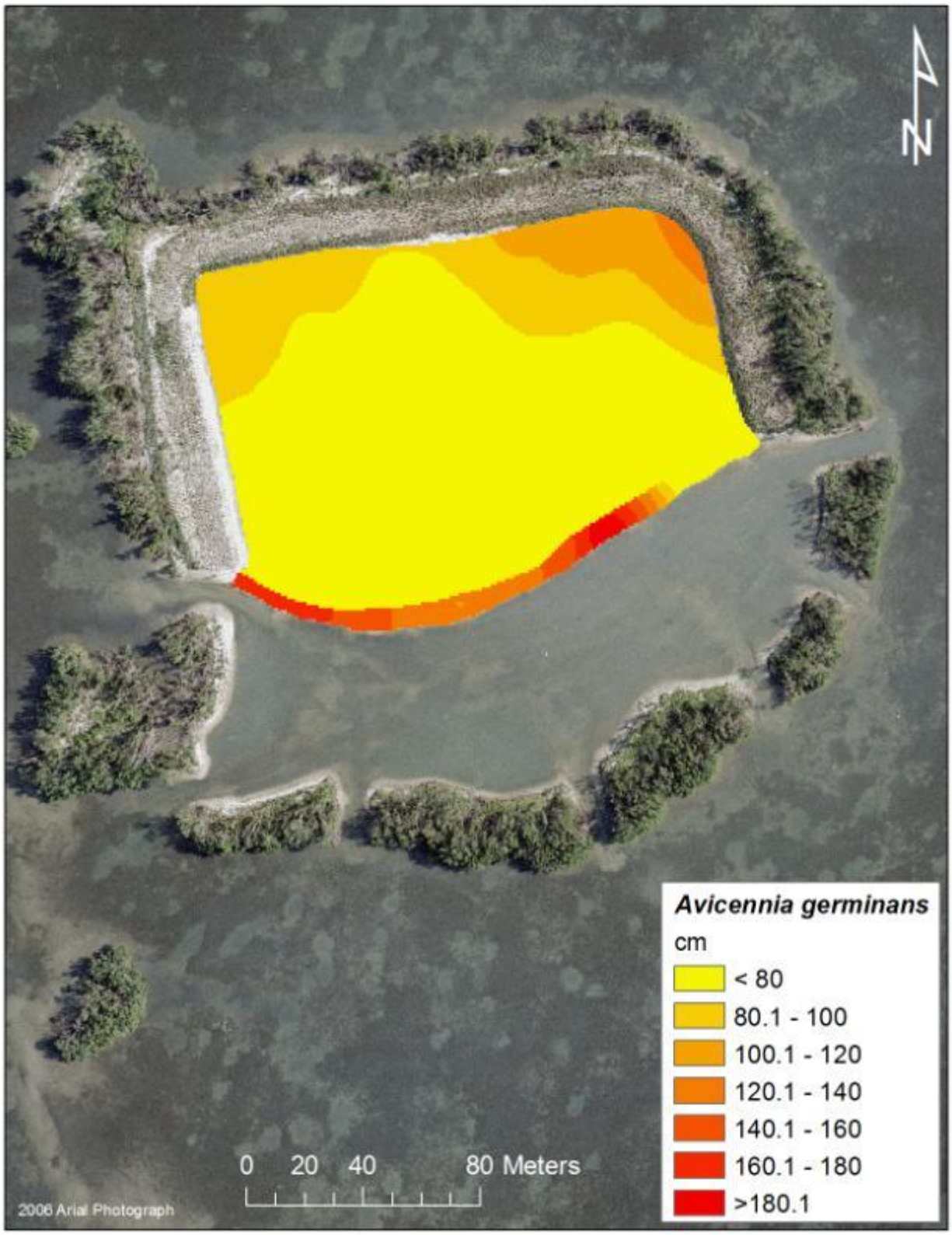

Figure 120. *A. germinans* maximum height for winter 2010.

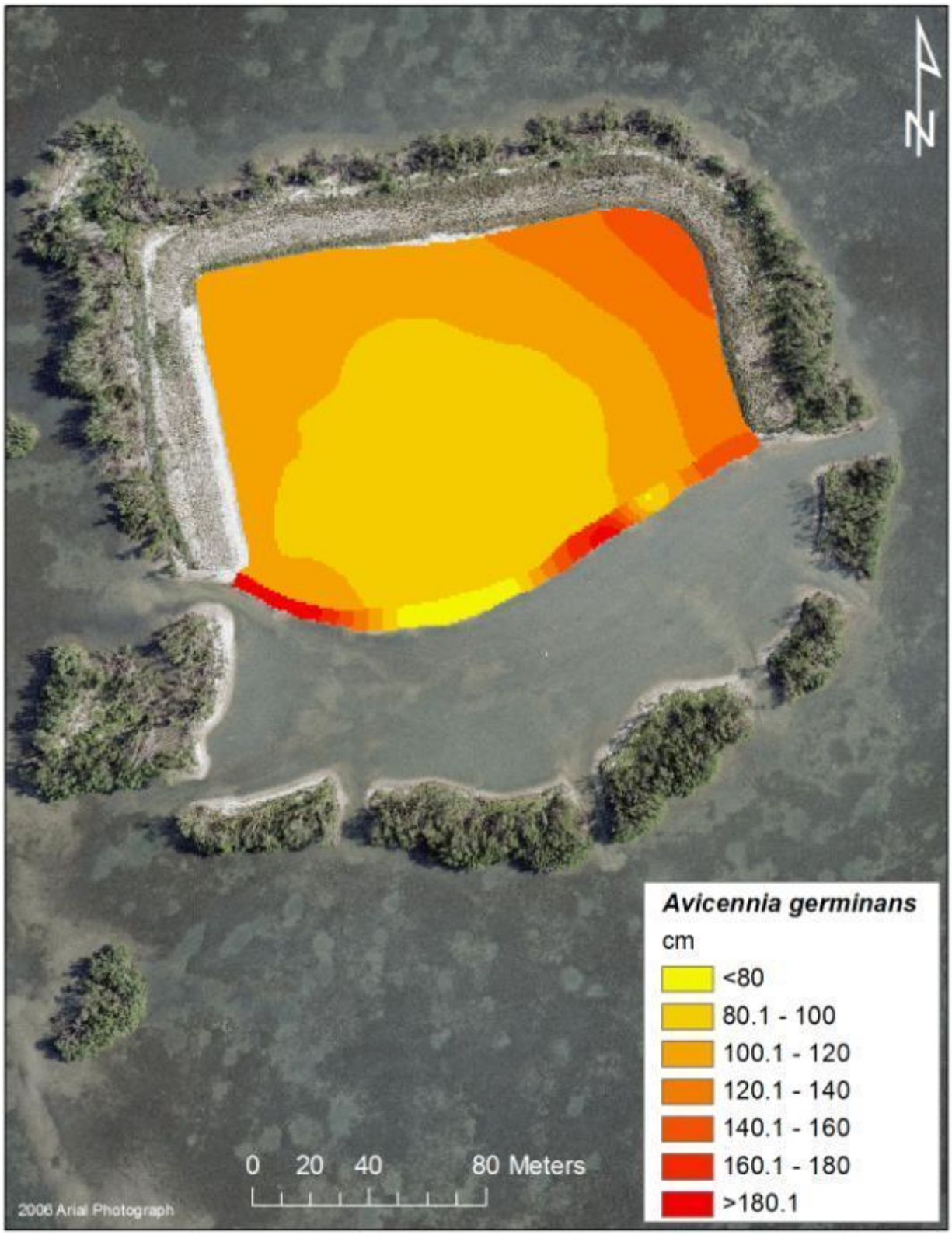

Figure 121. *A. germinans* maximum height for summer 2011.

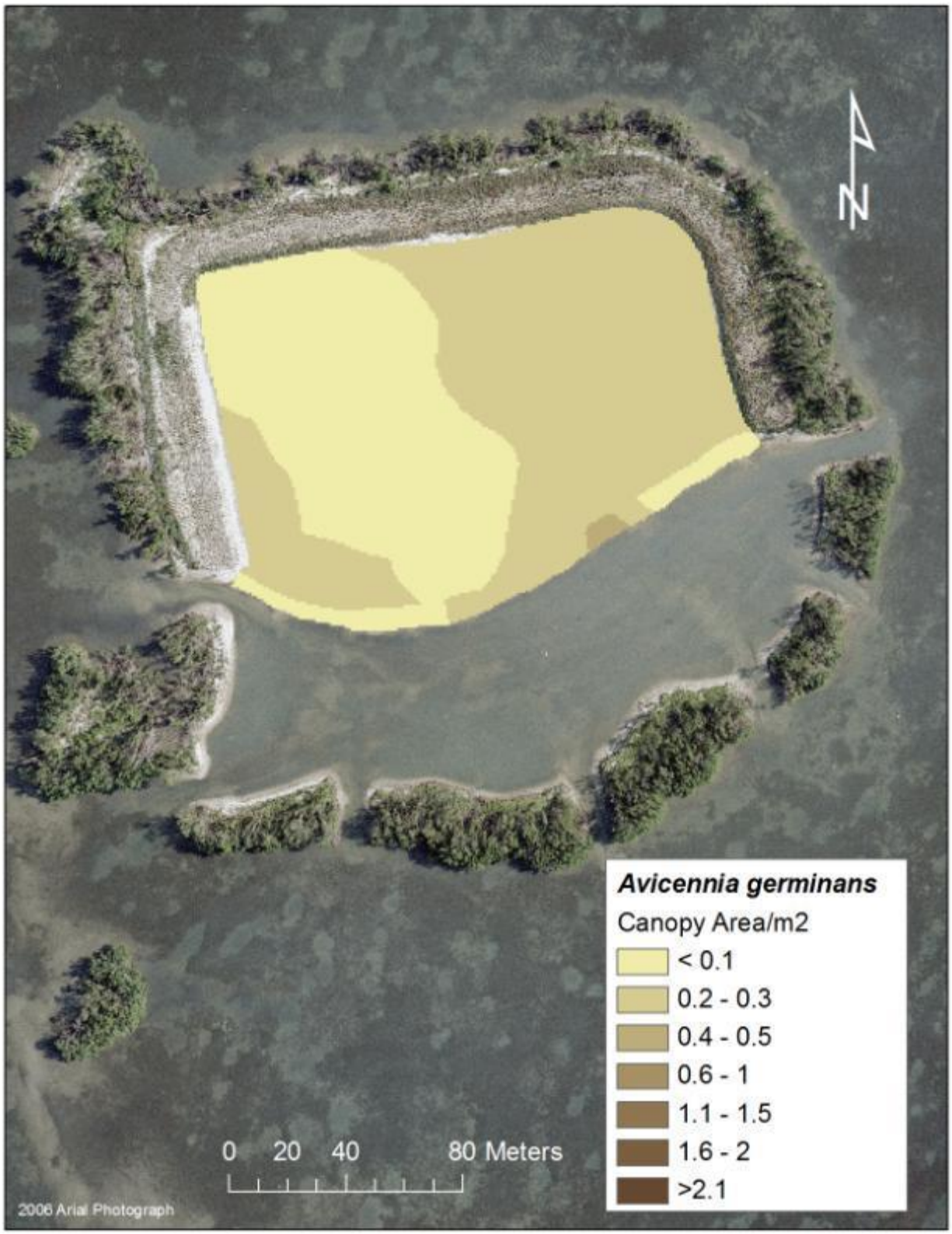

Figure 122. *A. germinans* calculated canopy area/m2 for summer 2011.

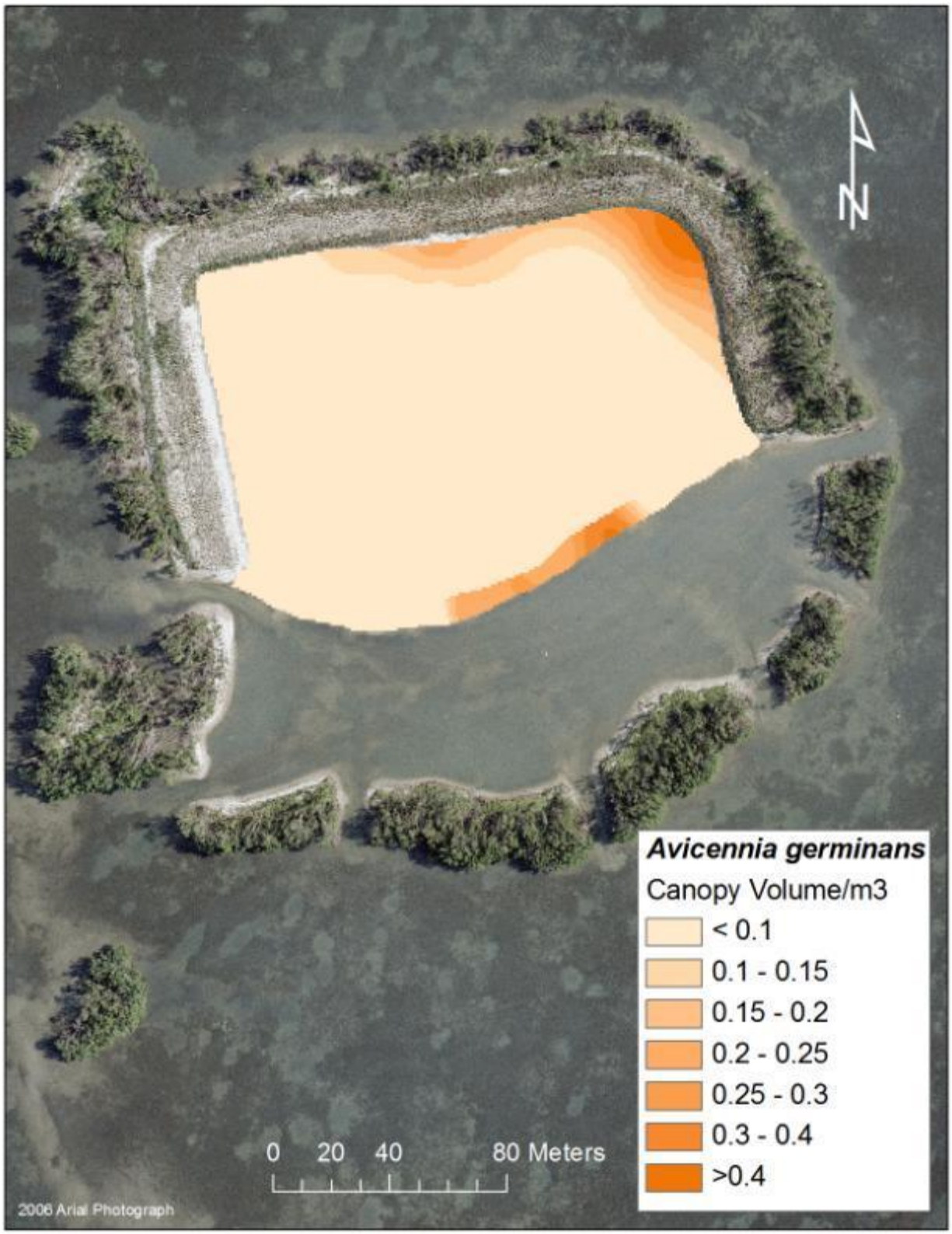

Figure 123. *A. germinans* calculated volume area/m3 for summer 2011.

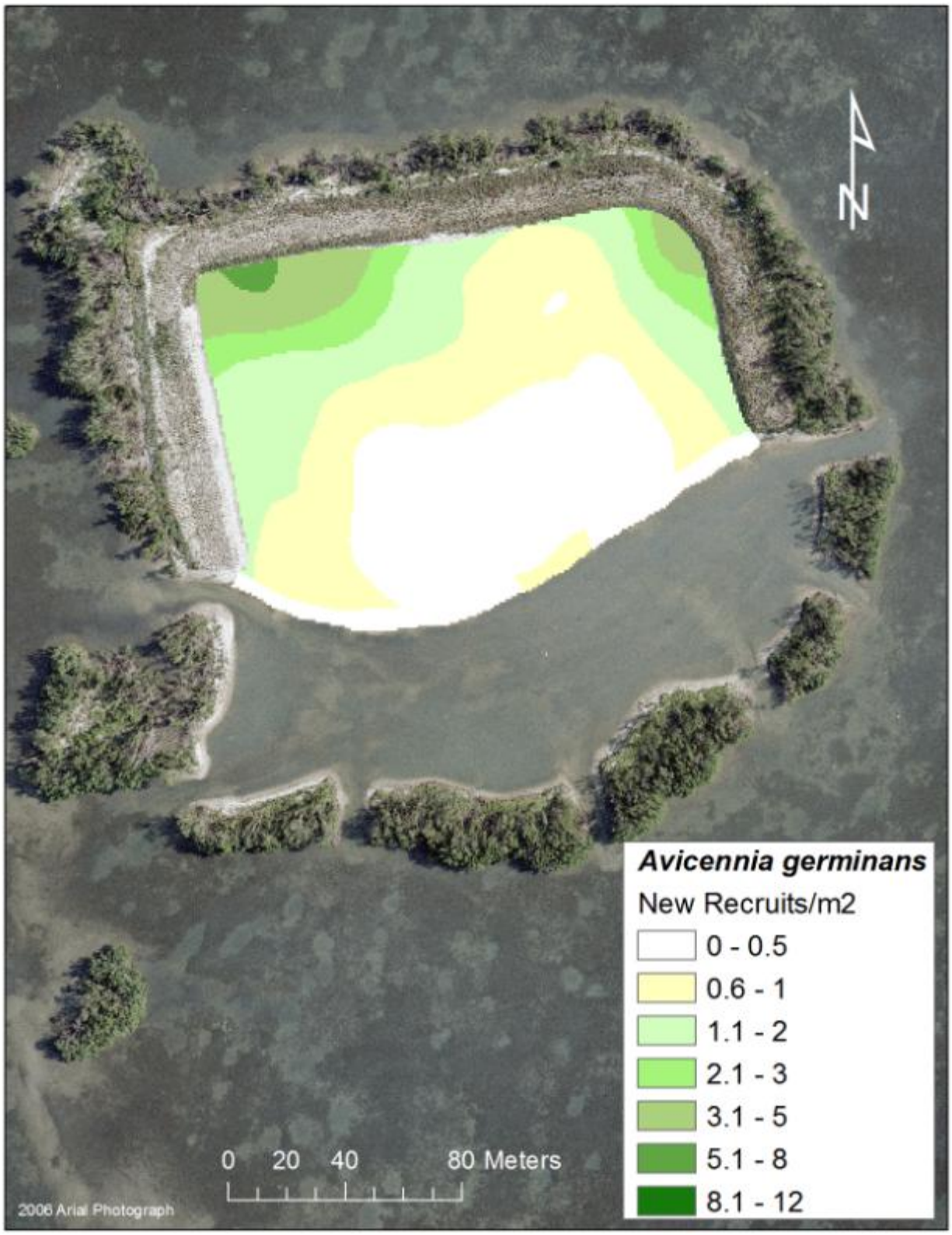

Figure 124. *A. germinans* juvenile new recruits in summer 2011.

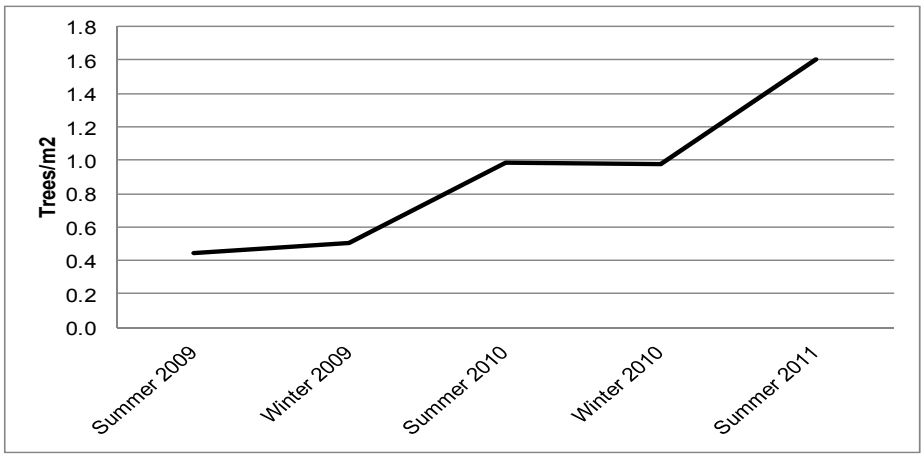

Figure 125. Average *A. germinans* mangrove density (trees/m2) from summer 2009 to summer 2011.

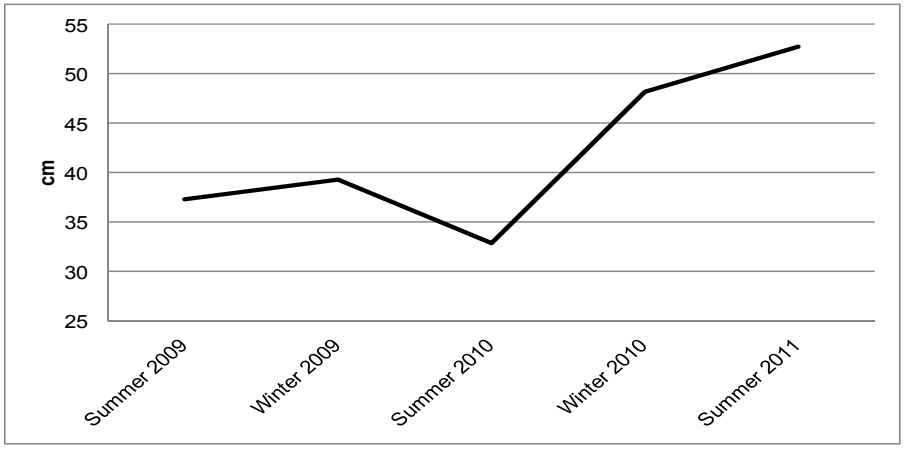

Figure 126. Average *A. germinans* mangrove height from summer 2009 to summer 2011.

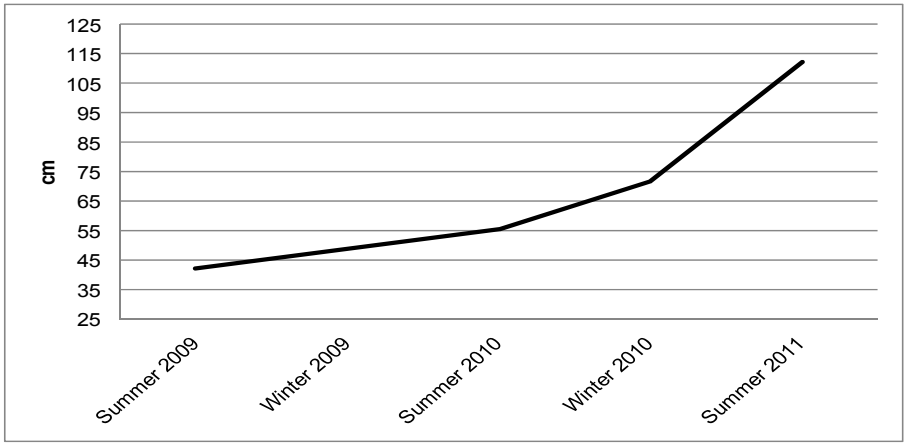

Figure 127. Average *A. germinans* mangrove height from summer 2009 to summer 2011.

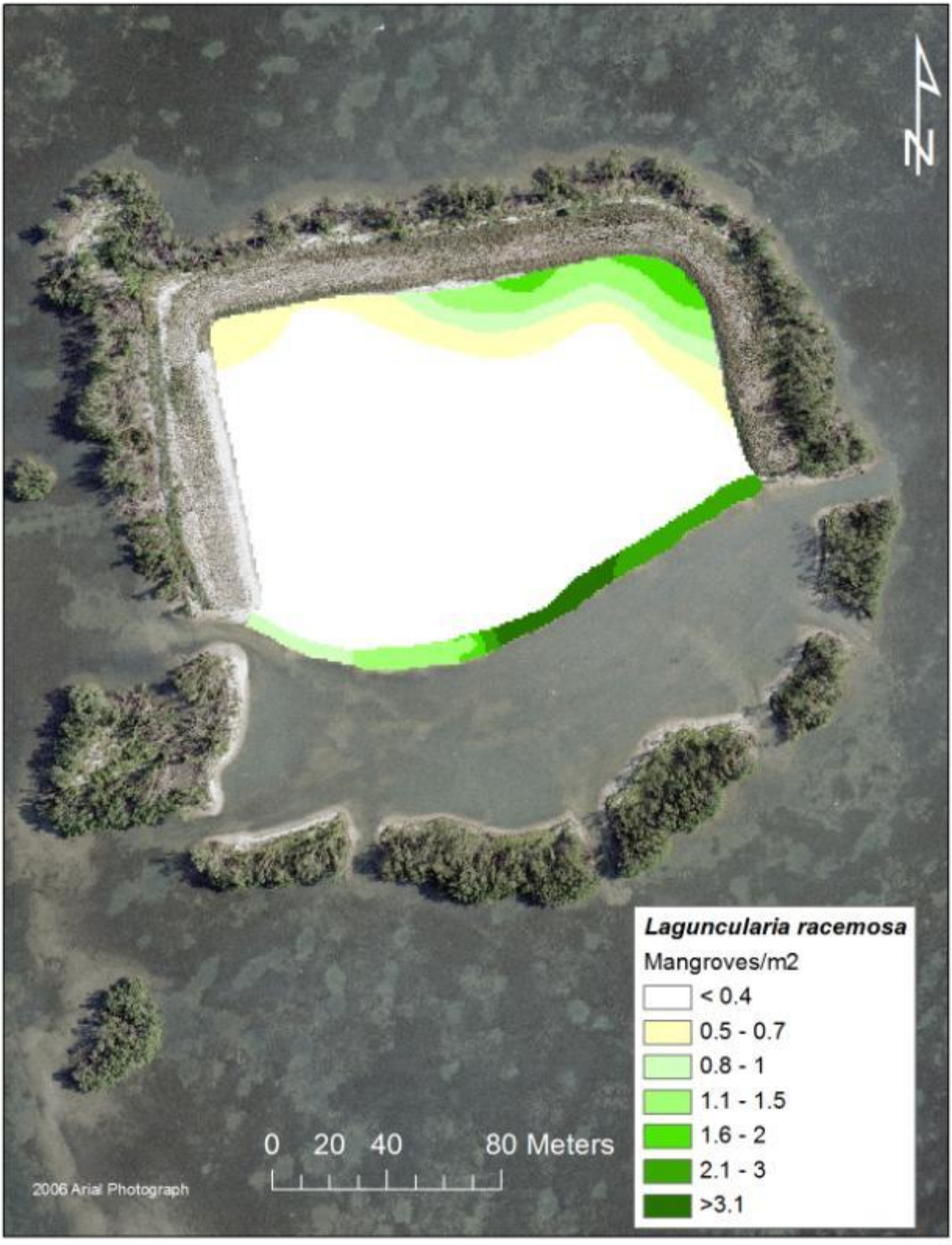

Figure 128. *L racemosa* density in the mangrove planter for summer 2009.

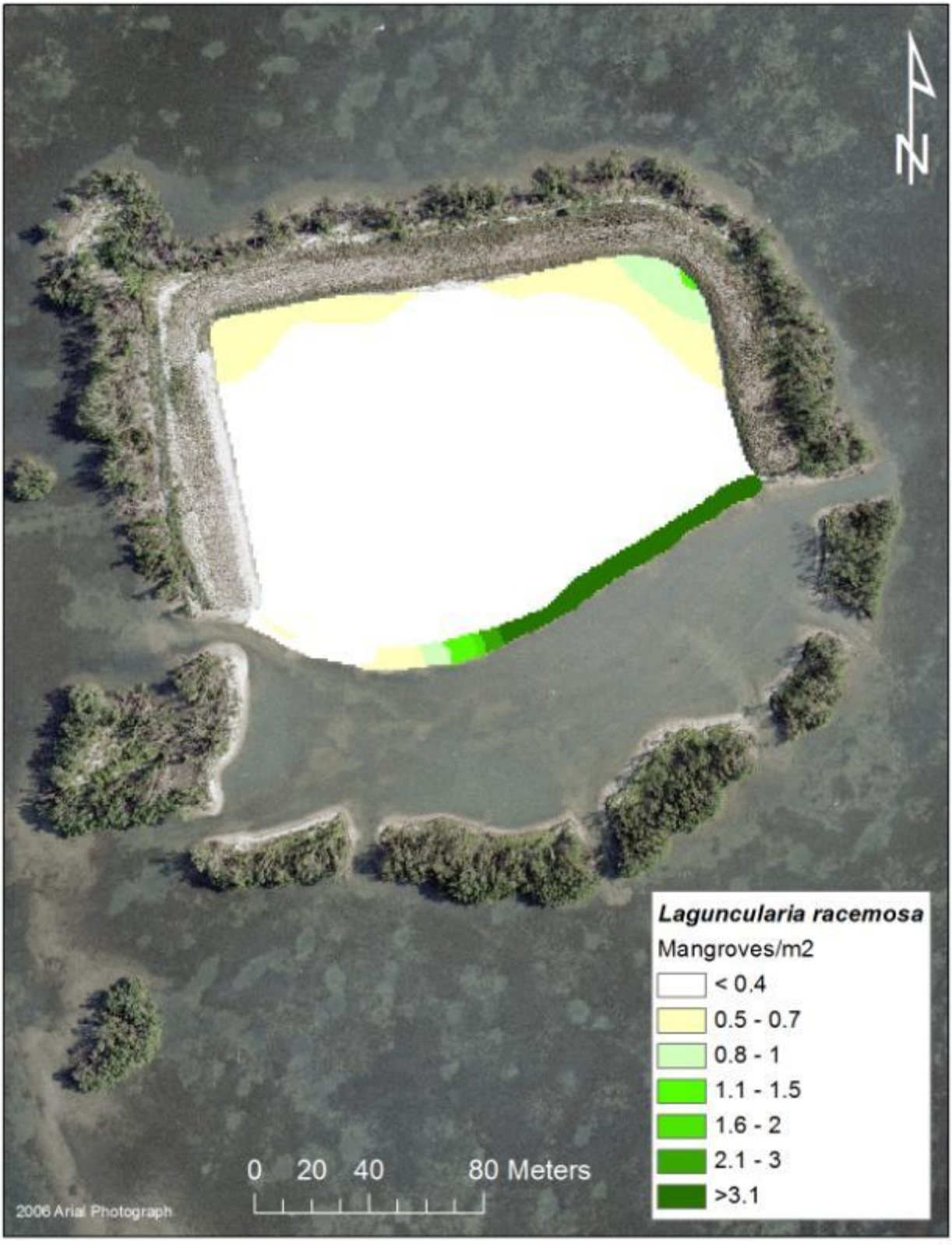

Figure 129. *L racemosa* density in the mangrove planter for winter 2009.

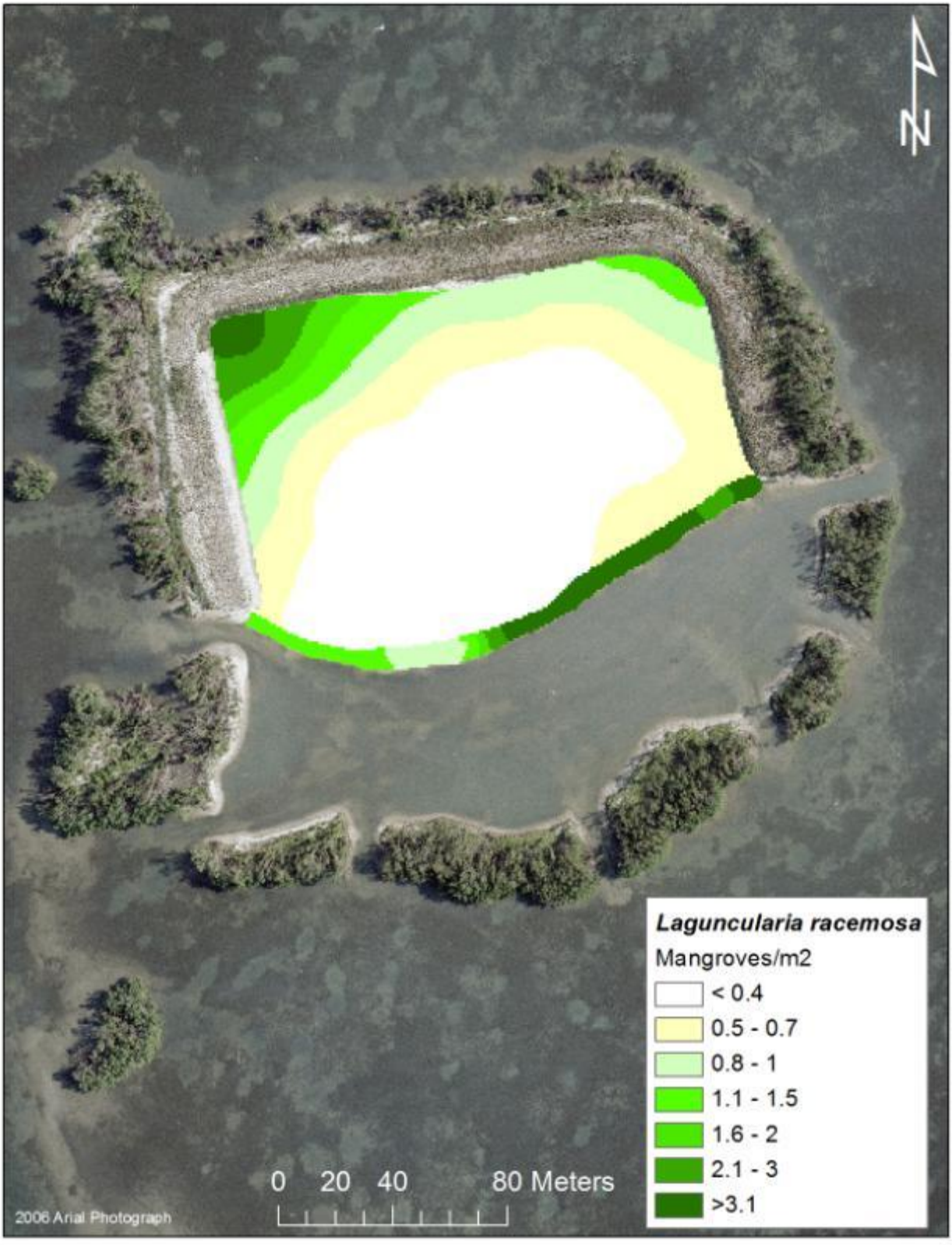

Figure 130. *L racemosa* density in the mangrove planter for summer 2010.

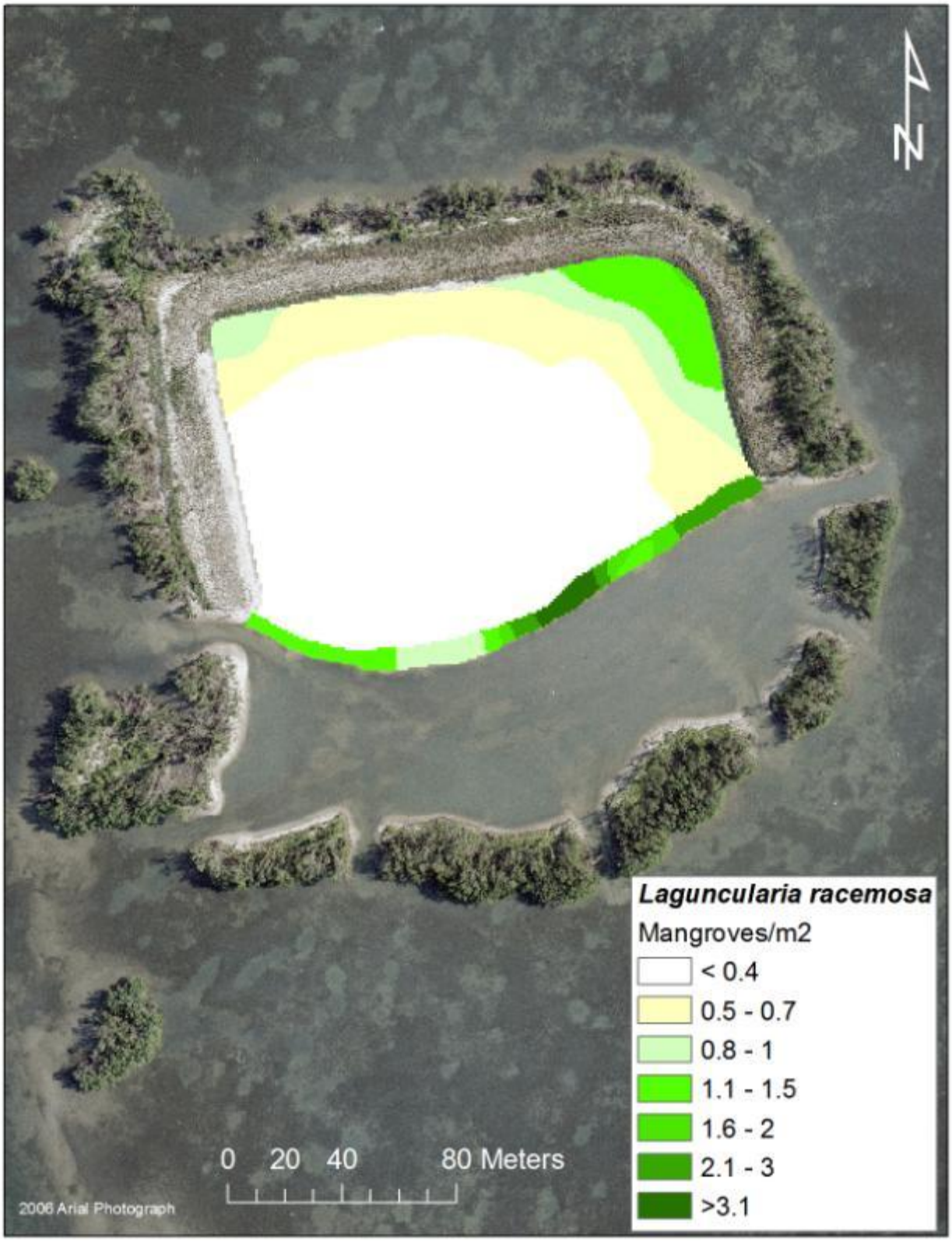

Figure 131. *L racemosa* density in the mangrove planter for winter 2010.

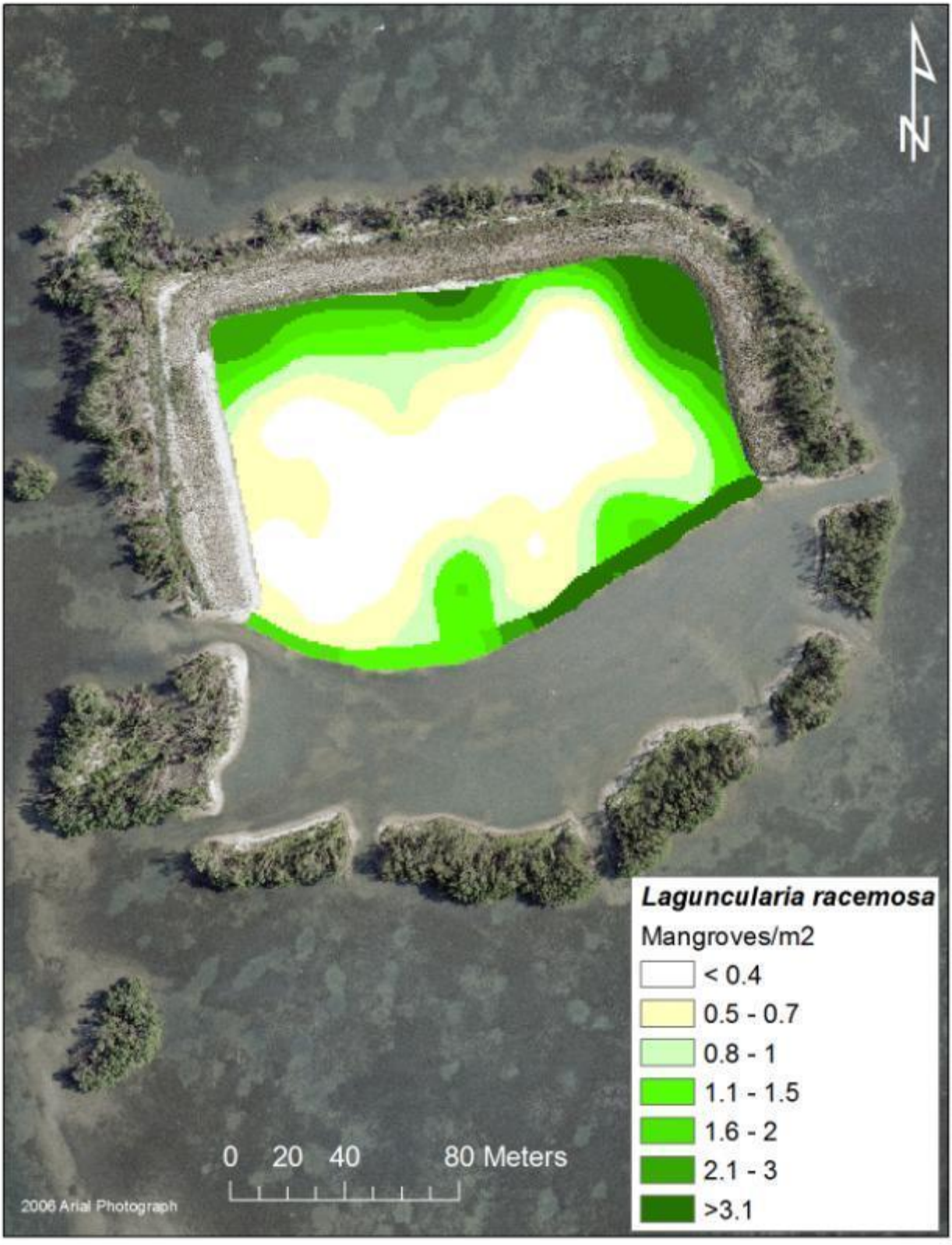

Figure 132. *L racemosa* density in the mangrove planter for summer 2011.

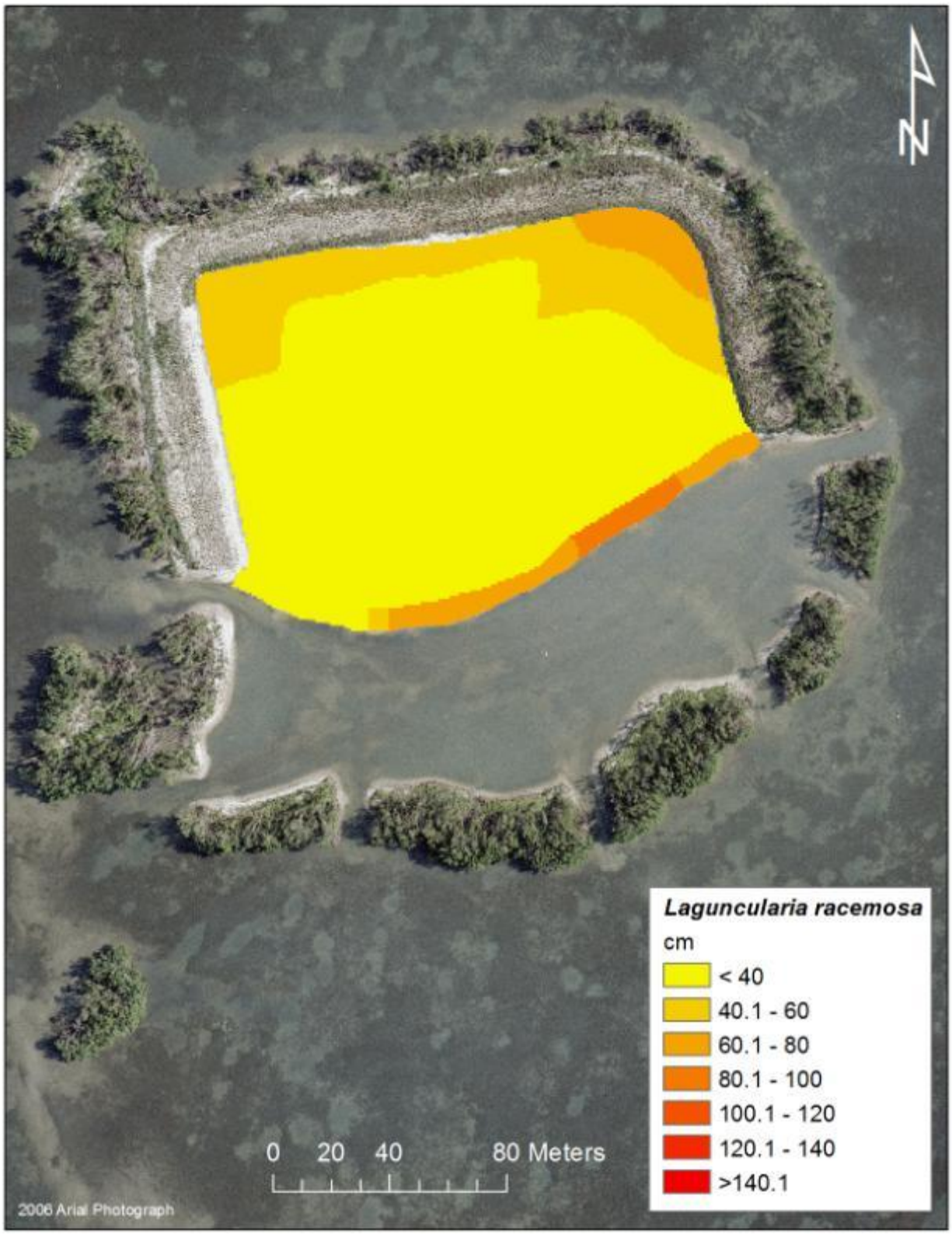

Figure 133. *L racemosa* average height for summer 2009.

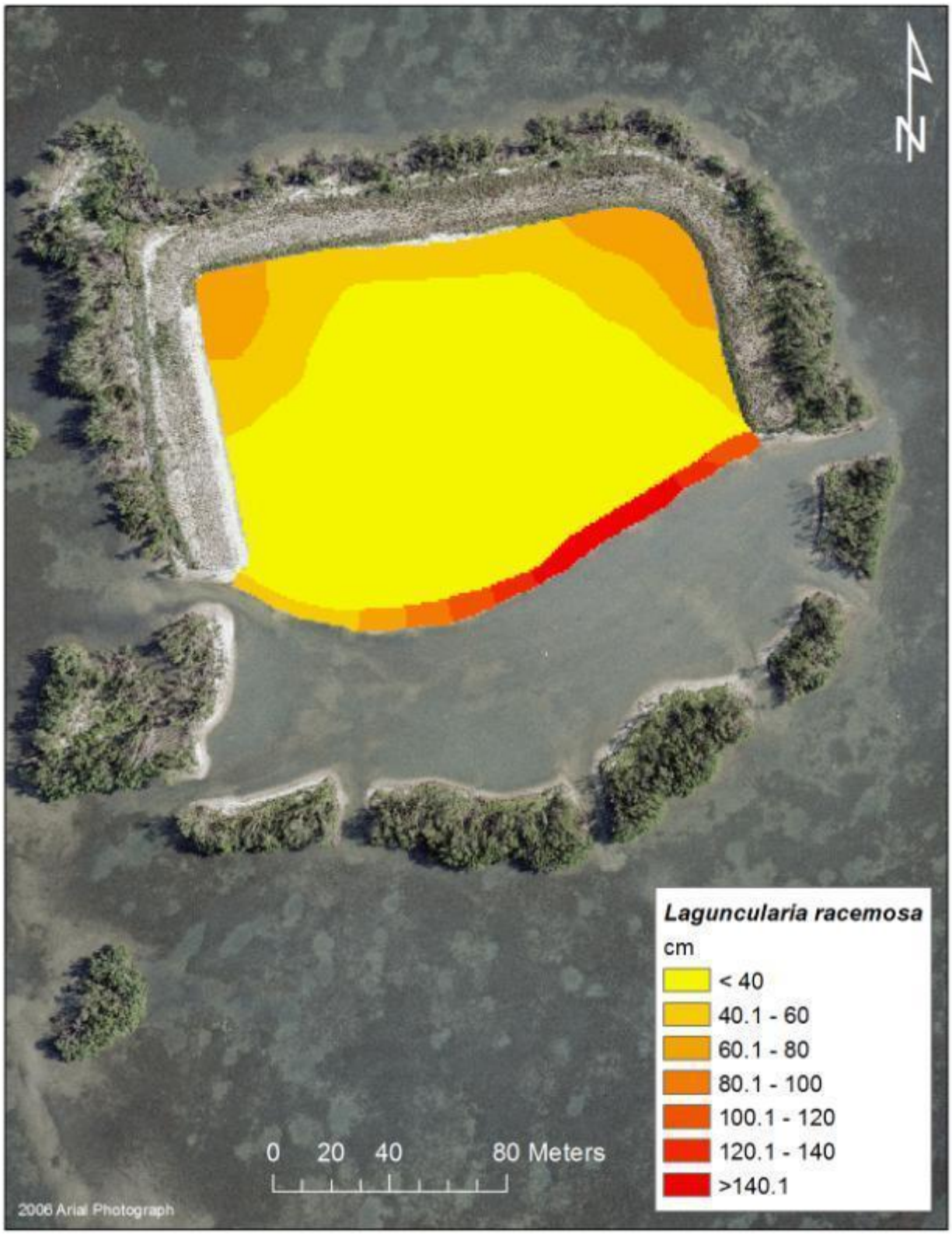

Figure 134. *L racemosa* density in the mangrove planter for winter 2009.

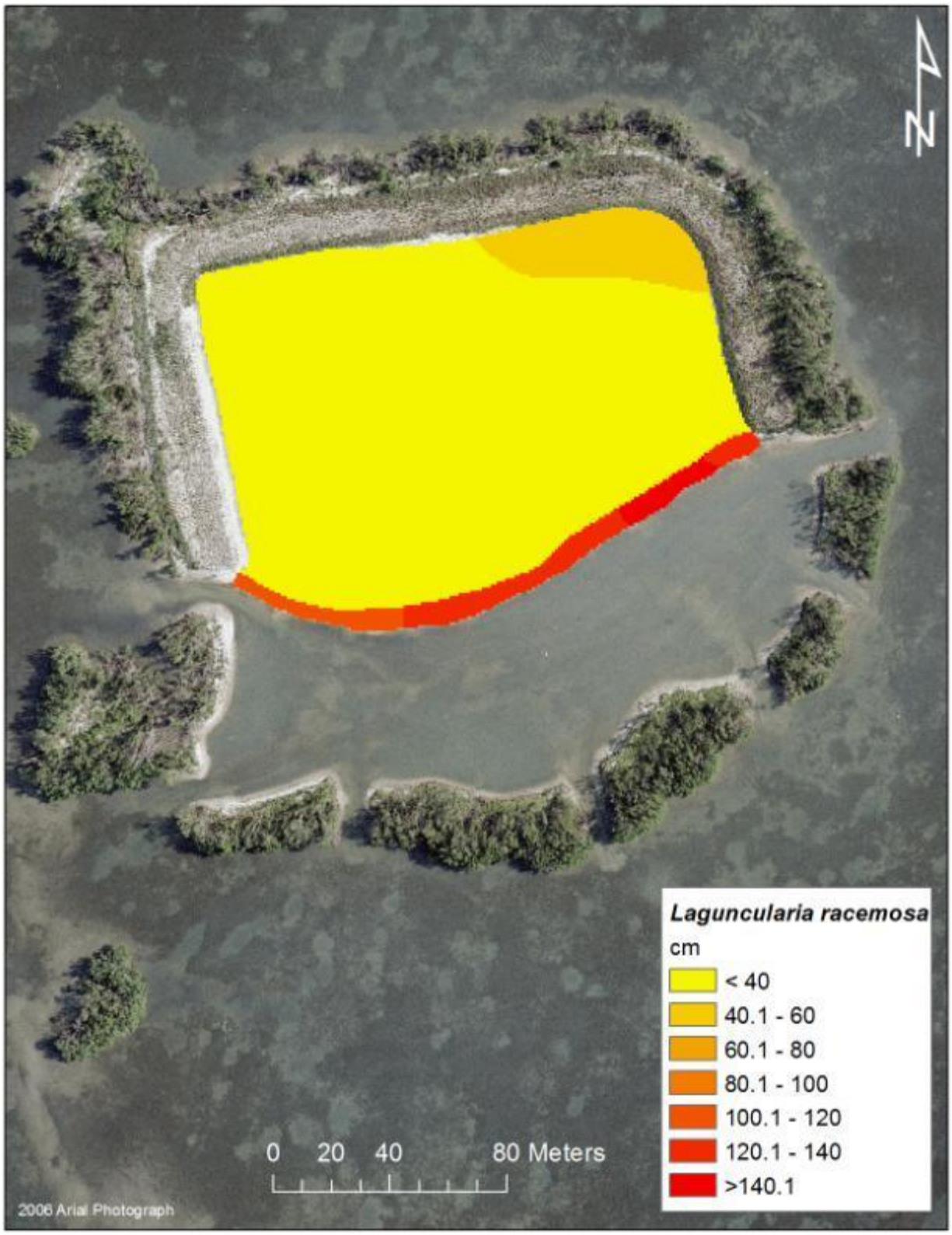

Figure 135. *L racemosa* density in the mangrove planter for summer 2010.

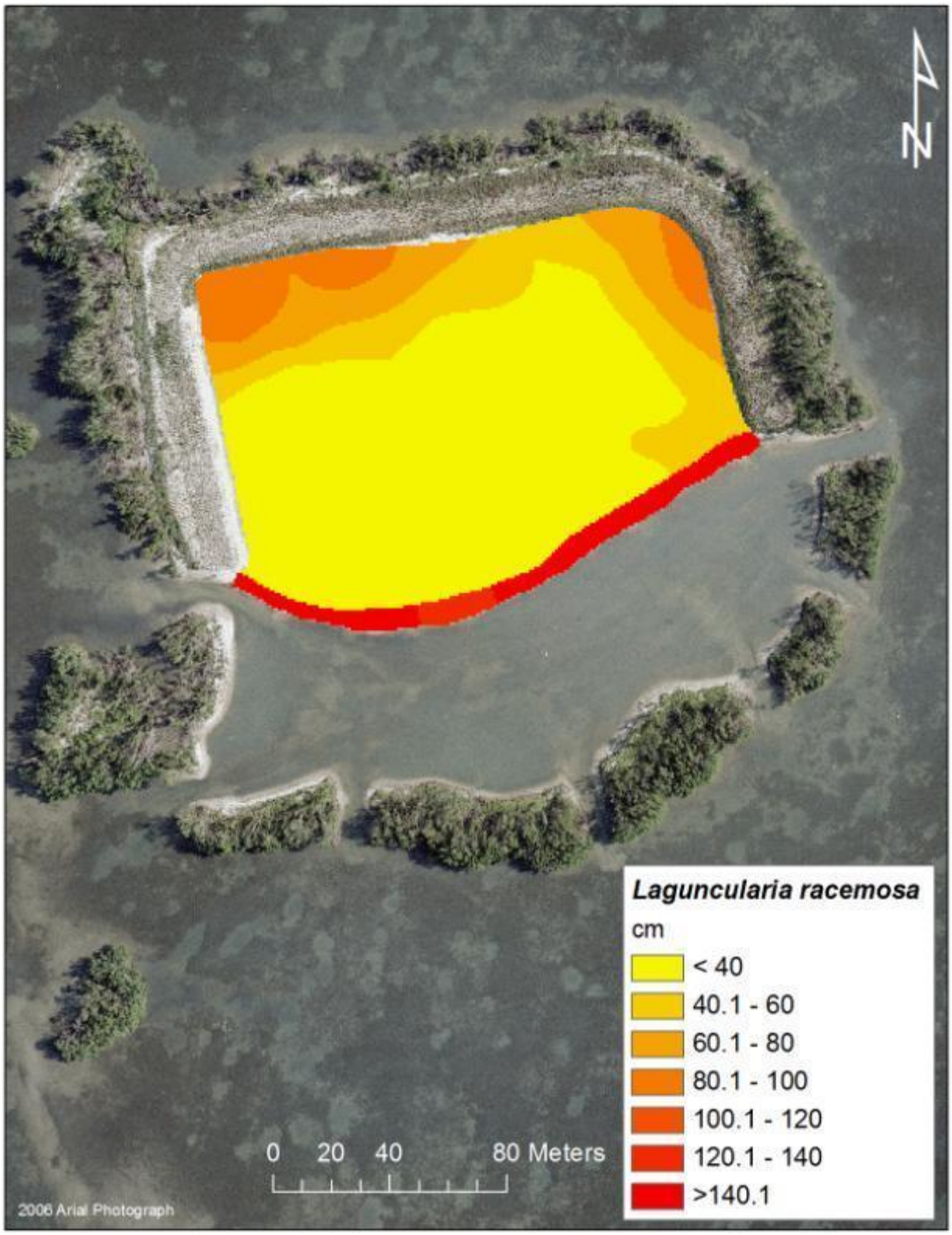

Figure 136. *L racemosa* density in the mangrove planter for winter 2009.

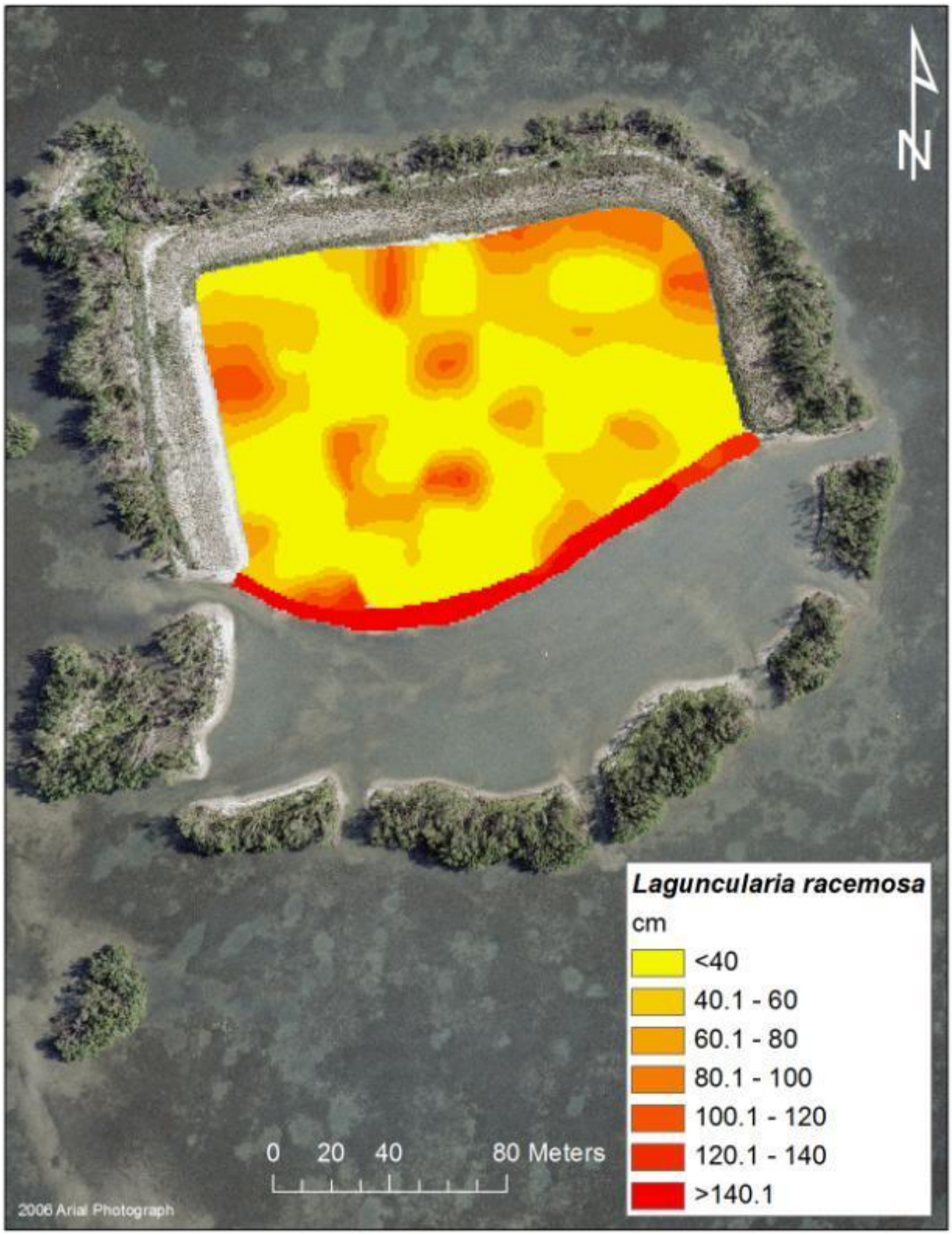

Figure 137. *L racemosa* density in the mangrove planter for summer 2011.

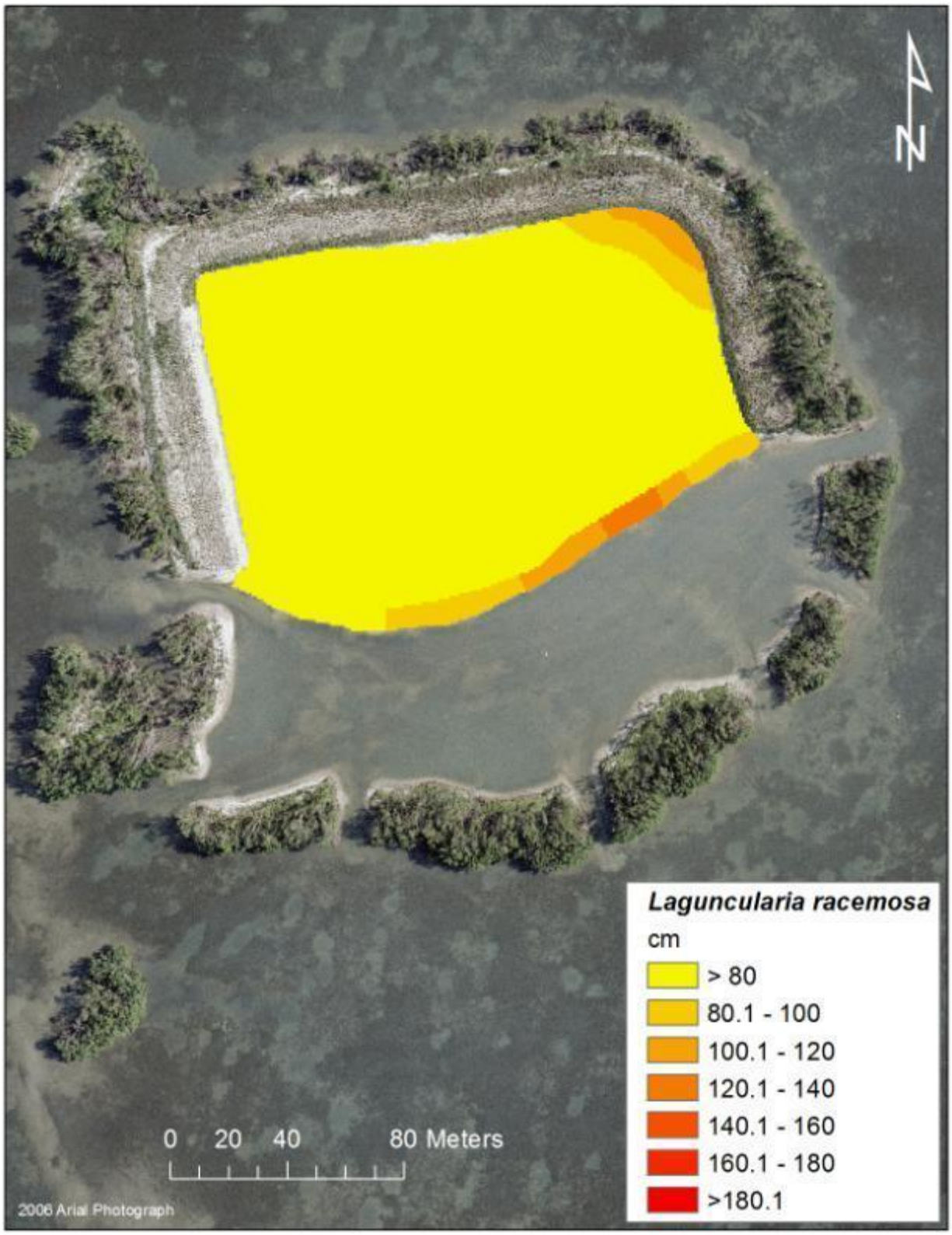

Figure 138. *L racemosa* maximum height for summer 2009.

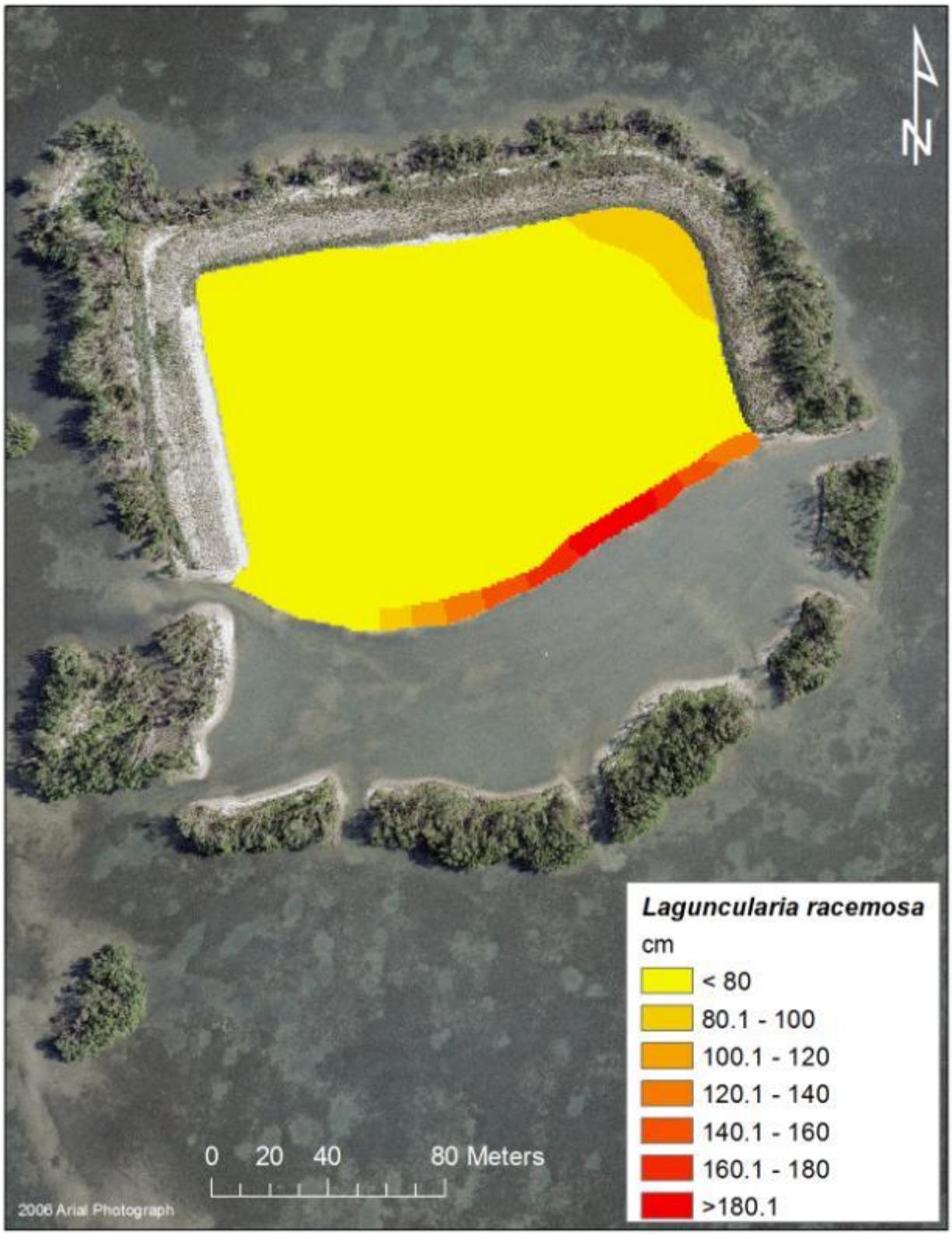

Figure 139. *L racemosa* maximum height for winter 2009.

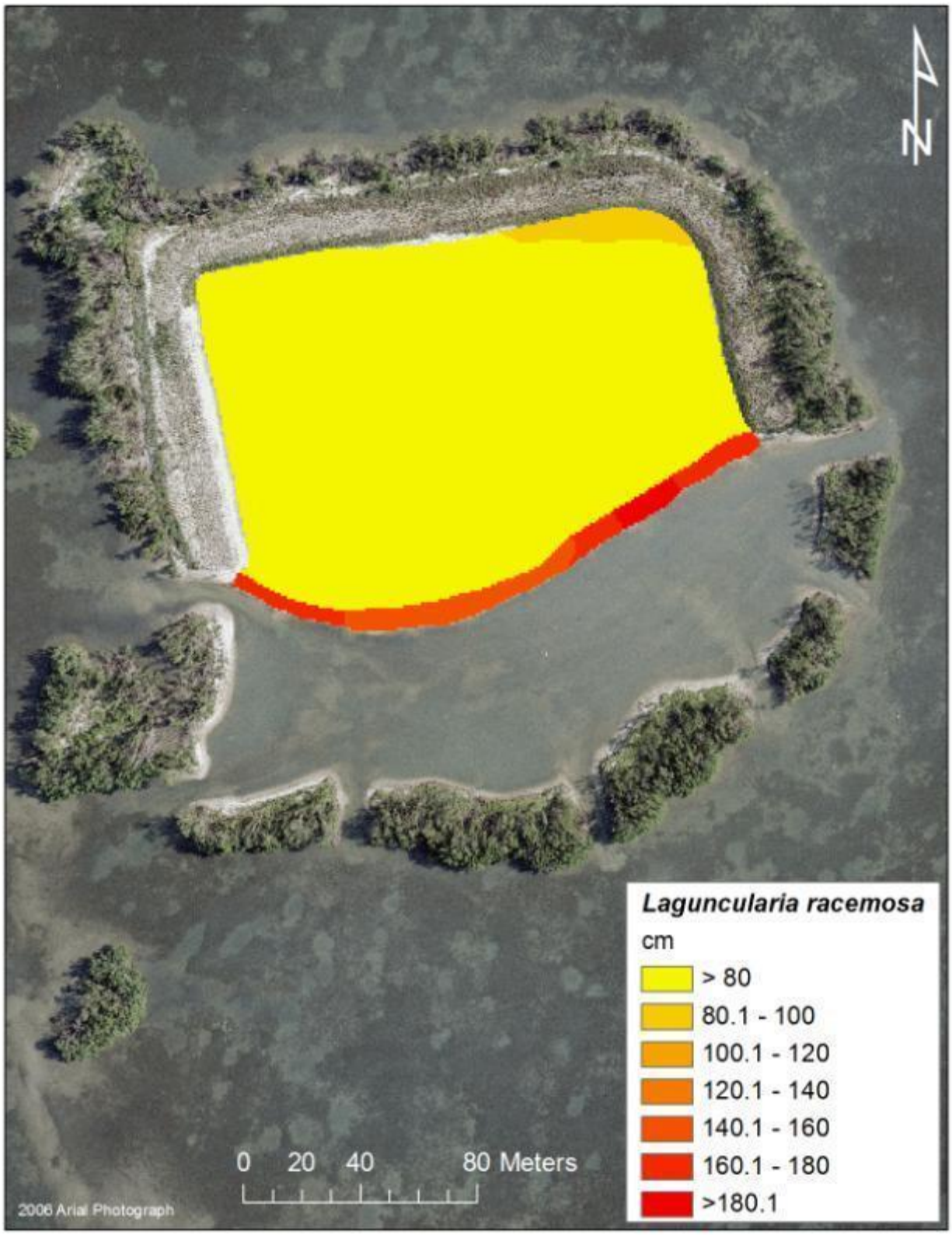

Figure 140. *L racemosa* maximum height for summer 2010.

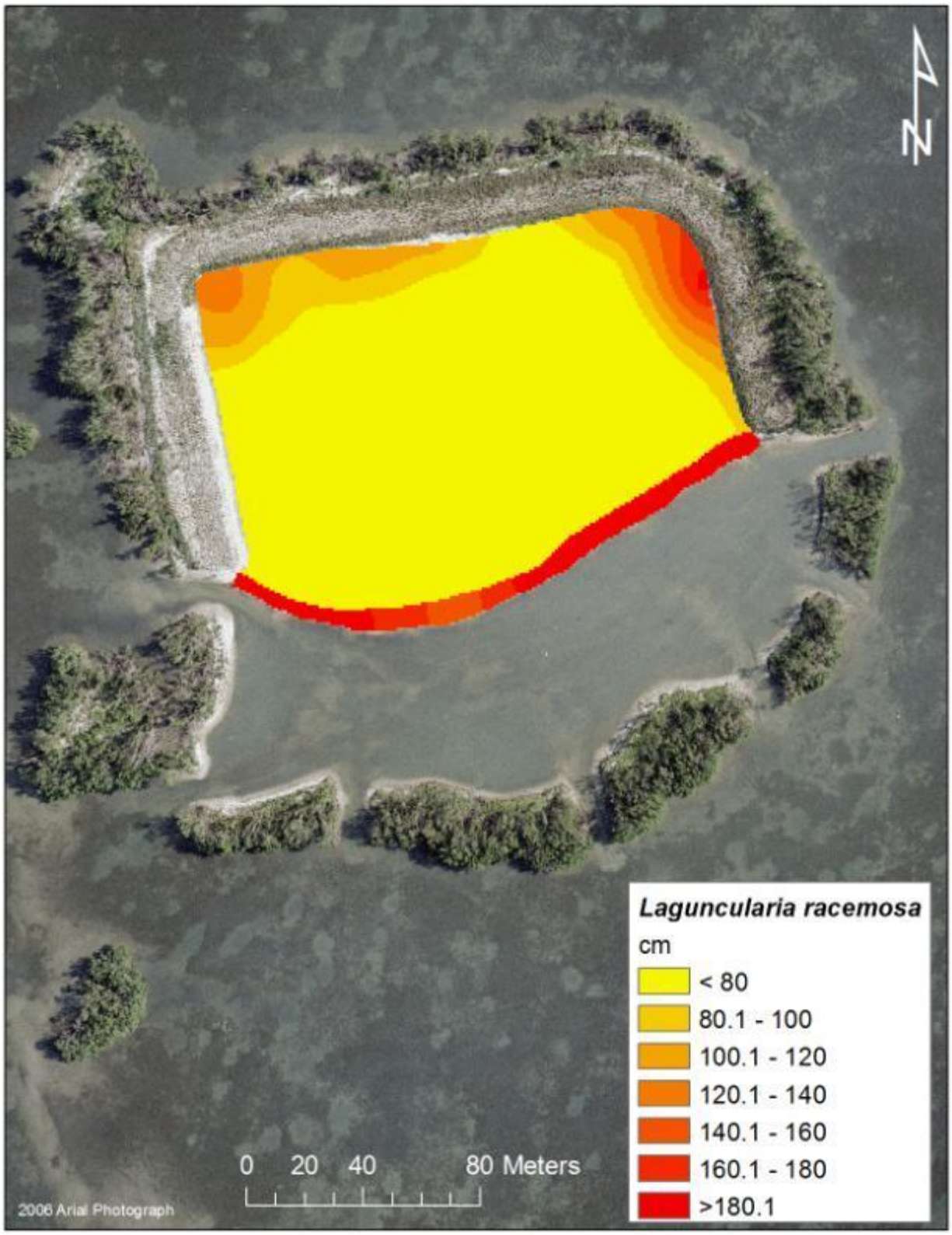

Figure 141. *L racemosa* maximum height for winter 2010.

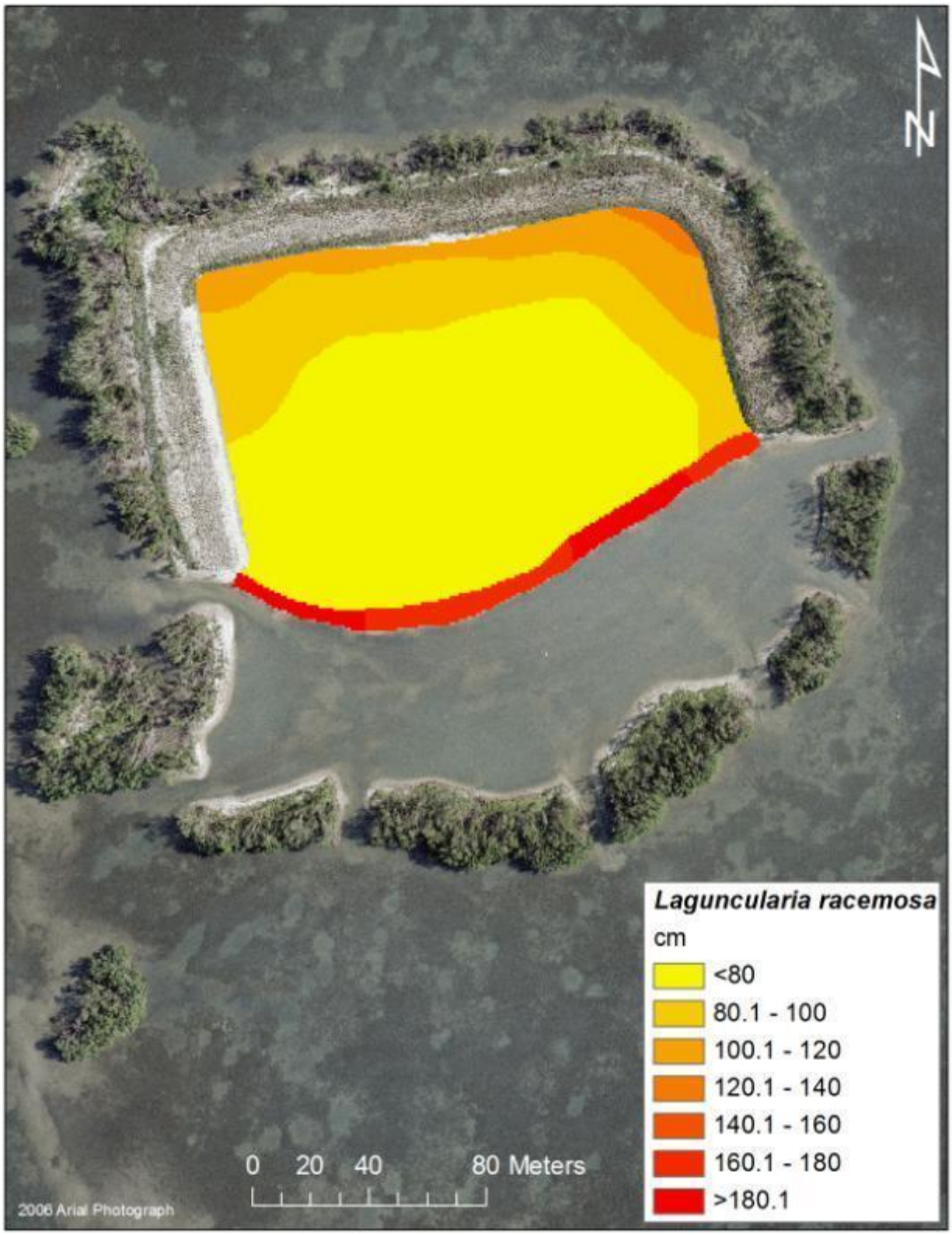

Figure 142. *L racemosa* maximum height for summer 2011.

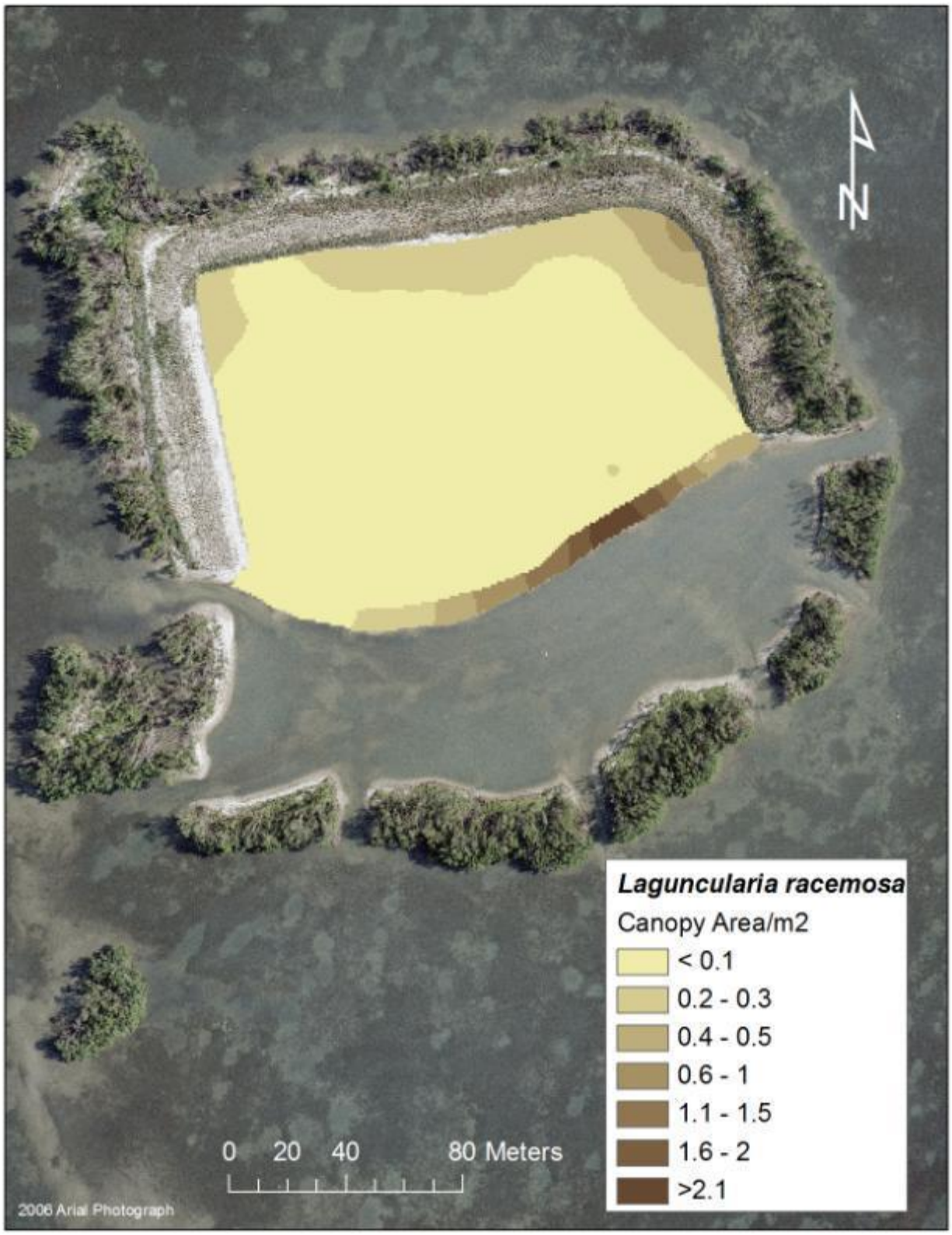

Figure 143. *L racemosa* calculated canopy area/m2 summer 2011.

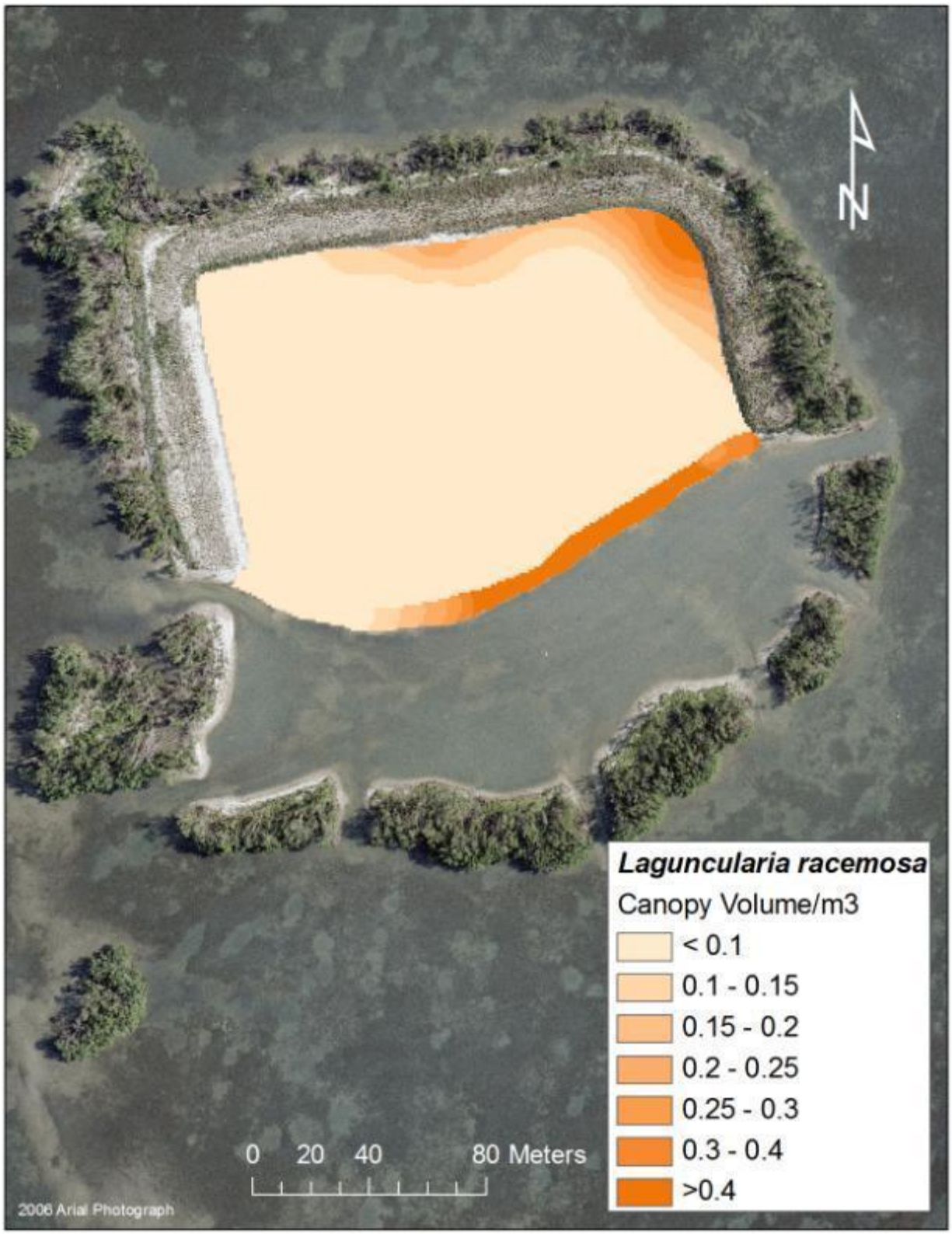

Figure 144. *L racemosa* calculated volume/m3 summer 2011.

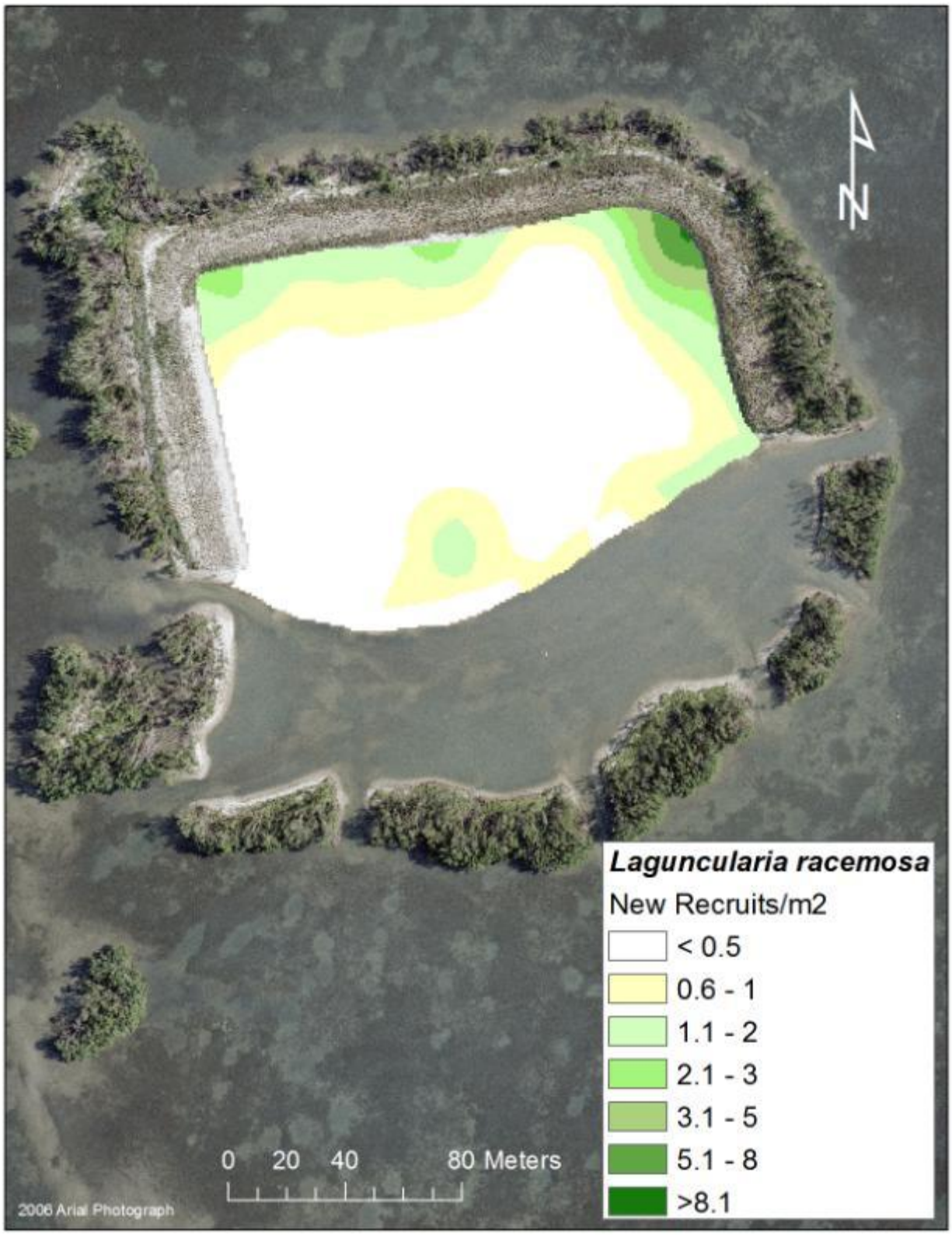

Figure 145. *L racemosa* juvenile mangrove recruits in summer 2011.

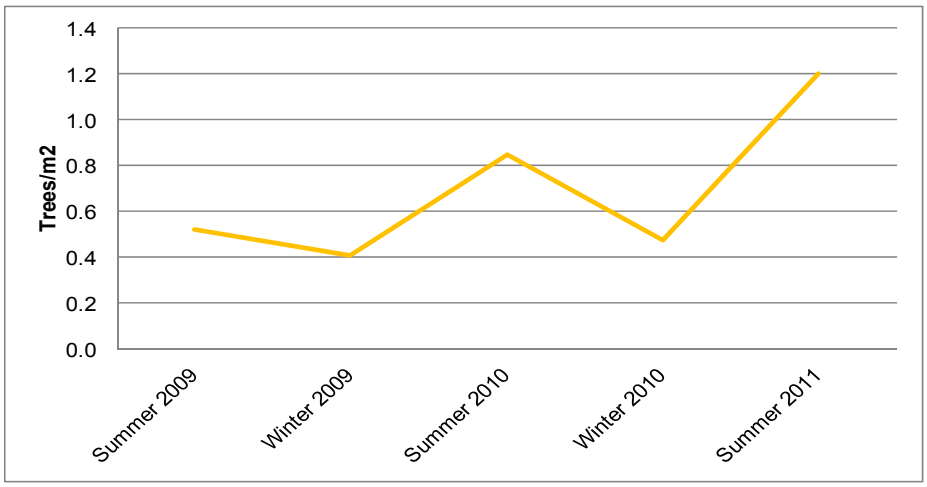

Figure 146. Average *L racemosa* density from summer 2009 to summer 2011.

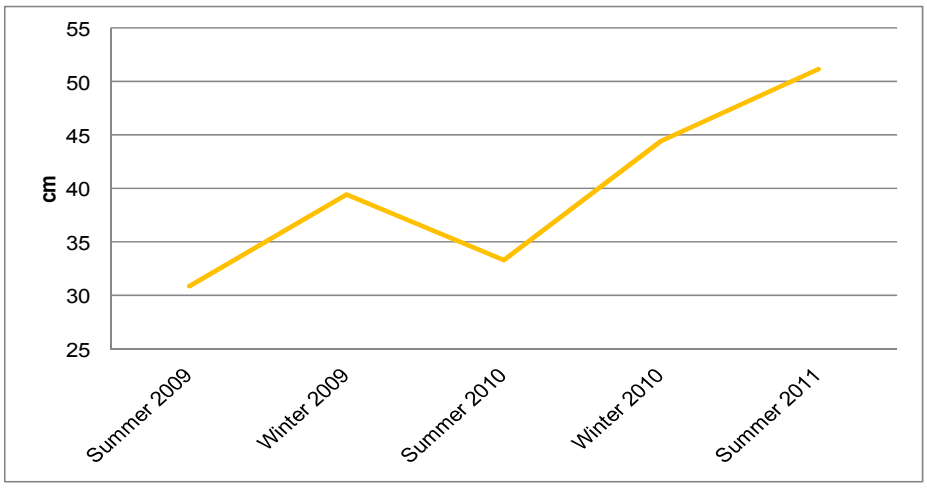

Figure 147. Average *L racemosa* height from summer 2009 to summer 2011.

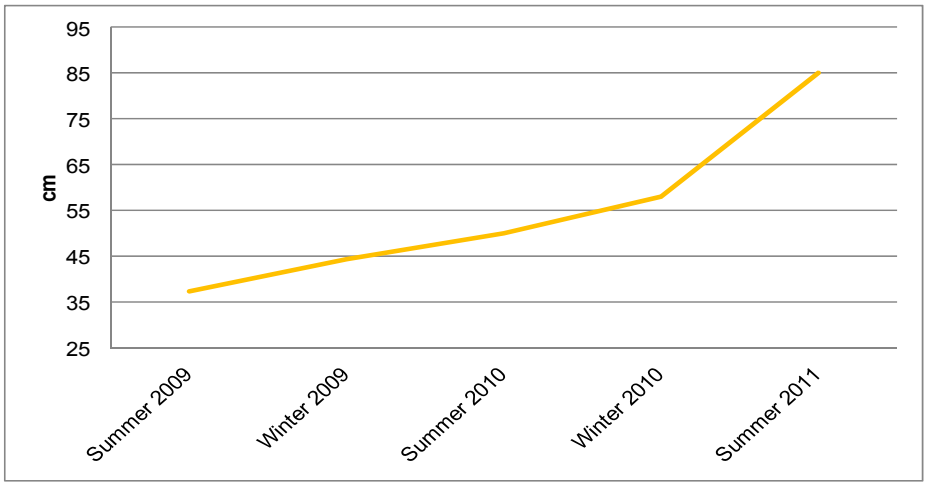

Figure 148. Average *L racemosa* maximum height from summer 2009 to summer 2011.

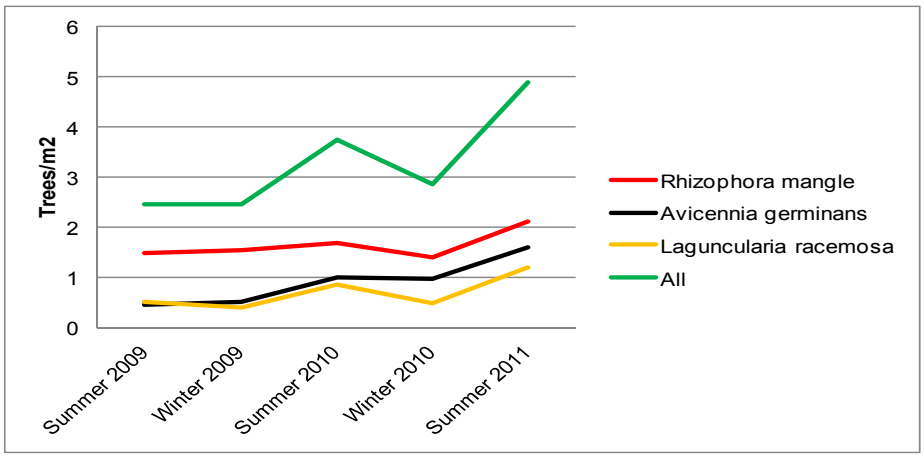

Figure 149. Average mangrove density for all species of mangrove from summer 2009 to summer 2011.

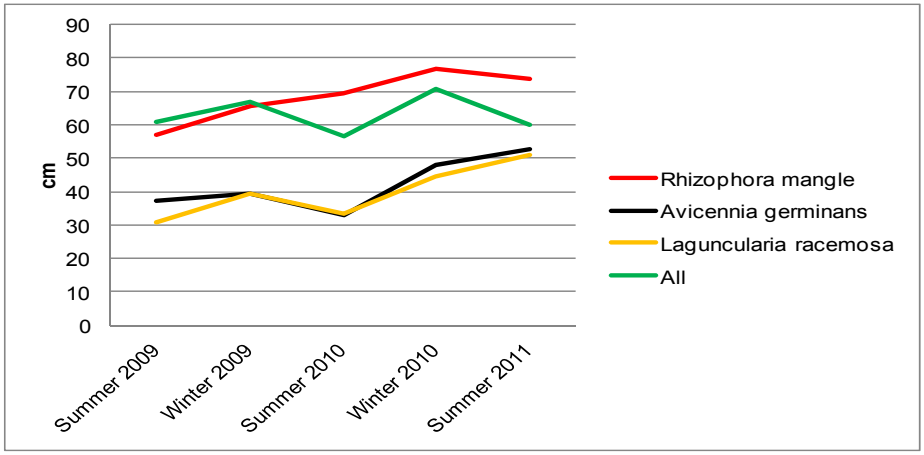

Figure 150. Average mangrove height for all species of mangroves from summer 2009 to summer 2011.

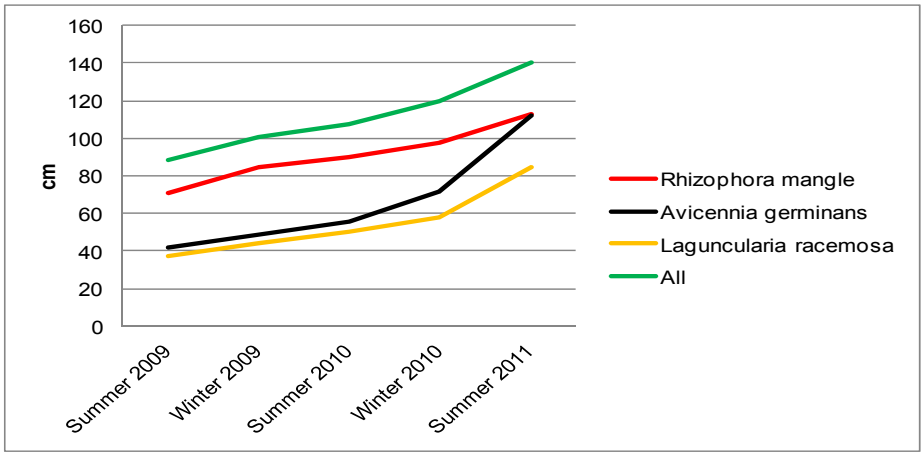

Figure 151. Average mangrove maximum height for all species of mangroves from summer 2009 to summer 2011.

A

B

Figure 152. Drift algae coverage of seagrass area (A) and an example depicting percent cover estimation of the algae (B).

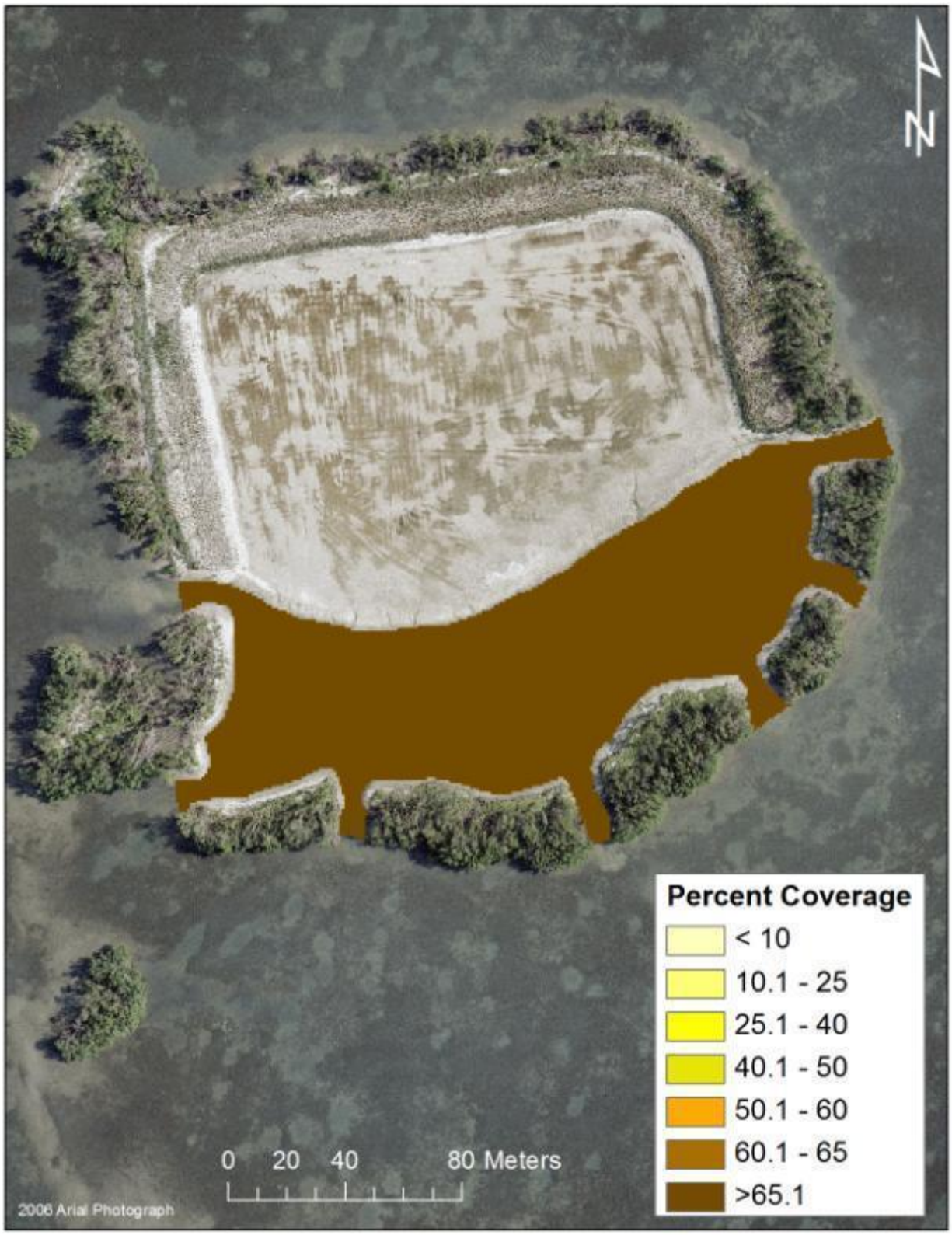

Figure 153. Algae coverage in the seagrass recruitment area during winter 2008.

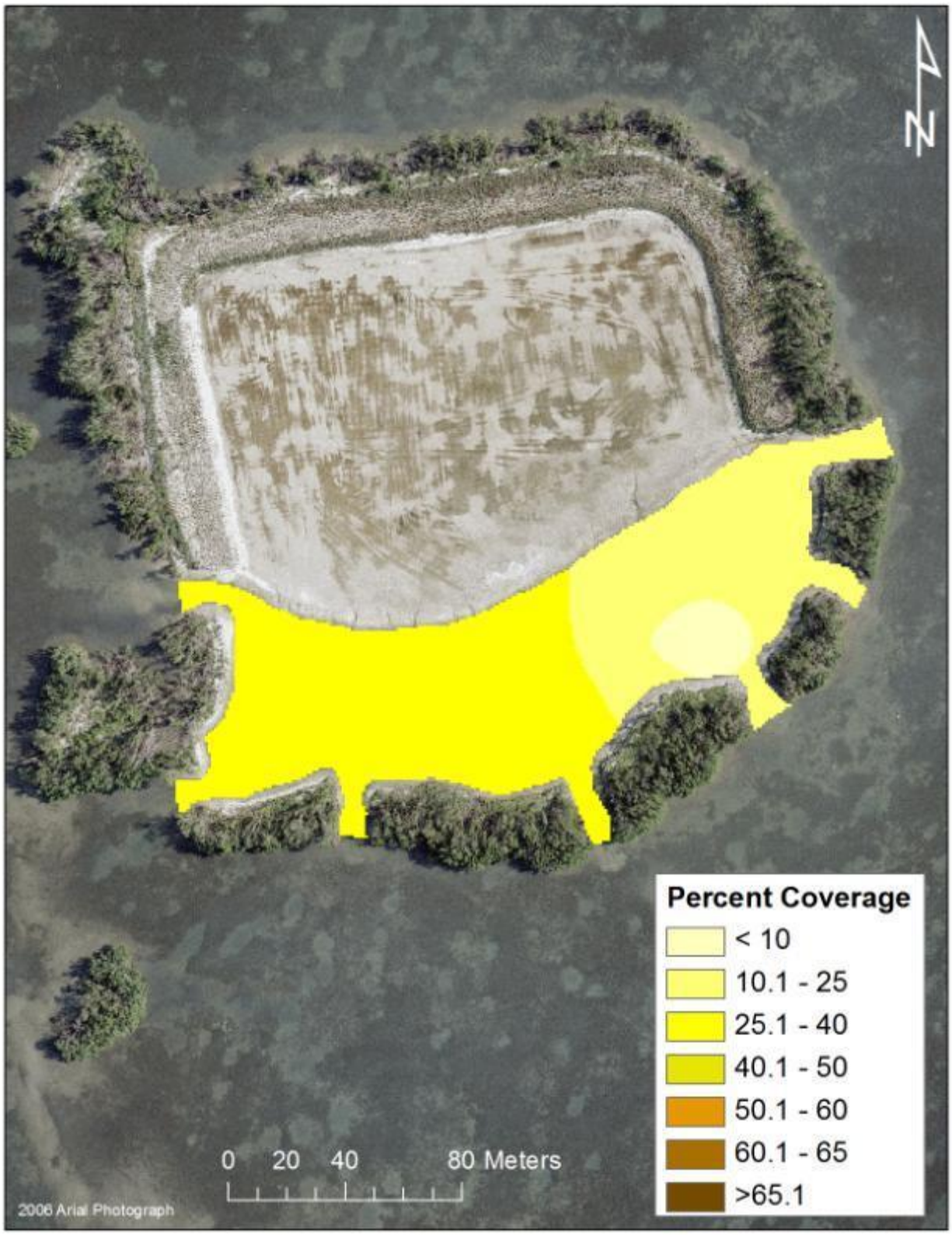

Figure 154. Algae coverage in the seagrass recruitment area during summer 2009.

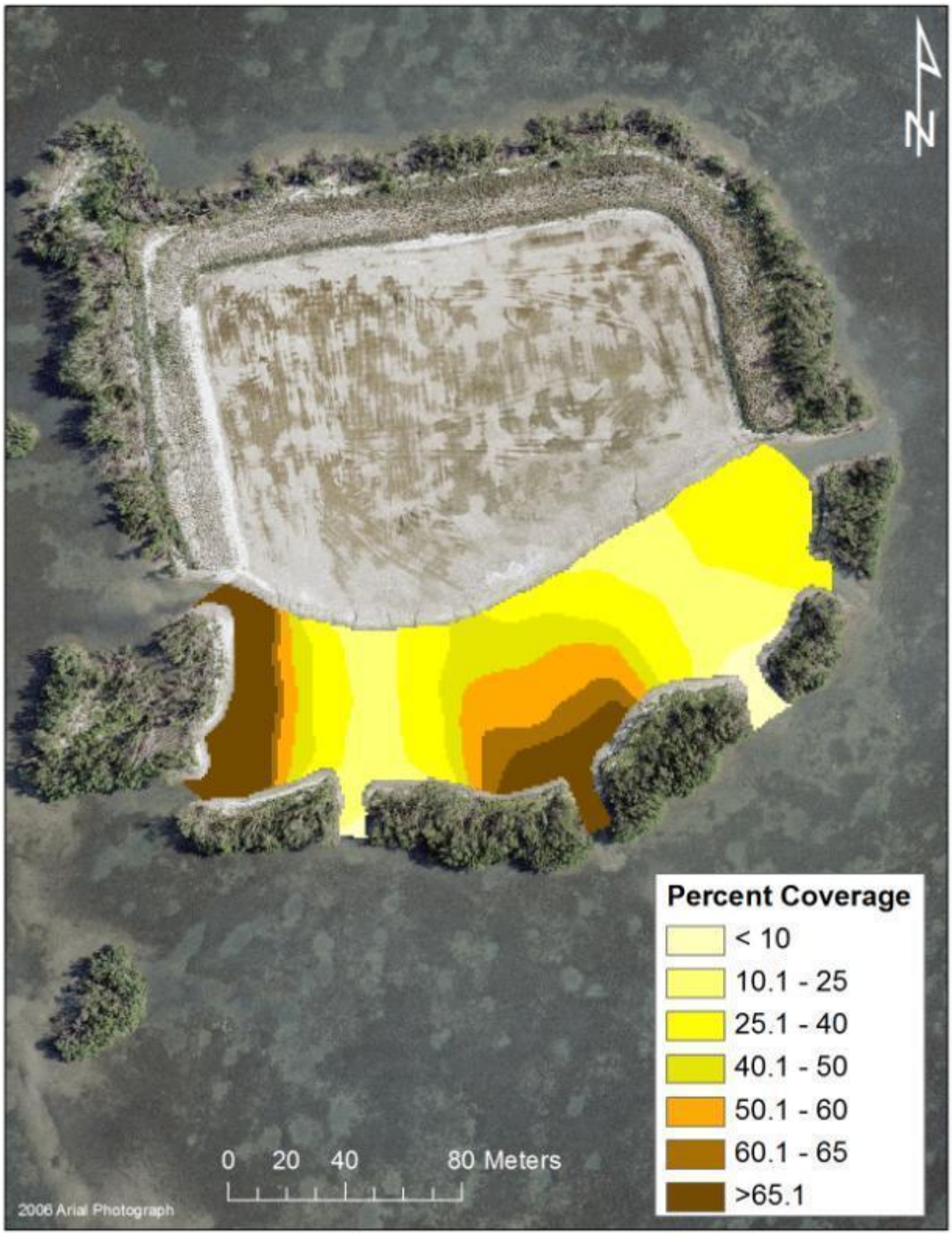

Figure 155. Algae coverage in the seagrass recruitment area during winter 2009.

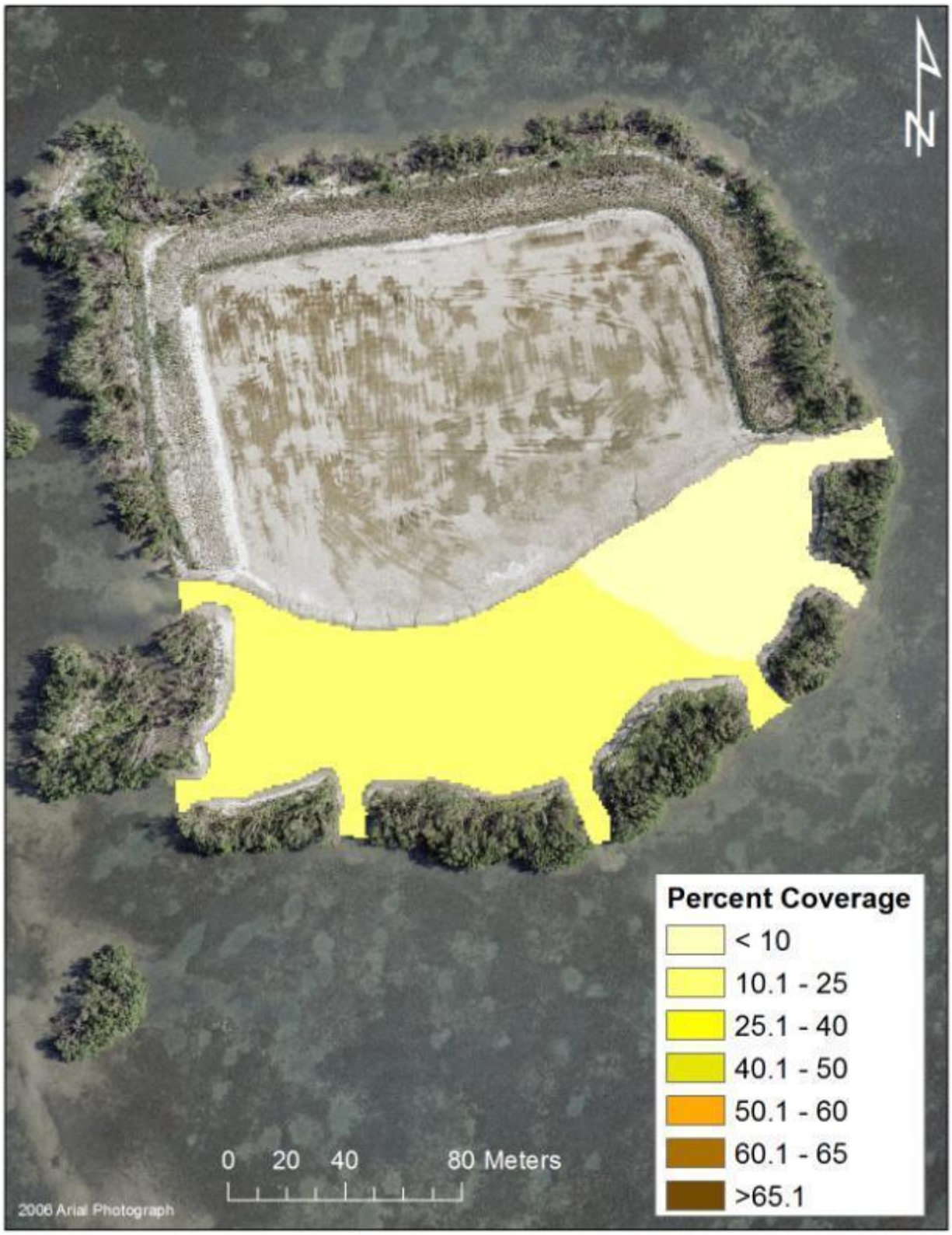

Figure 156. Algae coverage in the seagrass recruitment area during summer 2010.

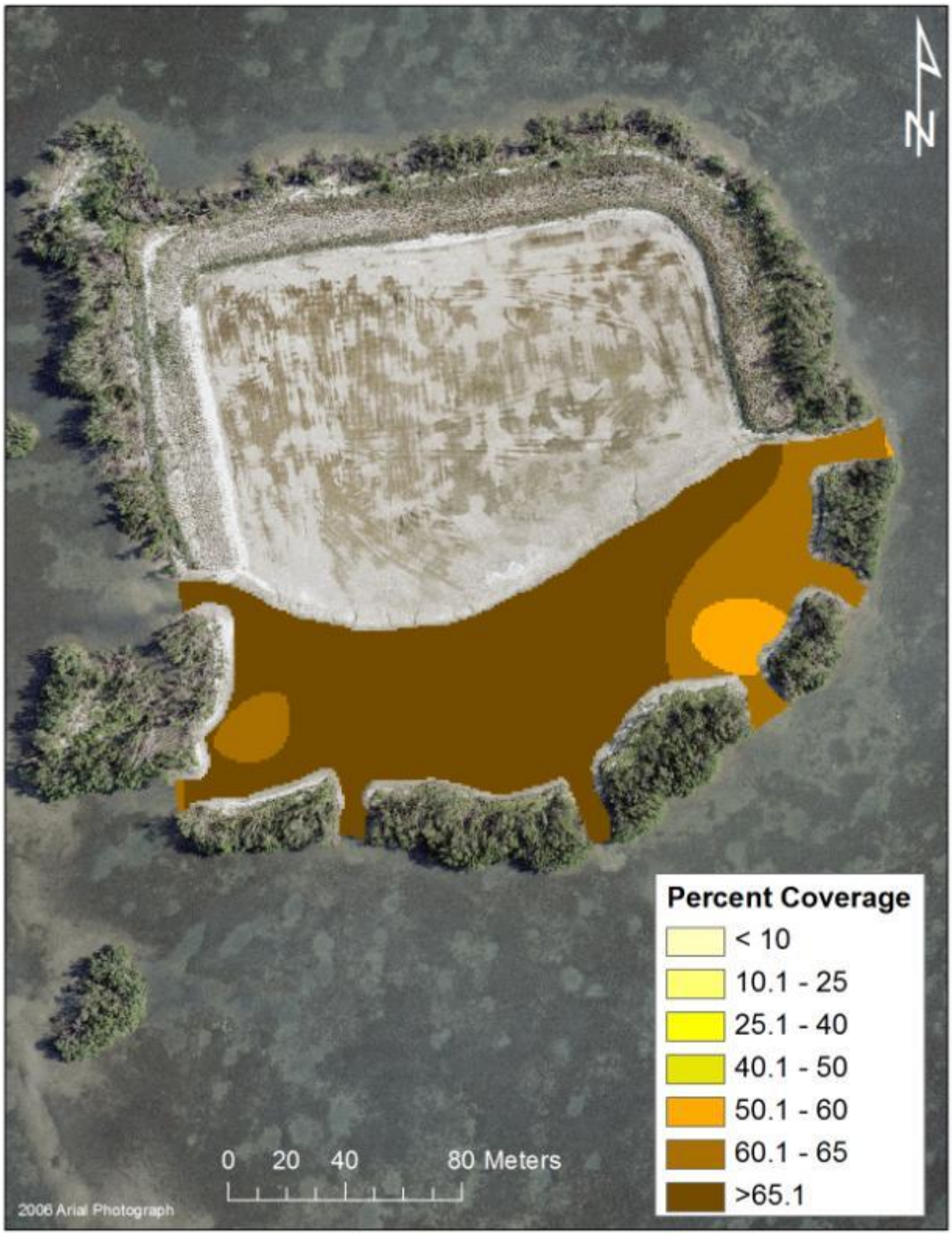

Figure 157. Algae coverage in the seagrass recruitment area during winter 2010.

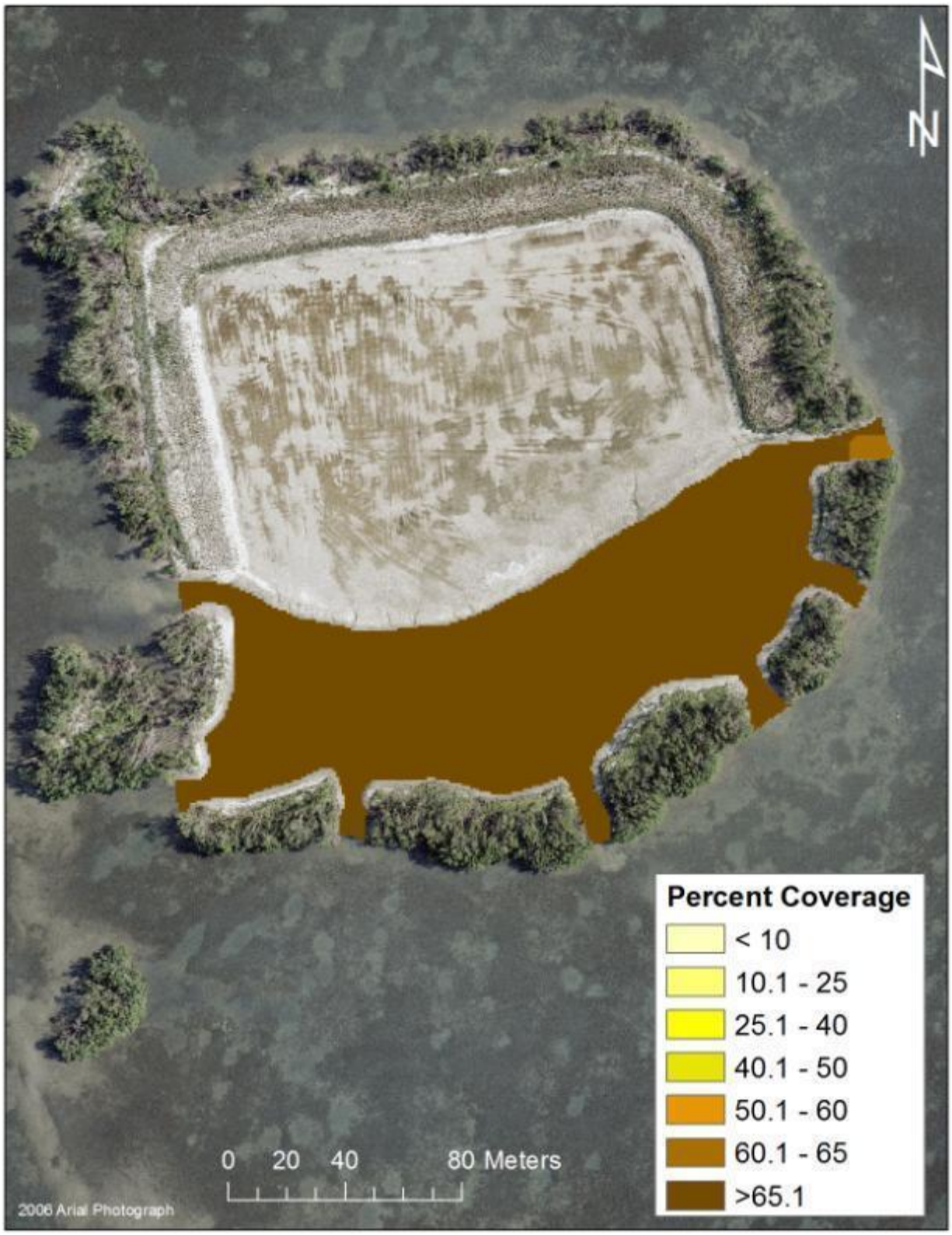

Figure 158. Algae coverage in the seagrass recruitment area during summer 2011.

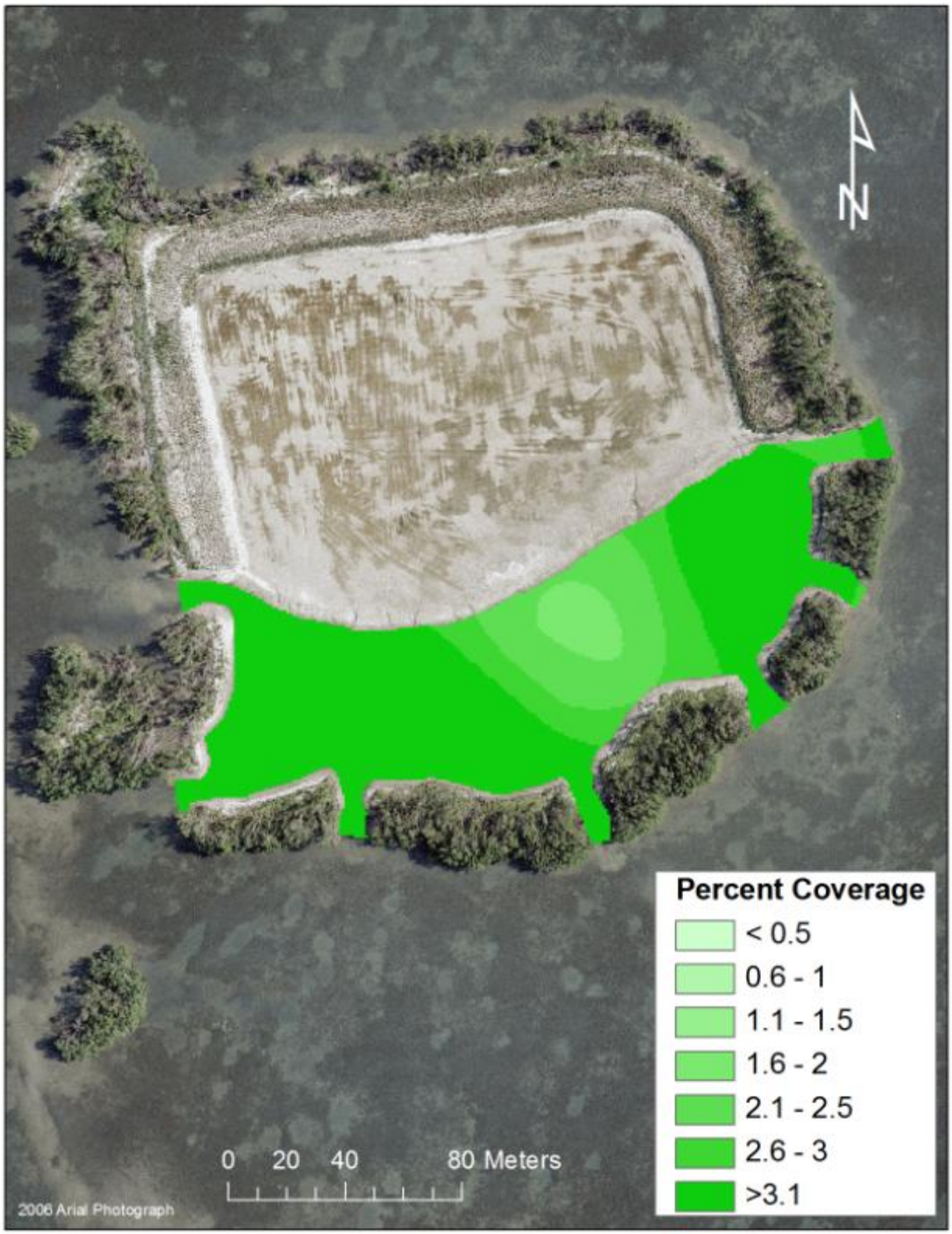

Figure 159. Seagrass coverage in the seagrass recruitment area for winter 2008.

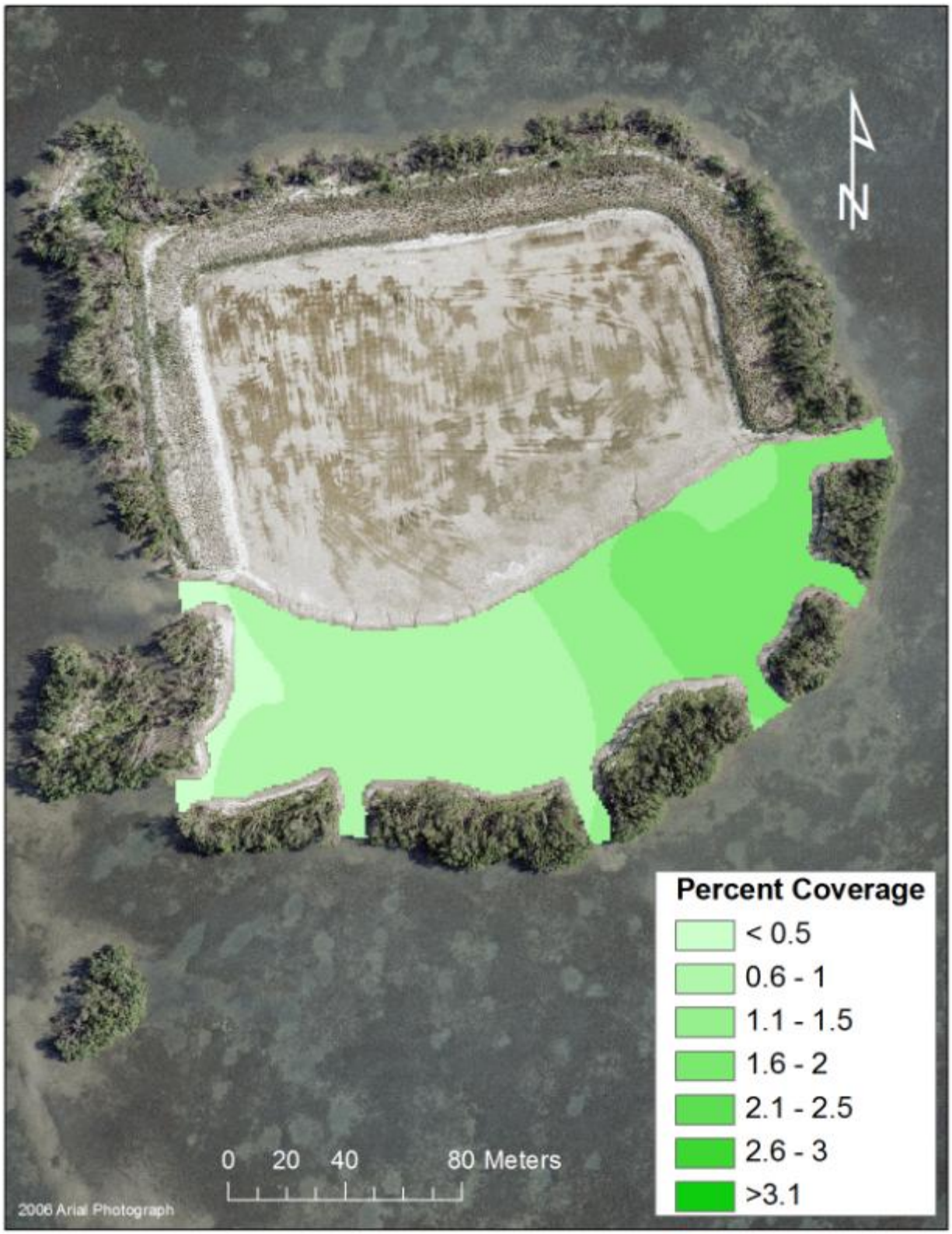

Figure 160. Seagrass coverage in the seagrass recruitment area for summer 2009.

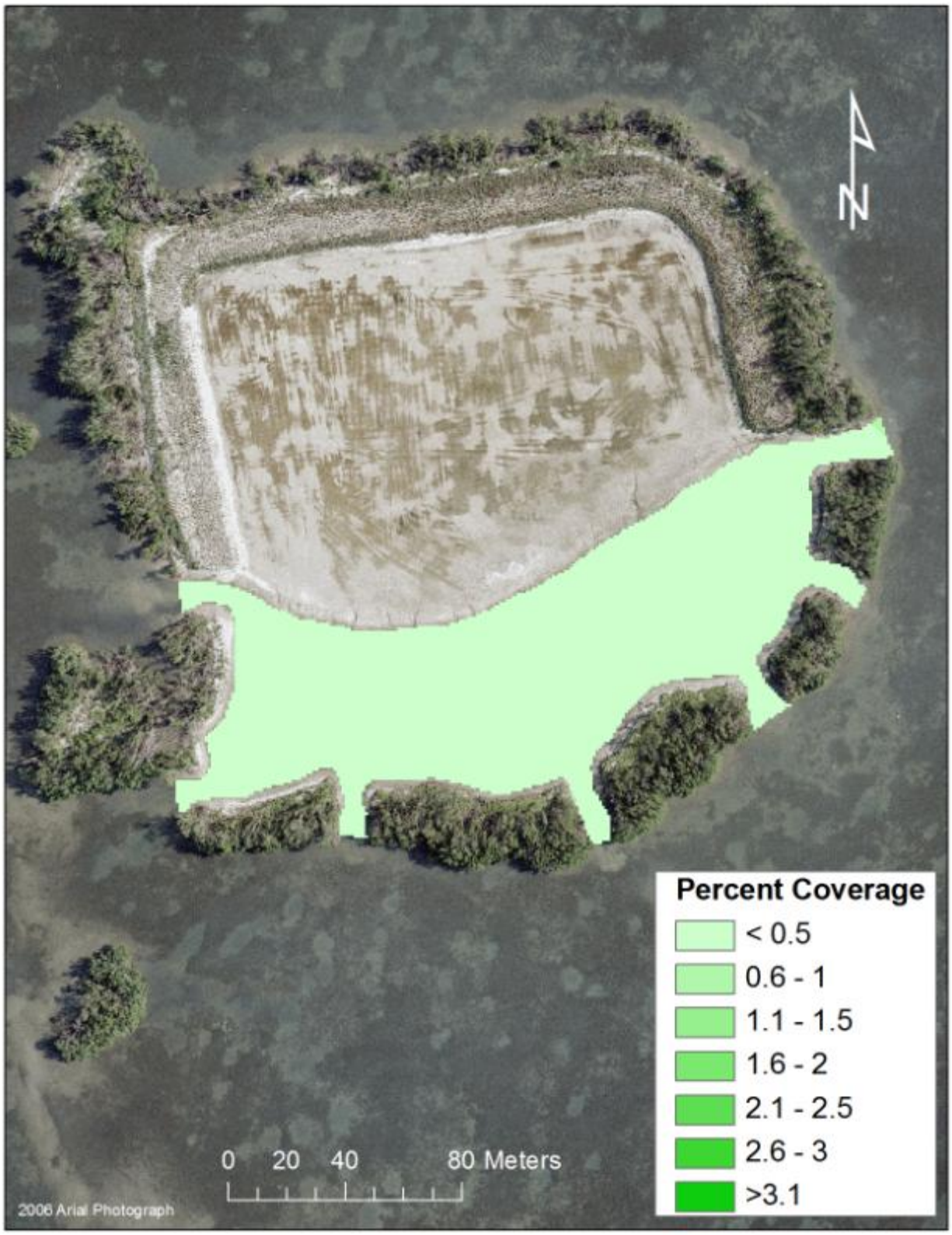

Figure 161. Seagrass coverage in the seagrass recruitment area for winter 2009.

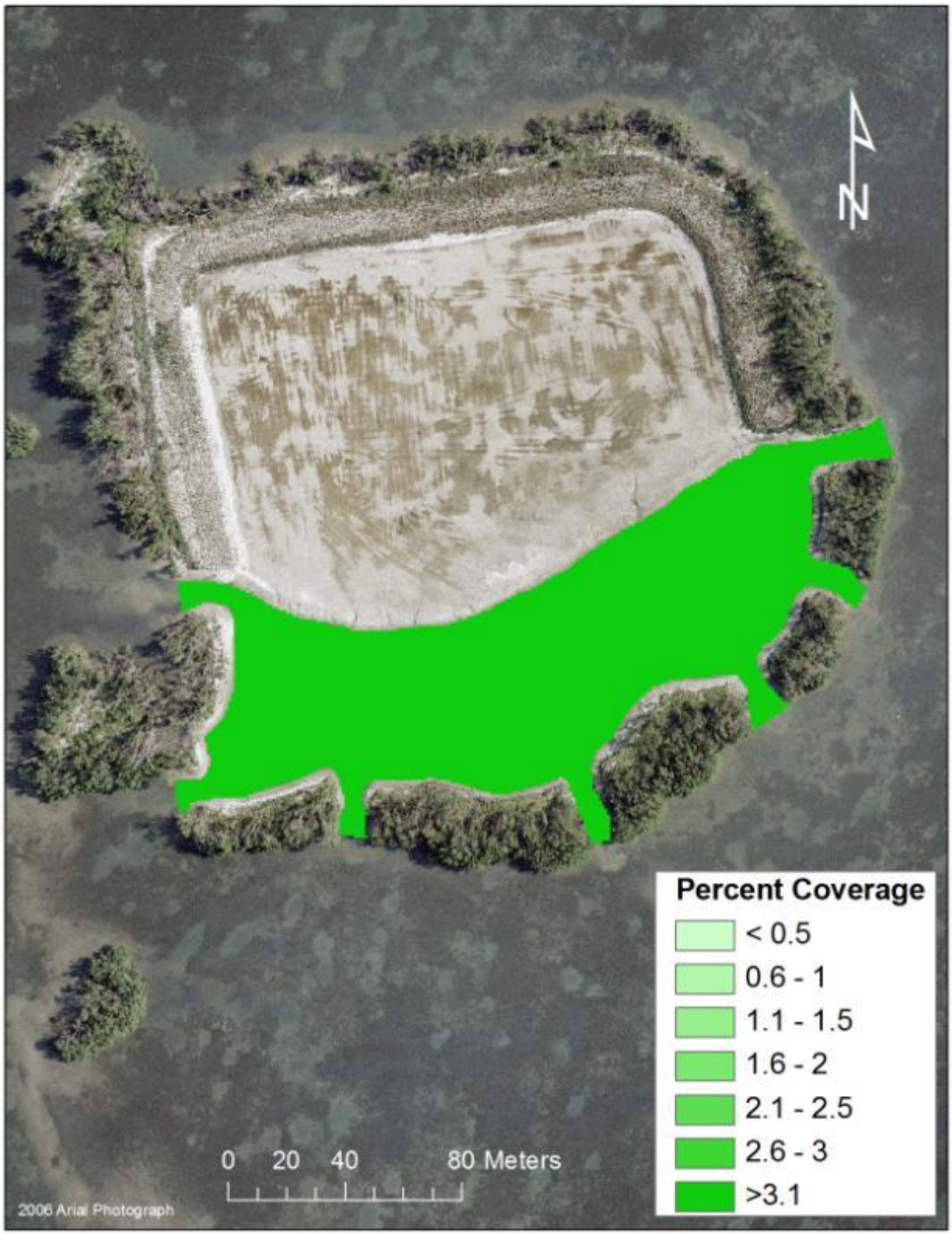

Figure 162. Seagrass coverage in the seagrass recruitment area for summer 2010.

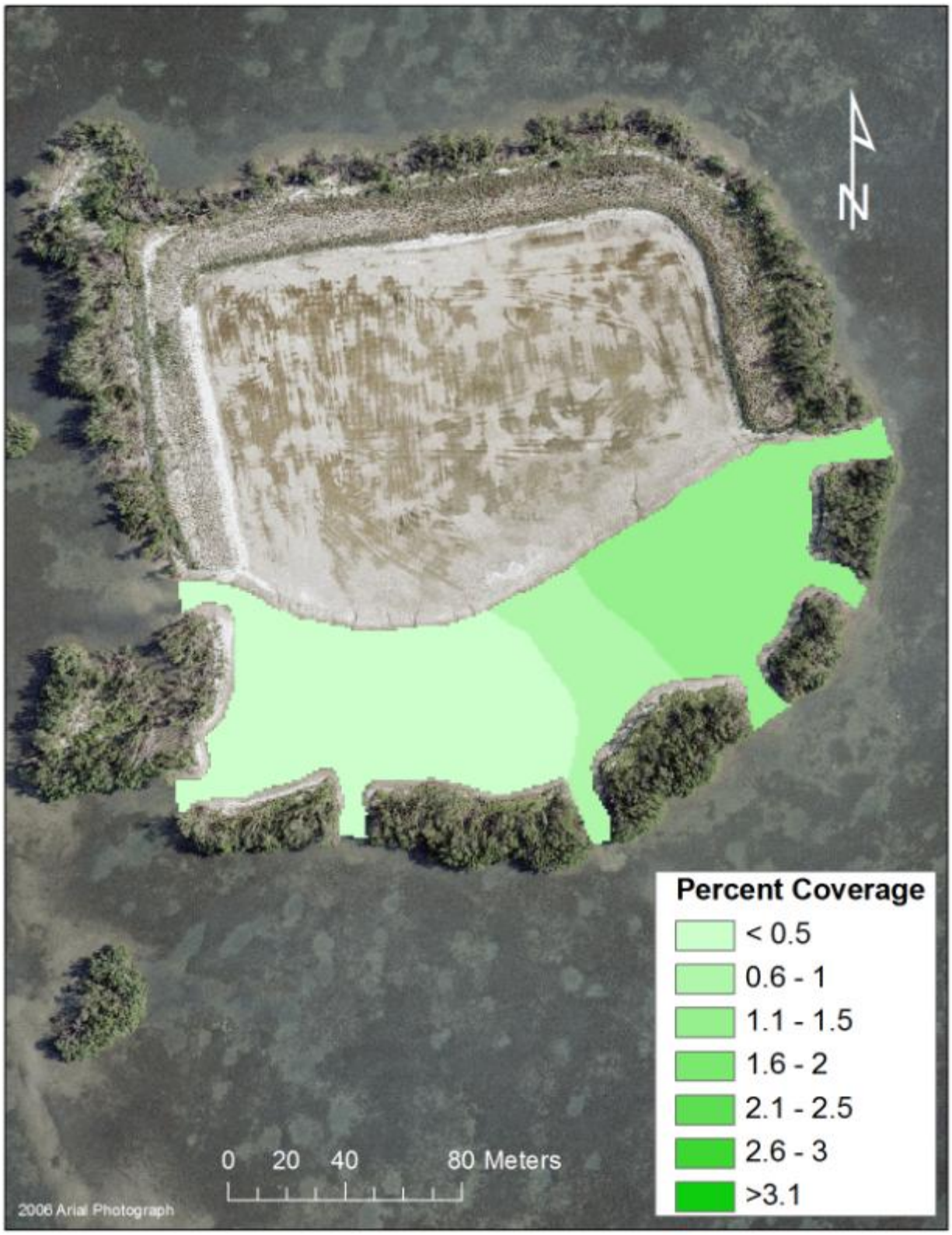

Figure 163. Seagrass coverage in the seagrass recruitment area for winter 2010.

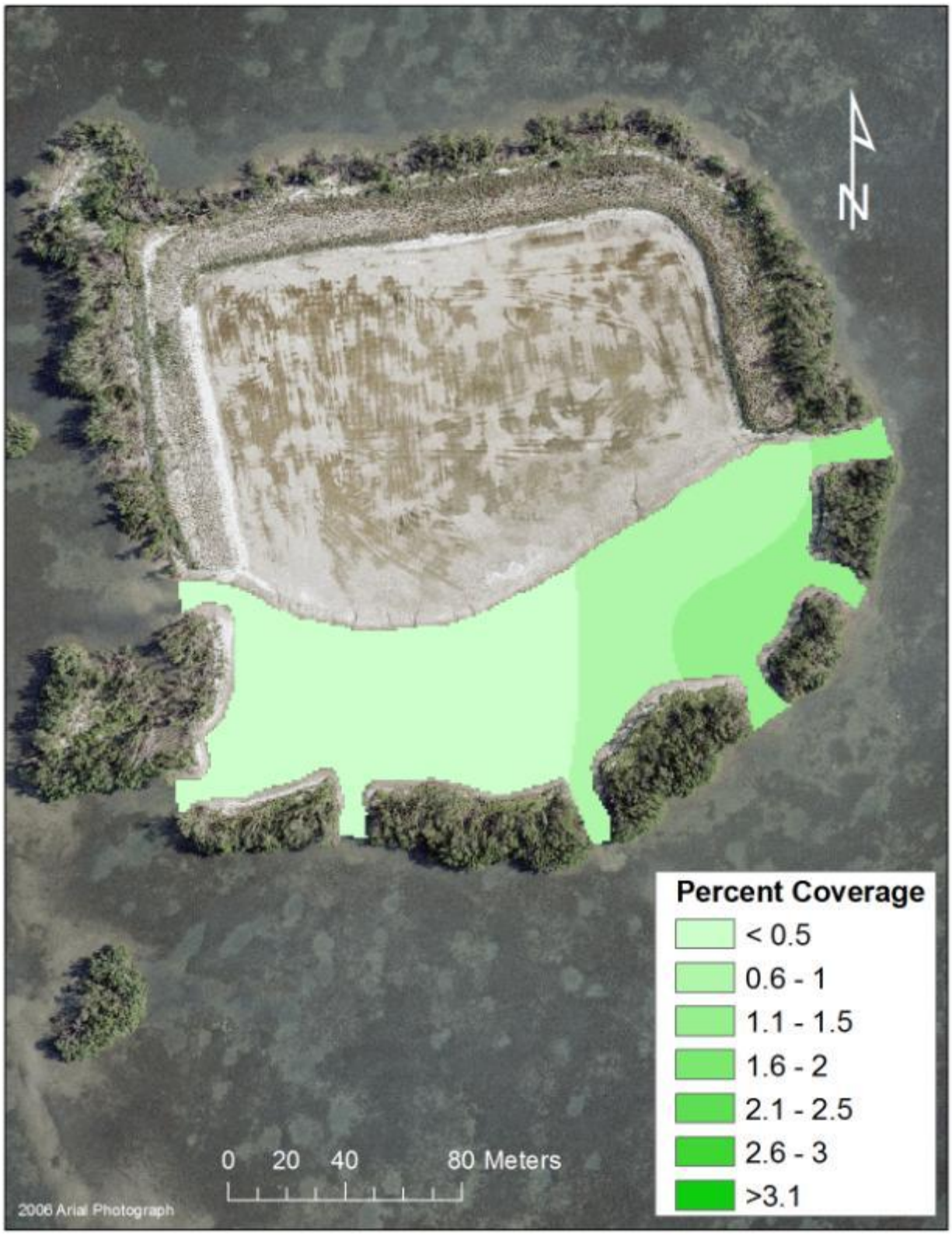

Figure 164. Seagrass coverage in the seagrass recruitment area for summer 2011.

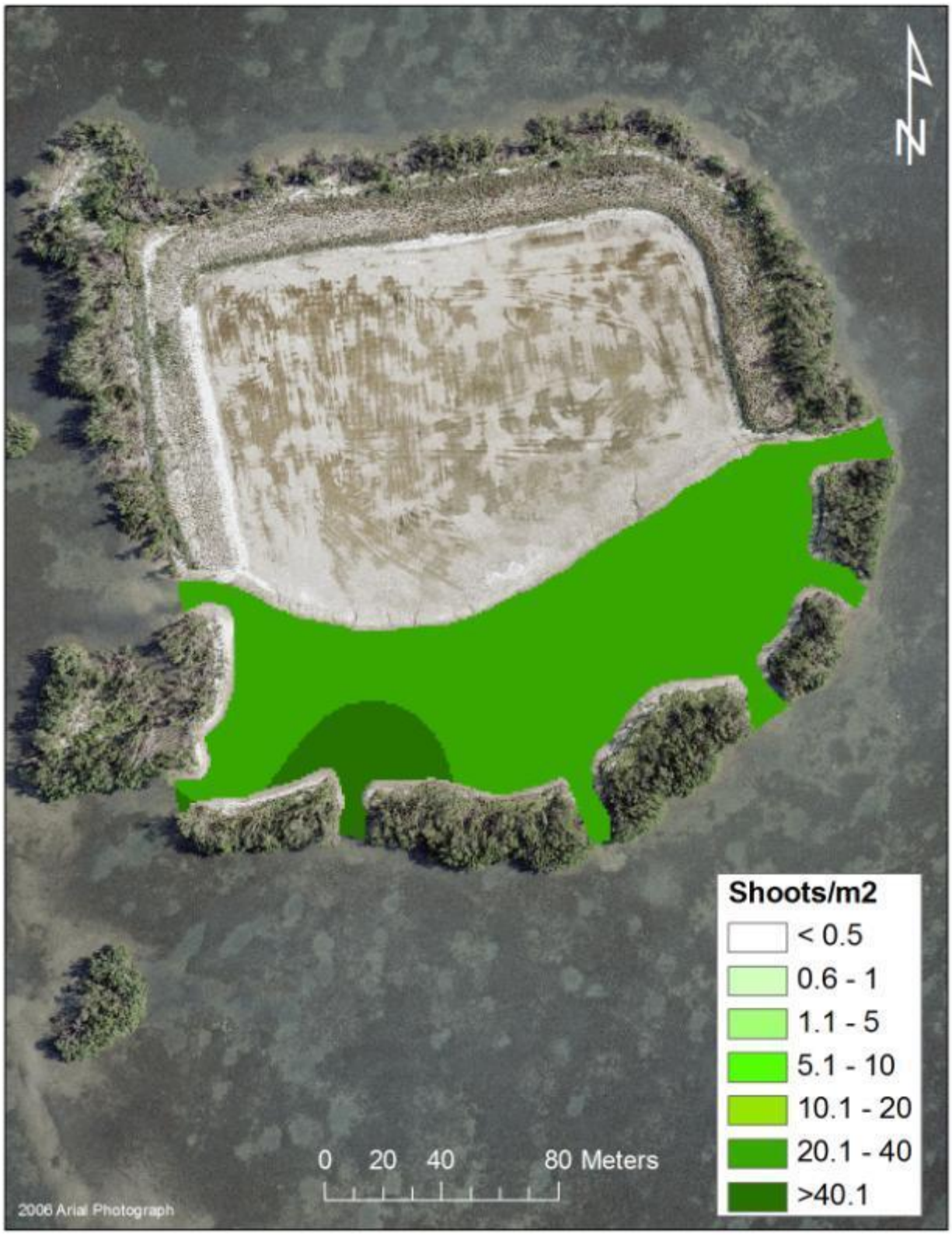

Figure 165. Seagrass density (shoots/m2) in the seagrass recruitment area during winter 2008.

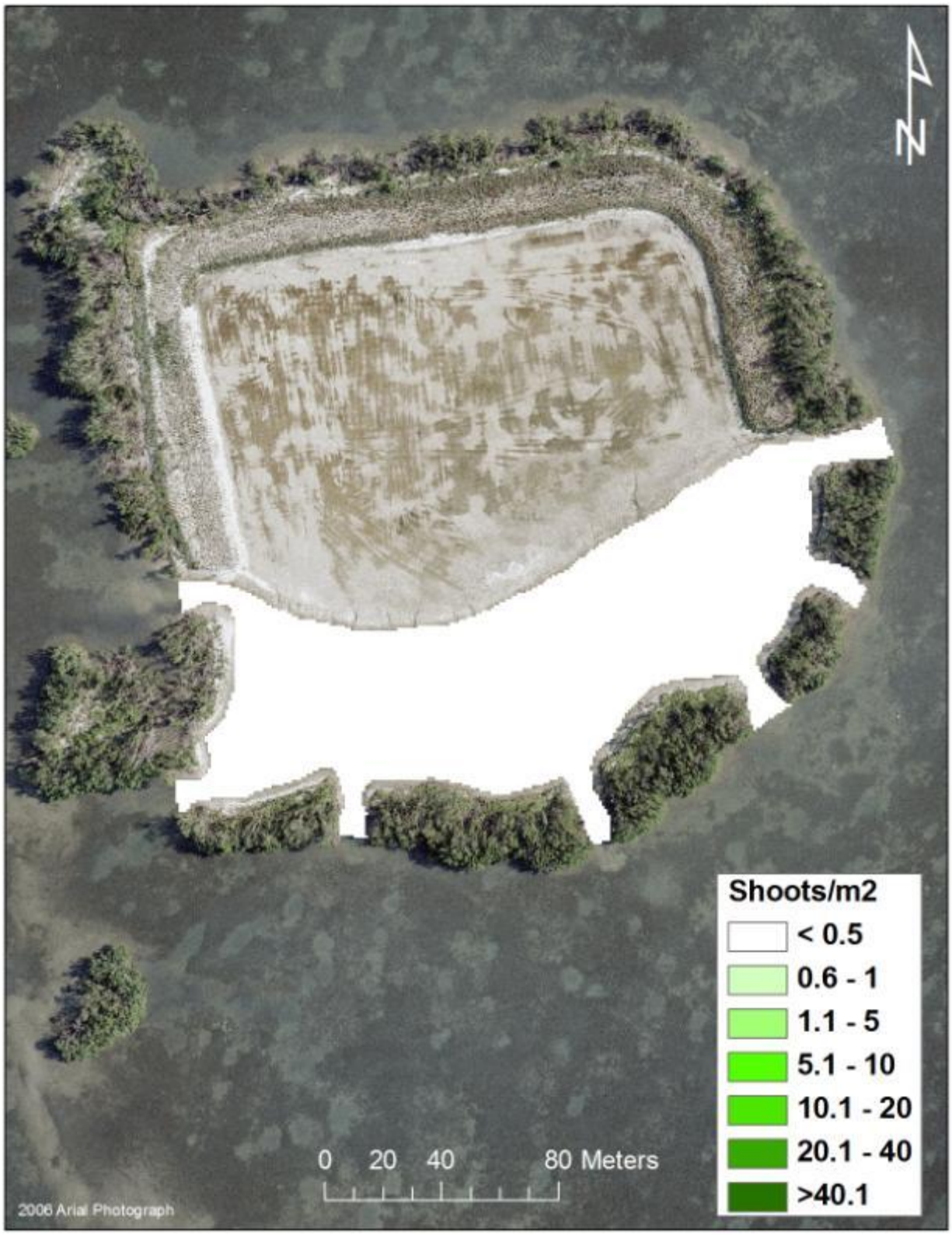

Figure 166. Seagrass density (shoots/m2) in the seagrass recruitment area during summer 2009.

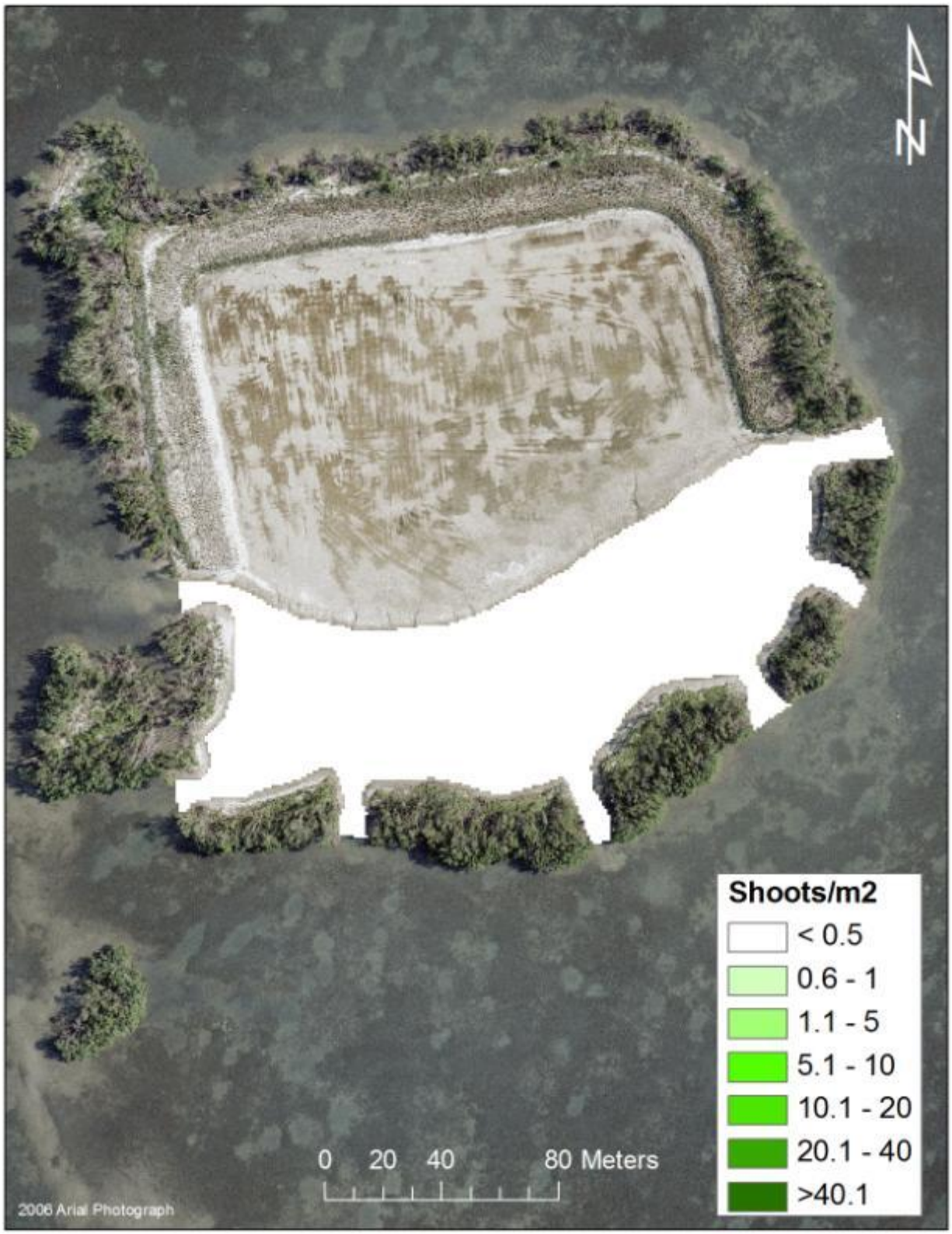

Figure 167. Seagrass density (shoots/m2) in the seagrass recruitment area during winter 2009.

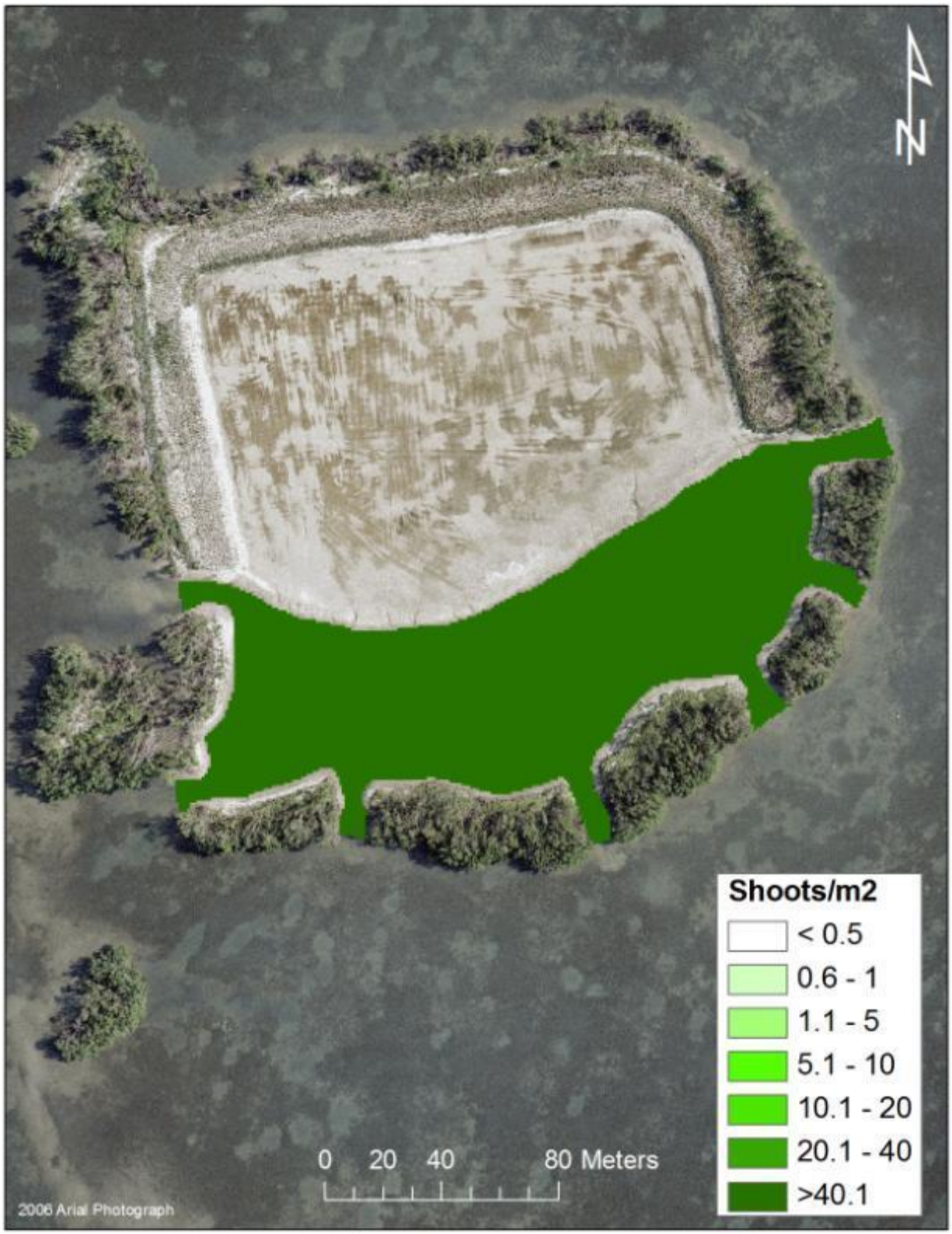

Figure 168. Seagrass density (shoots/m2) in the seagrass recruitment area during summer 2010.

Figure 169. Seagrass density (shoots/m2) in the seagrass recruitment area during winter 2010.

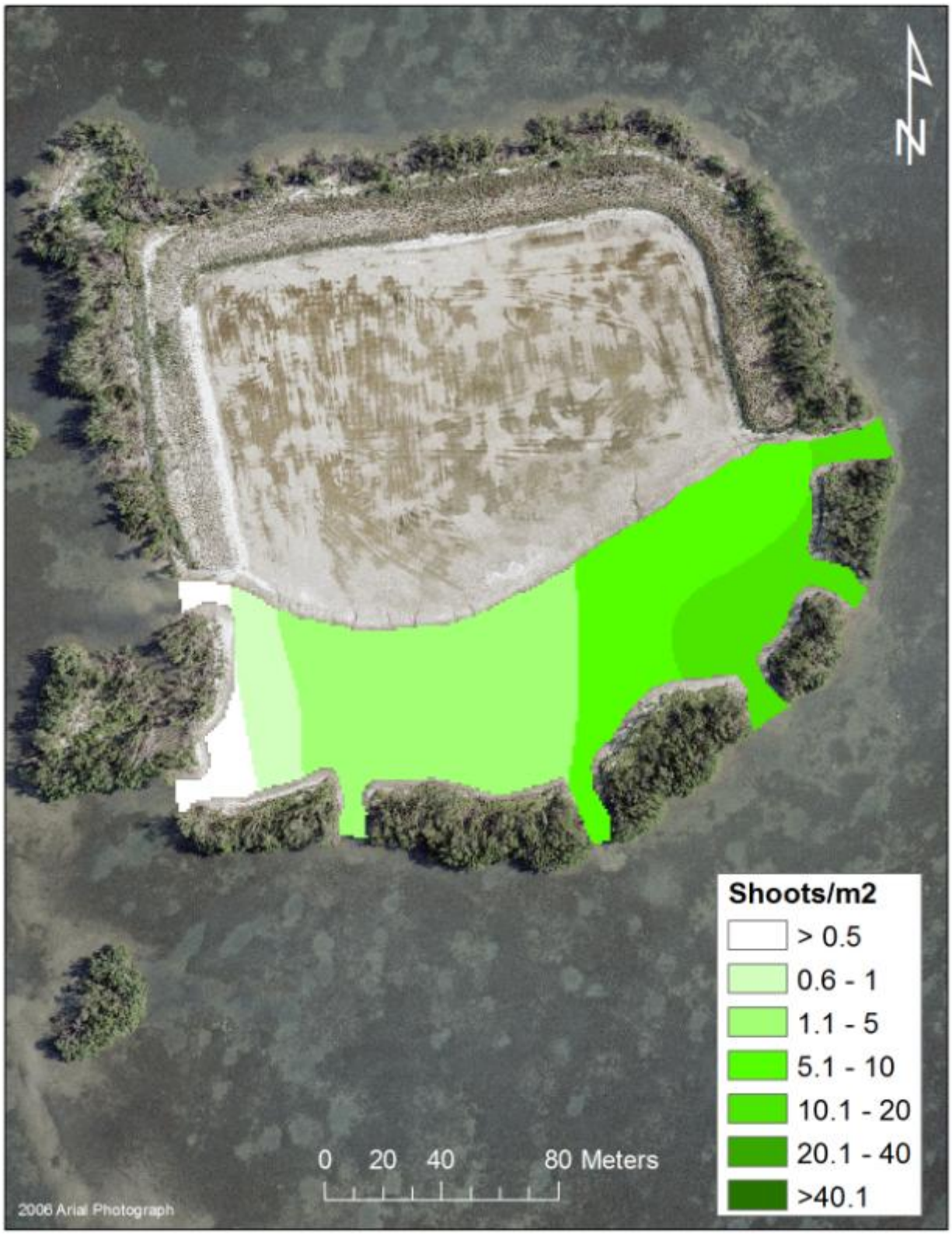

Figure 170. Seagrass density (shoots/m2) in the seagrass recruitment area during summer 2011.

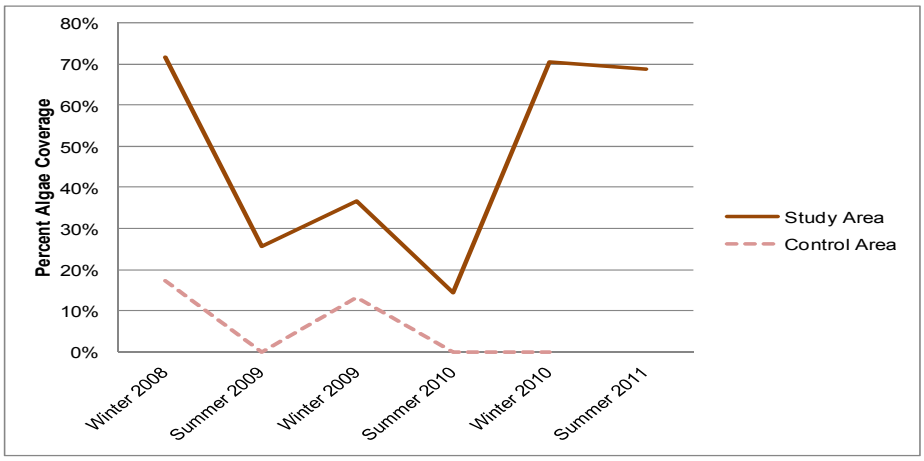

Figure 171. Average percent algae coverage for the seagrass recruitment area and the control area from winter2008 to summer 2011.

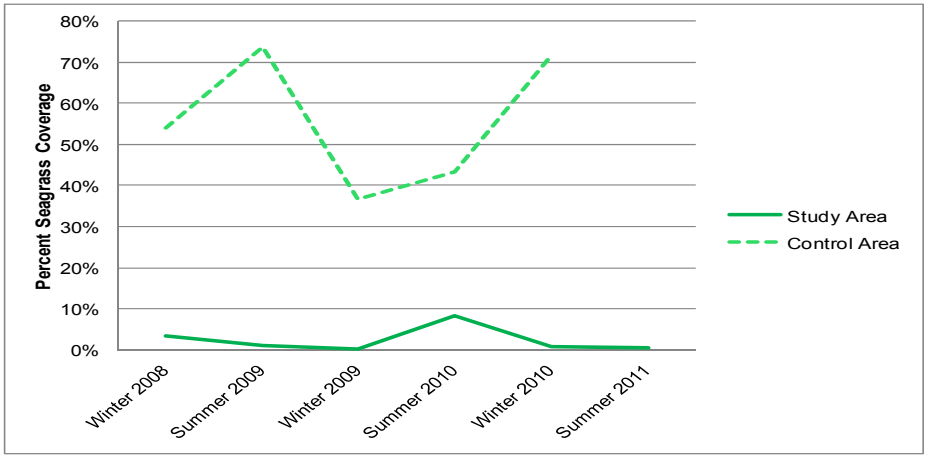

Figure 172. Average percent seagrass coverage for the seagrass recruitment area and the control area from winter2008 to summer 2011.

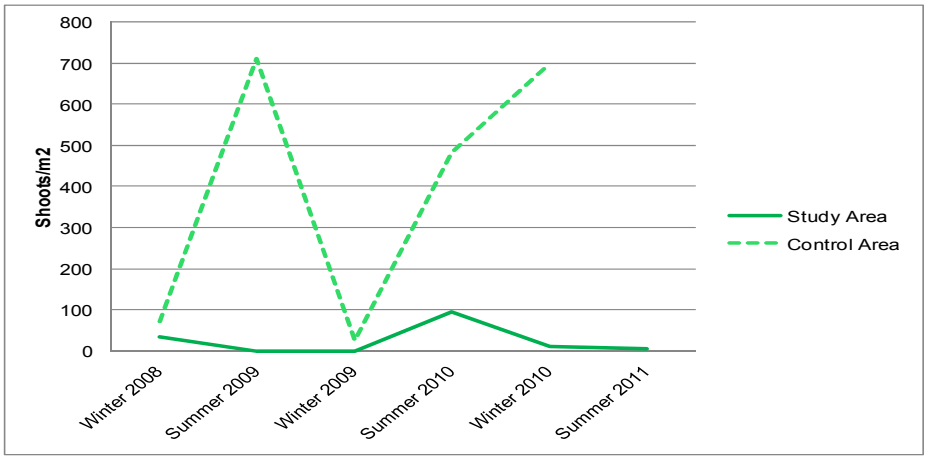

Figure 173. Average seagrass density for the seagrass recruitment area and the control area from winter 2008 to summer 2011.

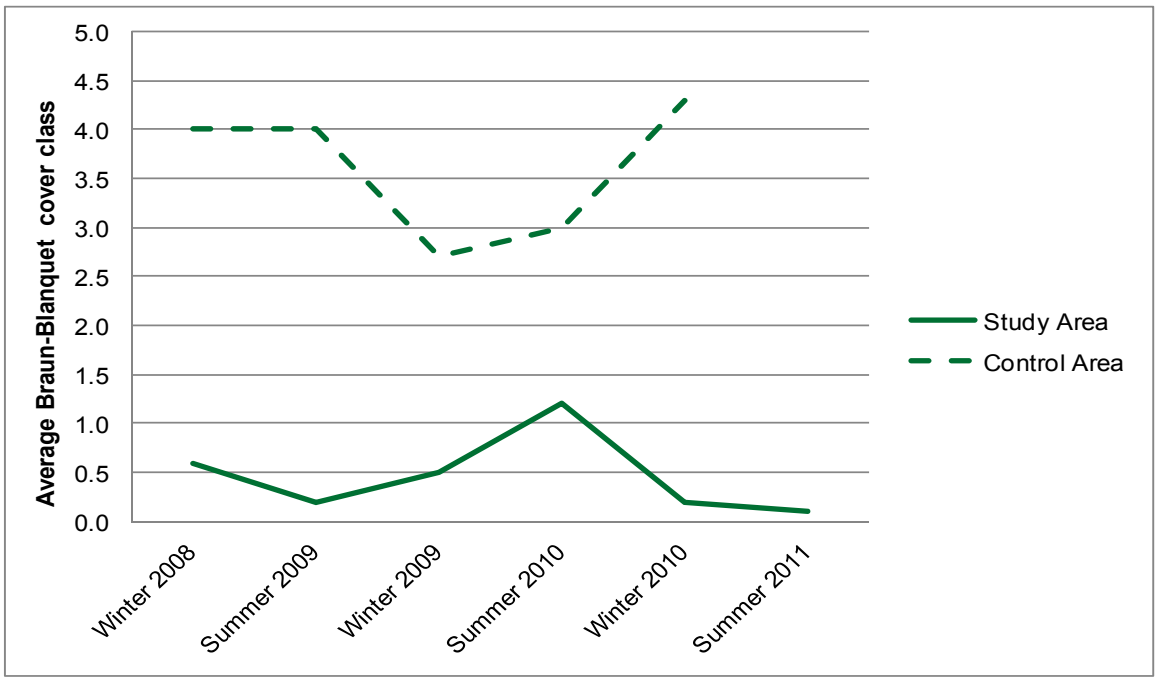

Figure 174. Average Braun-Blanquet cover class for the seagrass recruitment area and the control area from winter 2008 to summer 2011.

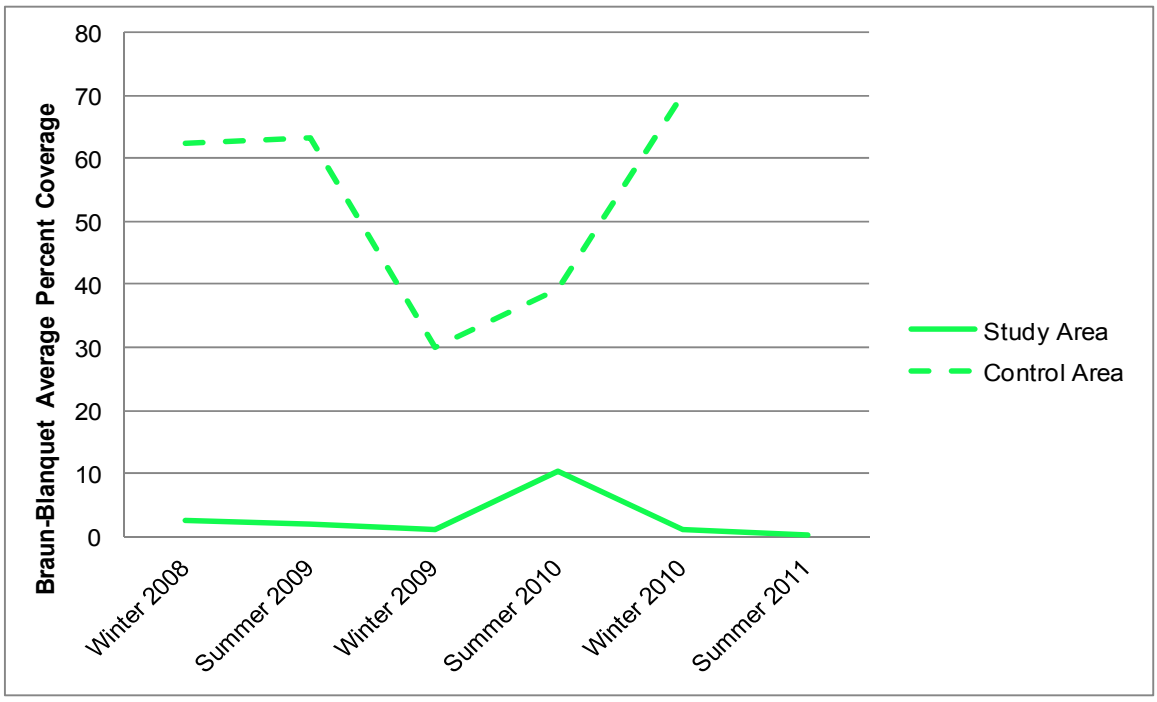

Figure 175. Average Braun-Blanquet percent coverage for the seagrass recruitment area and the control area from winter 2008 to summer 2011.

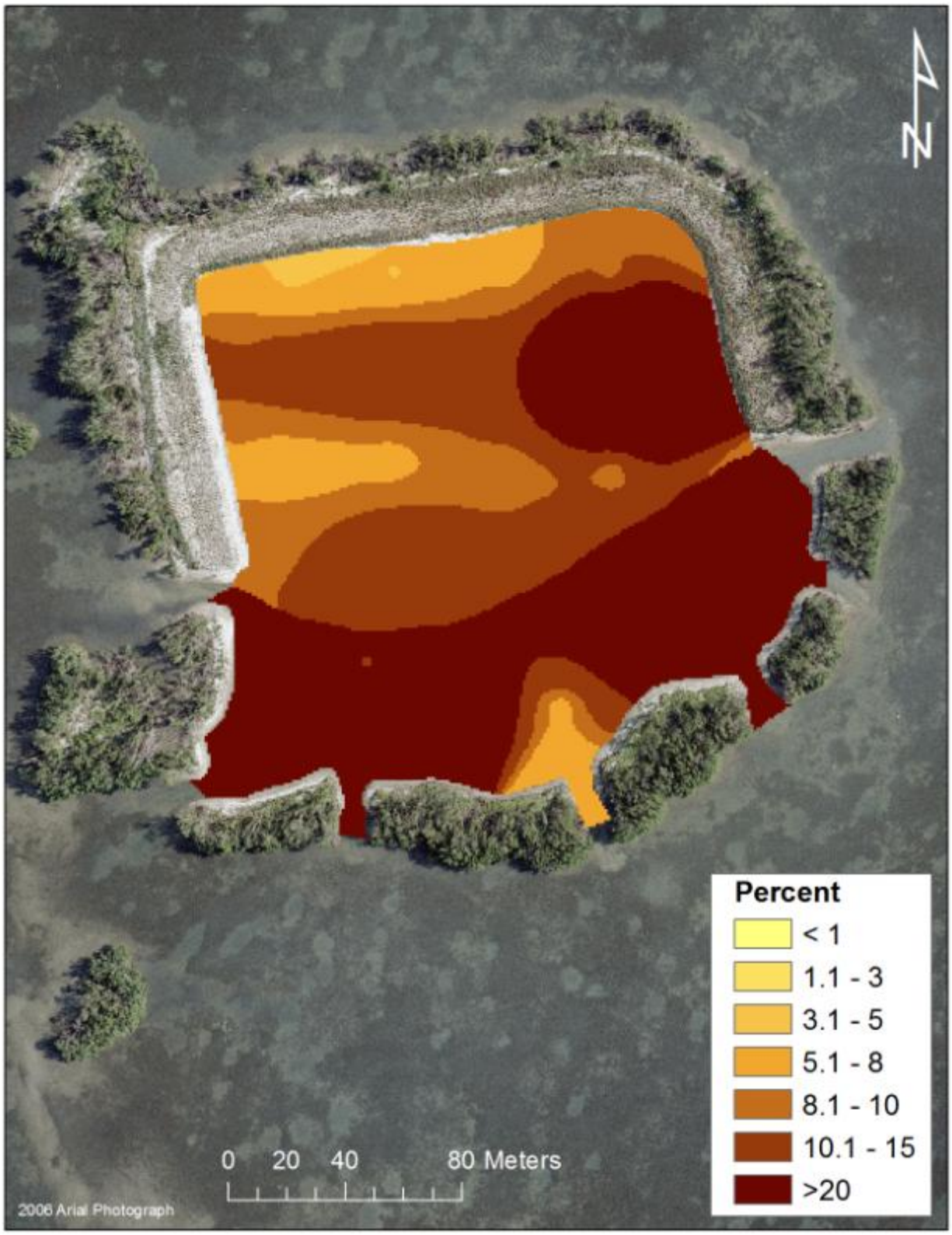

Figure 176. Percentage of soil sample with fine soil particles for winter 2008.

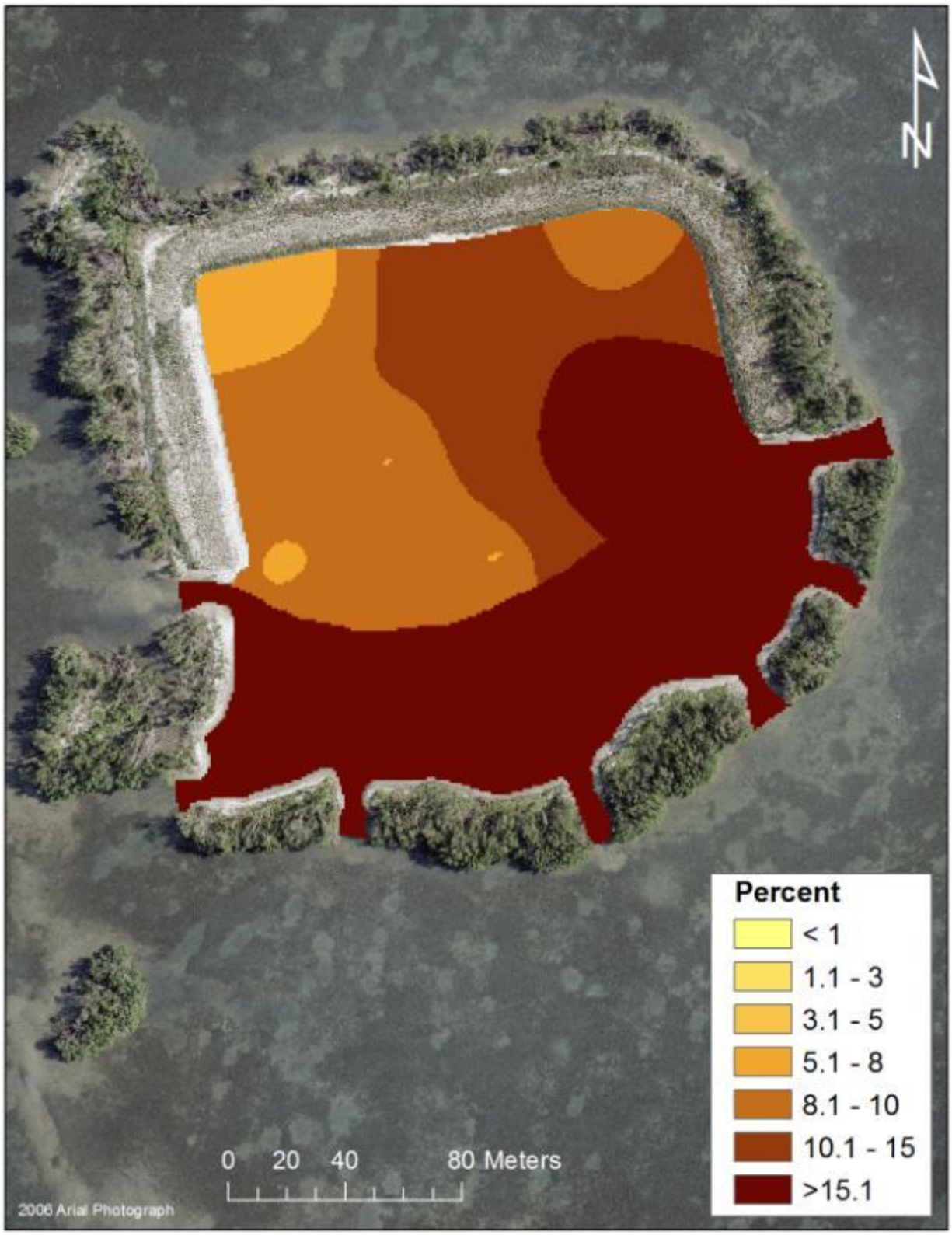

Figure 177. Percentage of soil sample with fine soil particles for summer 2009.

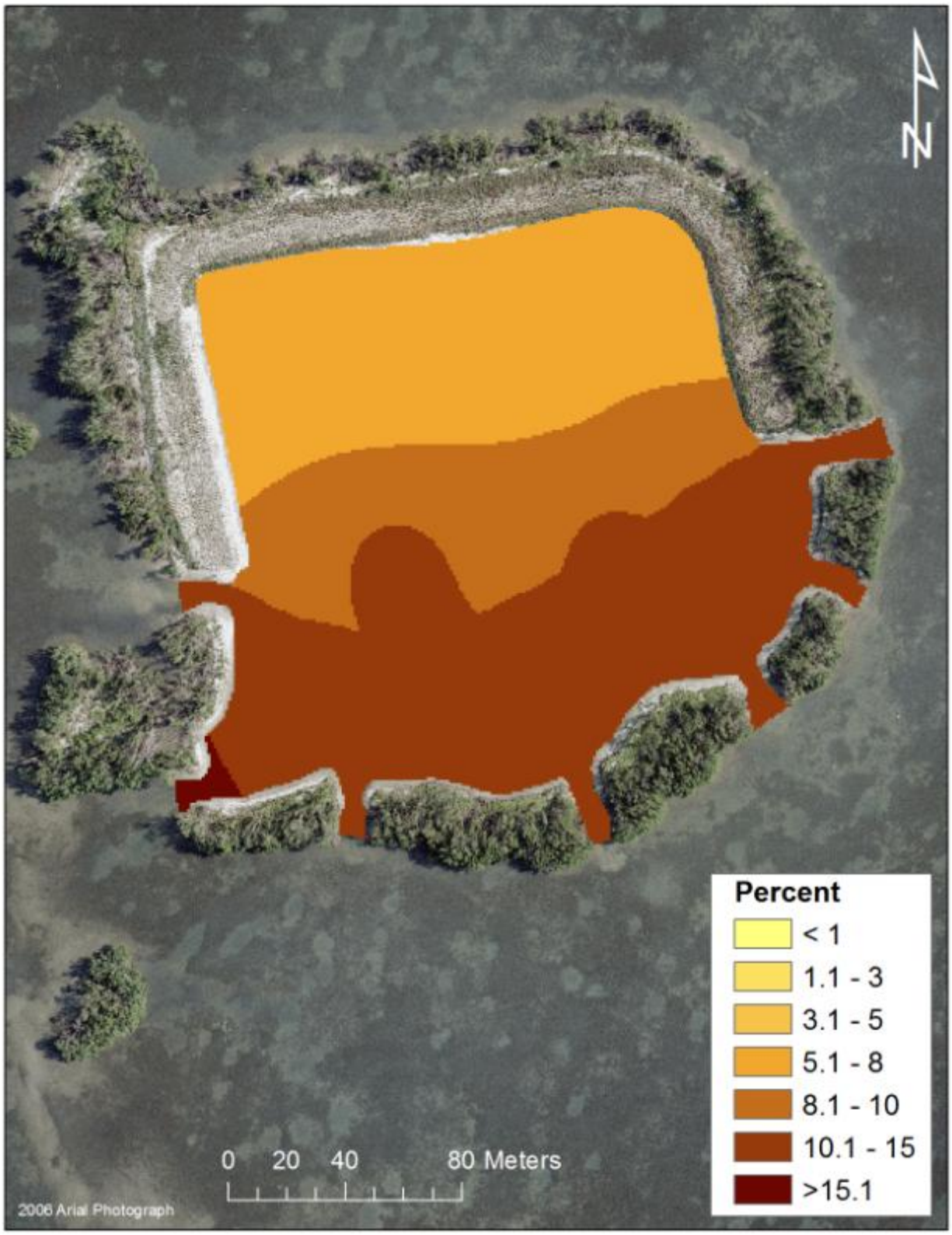

Figure 178. Percentage of soil sample with fine soil particles for winter 2009.

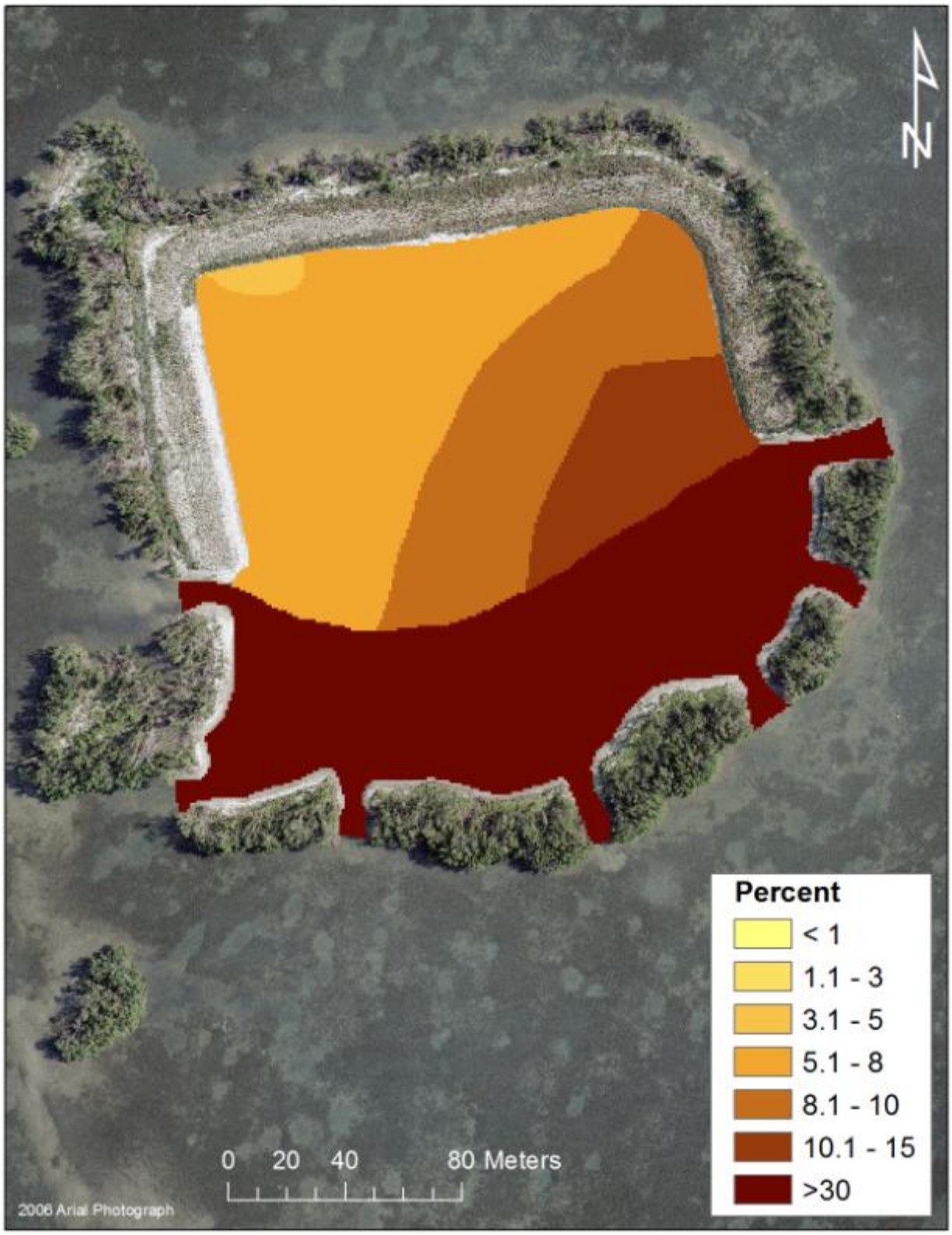

Figure 179. Percentage of soil sample with fine soil particles for summer 2010.

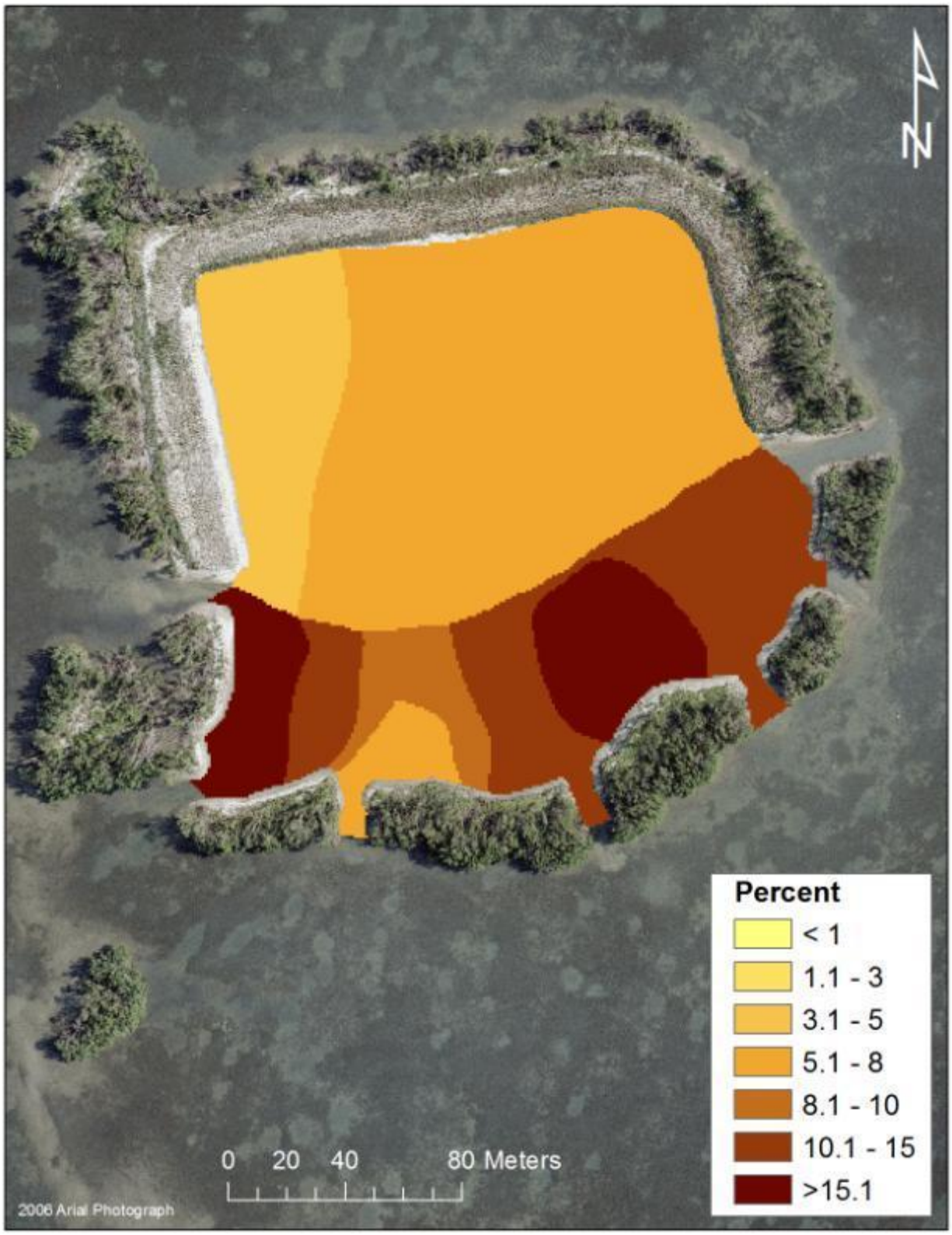

Figure 180. Percentage of soil sample with fine soil particles for winter 2010.

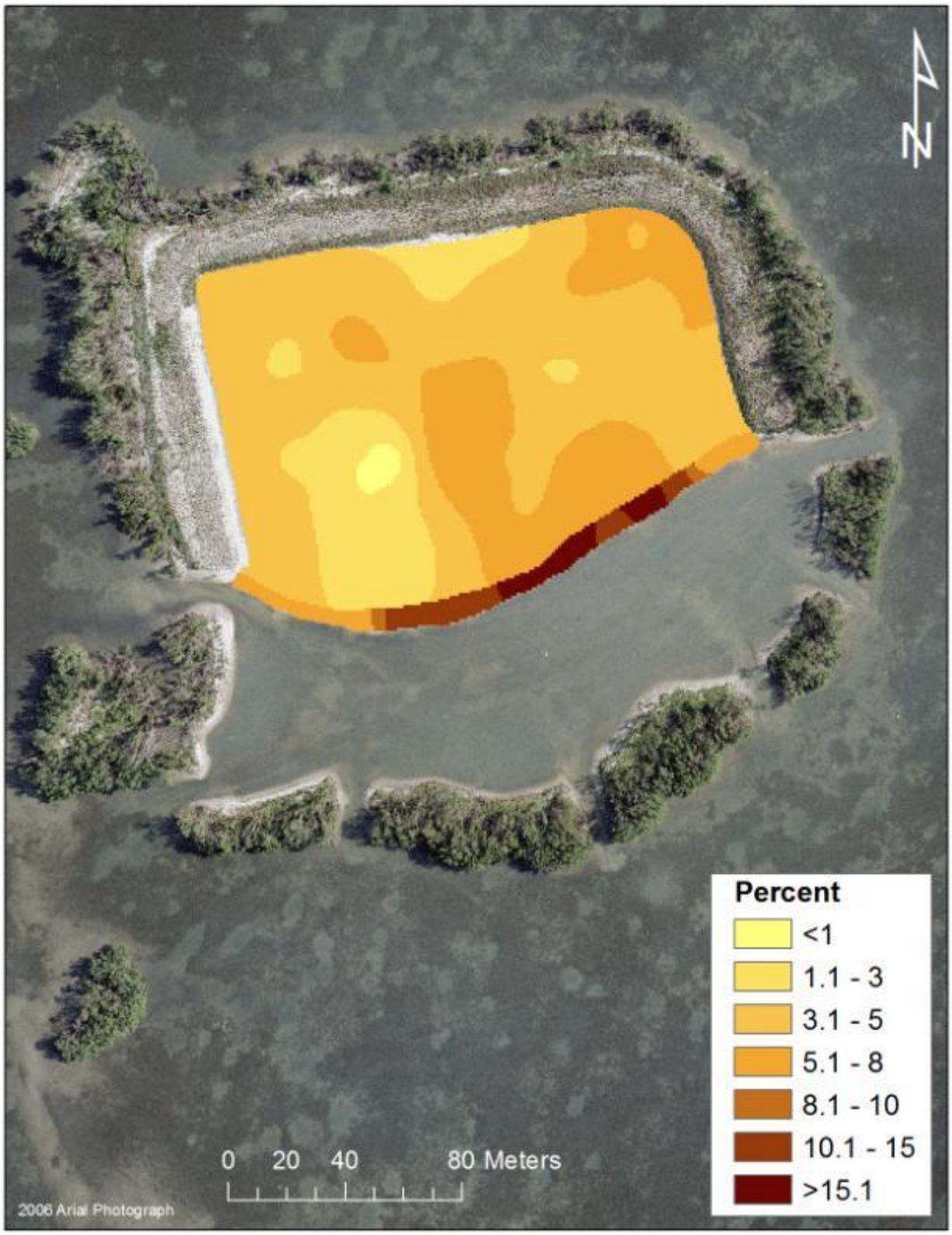

Figure 181. Percentage of soil sample with fine soil particles for summer 2011.

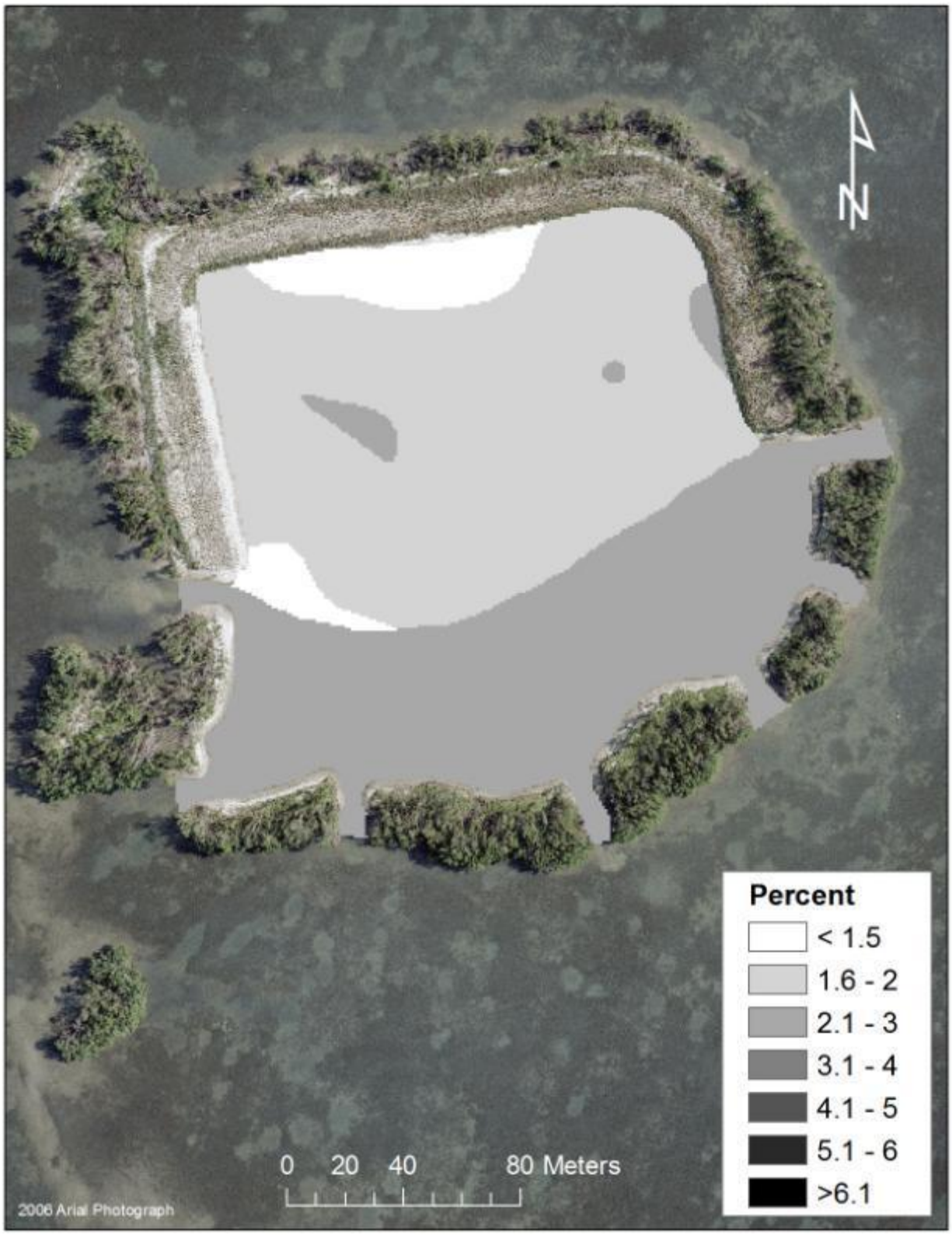

Figure 182. Percentage of organic matter within the soil for winter 2008.

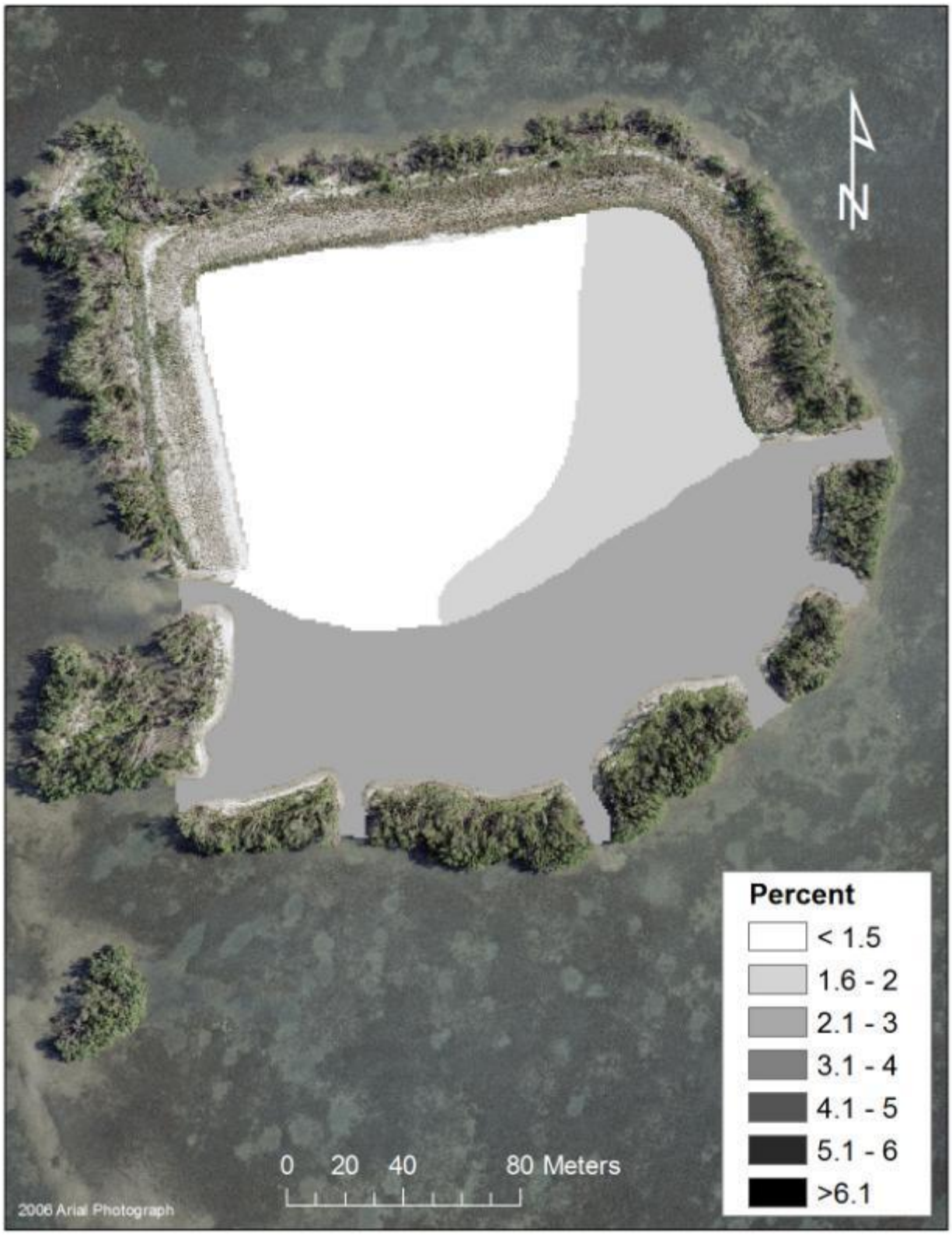

Figure 183. Percentage of organic matter within the soil for summer 2009.

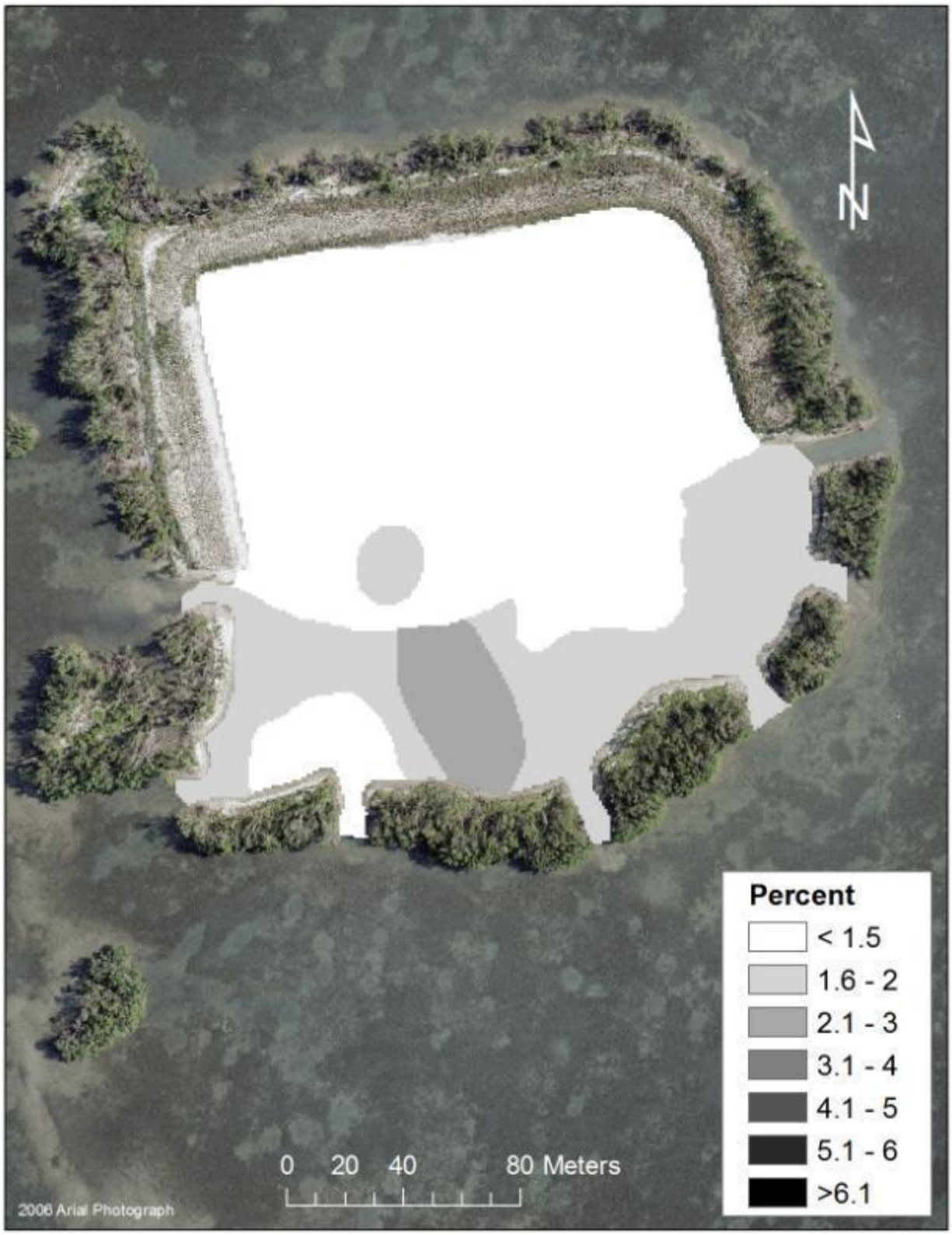

Figure 184. Percentage of organic matter within the soil for winter 2009.

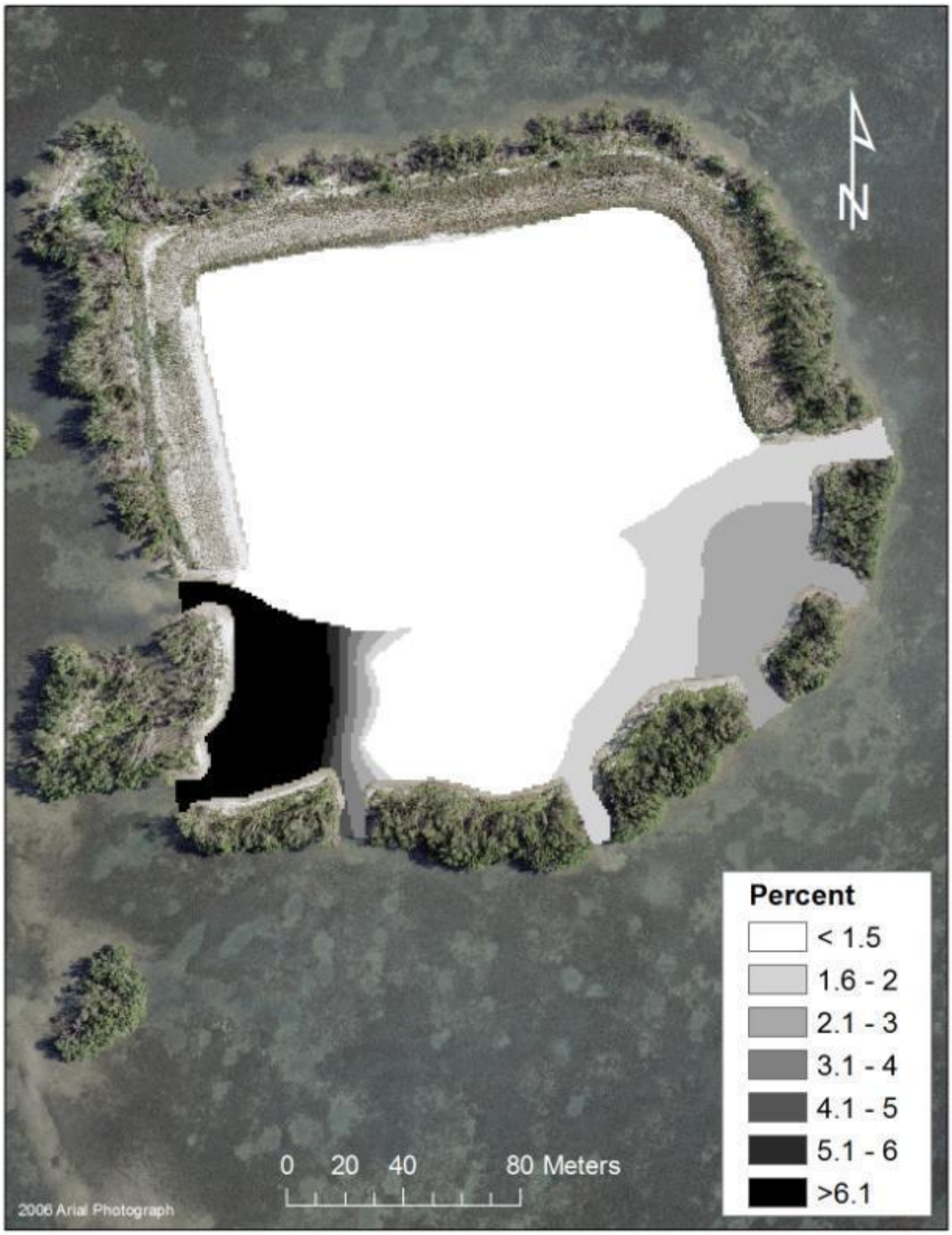

Figure 185. Percentage of organic matter within the soil for summer 2010.

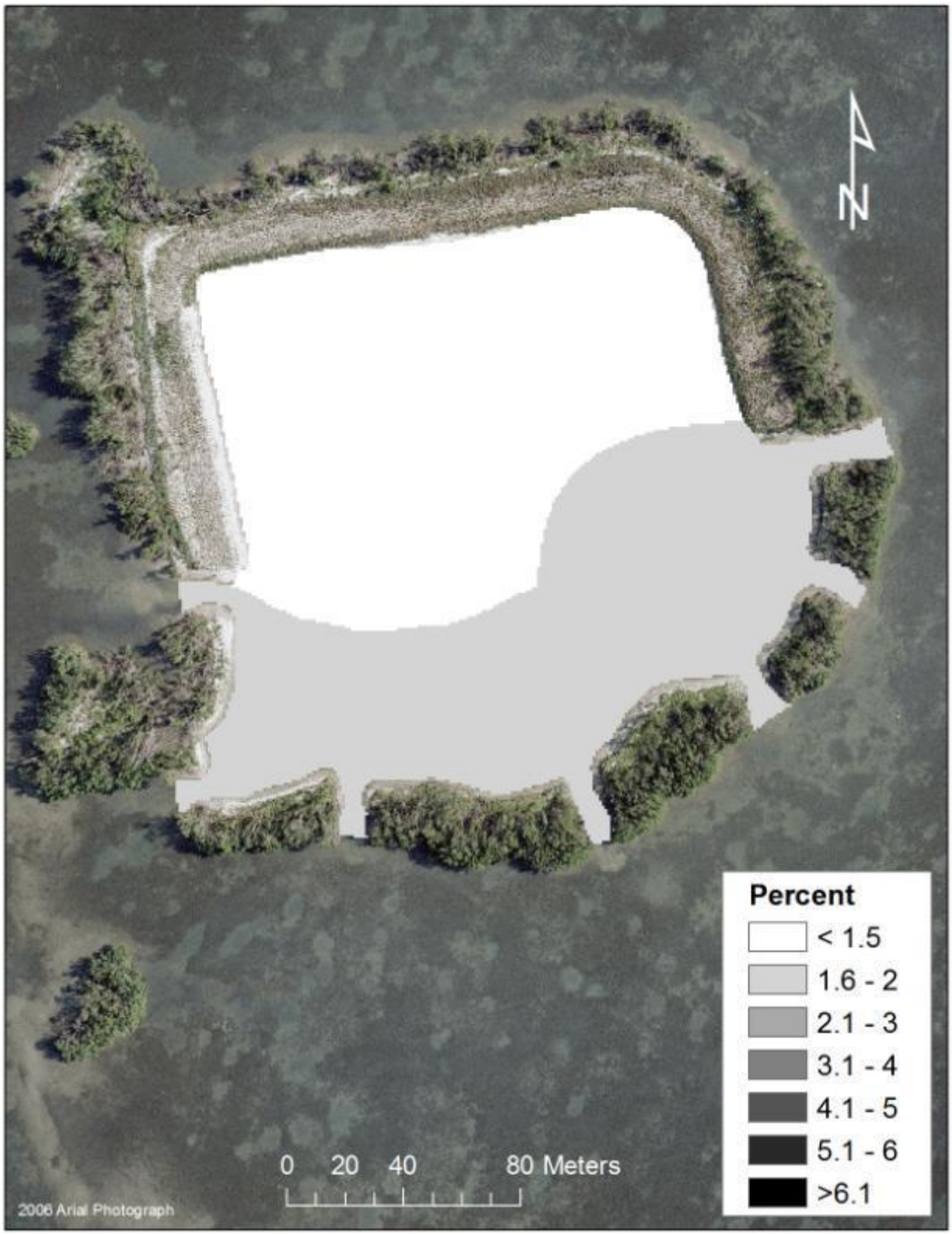

Figure 186. Percentage of organic matter within the soil for winter 2010.

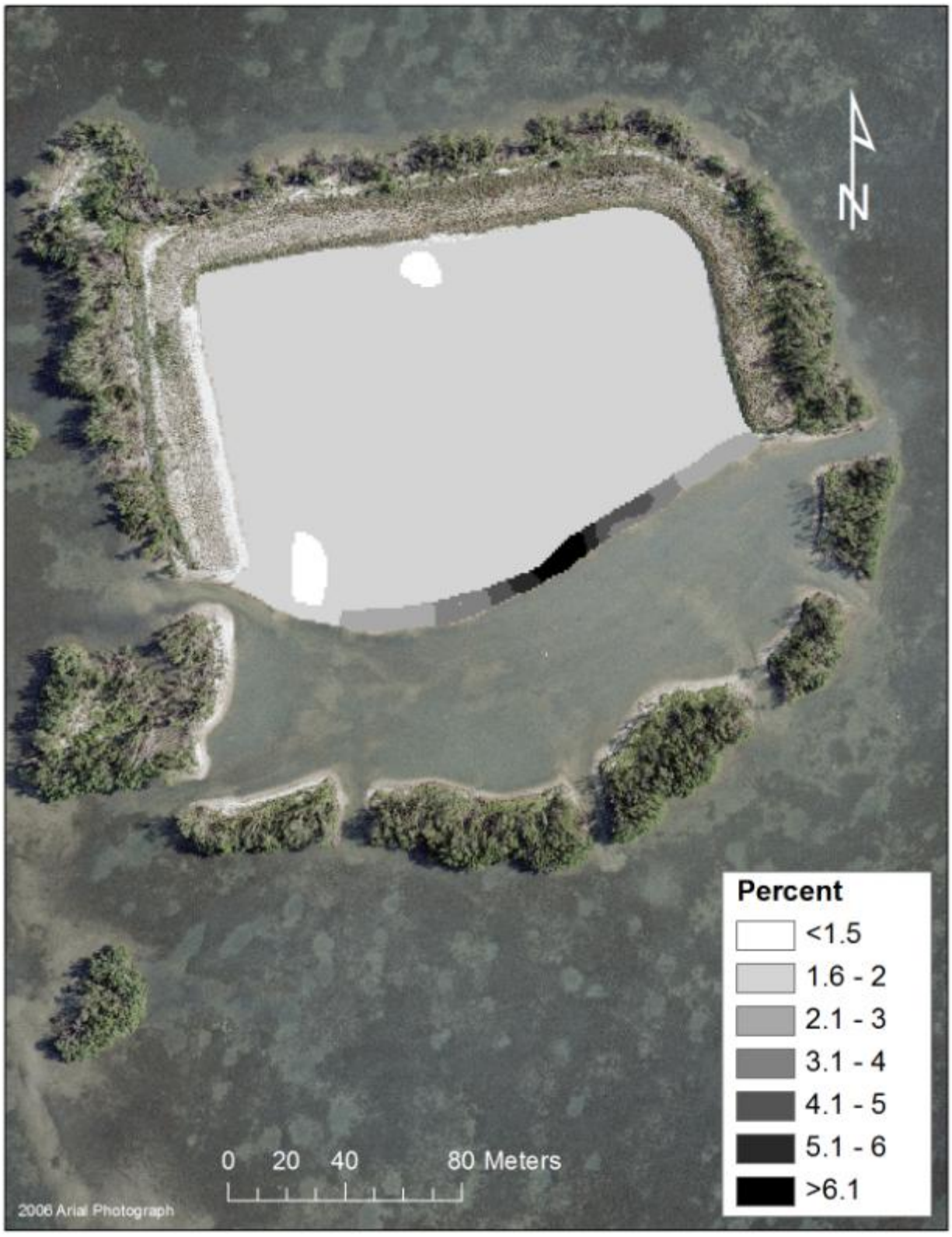

Figure 187. Percentage of organic matter within the soil for winter 2011.

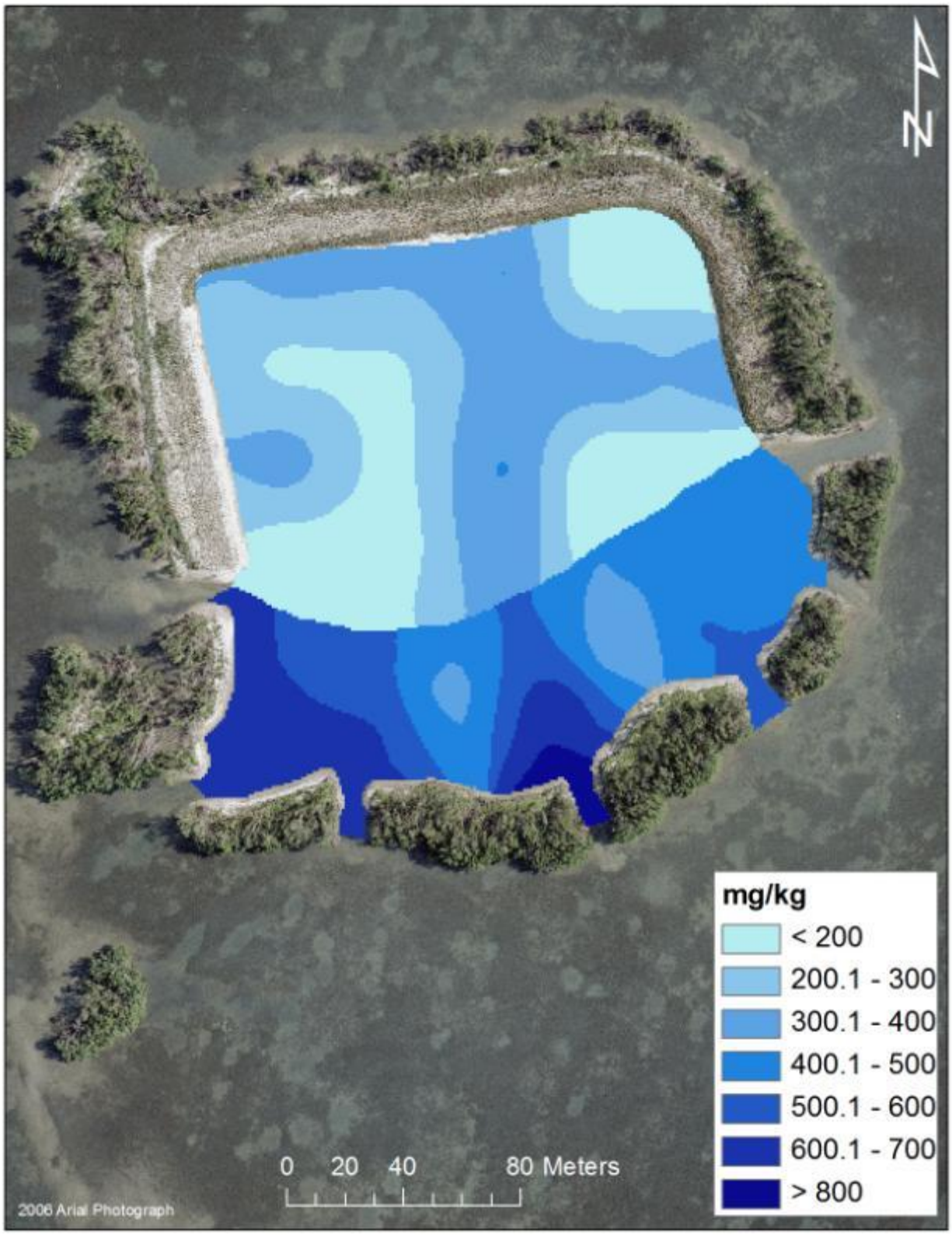

Figure 188. Amount of phosphorus within the soil in winter 2008.

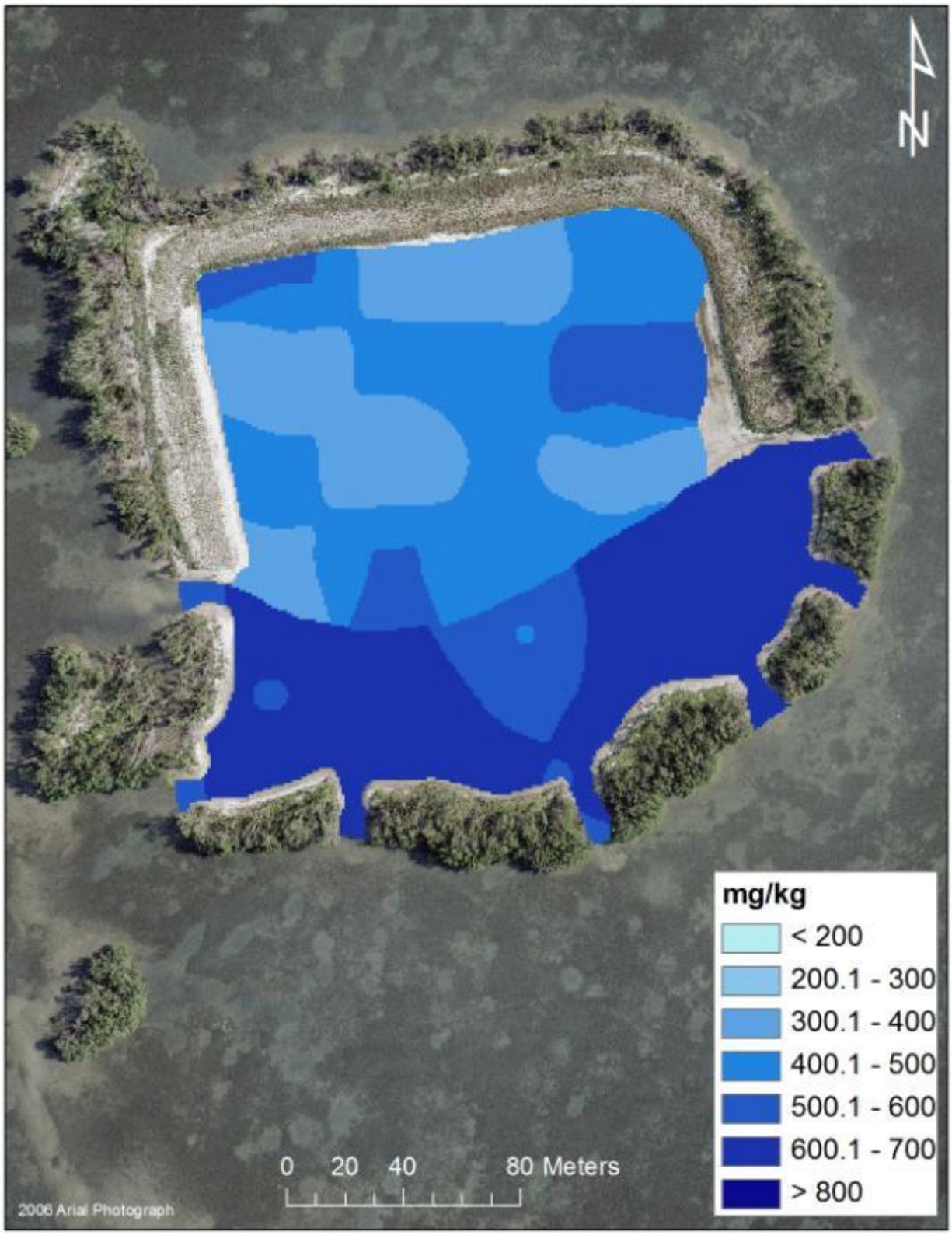

Figure 189. Amount of phosphorus within the soil in summer 2009.

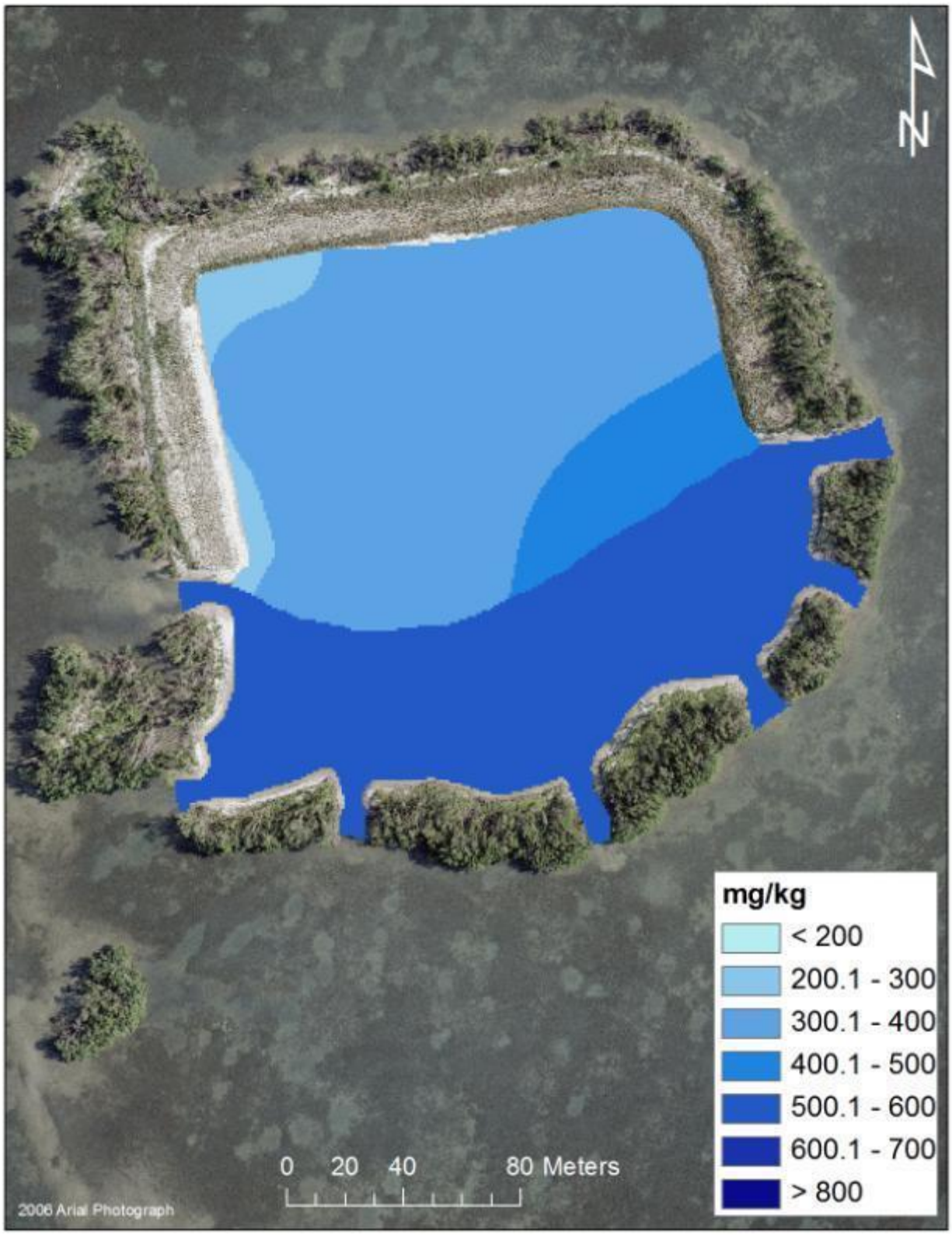

Figure 190. Amount of phosphorus within the soil in winter 2009.

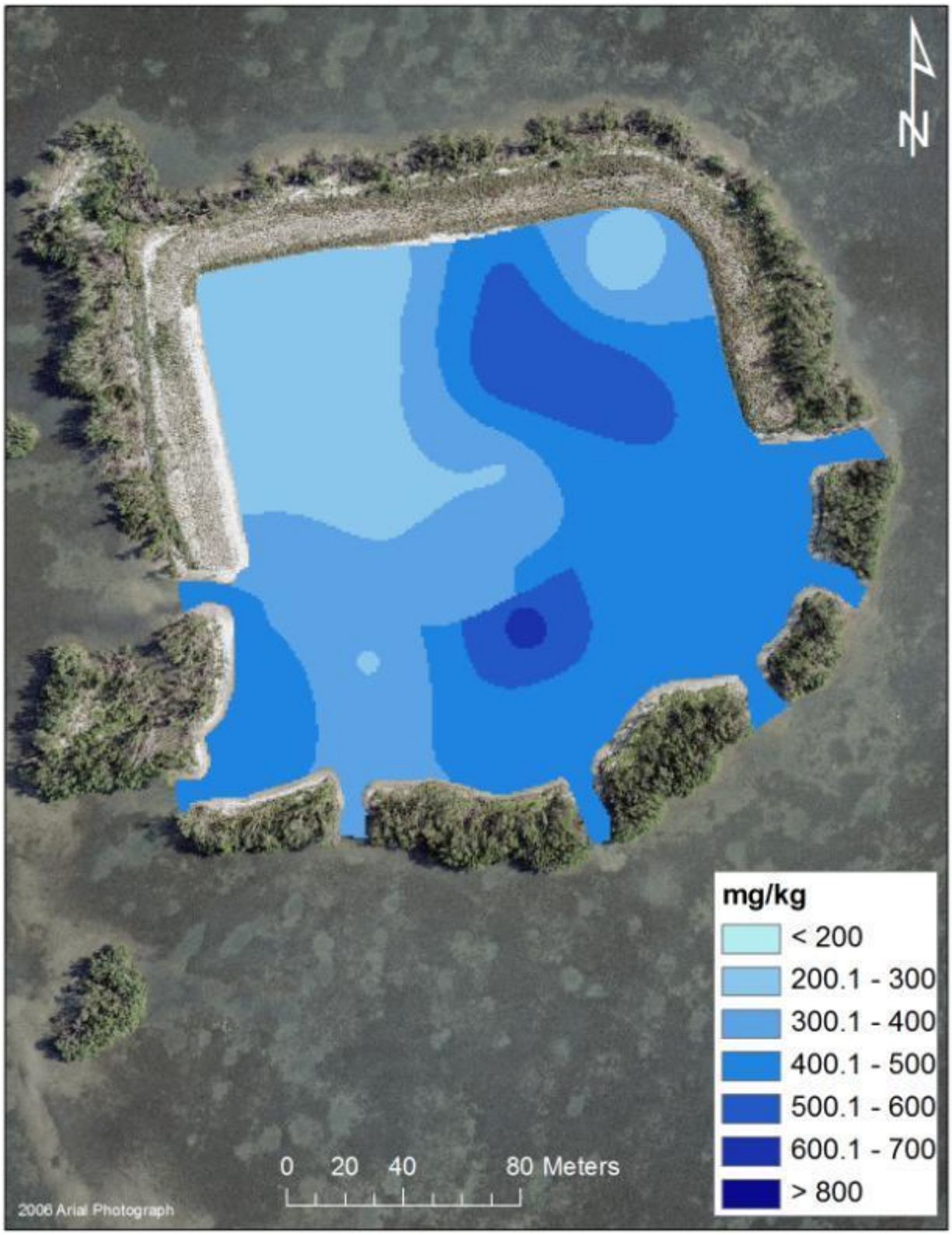

Figure 191. Amount of phosphorus within the soil in summer 2010.

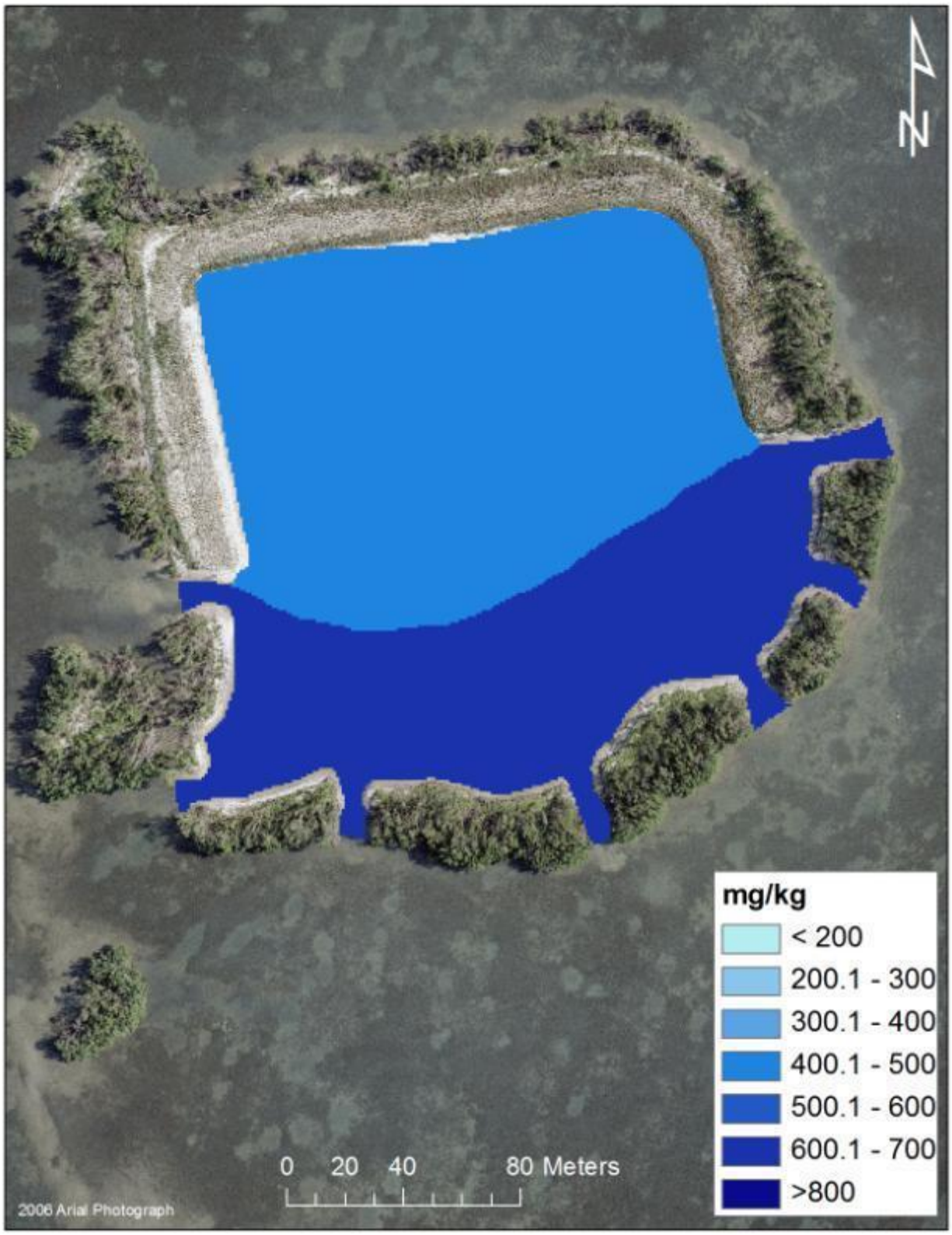

Figure 192. Amount of phosphorus within the soil in winter 2010.

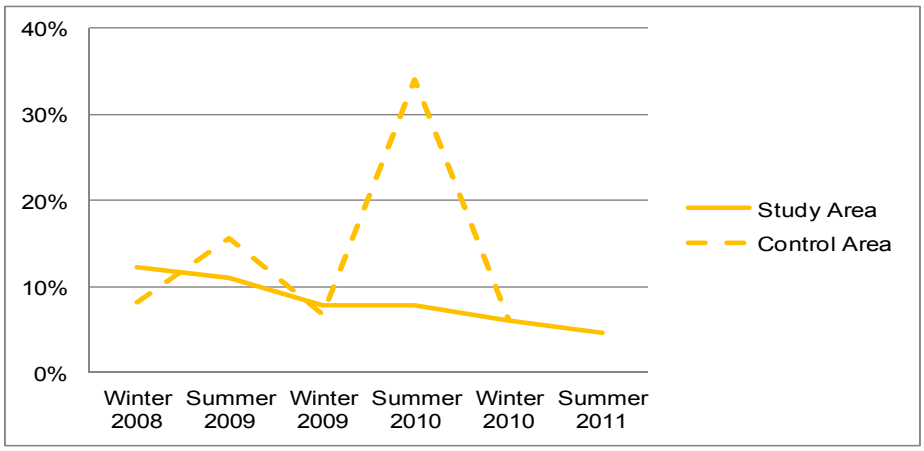

Figure 193. Average percent fine particles in the mangrove planter and upland control area form winter 2008 to winter 2010..

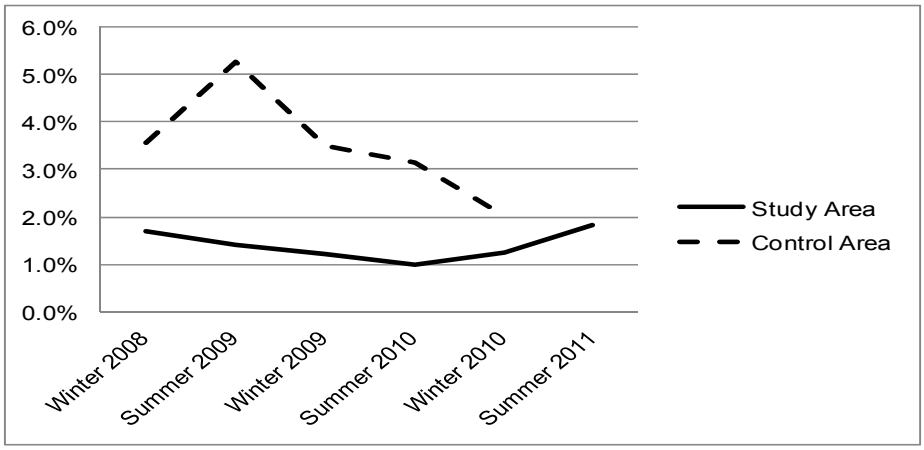

Figure 194. Average percent organic matter in the mangrove planter and upland control area form winter 2008 to winter 2010.

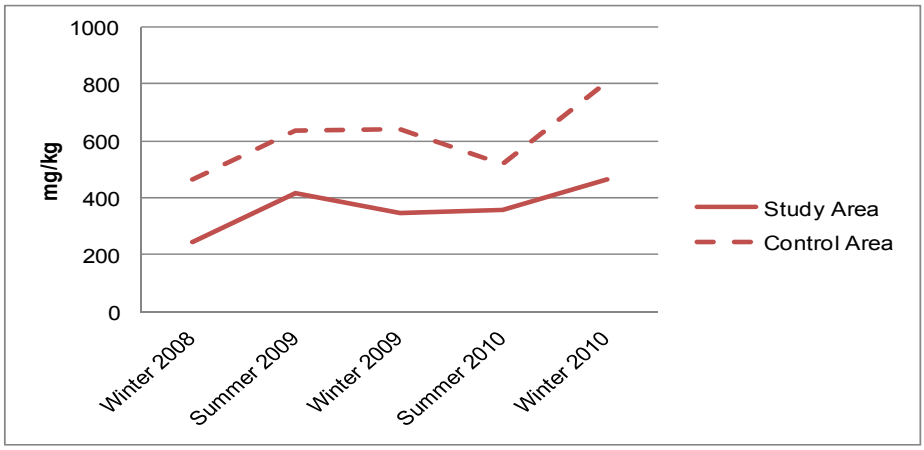

Figure 195. Average phosphorus in the mangrove planter and upland control area form winter 2008 to winter 2010.

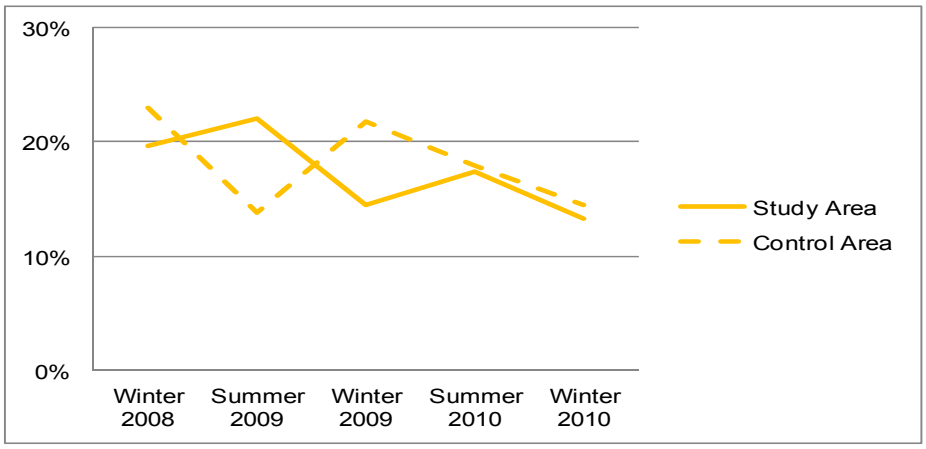

Figure 196. Average percent fine particles in the seagrass recruitment area and seagrass control area form winter 2008 to winter 2010.

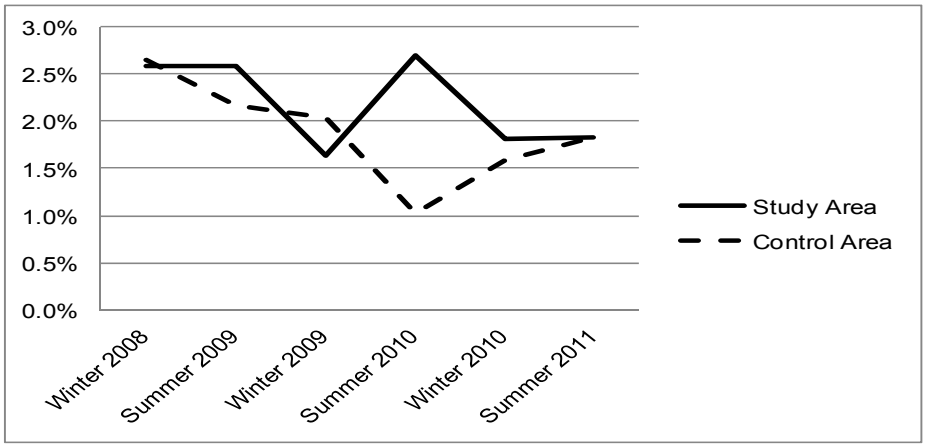

Figure 197. Average percent organic matter in the seagrass recruitment area and seagrass control area form winter 2008 to winter 2010.

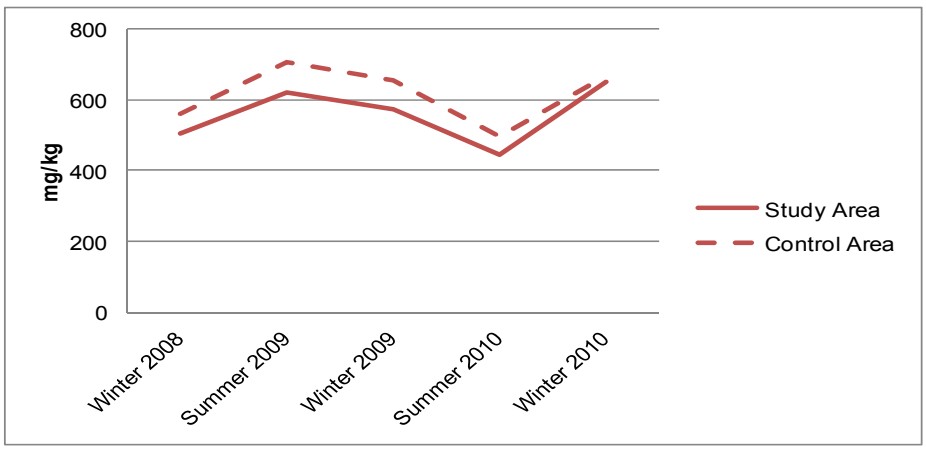

Figure 198. Average phosphorus in the seagrass recruitment area and seagrass control area form winter 2008 to winter 2010.

Table 25. Braun-Blanquet classification system for determining score and percent cover.

Braun Blanquet Density Scores	
Score	**Cover**
0	Taxa absent from quadrat
0.1	Taxa represented by a solitary shoot, <5% cover
0.5	Taxa represented by a few (<5) shoots, <5% cover
1	Taxa represented by many (>5) shoots, <5% cover
2	Taxa represented by many (>5) shoots, 5 - 25% cover
3	Taxa represented by many (>5) shoots, 25 - 50% cover
4	Taxa represented by many (>5) shoots, 50 - 75% cover
5	Taxa represented by many (>5) shoots, 75 - 100% cover

Table 26. Average mangrove density, height, maximum height, calculated canopy area, calculated canopy volume, and new recruit density of the study sites within the mangrove planting area.

Species	Season/Year	Average Density (Trees/m2)	Average Height (cm)	Average Maximum Height (cm)	Average Calculated Canopy Area m2/m2	Average Calculated Canopy Volume m3/m2	Average New Mangrove Recruit/m2
Rhizophora mangle	Summer 2009	1.5 (SD=1.7)	57.0 (SD=17.9)	71.0 (SD=26.2)	N/A	N/A	N/A
Rhizophora mangle	Winter 2009	1.5 (SD=2.5)	65.6 (SD=29.8)	84.6 (SD=39.2)	N/A	N/A	N/A
Rhizophora mangle	Summer 2010	1.7 (SD=2.8)	69.4 (SD=24.9)	89.6 (SD=36.5)	N/A	N/A	N/A
Rhizophora mangle	Winter 2010	1.4 (SD=1.3)	76.9 (SD=30.0)	97.3 (SD=38.0)	N/A	N/A	N/A
Rhizophora mangle	Summer 2011	2.1 (SD=4.2)	73.9 (SD=18.7)	112.6 (SD=32.2)	0.3 (SD=0.2)	0.1 (SD=0.1)	0.5 (SD=1.2)
Avicennia germinans	Summer 2009	0.4 (SD=0.6)	37.3 (SD=47.8)	42.1 (SD=53.2)	N/A	N/A	N/A
Avicennia germinans	Winter 2009	0.5 (SD=0.7)	39.3 (SD=47.2)	48.6 (SD=56.3)	N/A	N/A	N/A
Avicennia germinans	Summer 2010	1.0 (SD=1.6)	32.8 (SD=39.2)	55.2 (SD=59.4)	N/A	N/A	N/A
Avicennia germinans	Winter 2010	1.0 (SD=1.8)	48.1 (SD=41.7)	71.9 (SD=65.9)	N/A	N/A	N/A
Avicennia germinans	Summer 2011	1.6 (SD=2.1)	52.7 (SD=36.2)	112.1 (SD=60.6)	0.1 (SD=0.2)	0.1 (SD=0.2)	1.3 (SD=2.0)
Laguncularia racemosa	Summer 2009	0.5 (SD=1.3)	30.9 (SD=38.1)	37.2 (SD=47.2)	N/A	N/A	N/A
Laguncularia racemosa	Winter 2009	0.4 (SD=1.1)	39.3 (SD=51.3)	44.3 (SD=57.4)	N/A	N/A	N/A
Laguncularia racemosa	Summer 2010	0.8 (SD=1.8)	33.3 (SD=42.6)	50.0 (SD=59.2)	N/A	N/A	N/A
Laguncularia racemosa	Winter 2010	0.5 (SD=0.8)	44.5 (SD=55.1)	57.8 (SD=74.2)	N/A	N/A	N/A
Laguncularia racemosa	Summer 2011	1.2 (SD=3.3)	51.2 (SD=45.7)	84.9 (SD=61.2)	0.1 (SD=0.4)	0.1 (SD=0.4)	0.7 (SD=2.1)
All	Summer 2009	2.4 (SD=3.2)	60.8 (SD=19.5)	88.1 (SD=35.1)	N/A	N/A	N/A
All	Winter 2009	2.4 (SD=3.9)	66.9 (SD=27.7)	100.9 (SD=44.0)	N/A	N/A	N/A
All	Summer 2010	3.7 (SD=5.1)	56.6 (SD=26.7)	107.4 (SD=41.1)	N/A	N/A	N/A
All	Winter 2010	2.8 (SD=3.0)	70.7 (SD=27.0)	119.3 (SD=53.4)	N/A	N/A	N/A
All	Summer 2011	4.9 (SD=7.4)	60.1 (SD=24.8)	140.1 (SD=41.5)	0.5 (SD=0.5)	0.4 (SD=0.5)	2.6 (SD=4.3)

Table 27. Average algae coverage, seagrass coverage, and seagrass density for sites within the seagrass embayment and control.

Season/Year	Average Algae Coverage		Average Seagrass Coverage		Average Seagrass Desnity Shoots/m2	
	Study Area	Control Sites	Study Area	Control Sites	Study Area	Control Sites
Winter 2008	71.8 (SD=24.4)	17.3 (SD=14.2)	3.5 (SD=6.5)	54.0 (SD=5.3)	34.4 (SD=41.0)	71.7 (SD=25.7)
Summer 2009	25.7 (SD=11.7)	0.0 (SD=0.0)	1.0 (SD=1.2)	73.7 (SD=45.6)	0.0 (SD=0.1)	710.0 (SD=255.9)
Winter 2009	36.8 (SD=29.5)	13.3 (SD=23.1)	0.3 (SD=0.5)	36.7 (SD=23.1)	0.0 (SD=0.1)	24.0 (SD=24.8)
Summer 2010	14.5 (SD=13.0)	0.0 (SD=0.0)	8.3 (SD=10.7)	43.3 (SD=49.3)	95.0 (SD=144.7)	483.3 (SD=144.3)
Winter 2010	70.5 (SD=26.5)	0.0 (SD=0.0)	0.7 (SD=1.1)	71.7 (SD=36.2)	10.0 (SD=31.6)	700.0 (SD=204.6)
Summer 2011	68.7 (SD=27.3)	N/A	0.5 (SD=1.6)	N/A	5.0 (SD=15.8)	N/A

Table 28. Average Braun-Blanquet cover class and percent coverage for sites within the seagrass embayment and control area.

Season/Year	Average Braun-Blanquet cover class		Braun-Blanquet Average Percent Coverage	
	Study Area	Control Sites	Study Area	Control Sites
Winter 2008	0.6 (SD=0.7)	4.0 (SD=0.0)	2.5 (SD=5.6)	62.5 (SD=0.0)
Summer 2009	0.2 (SD=0.2)	4.0 (SD=1.7)	2.0 (SD=1.1)	63.3 (SD=41.9)
Winter 2009	0.5 (SD=0.2)	2.7 (SD=0.6)	1.2 (SD=0.8)	30.0 (SD=13.0)
Summer 2010	1.2 (SD=1.1)	3.0 (SD=1.7)	10.3 (SD=11.9)	39.2 (SD=41.9)
Winter 2010	0.2 (SD=0.2)	4.3 (SD=1.2)	1.0 (SD=1.3)	70.1 (SD=28.9)
Summer 2011	0.1 (SD=0.2)	N/A	0.3 (SD=0.8)	N/A

Table 29. Seagrass composition density of sites within the seagrass embayment and the control areas.

Season/Year	Average *Halophila johnsonii* Denisty Shoots/m2		Average *Halodule wrightii* Denisty Shoots/m2		Average *Syringodium filiforme* Denisty Shoots/m2		Average *Thalassia testudinum* Denisty Shoots/m2	
	Study Area	Control Sites	Study Area	Control Sites	Study Area	Control Sites	Study Area	Control Sites
Winter 2008	30.6 (SD=40.8)	0.0 (SD=0.0)	3.75 (SD=7.9)	0.0 (SD=0.0)	0.0 (SD=0.0)	58.0 (SD=20.4)	0.0 (SD=0.0)	13.7 (SD=23.7)
Summer 2009	0.0 (SD=0.1)	0.0 (SD=0.0)	0.0 (SD=0.1)	0.0 (SD=0.0)	0.0 (SD=0.0)	698.0 (SD=203.9)	0.0 (SD=0.0)	12.0 (SD=20.8)
Winter 2009	0.0 (SD=0.1)	0.0 (SD=0.0)	0.0 (SD=0.1)	0.0 (SD=0.0)	0.0 (SD=0.0)	15.0 (SD=10.0)	0.0 (SD=0.0)	9.0 (SD=15.6)
Summer 2010	87.5 (SD=143.5)	0.0 (SD=0.0)	7.5 (SD=16.9)	0.0 (SD=0.0)	0.0 (SD=0.0)	450.0 (SD=86.6)	0.0 (SD=0.0)	33.3 (SD=57.7)
Winter 2010	0.0 (SD=0.0)	0.0 (SD=0.0)	10.0 (SD=31.6)	0.0 (SD=0.0)	0.0 (SD=0.0)	683.3 (SD=200.5)	0.0 (SD=0.0)	16.7 (SD=28.9)
Summer 2011	0.0 (SD=0.0)	N/A	5.0 (SD=15.8)	N/A	0.0 (SD=0.0)	N/A	0.0 (SD=0.0)	N/A

Table 30. Average percent sand, percent fine, percent organic matter, bulk density, and total phosphorus for soil samples within the mangrove planter and the upland control sites.

Season/Year	Average Percent Sand Soil Particles		Average Percent Fine Soil Particles		Average Percent Orangic Soil Matter		Average Bulk Density mg/cm3		Average Total Phophorus mg/kg	
	Mangrove Planter	Upland Control	Mangrove Planter	Upland Control	Mangrove Planter	Upland Control	Mangrove Planter	Upland Control	Mangrove Planter	Upland Control
Winter 2008	87.8 (SD=10.8)	91.8 (SD=2.6)	12.2 (SD=10.8)	8.2 (SD=2.6)	1.7 (SD=0.5)	3.6 (SD=1.2)	1.1 (SD=0.2)	0.8 (SD=0.0)	247.4 (SD=131.9)	463.7 (SD=378.4)
Summer 2009	89.1 (SD=4.1)	84.5 (SD=10.5)	10.9 (SD=4.1)	15.5 (SD=10.5)	1.4 (SD=0.5)	5.3 (SD=2.9)	1.3 (SD=0.2)	1.1 (SD=0.1)	418.0 (SD=74.3)	636.0 (SD=98.4)
Winter 2009	92.2 (SD=3.5)	93.3 (SD=4.6)	7.8 (SD=3.5)	6.7 (SD=4.6)	1.2 (SD=0.3)	3.5 (SD=3.4)	1.6 (SD=0.1)	1.0 (SD=0.5)	348.7 (SD=137.6)	644.3 (SD=61.1)
Summer 2010	92.1 (SD=3.2)	66.0 (SD=25.2)	7.9 (SD=3.2)	34.0 (SD=25.2)	1.0 (SD=0.2)	3.1 (SD=1.5)	1.6 (SD=0.2)	0.4 (SD=0.2)	356.1 (SD=123.9)	519.3 (SD=261.8)
Winter 2010	94.0 (SD=3.0)	93.8 (SD=1.7)	6.0 (SD=3.0)	6.2 (SD=1.7)	1.2 (SD=0.3)	2.0 (SD=1.2)	1.4 (SD=0.2)	1.7 (SD=0.0)	463.4 (SD=117.4)	807.1 (SD=250.5)
Summer 2011	95.3 (SD=3.5)	N/A	4.7 (SD=3.5)	N/A	1.8 (SD=0.8)	N/A	1.5 (SD=0.1)	N/A	N/A	N/A

Table 31. Average percent sand, percent fine, percent organic matter, bulk density, and total phosphorus for oil samples within the seagrass embayment and the control sites.

Season/Year	Average Percent Sand Soil Particles		Average Percent Fine Soil Particles		Average Percent Orangic Soil Matter		Average Bulk Density mg/cm3		Average Total Phophorus mg/kg	
	Seagrass Recruitment Area	Seagrass control Sites	Seagrass Recruitment Area	Seagrass control Sites	Seagrass Recruitment Area	Seagrass control Sites	Seagrass Recruitment Area	Seagrass control Sites	Seagrass Recruitment Area	Seagrass control Sites
Winter 2008	80.4 (SD=7.8)	77.1 (SD=9.8)	19.6 (SD=7.8)	22.9 (SD=9.8)	2.6 (SD=0.7)	2.7 (SD=1.4)	0.8 (SD=0.2)	0.6 (SD=0.2)	503.3 (SD=128.7)	560.6 (SD=172.6)
Summer 2009	77.9 (SD=6.1)	86.2 (SD=6.1)	22.1 (SD=6.1)	13.8 (SD=6.1)	2.6 (SD=0.8)	2.2 (SD=1.0)	1.1 (SD=0.5)	1.0 (SD=0.1)	619.8 (SD=52.7)	706.7 (SD=25.9)
Winter 2009	85.4 (SD=5.2)	78.2 (SD=9.8)	14.6 (SD=5.2)	21.8 (SD=9.8)	1.6 (SD=0.5)	2.0 (SD=0.8)	1.3 (SD=0.3)	1.0 (SD=0.3)	572.2 (SD=52.8)	655.8 (SD=9.5)
Summer 2010	82.7 (SD=6.3)	82.1 (SD=1.5)	17.3 (SD=6.3)	17.9 (SD=1.5)	2.7 (SD=4.1)	1.0 (SD=0.3)	1.2 (SD=0.3)	1.2 (SD=0.2)	443.6 (SD=93.2)	496.2 (SD=175.9)
Winter 2010	86.7 (SD=7.3)	85.5 (SD=4.3)	13.3 (SD=7.3)	14.5 (SD=4.3)	1.8 (SD=0.7)	1.6 (SD=0.2)	1.2 (SD=0.3)	1.2 (SD=0.1)	649.9 (SD=128.7)	666.8 (SD=8.5)
Summer 2011	N/A	N/A	N/A	N/A	1.8 (SD=0.7)	1.8 (SD=0.7)	1.4 (SD=0.1)	N/A	N/A	N/A

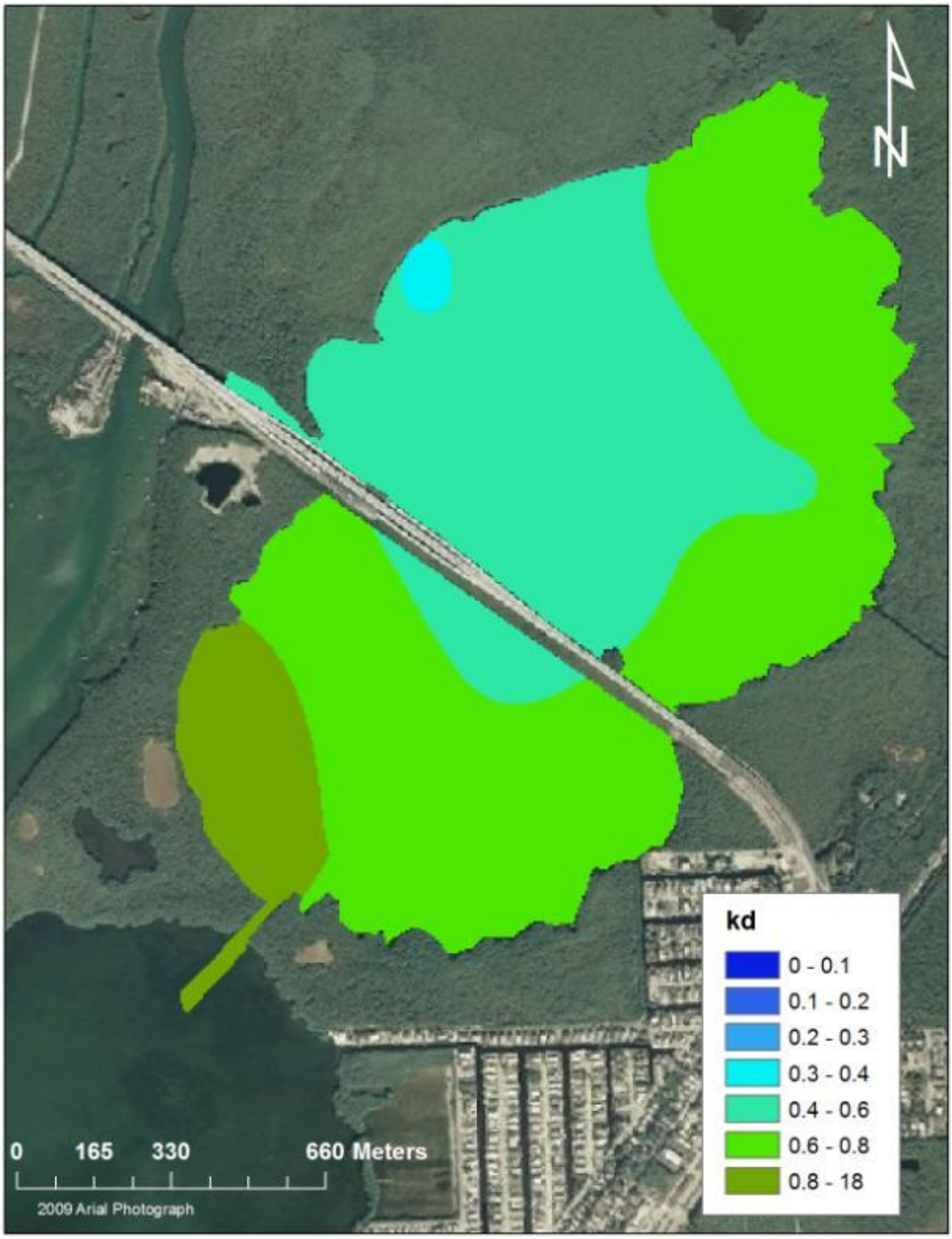

Figure 199. Water column downward light extinction coefficient (Kd) (winter 2008).

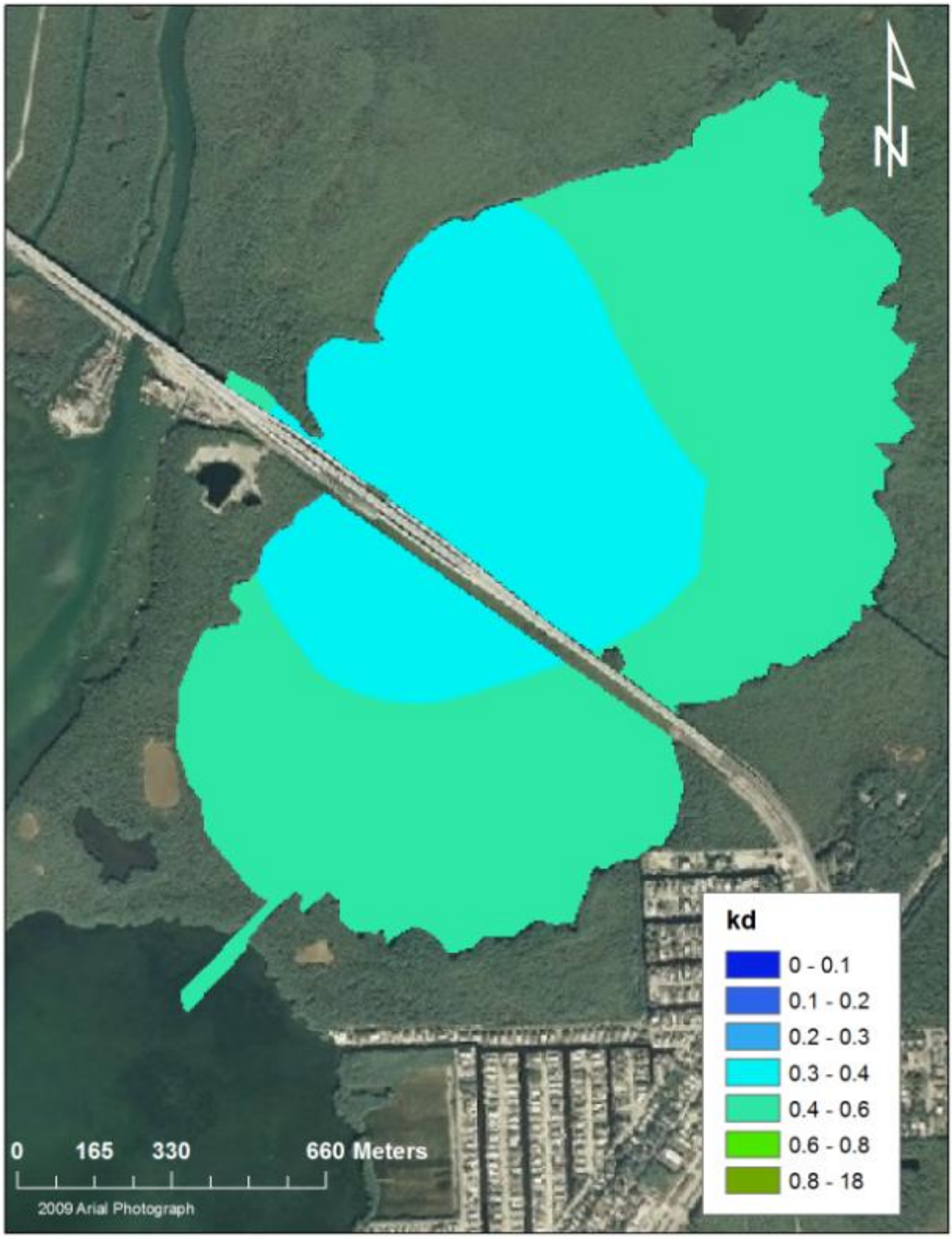

Figure 200. Water column downward light extinction coefficient (Kd) (summer 2009).

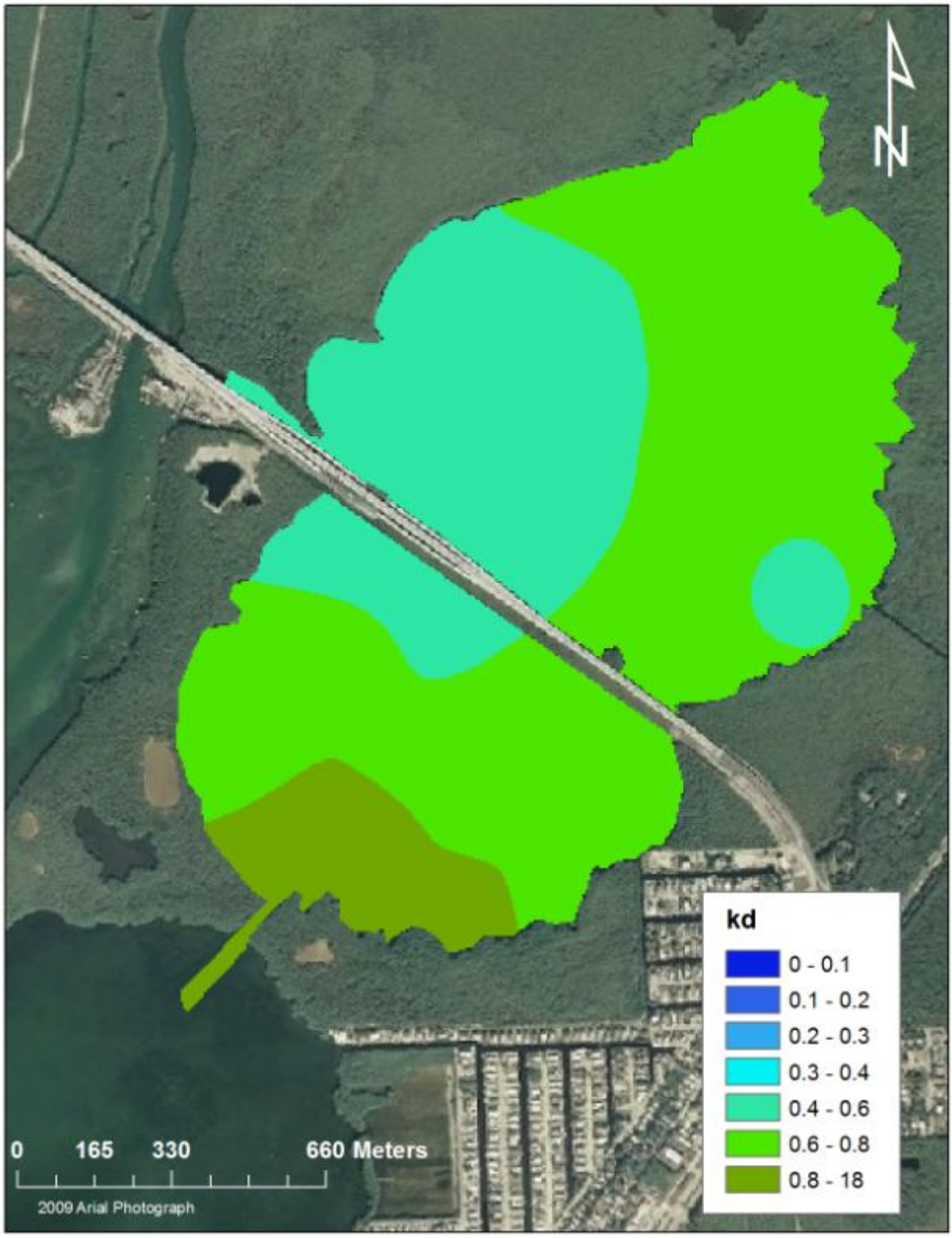
N
kd
0 - 0.1
0.1 - 0.2
0.2 - 0.3
0.3 - 0.4
0.4 - 0.6
0.6 - 0.8
0.8 - 18
0
165
330
660 Meters
2009 Arial Photograph

Figure 201. Water column downward light extinction coefficient (Kd) (winter 2009).

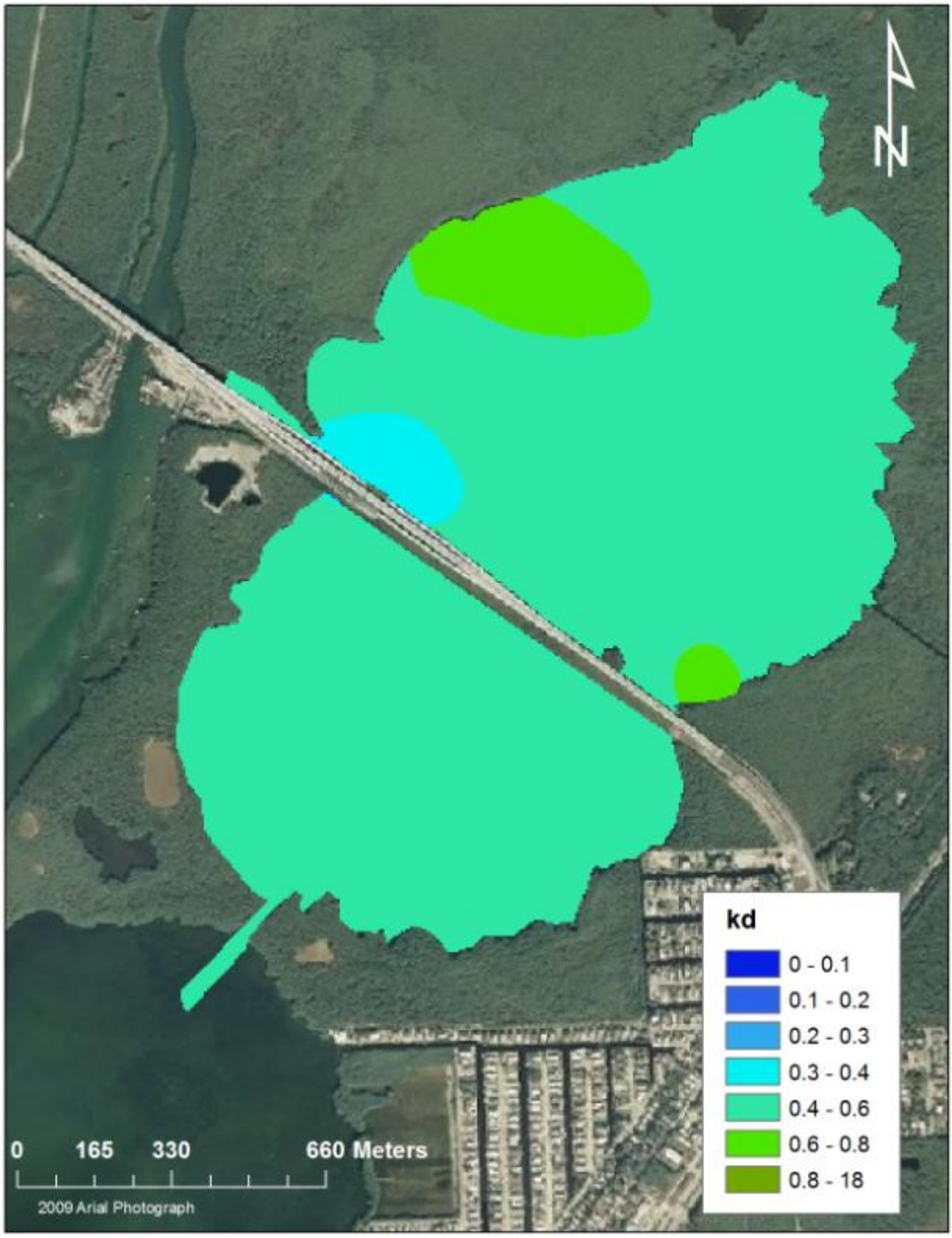
N
kd
0 - 0.1
0.1 - 0.2
0.2 - 0.3
0.3 - 0.4
0.4 - 0.6
0.6 - 0.8
0.8 - 18
0
165
330
660 Meters
2009 Arial Photograph

Figure 202. Water column downward light extinction coefficient (Kd) (summer 2010).

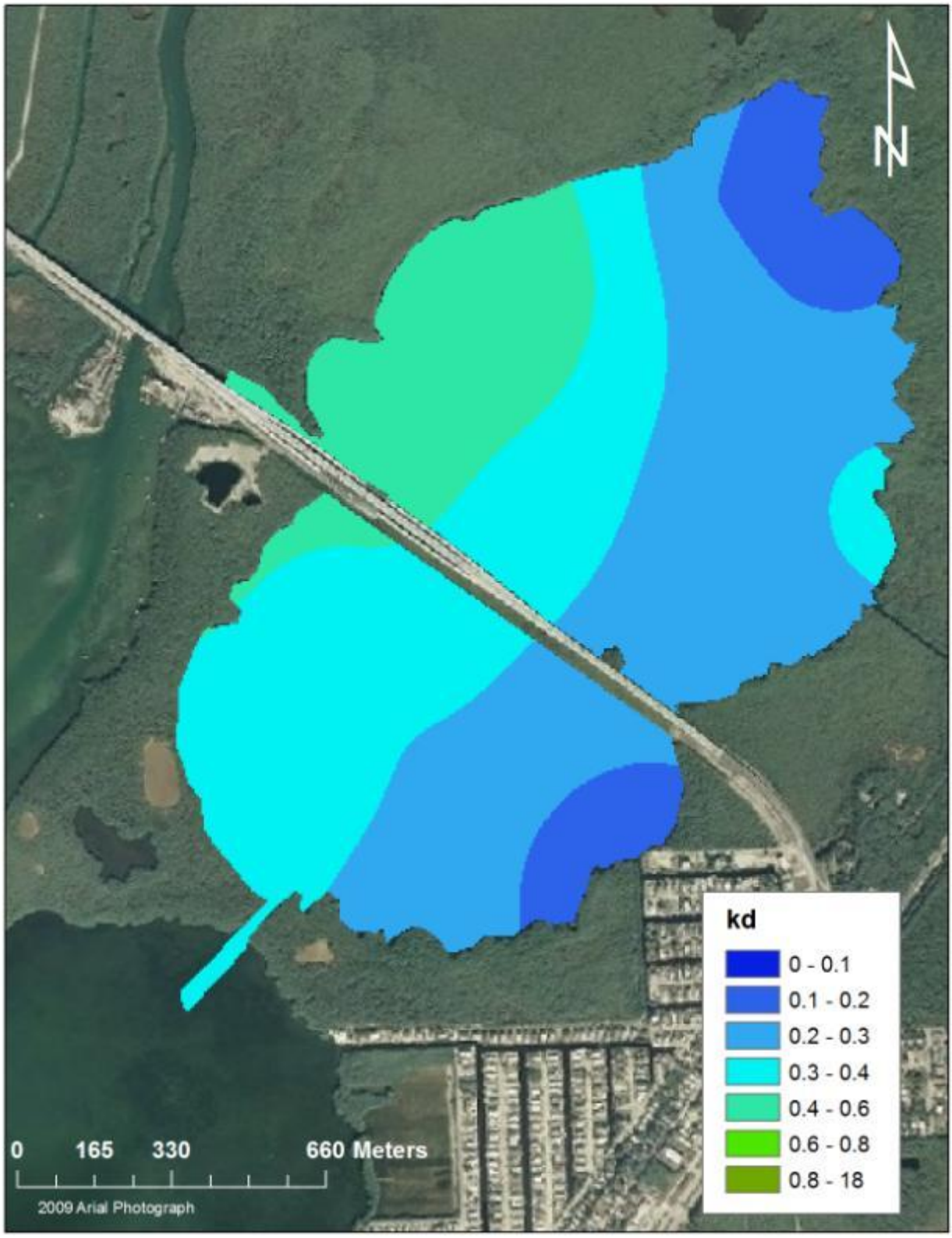
N
kd
0 - 0.1
0.1 - 0.2
0.2 - 0.3
0.3 - 0.4
0.4 - 0.6
0.6 - 0.8
0.8 - 18
0
165
330
660 Meters
2009 Arial Photograph

Figure 203. Water column downward light extinction coefficient (Kd) (winter 2010).

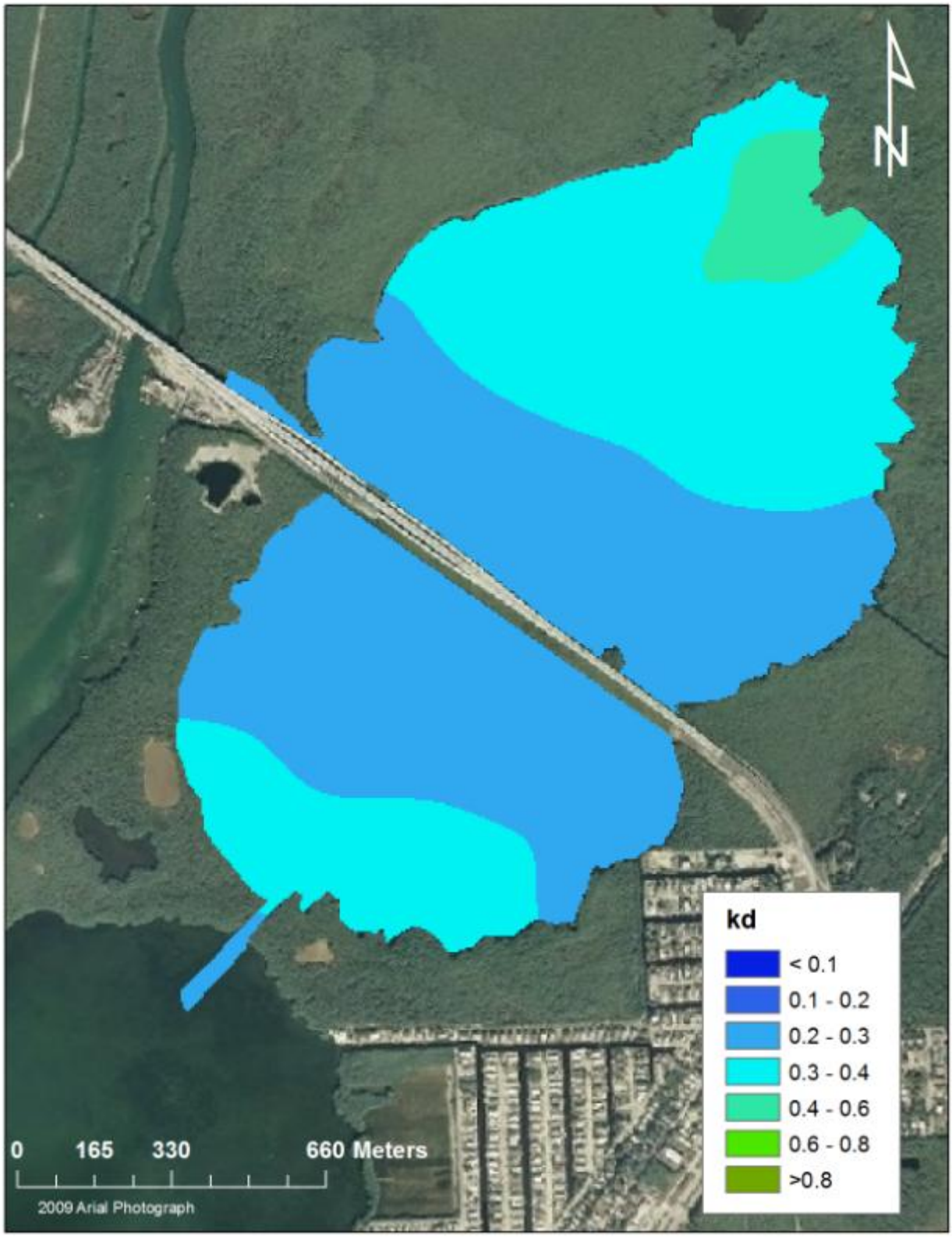
N
kd
< 0.1
0.1 - 0.2
0.2 - 0.3
0.3 - 0.4
0.4 - 0.6
0.6 - 0.8
>0.8
0
165
330
660 Meters
2009 Arial Photograph

Figure 204. Water column downward light extinction coefficient (Kd) (summer 2011).


N
mg/L
< 4
4.1 - 5
5.1 - 6
6.1 - 8
8.1 - 10
10.1 - 12
>12.1
0
165
330
660 Meters
2009 Arial Photograph

Figure 205. Water column dissolved oxygen (summer 2009).

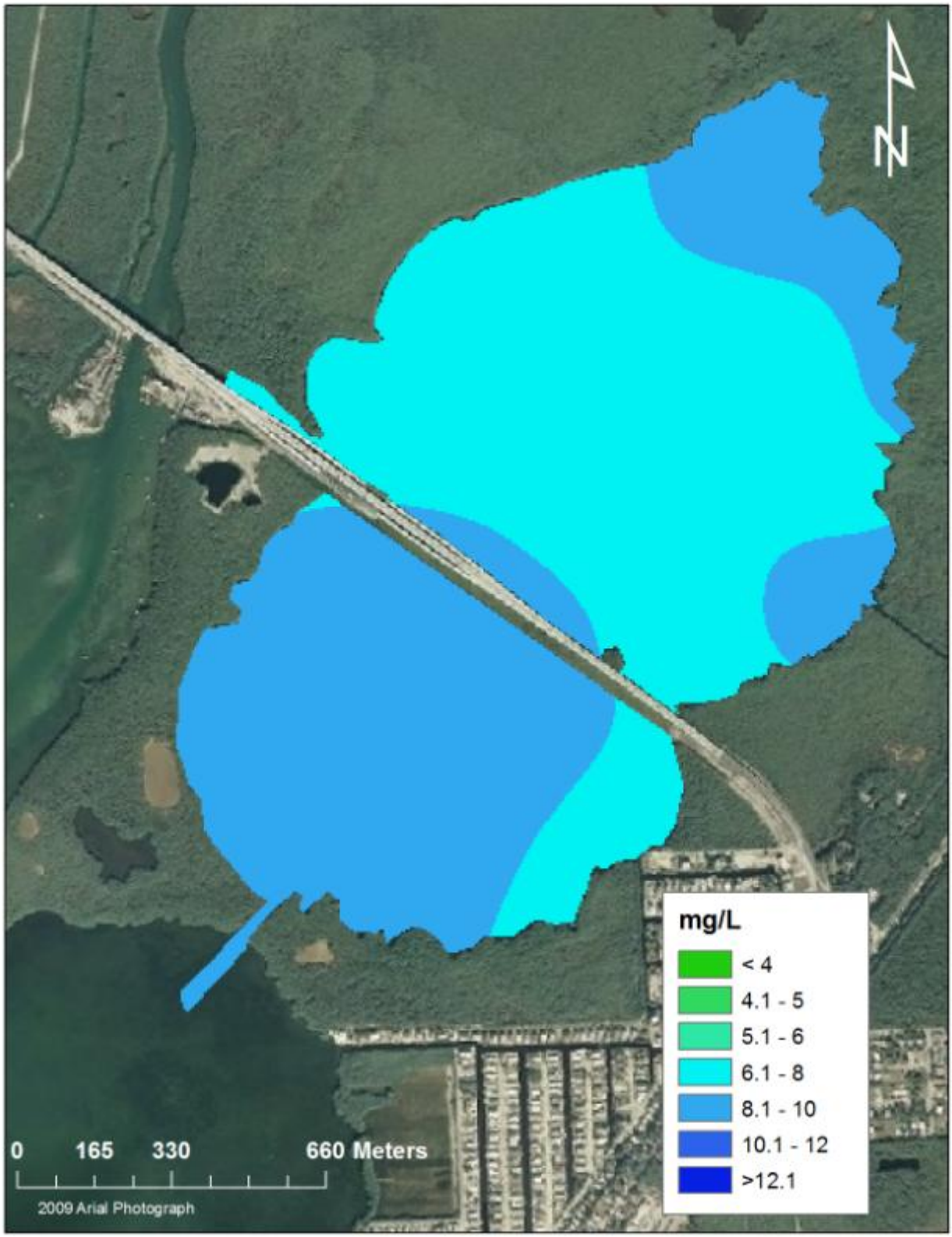
N
mg/L
< 4
4.1 - 5
5.1 - 6
6.1 - 8
8.1 - 10
10.1 - 12
>12.1
0
165
330
660 Meters
2009 Arial Photograph

Figure 206. Water column dissolved oxygen (winter 2009).

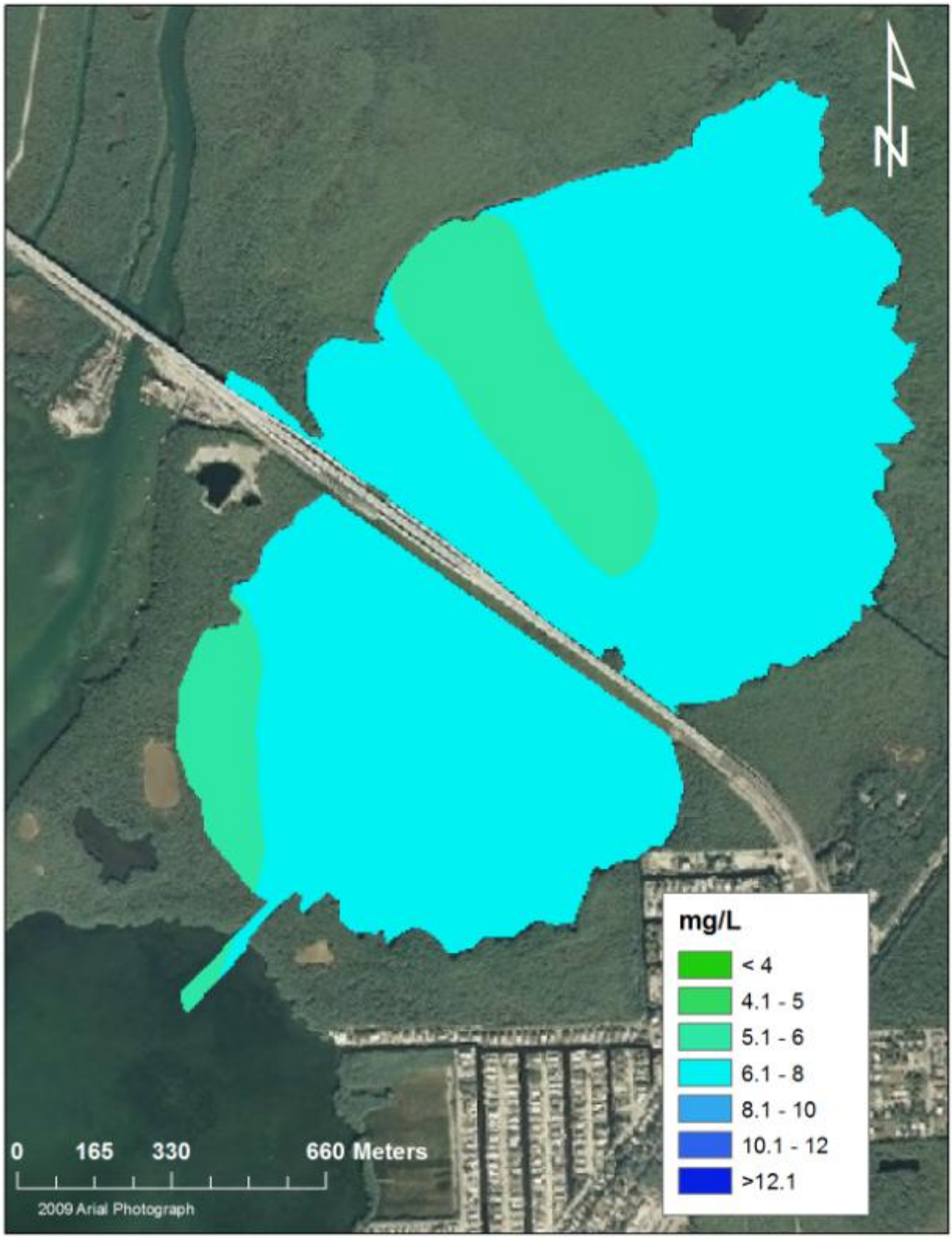

Figure 207. Water column dissolved oxygen (summer 2010).

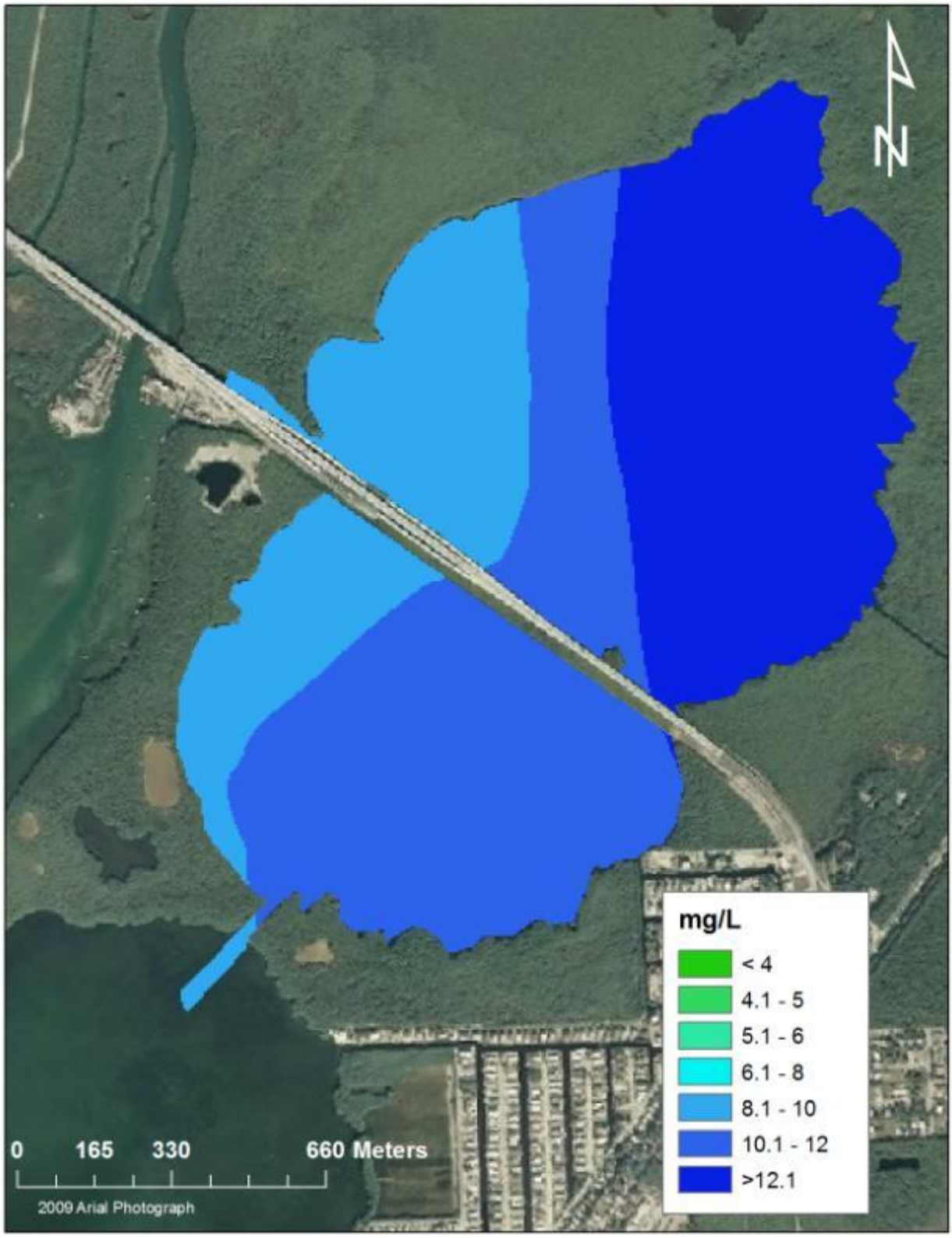
N
mg/L
< 4
4.1 - 5
5.1 - 6
6.1 - 8
8.1 - 10
10.1 - 12
>12.1
0
165
330
660 Meters
2009 Arial Photograph

Figure 208. Water column dissolved oxygen (winter 2010).

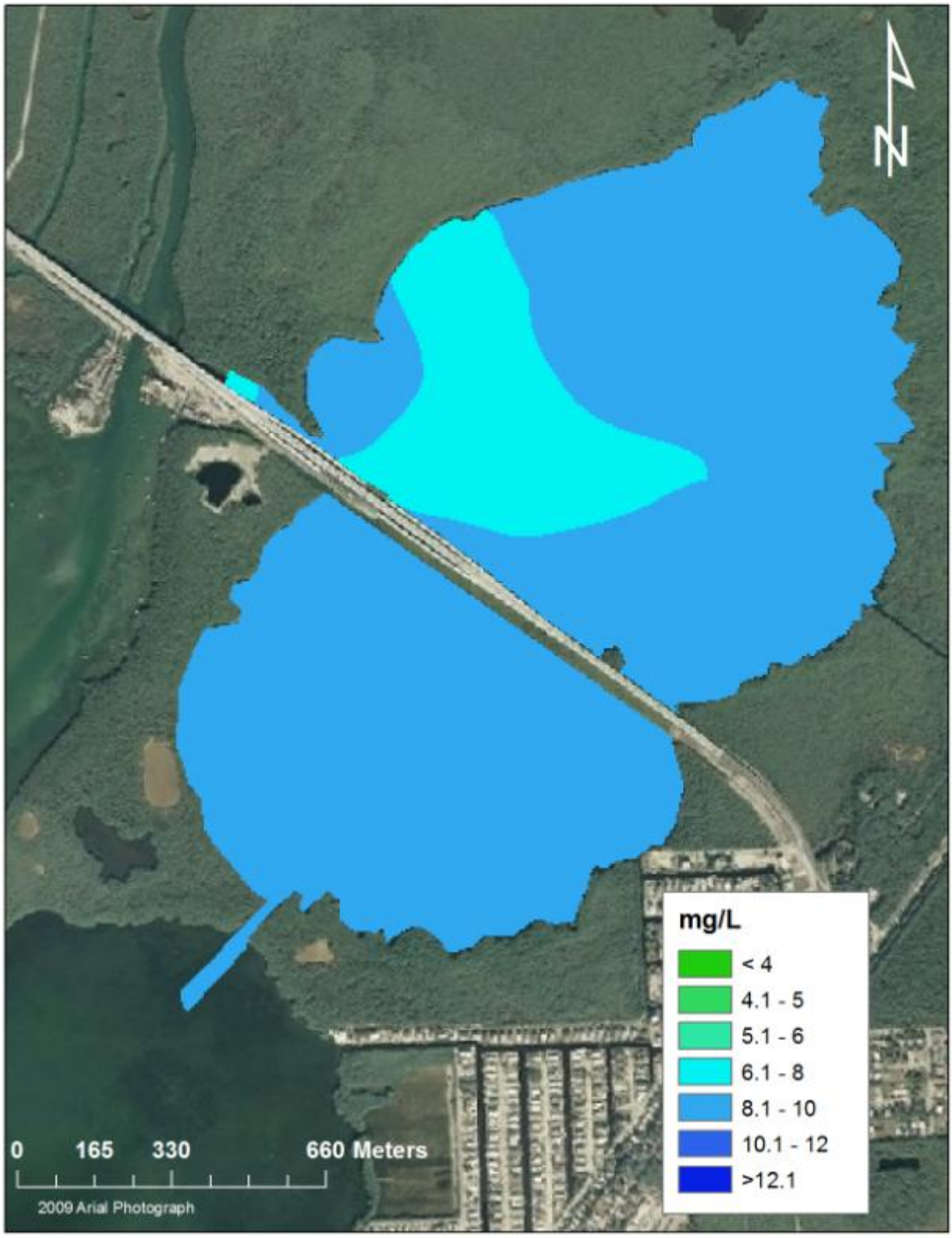
N
mg/L
< 4
4.1 - 5
5.1 - 6
6.1 - 8
8.1 - 10
10.1 - 12
>12.1
0
165
330
660 Meters
2009 Arial Photograph

Figure 209. Water column dissolved oxygen (summer 2011).

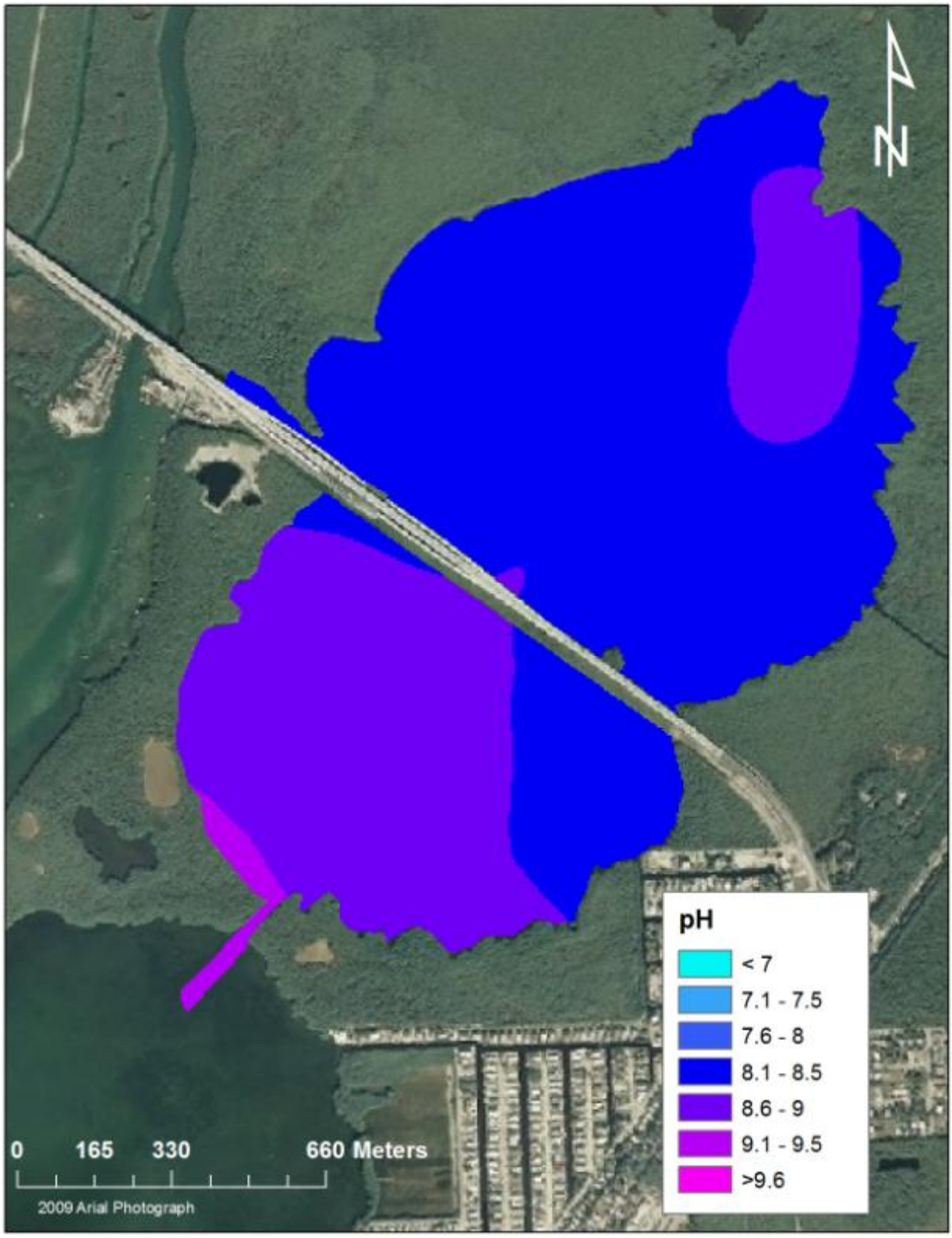
N
pH
< 7
7.1 - 7.5
7.6 - 8
8.1 - 8.5
8.6 - 9
9.1 - 9.5
>9.6
0
165
330
660 Meters
2009 Arial Photograph

Figure 210. Water column pH (summer 2009).

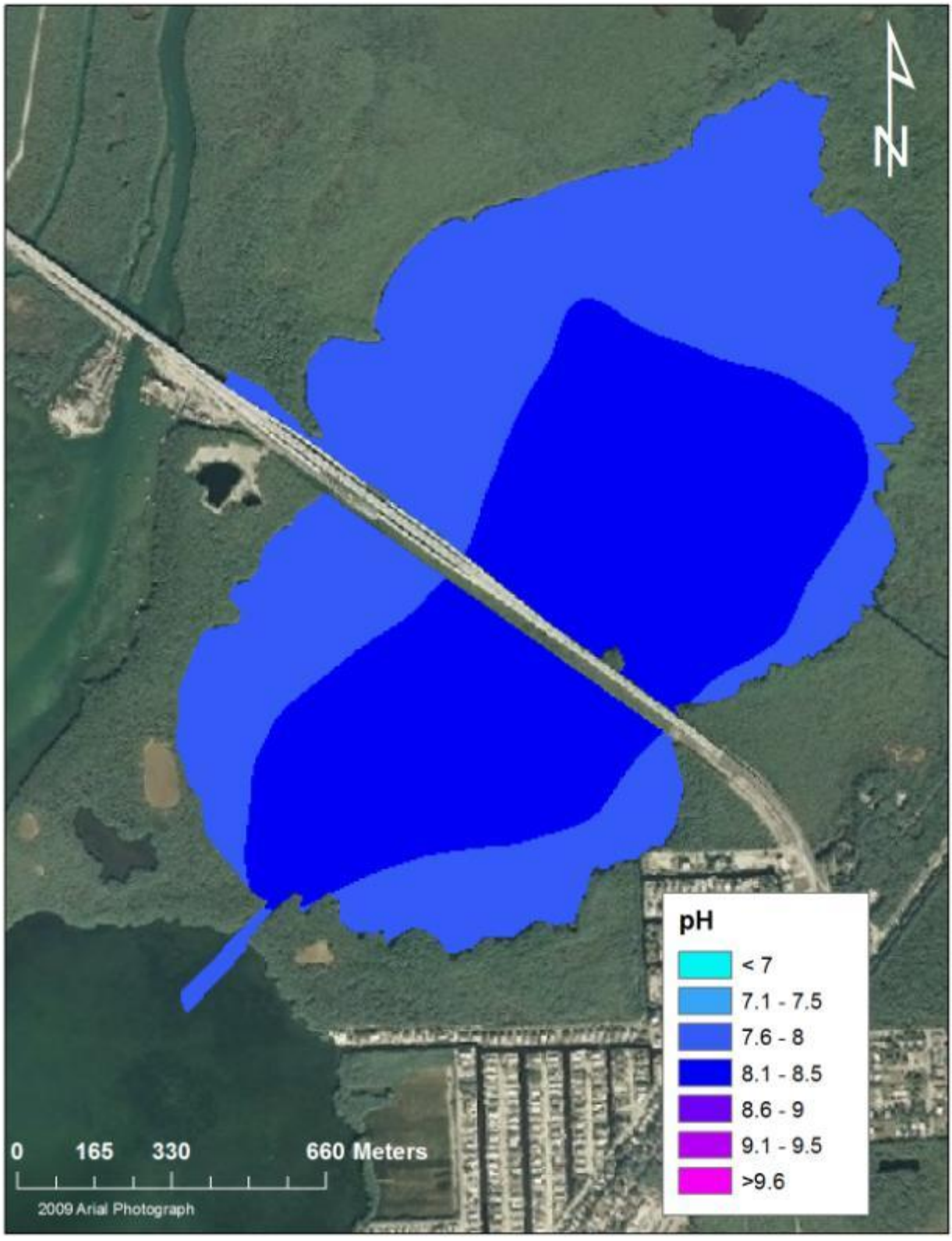

Figure 211. Water column pH (winter 2009).

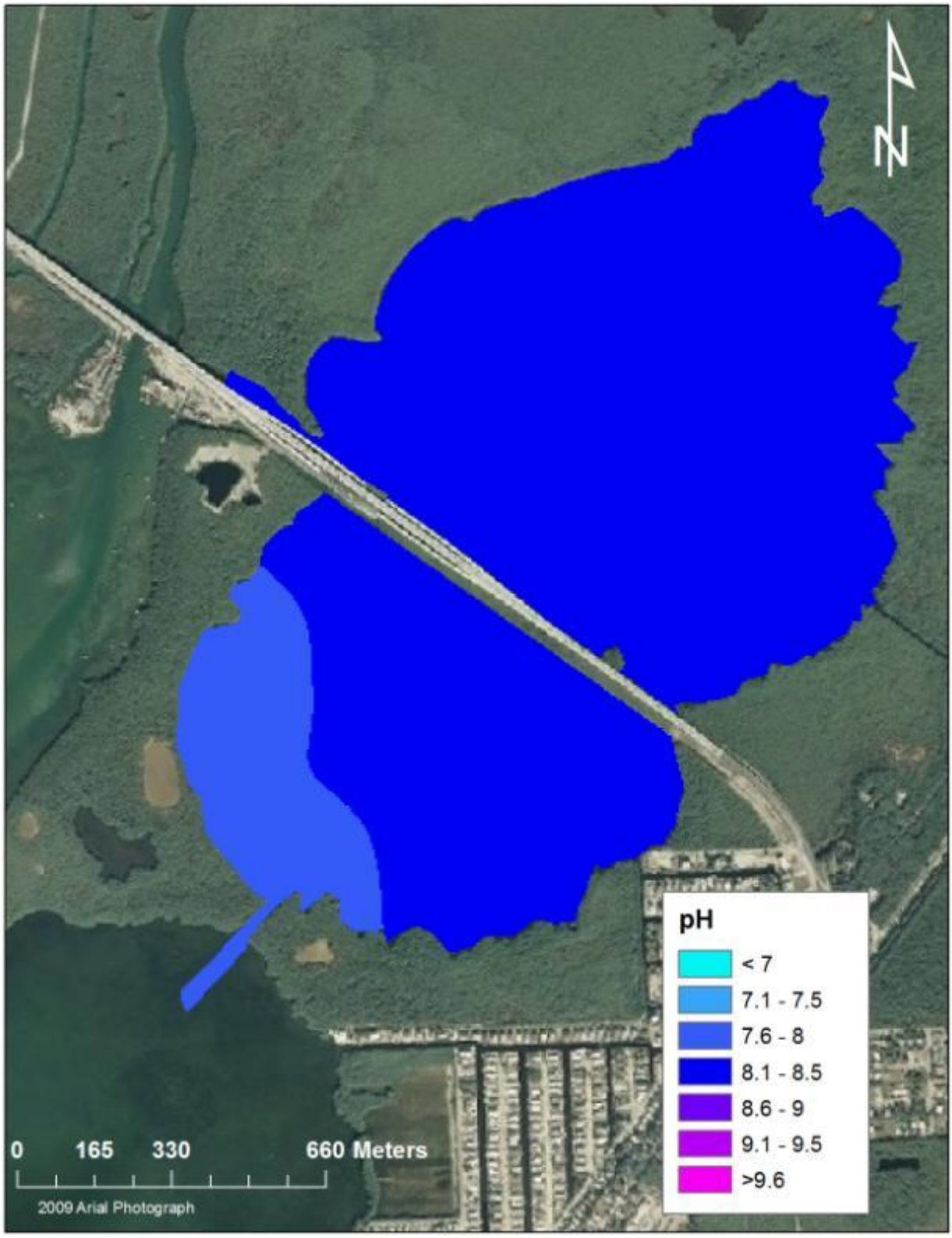
N
pH
< 7
7.1 - 7.5
7.6 - 8
8.1 - 8.5
8.6 - 9
9.1 - 9.5
>9.6
0
165
330
660 Meters
2009 Arial Photograph

Figure 212. Water column pH (summer 2010).

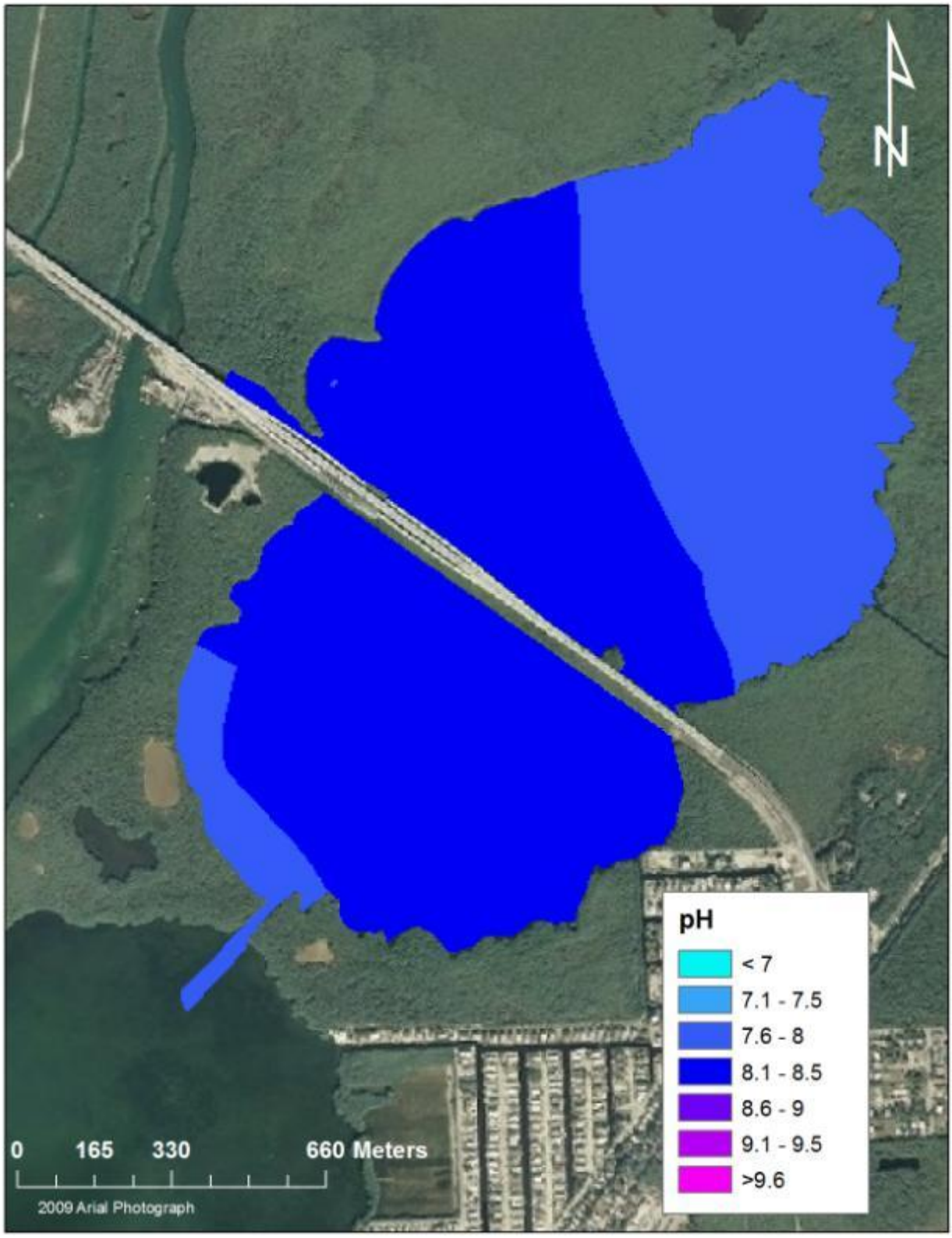
N
pH
< 7
7.1 - 7.5
7.6 - 8
8.1 - 8.5
8.6 - 9
9.1 - 9.5
>9.6
0
165
330
660 Meters
2009 Arial Photograph

Figure 213. Water column pH (winter 2010).

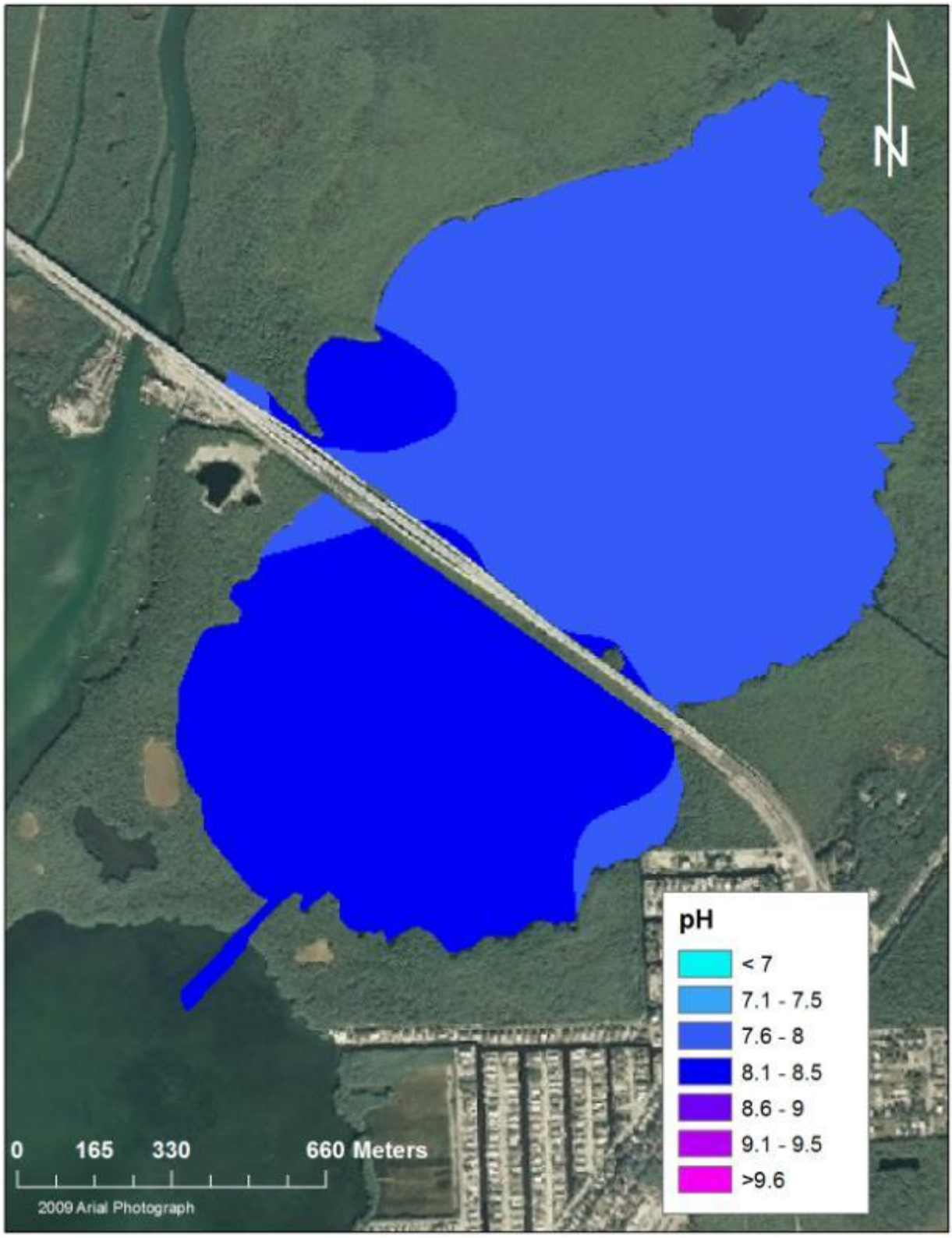
N
pH
< 7
7.1 - 7.5
7.6 - 8
8.1 - 8.5
8.6 - 9
9.1 - 9.5
>9.6
0
165
330
660 Meters
2009 Arial Photograph

Figure 214. Water column pH (summer 2011).


N
ppt
< 50
50.1 - 53
53.1 - 56
56.1 - 59
59.1 - 62
62.1 - 65
>65.1
0
165
330
660 Meters
2009 Arial Photograph

Figure 215. Water column salinity (summer 2009).

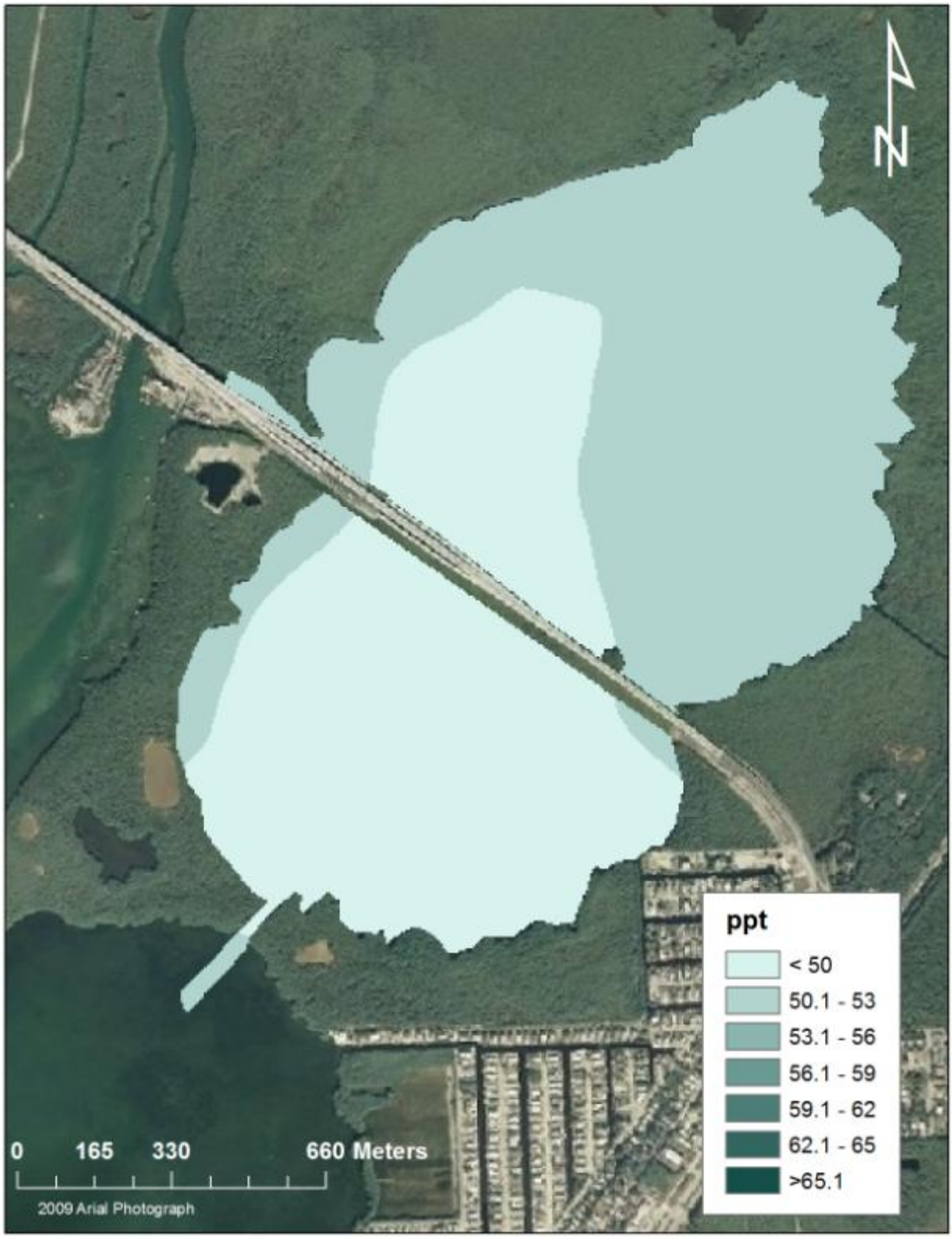

Figure 216. Water column salinity (winter 2009).

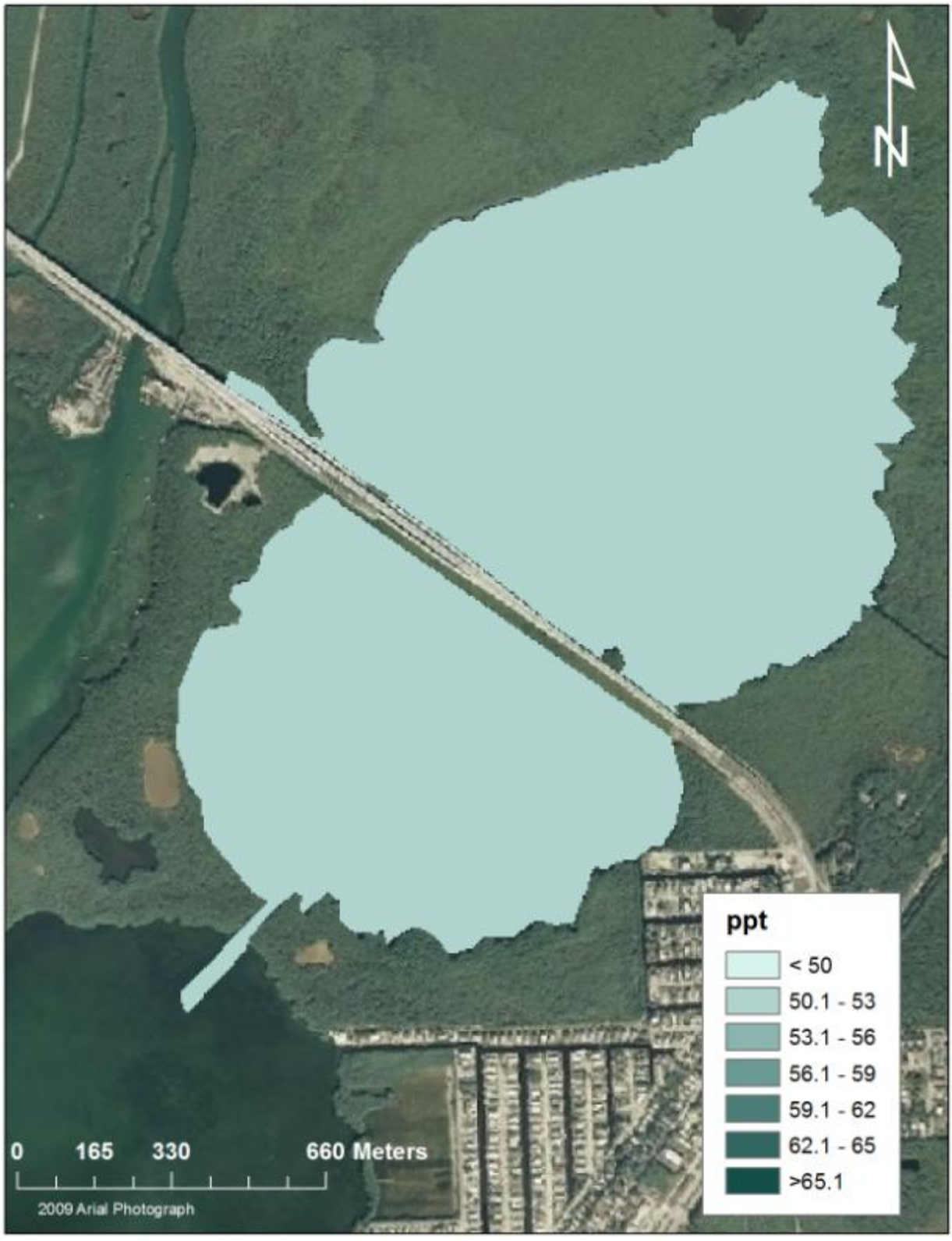

Figure 217. Water column salinity (summer 2010).

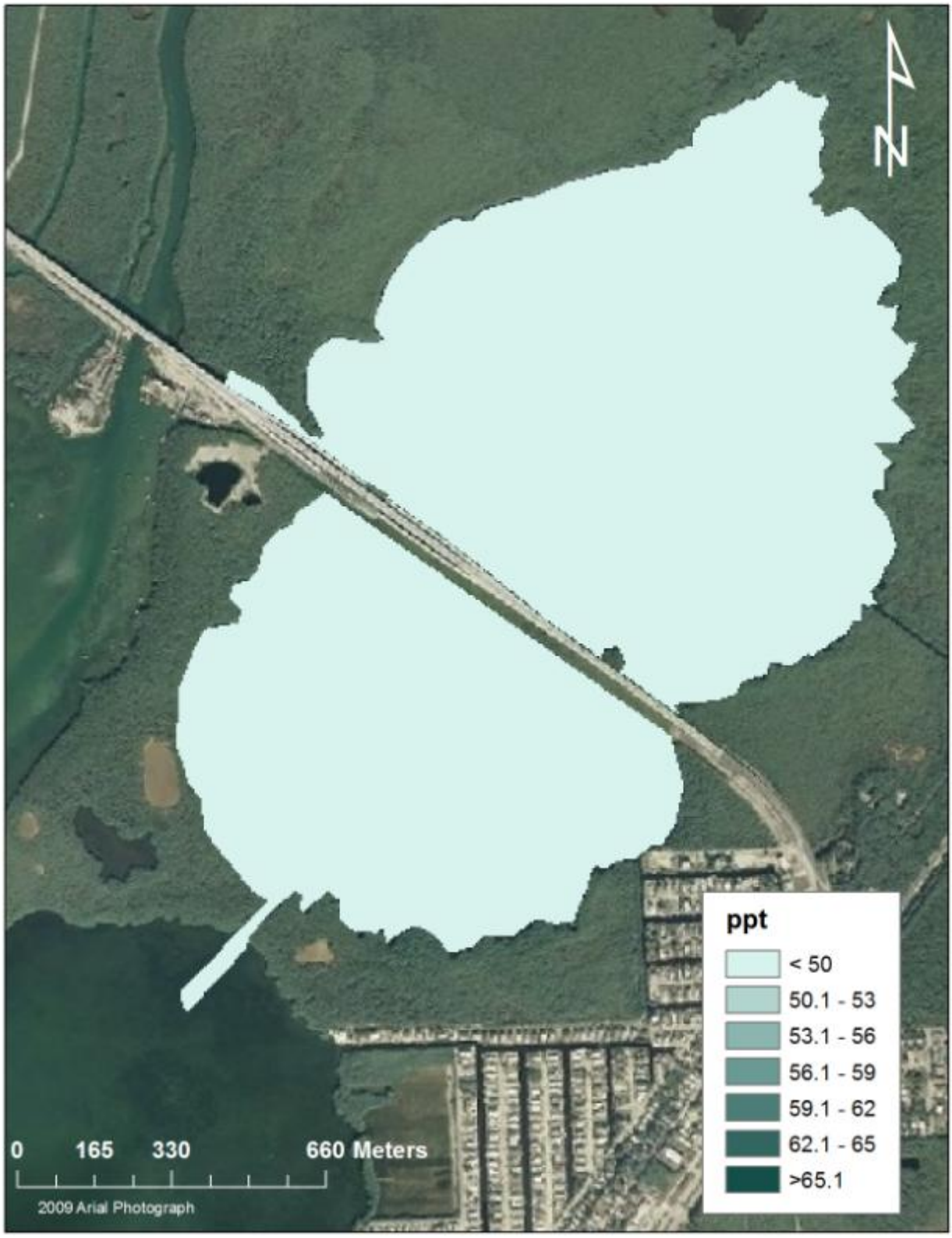
N
ppt
< 50
50.1 - 53
53.1 - 56
56.1 - 59
59.1 - 62
62.1 - 65
>65.1
0 165 330 660 Meters
2009 Arial Photograph

Figure 218. Water column salinity (winter 2010).


N
ppt
< 50
50.1 - 53
53.1 - 56
56.1 - 59
59.1 - 62
62.1 - 65
>65.1
0
165
330
660 Meters
2009 Arial Photograph

Figure 219. Water column salinity (summer 2011).

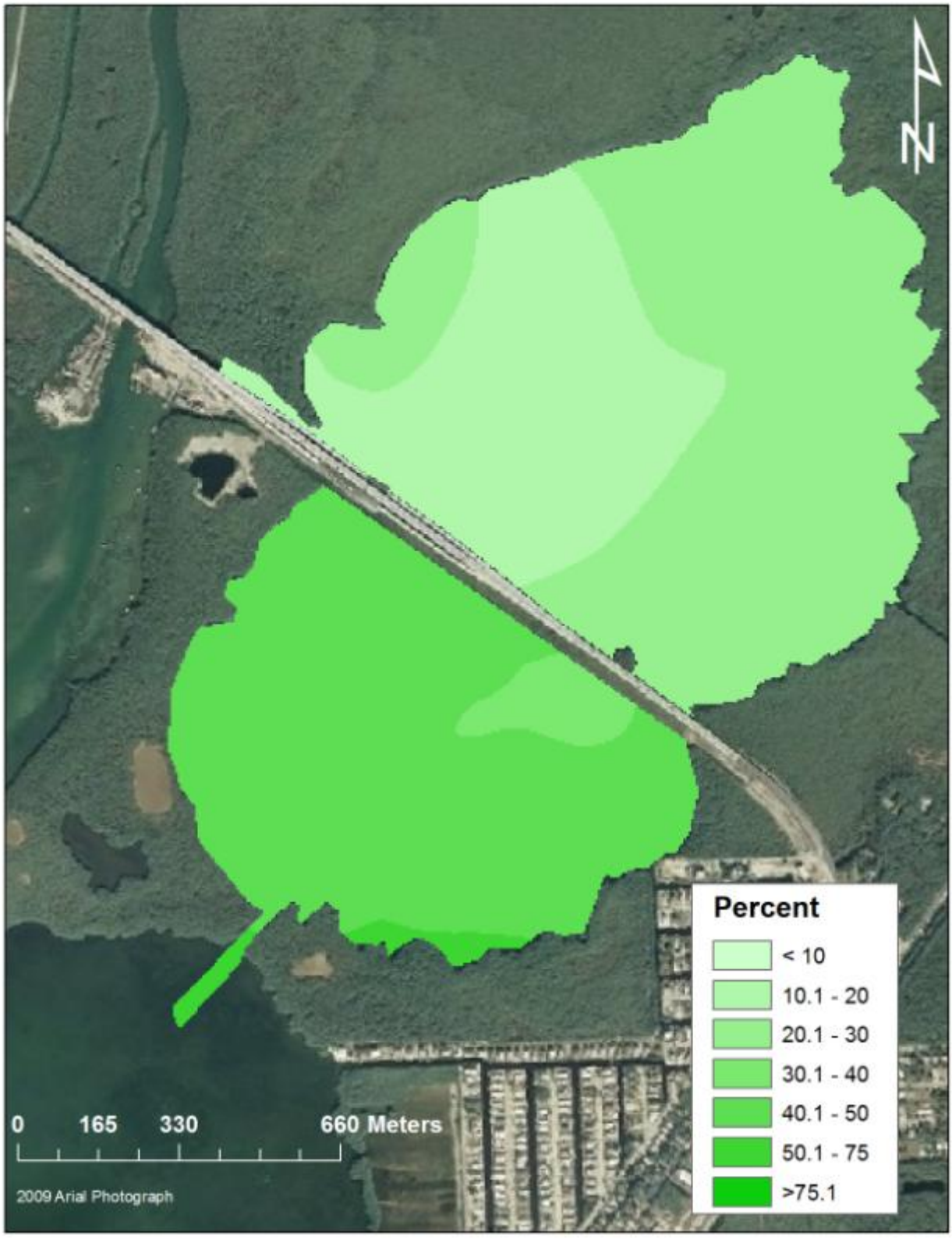

Figure 220. Seagrass percent cover (winter 2008).

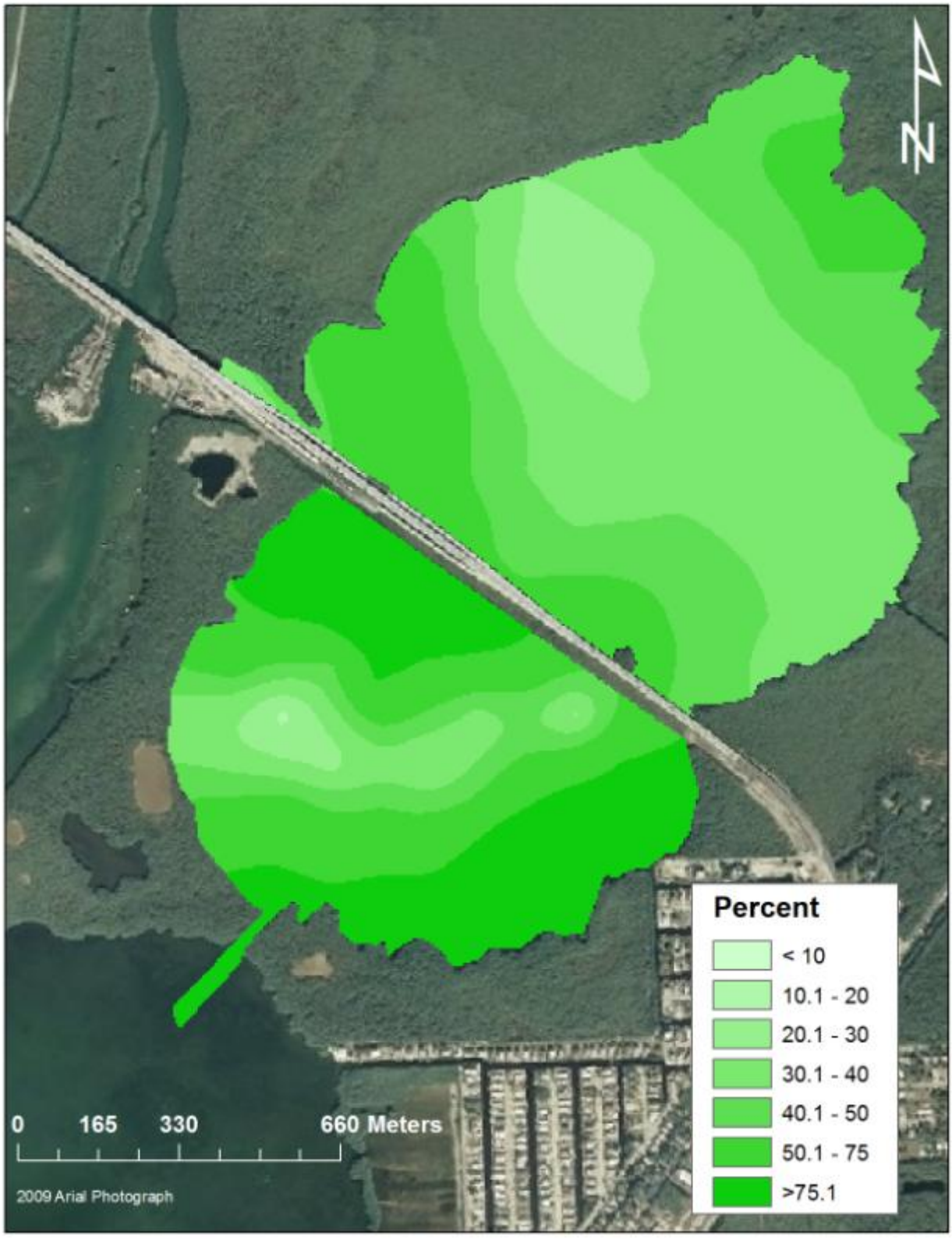
N
Percent
< 10
10.1 - 20
20.1 - 30
30.1 - 40
40.1 - 50
50.1 - 75
>75.1
0
165
330
660 Meters
2009 Arial Photograph

Figure 221. Seagrass percent cover (summer 2009).

Figure 222. Seagrass percent cover (winter 2009).

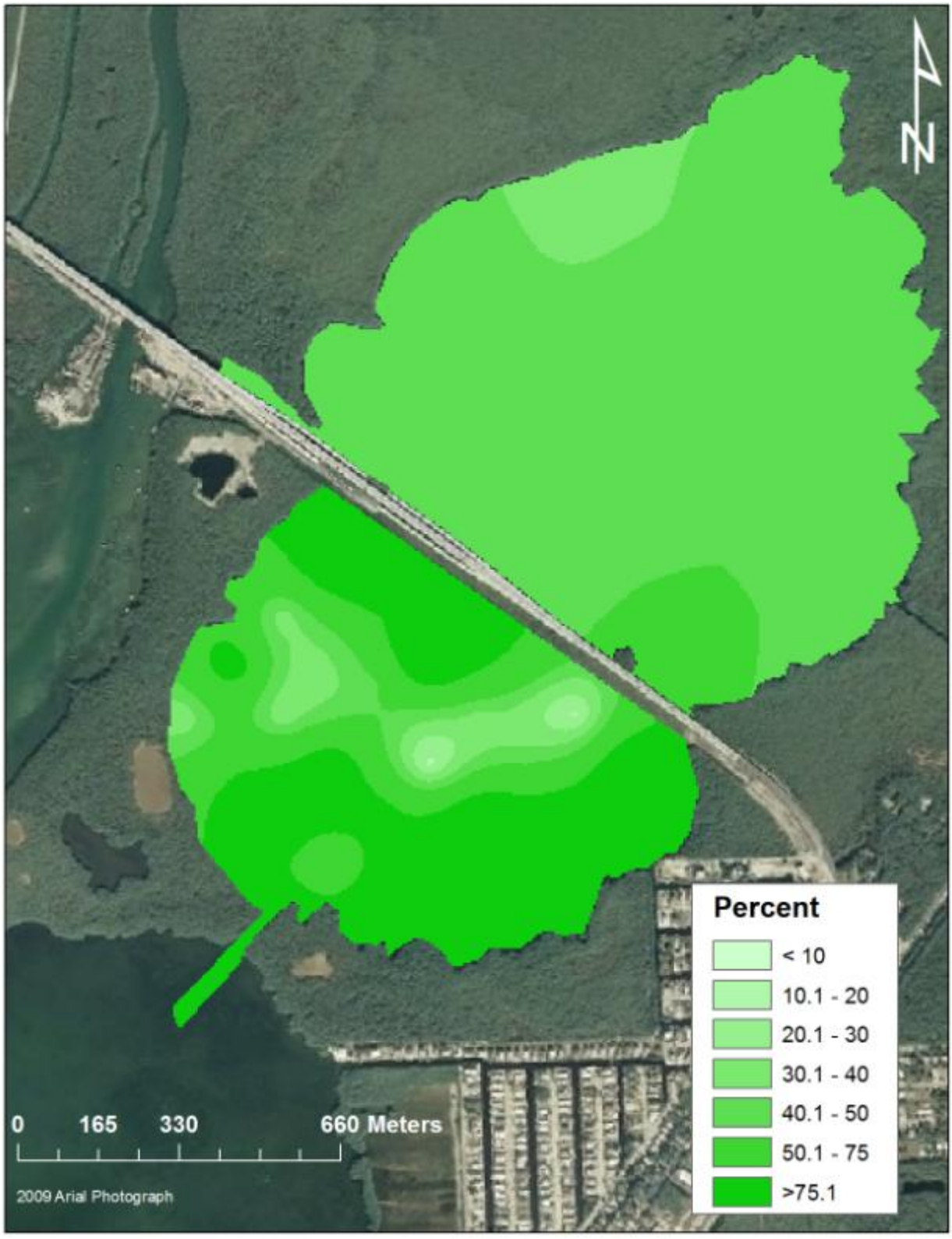

Figure 223. Seagrass percent cover (summer 2010).

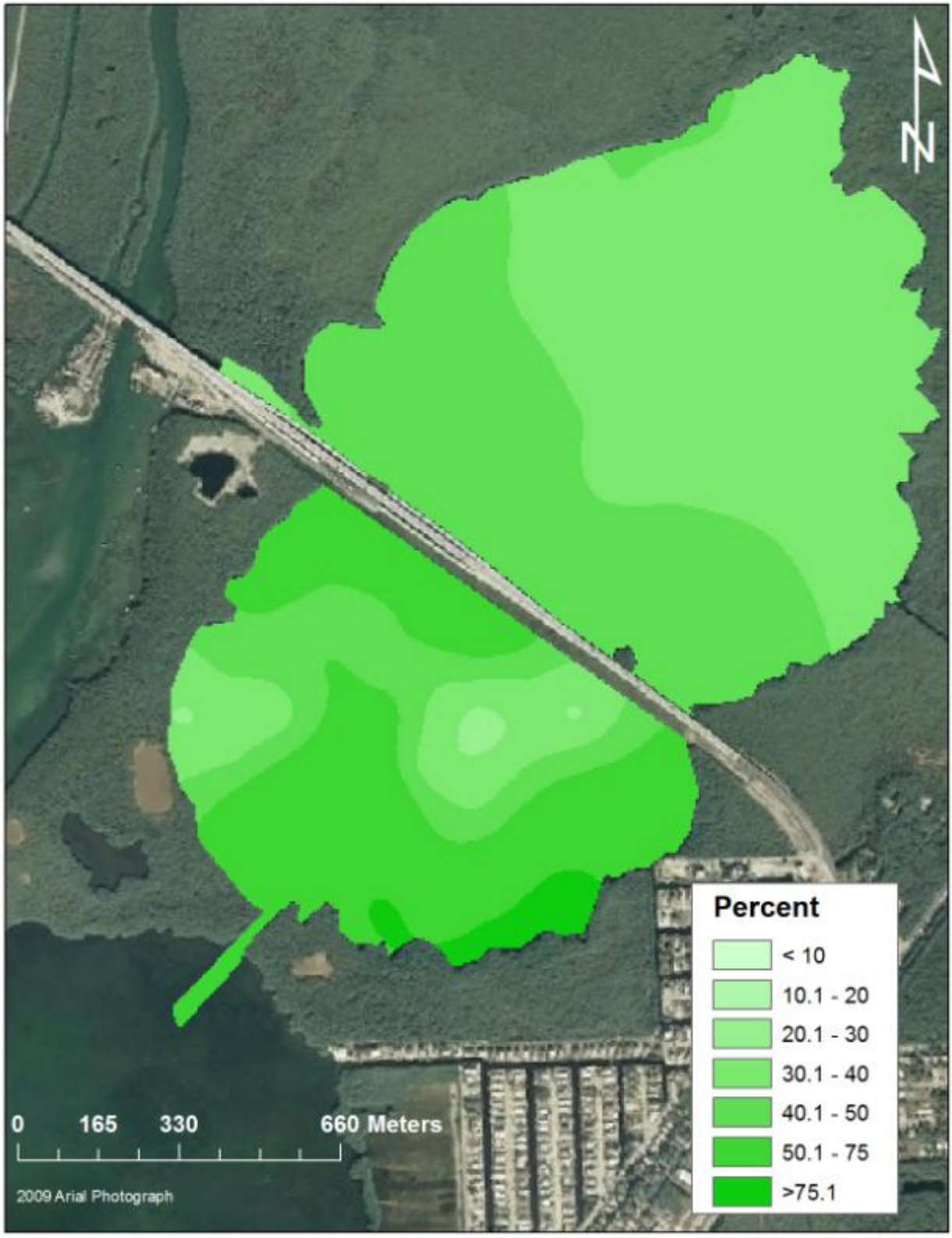

Figure 224. Seagrass percent cover (winter 2010)

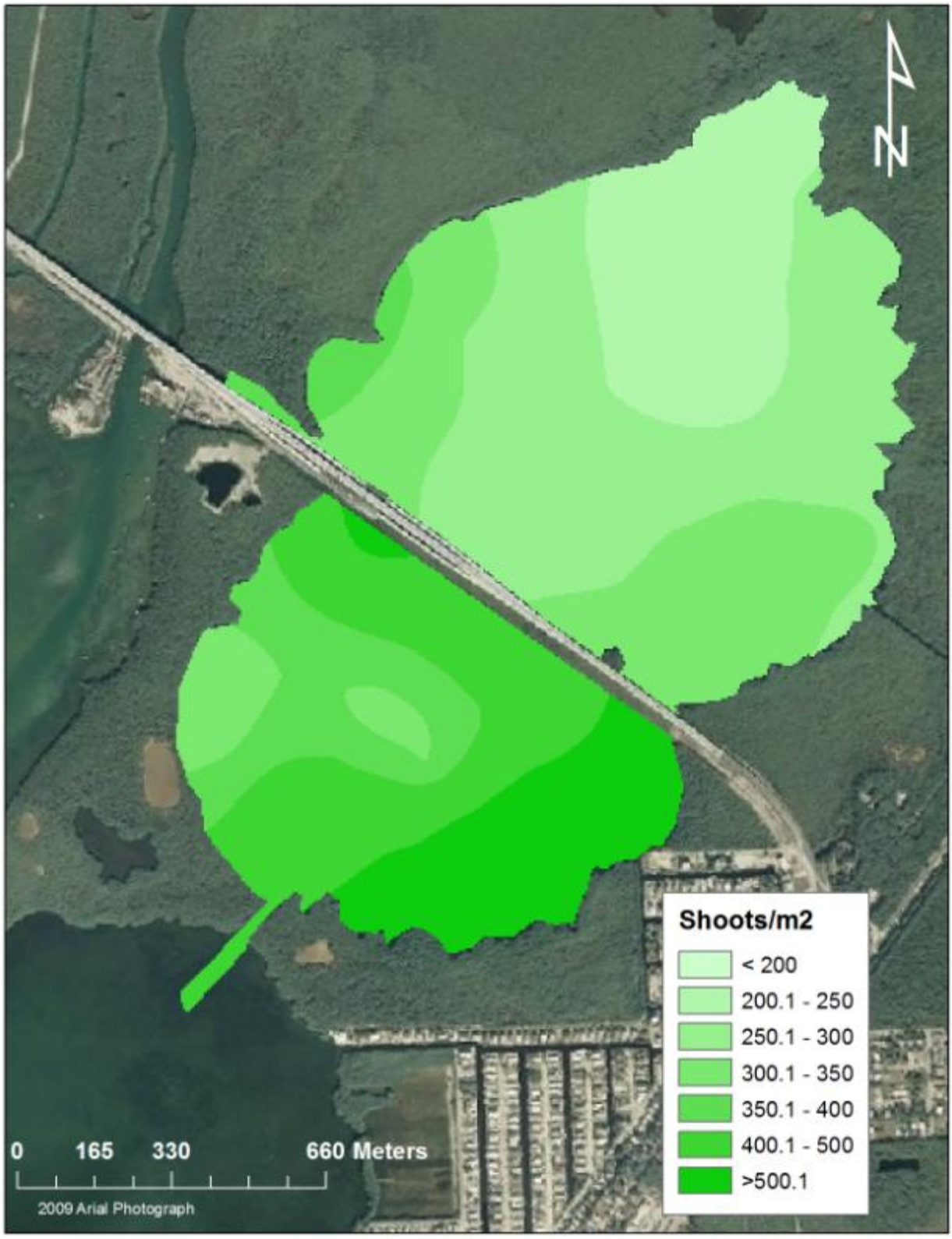
N
Shoots/m2
< 200
200.1 - 250
250.1 - 300
300.1 - 350
350.1 - 400
400.1 - 500
>500.1
0
165
330
660 Meters
2009 Arial Photograph

Figure 225. Seagrass percent cover (summer 2011).


N
Percent
< 10
10.1 - 20
20.1 - 30
30.1 - 40
40.1 - 50
50.1 - 75
>75.1
0
165
330
660 Meters
2009 Arial Photograph

Figure 226. *Thalassia testudinum* percent cover (winter 2008).

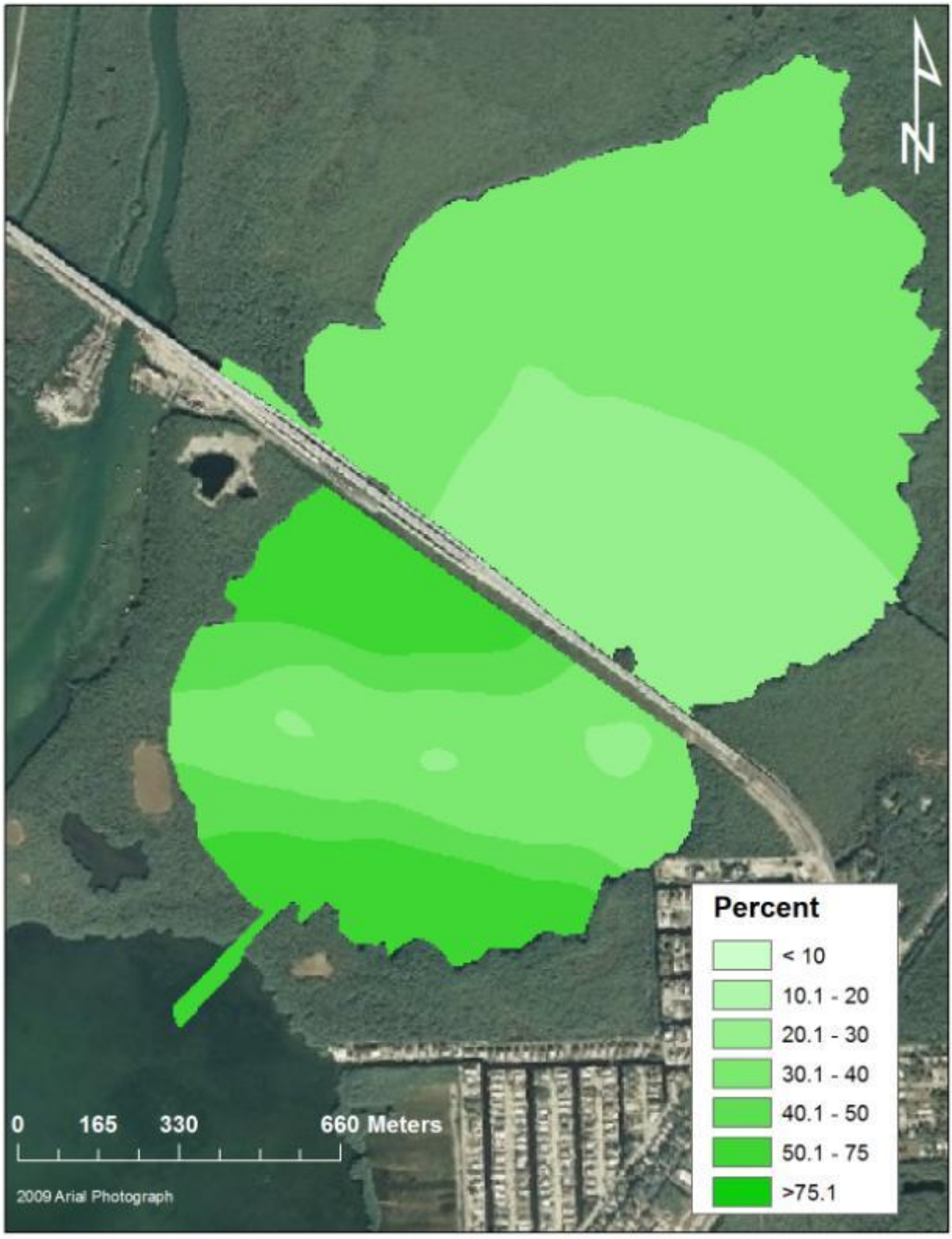

Figure 227. *Thalassia testudinum* percent cover (summer 2009).


N
Percent
< 10
10.1 - 20
20.1 - 30
30.1 - 40
40.1 - 50
50.1 - 75
>75.1
0
165
330
660 Meters
2009 Arial Photograph

Figure 228. *Thalassia testudinum* percent cover (winter 2009).

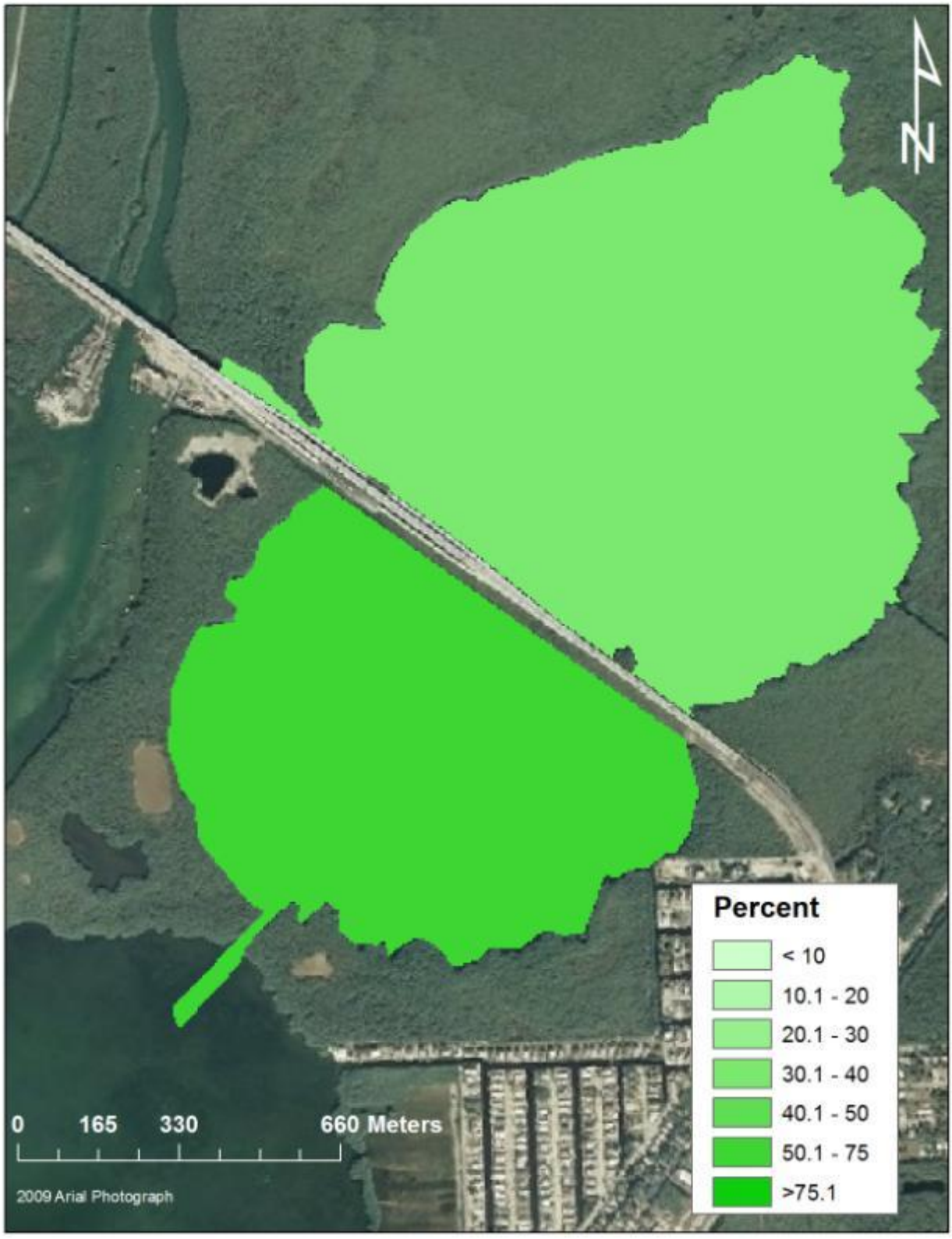
N
Percent
< 10
10.1 - 20
20.1 - 30
30.1 - 40
40.1 - 50
50.1 - 75
>75.1
0
165
330
660 Meters
2009 Arial Photograph

Figure 229. *Thalassia testudinum* percent cover (summer 2010).

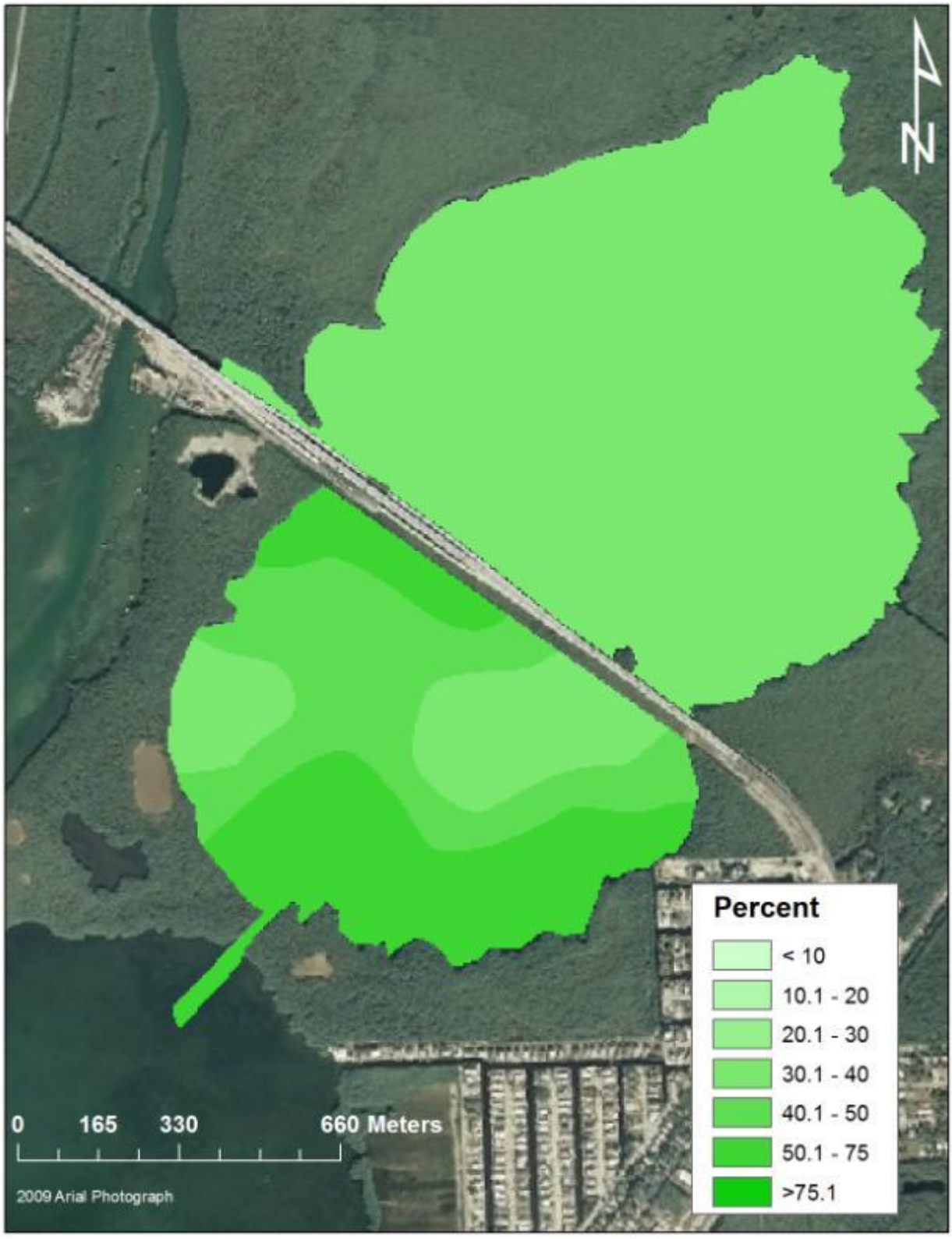

Figure 230. *Thalassia testudinum* percent cover (winter 2010).

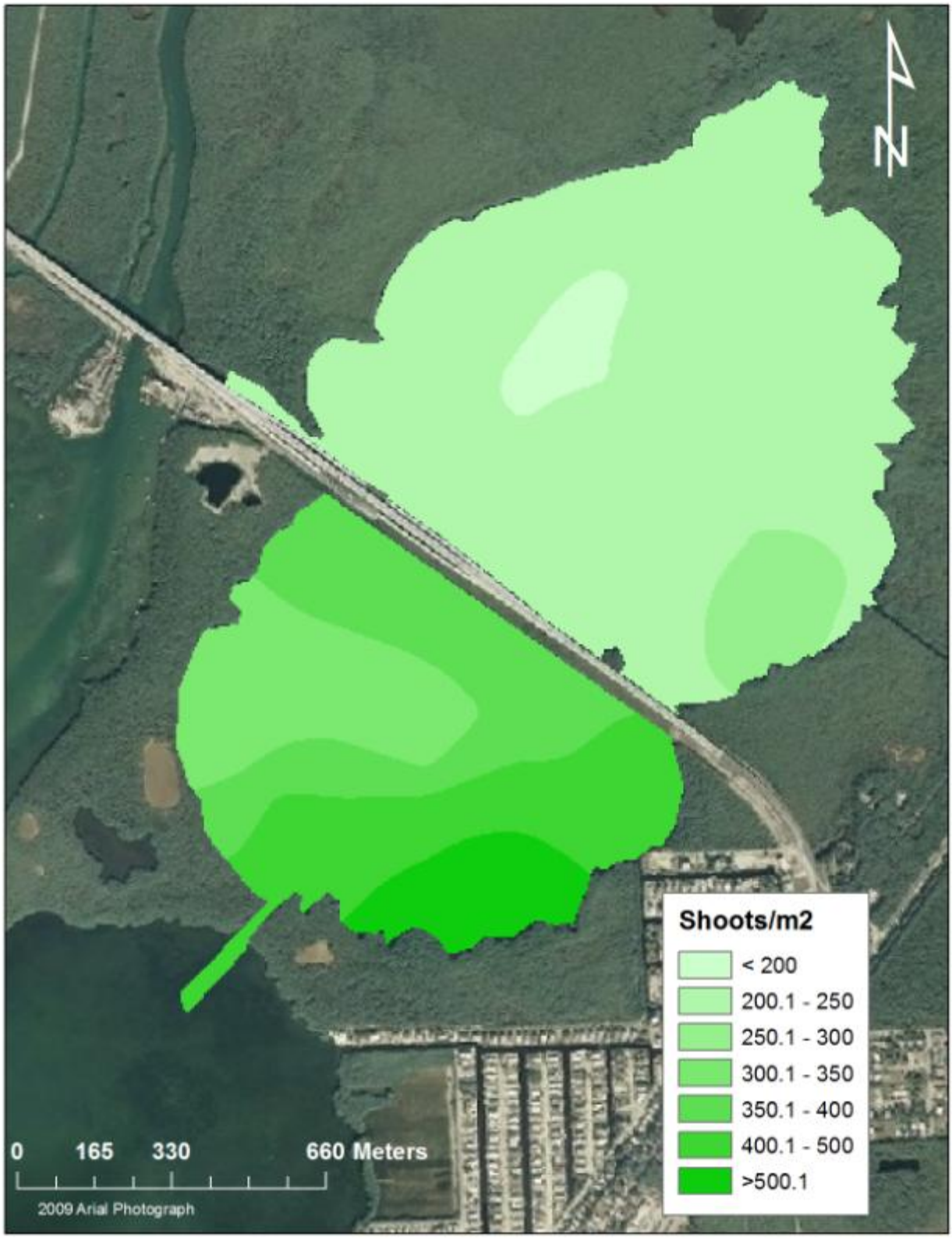

Figure 231. *Thalassia testudinum* percent cover (winter 2011).

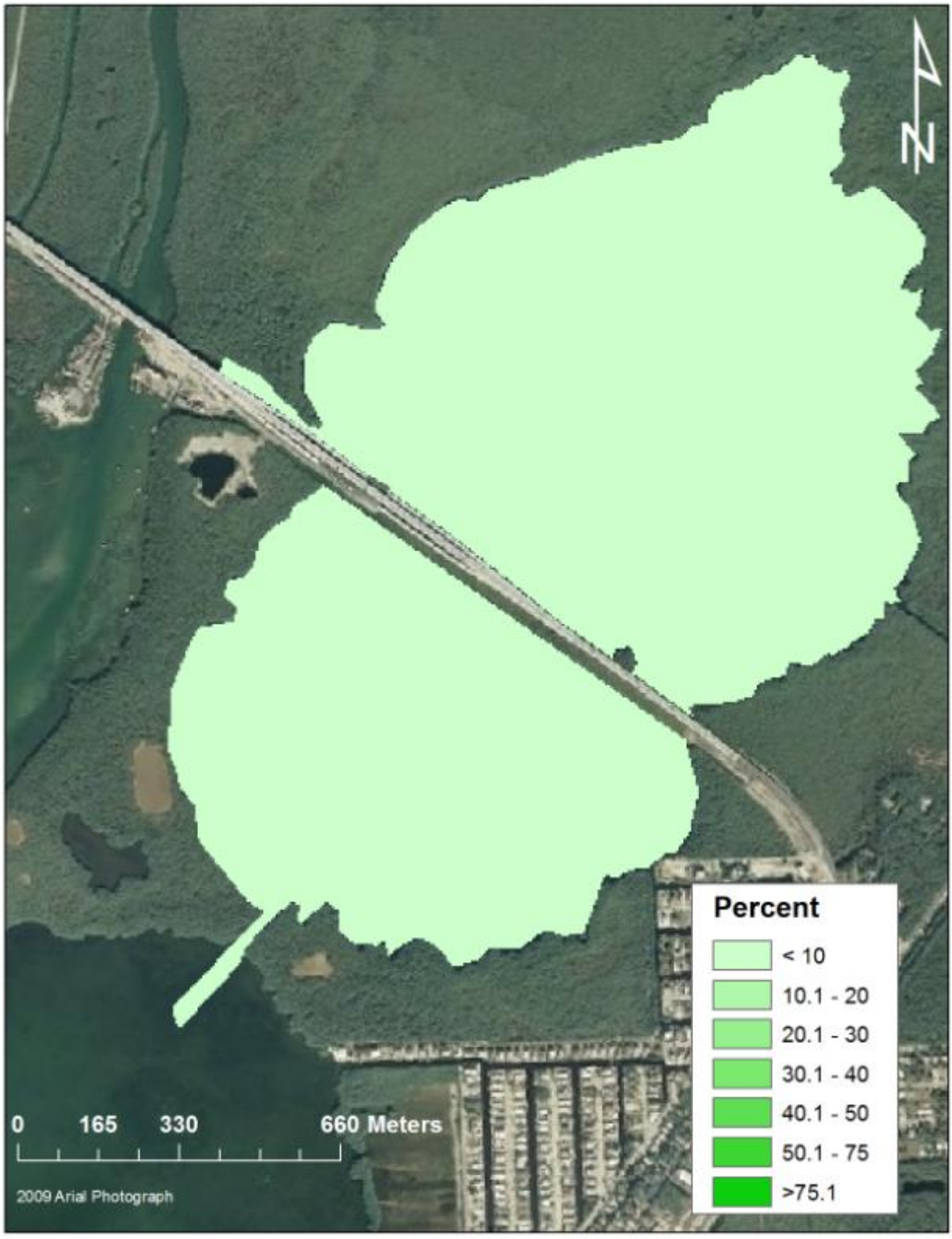
N
Percent
< 10
10.1 - 20
20.1 - 30
30.1 - 40
40.1 - 50
50.1 - 75
>75.1
0
165
330
660 Meters
2009 Arial Photograph

Figure 232. *Halodule wrightii* percent cover (winter 2008).

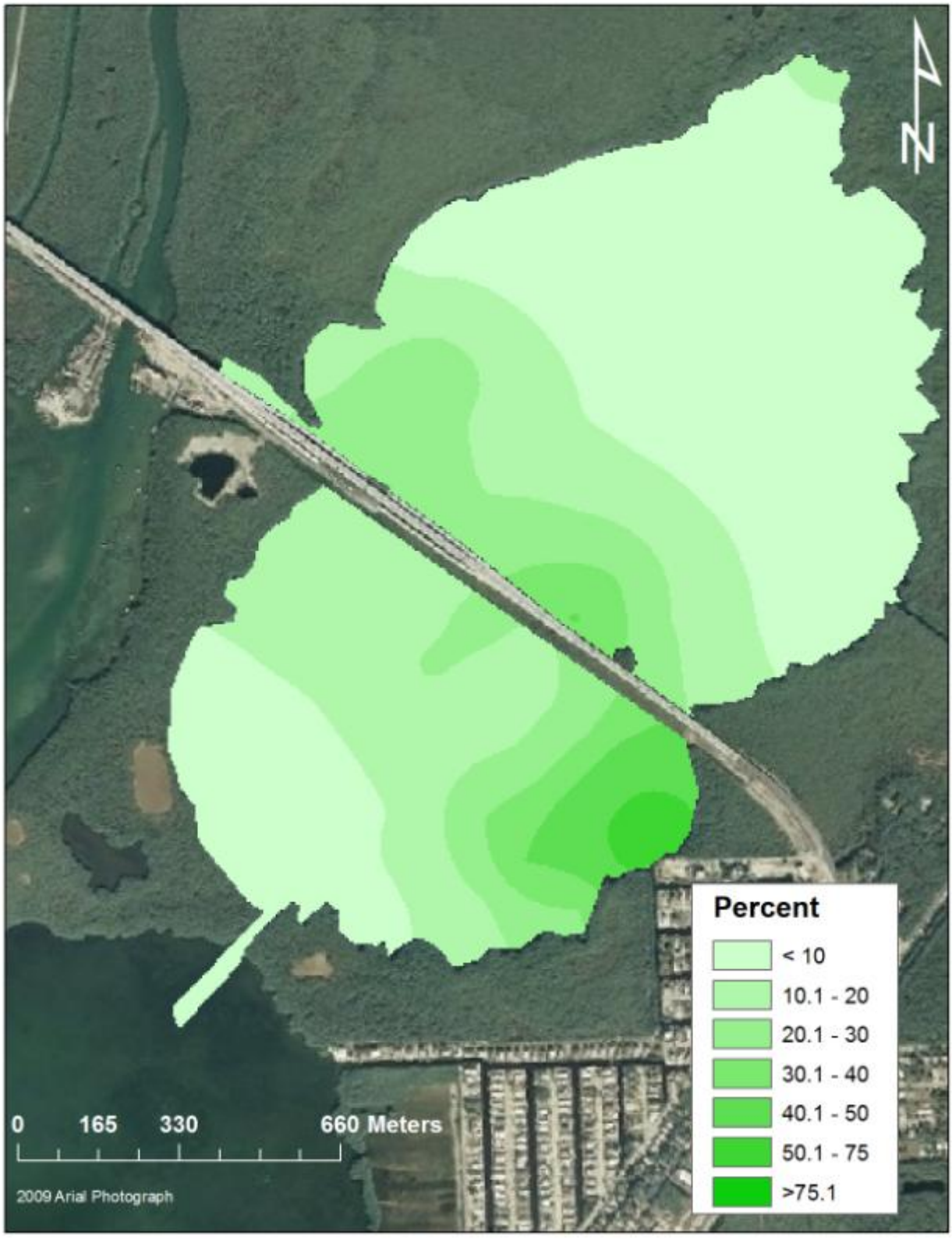

Figure 233. *Halodule wrightii* percent cover (summer 2009).

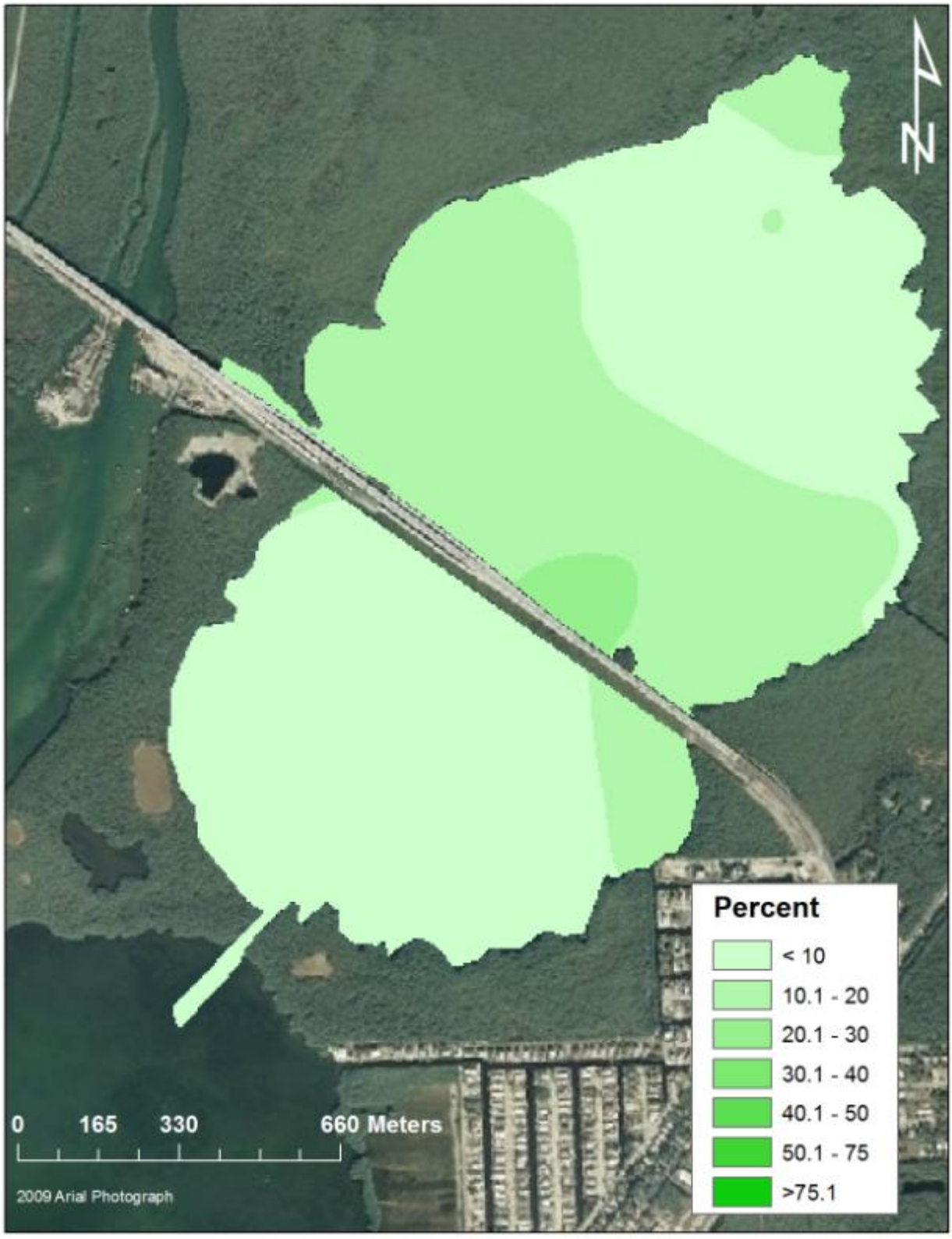
N
Percent
< 10
10.1 - 20
20.1 - 30
30.1 - 40
40.1 - 50
50.1 - 75
>75.1
0
165
330
660 Meters
2009 Arial Photograph

Figure 234. *Halodule wrightii* percent cover (winter 2009).

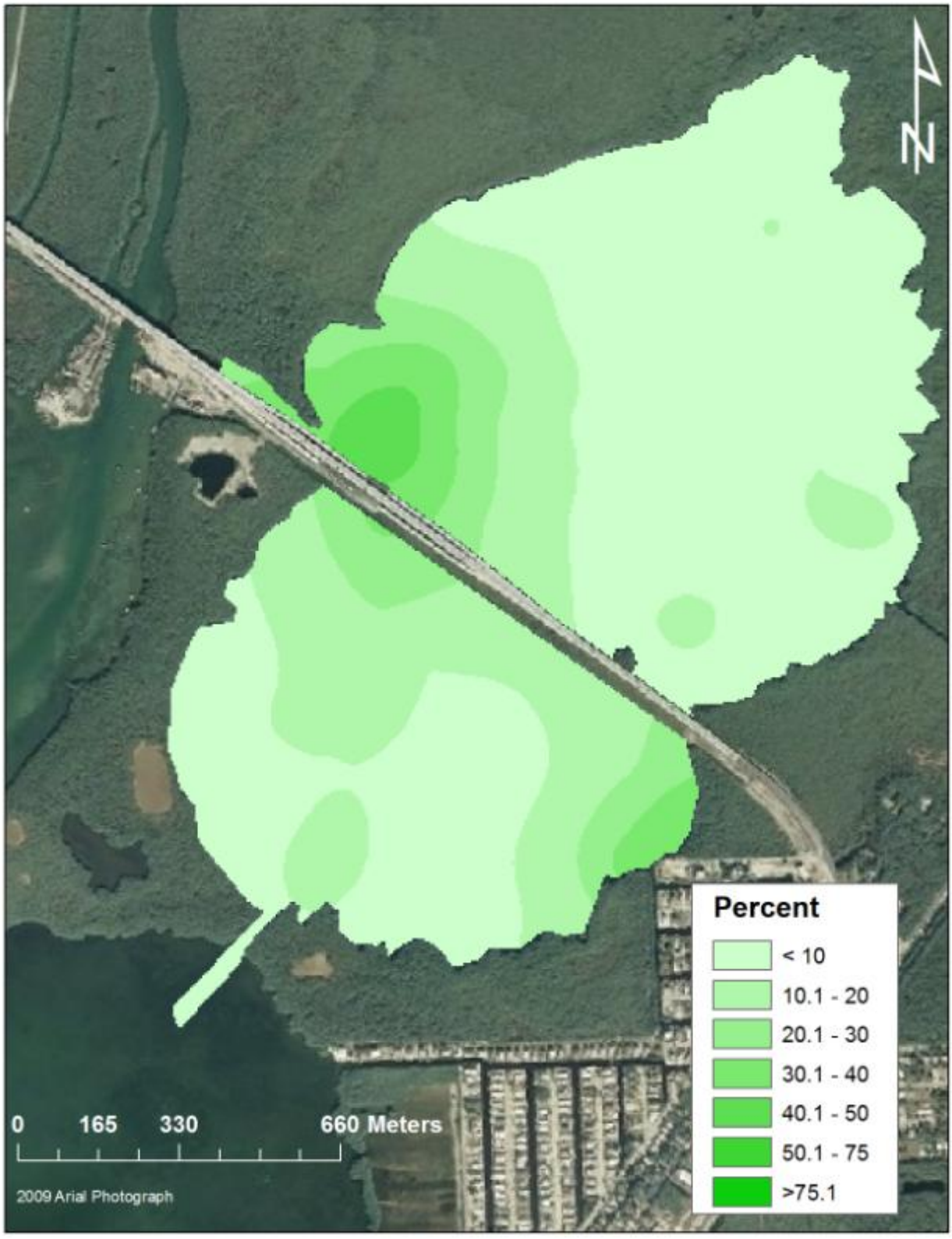
N
Percent
< 10
10.1 - 20
20.1 - 30
30.1 - 40
40.1 - 50
50.1 - 75
>75.1
0
165
330
660 Meters
2009 Arial Photograph

Figure 235. *Halodule wrightii* percent cover (summer 2010).

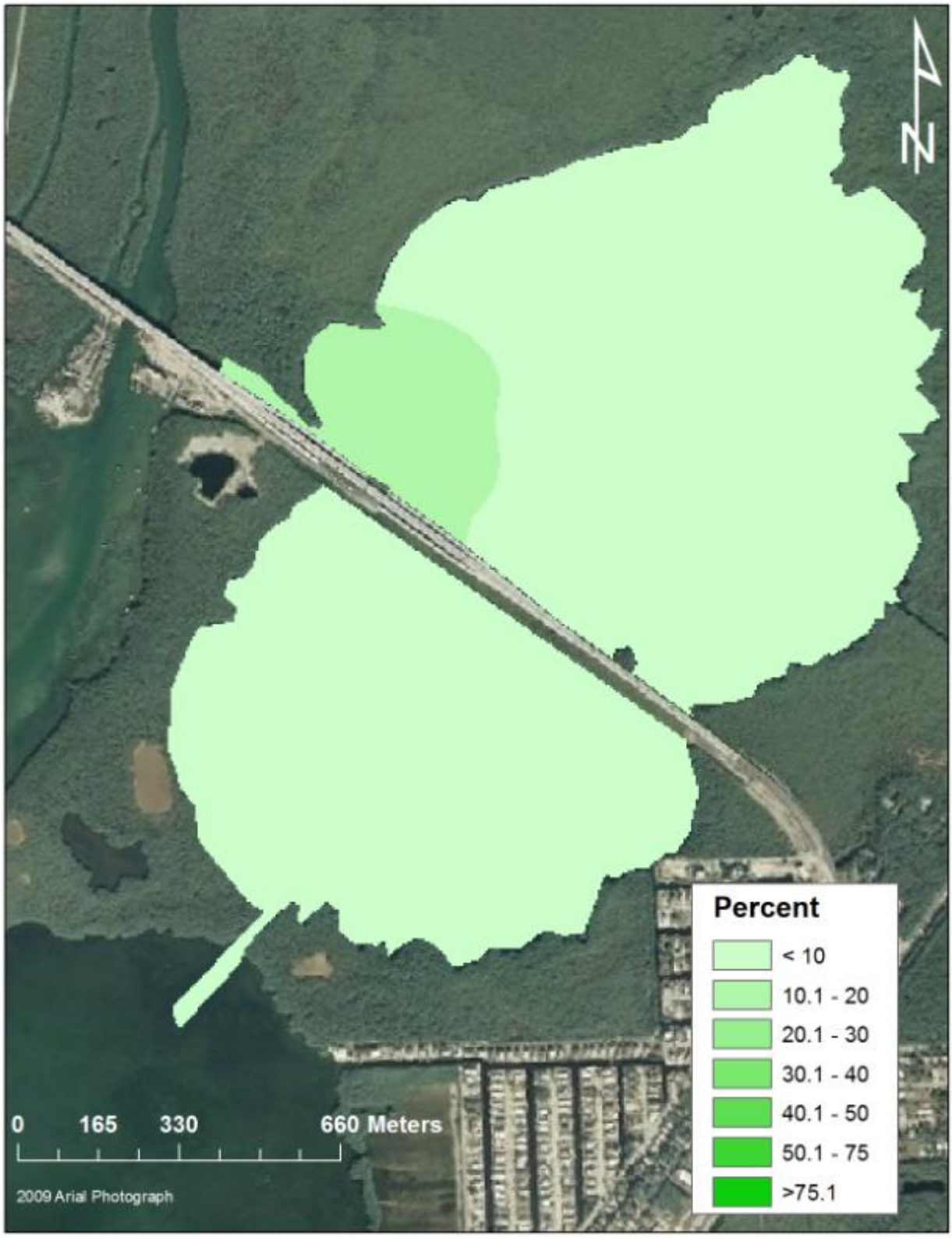

Figure 236. *Halodule wrightii* percent cover (winter 2010).

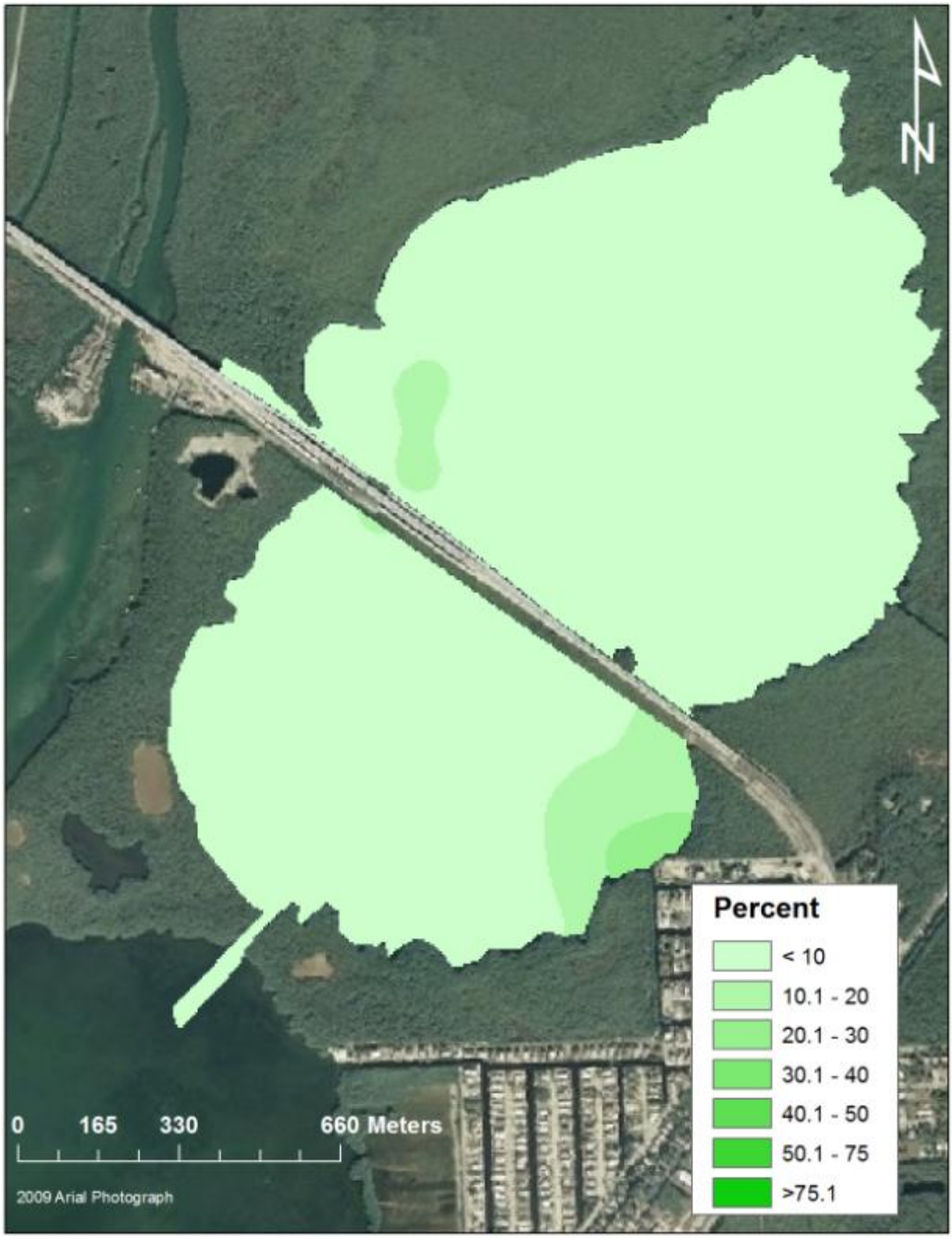

Figure 237. *Halodule wrightii* percent cover (summer 2011).

Figure 238. Seagrass shoot density (winter 2008).)

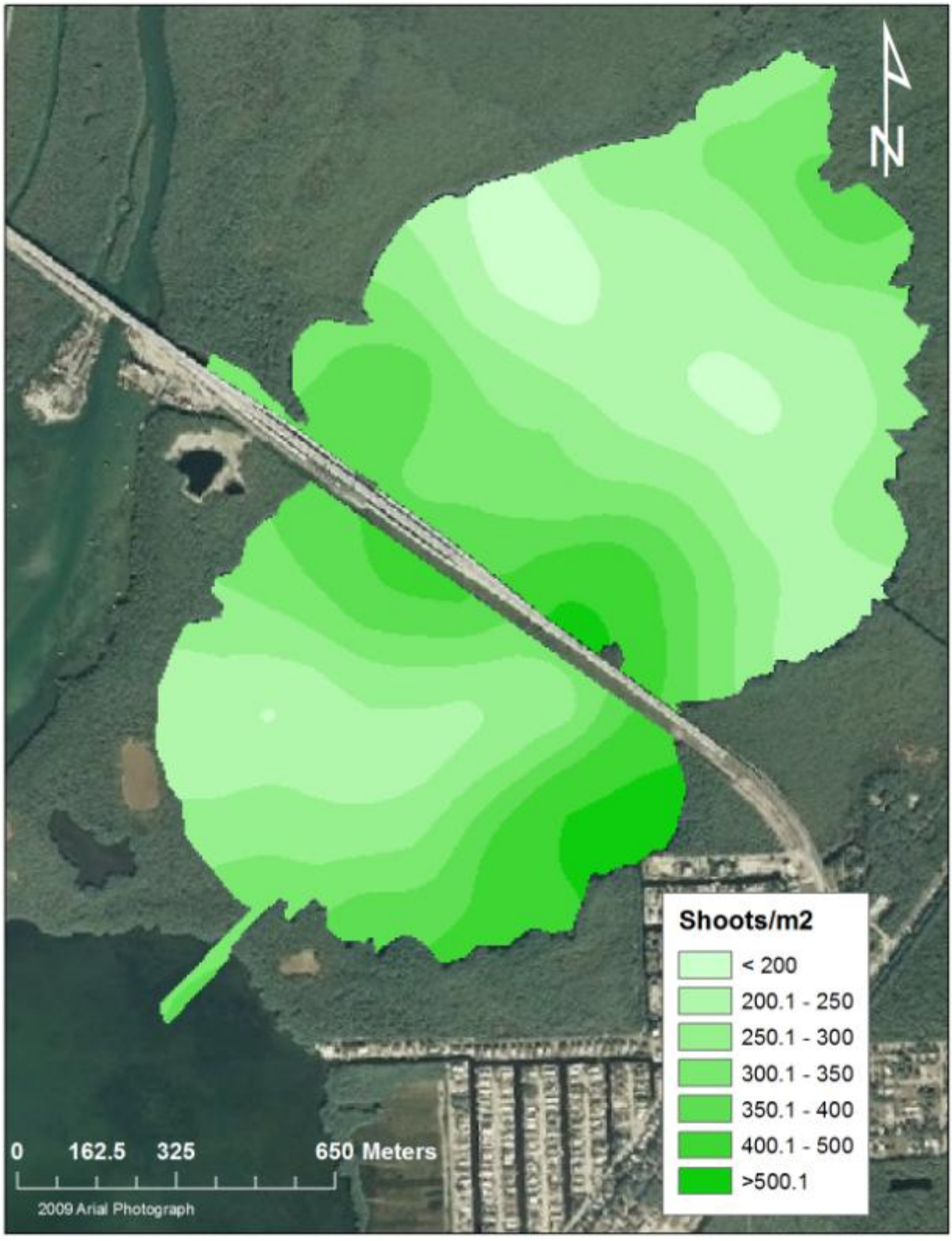

Figure 239. Seagrass shoot density (summer 2009).

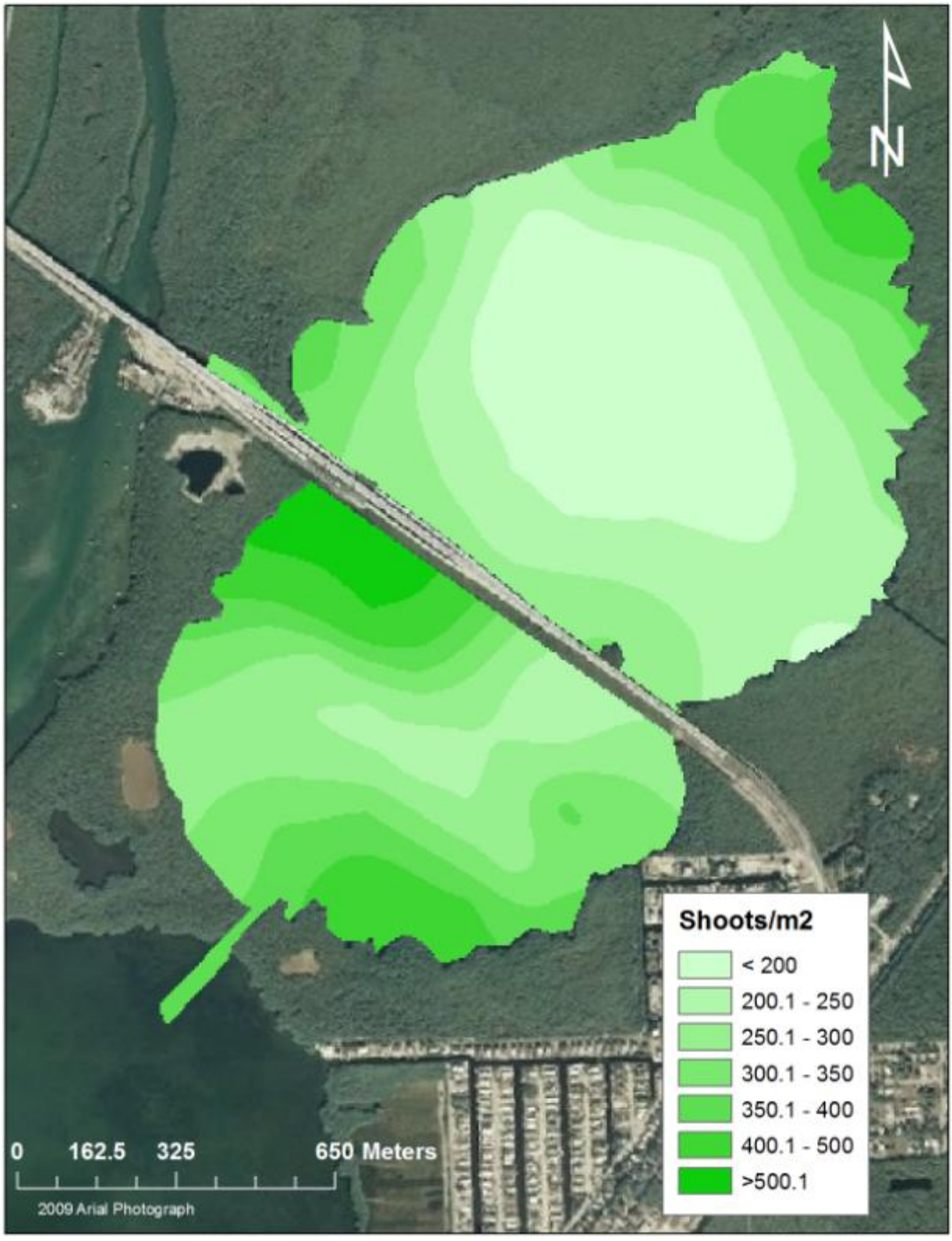

Figure 240. Seagrass shoot density (winter 2009).

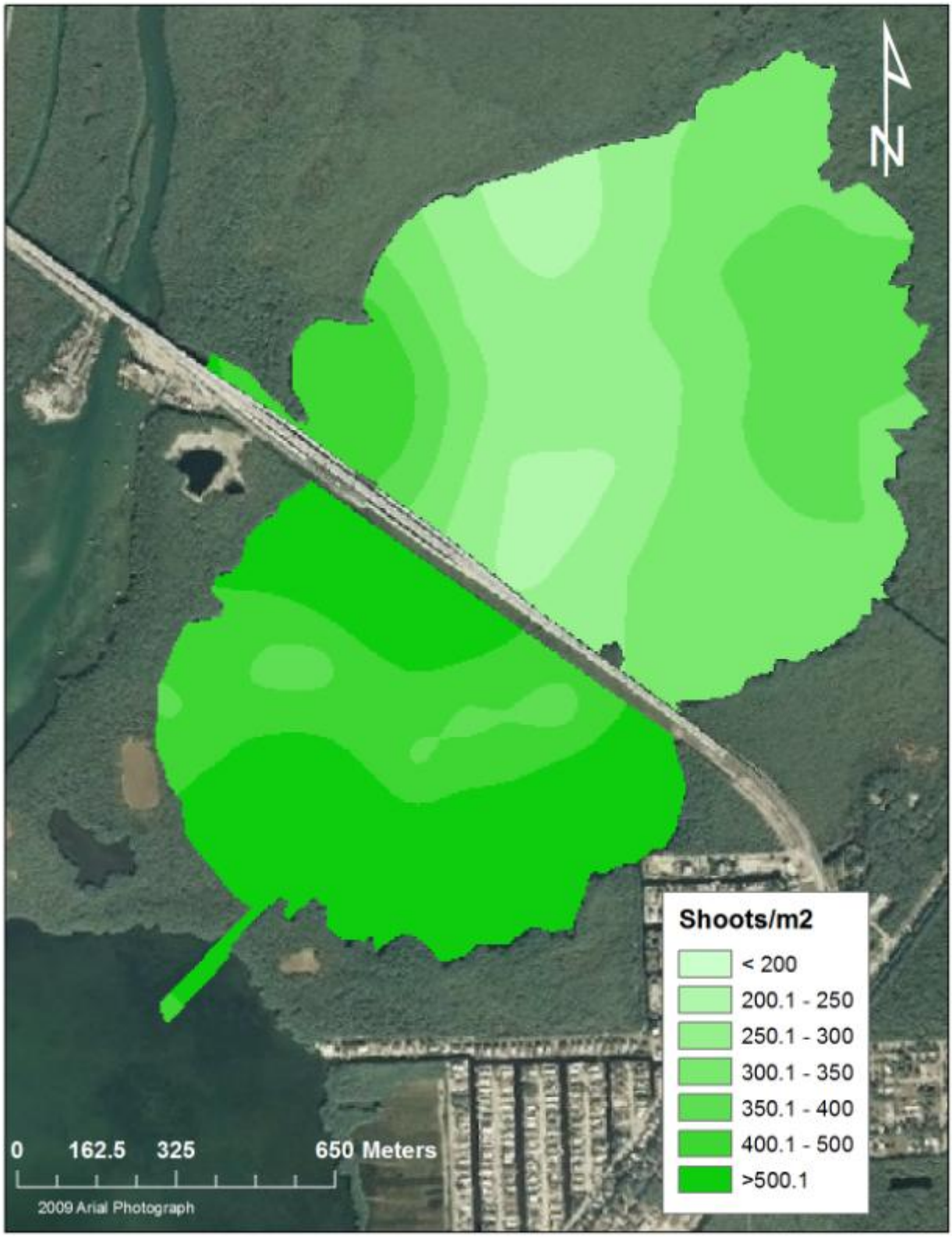
N
Shoots/m2
< 200
200.1 - 250
250.1 - 300
300.1 - 350
350.1 - 400
400.1 - 500
>500.1
0
162.5
325
650 Meters
2009 Arial Photograph

Figure 241. Seagrass shoot density (summer 2010).

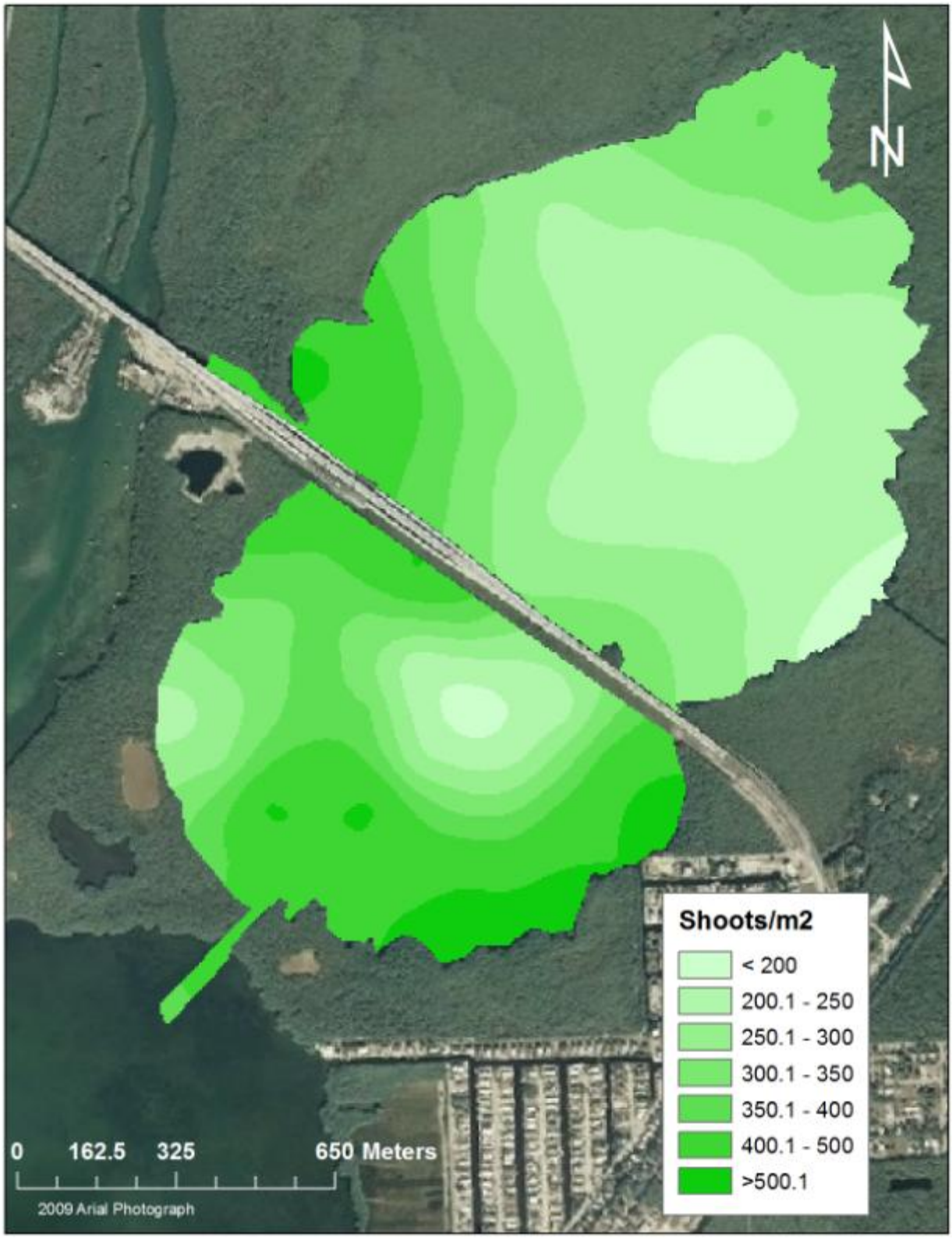

Figure 242. Seagrass shoot density (winter 2010).

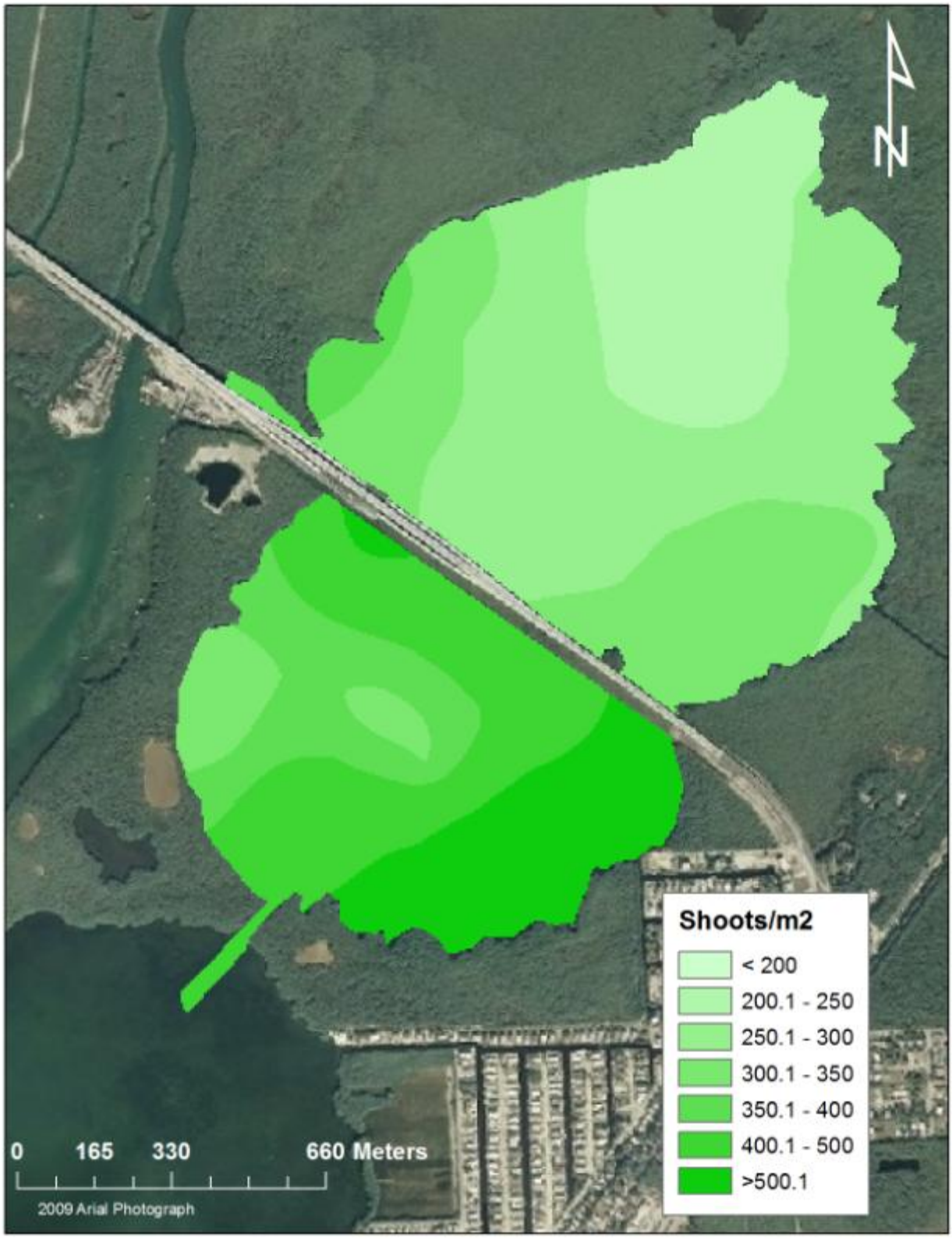

Figure 243. Seagrass shoot density (summer 2011).

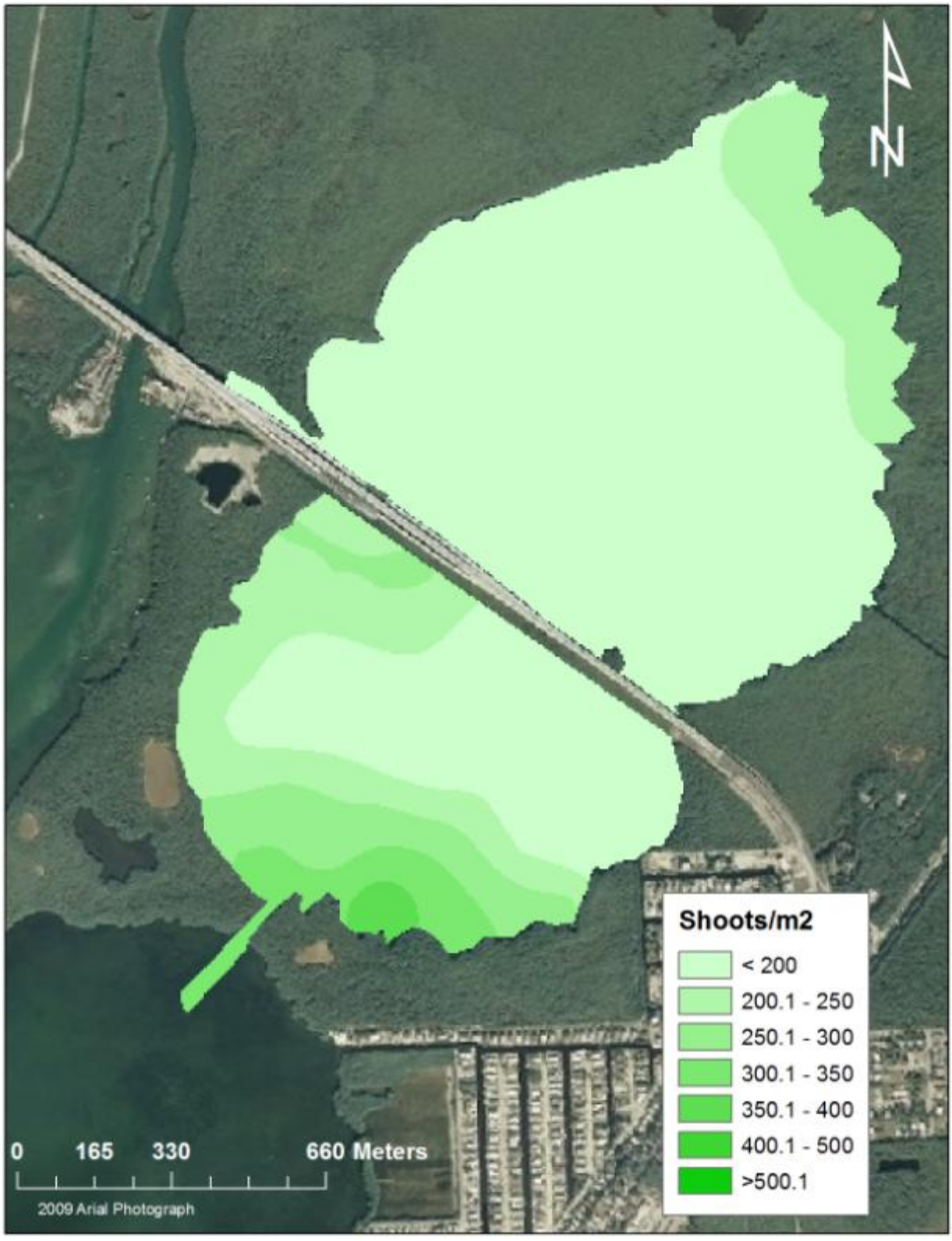
N
Shoots/m2
< 200
200.1 - 250
250.1 - 300
300.1 - 350
350.1 - 400
400.1 - 500
>500.1
0 165 330 660 Meters
2009 Arial Photograph

Figure 244. *Thalassia testudinum* shoot density (winter 2008).

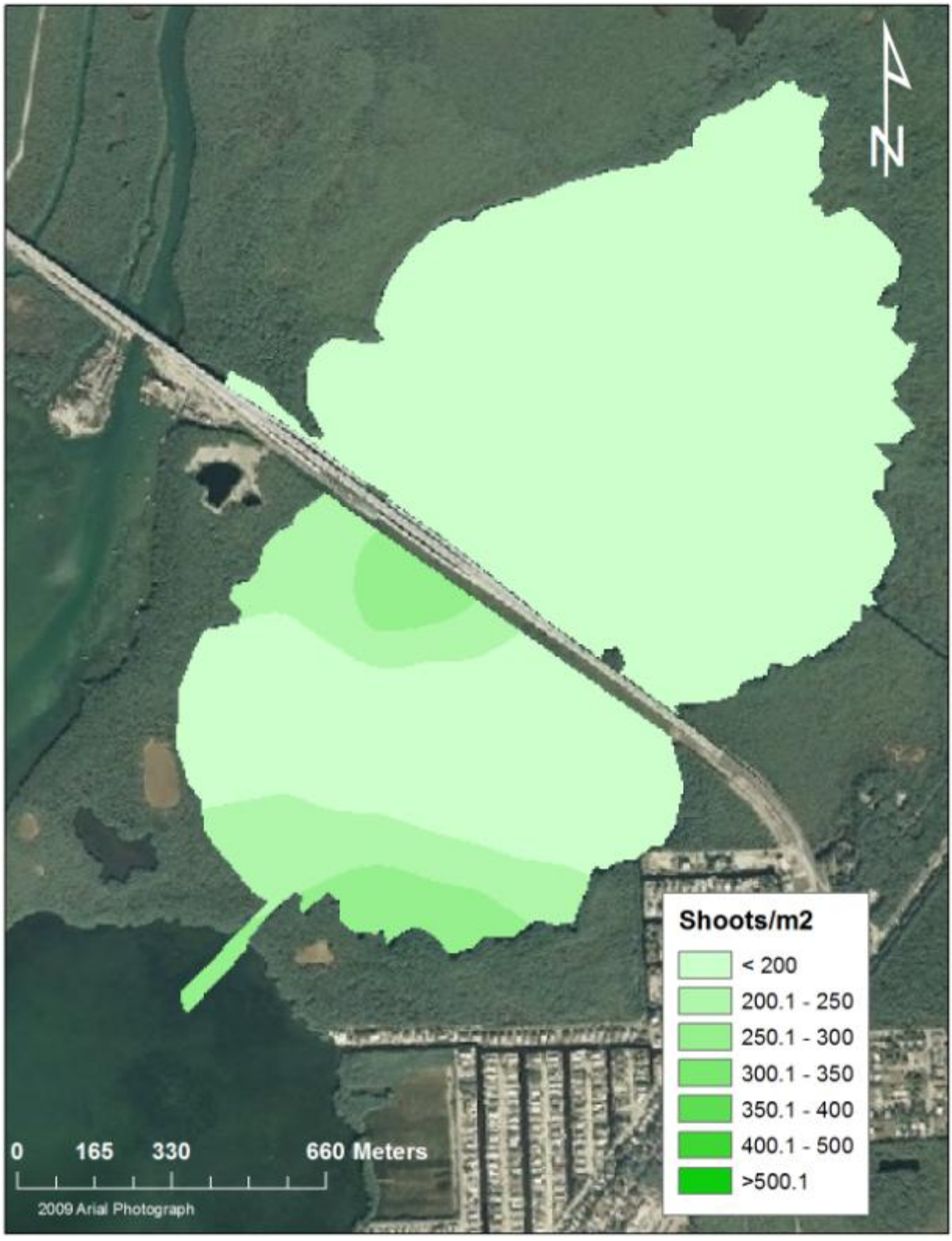

Figure 245. *Thalassia testudinum* shoot density (summer 2009).

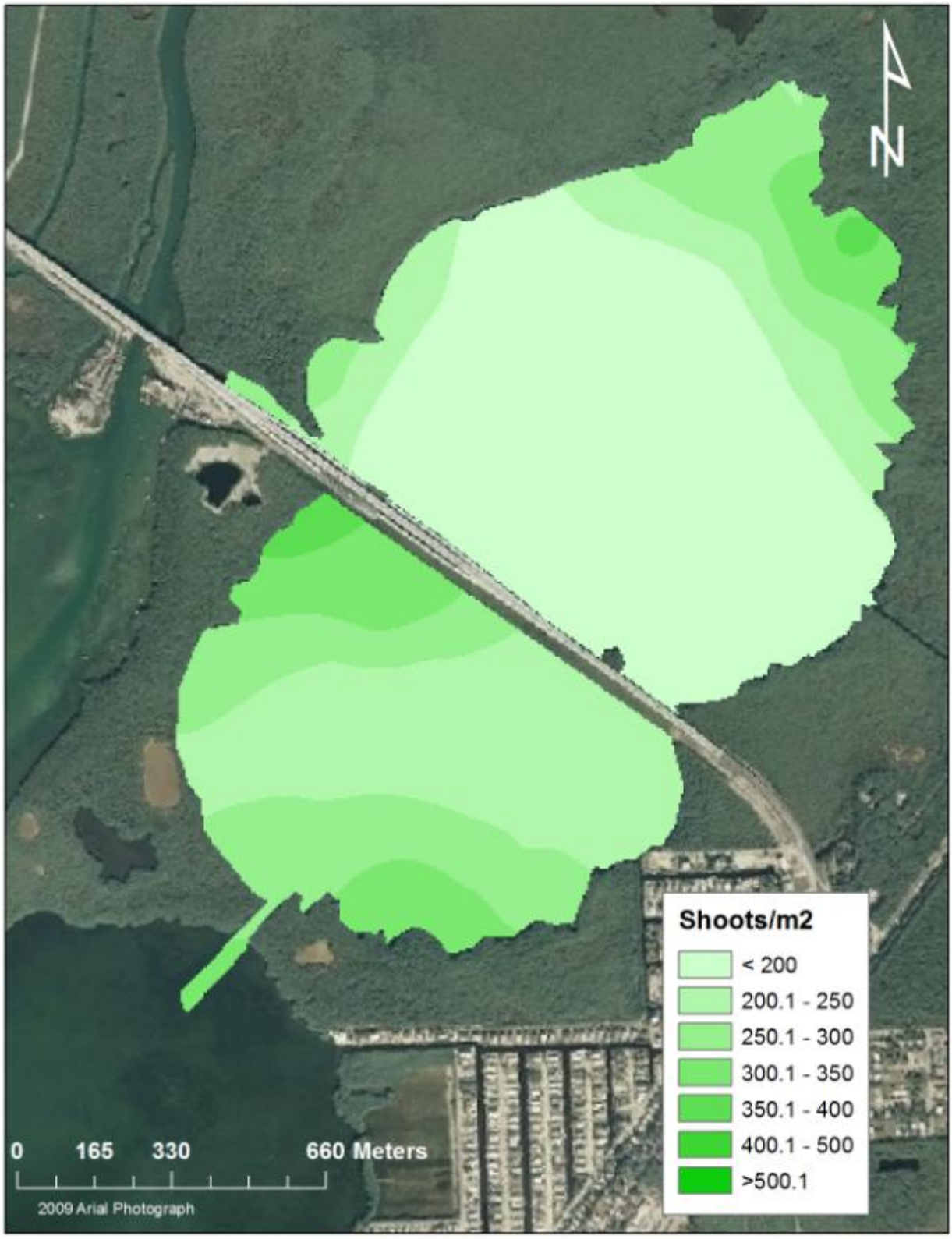

Figure 246. *Thalassia testudinum* shoot density (winter 2009).

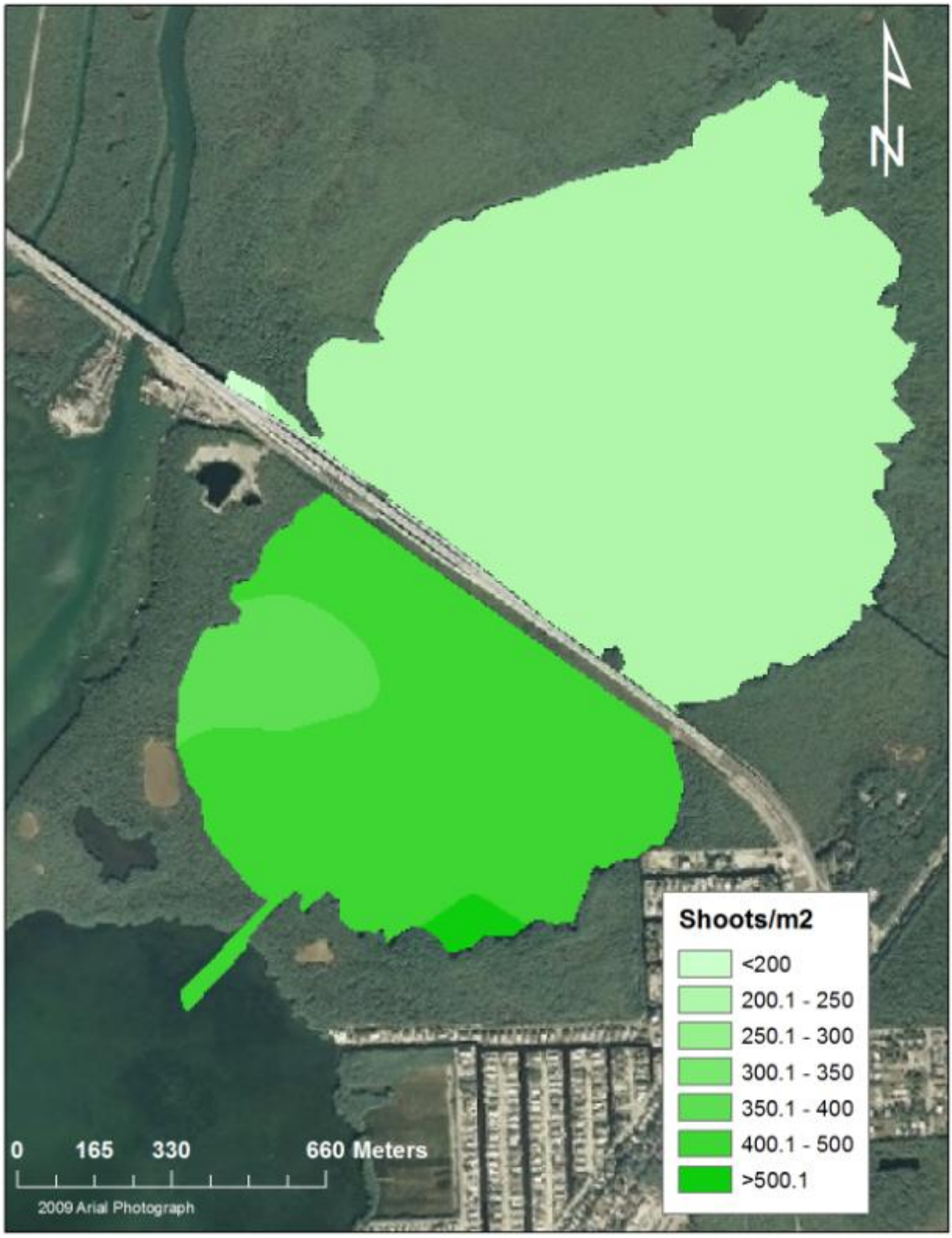

Figure 247. *Thalassia testudinum* shoot density (summer 2010).

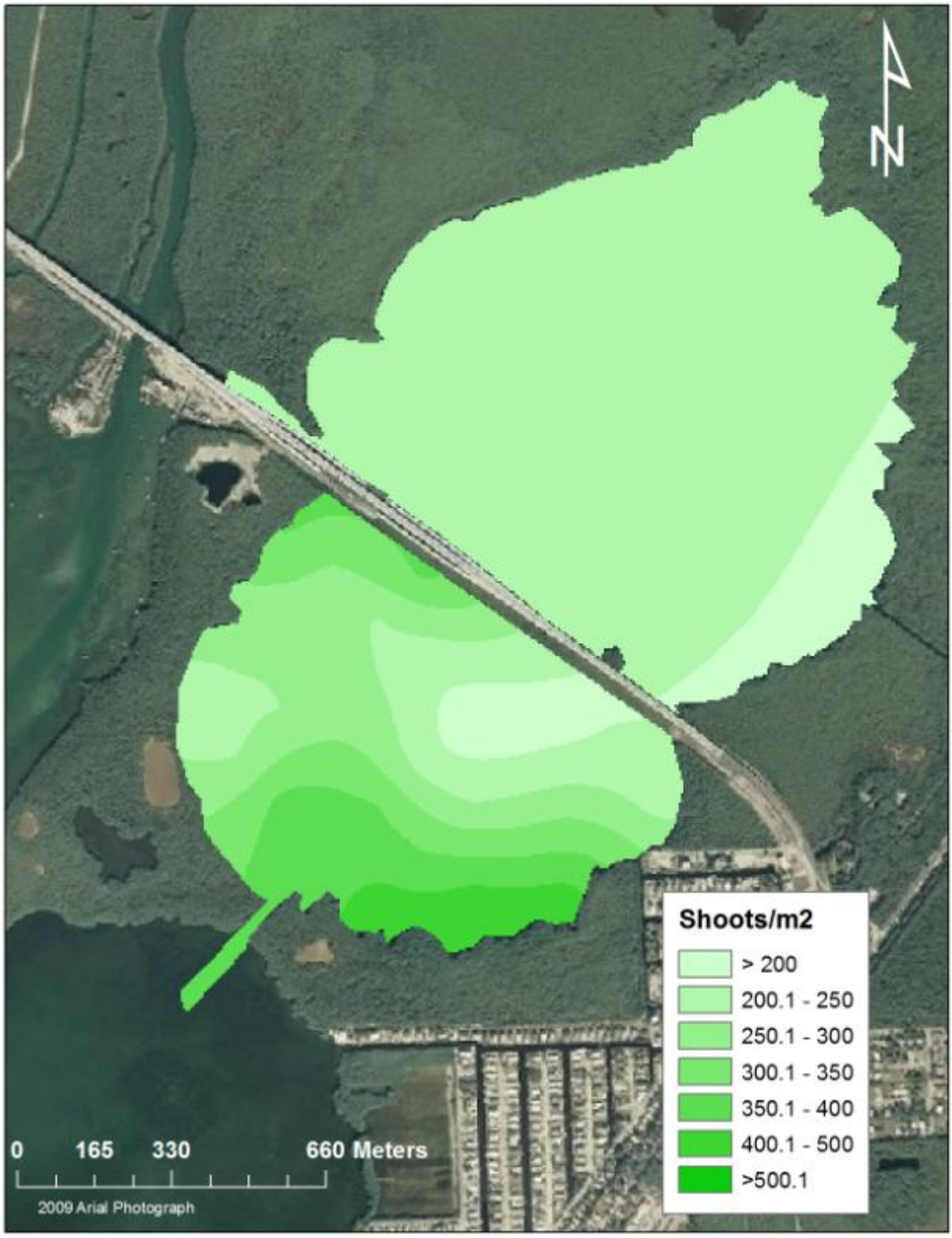

Figure 248. *Thalassia testudinum* shoot density (winter 2010).

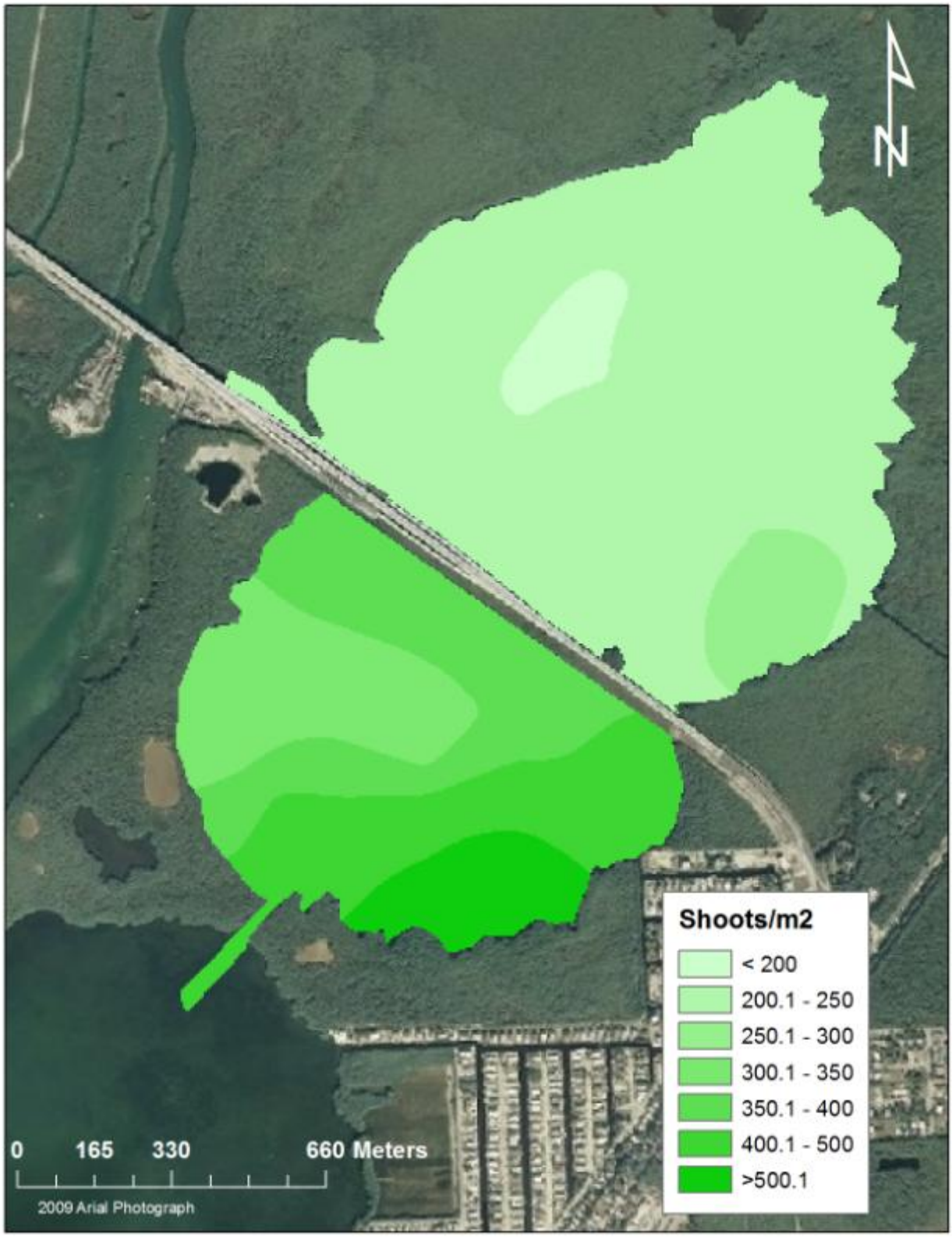

Figure 249. *Thalassia testudinum* shoot density (summer 2011).

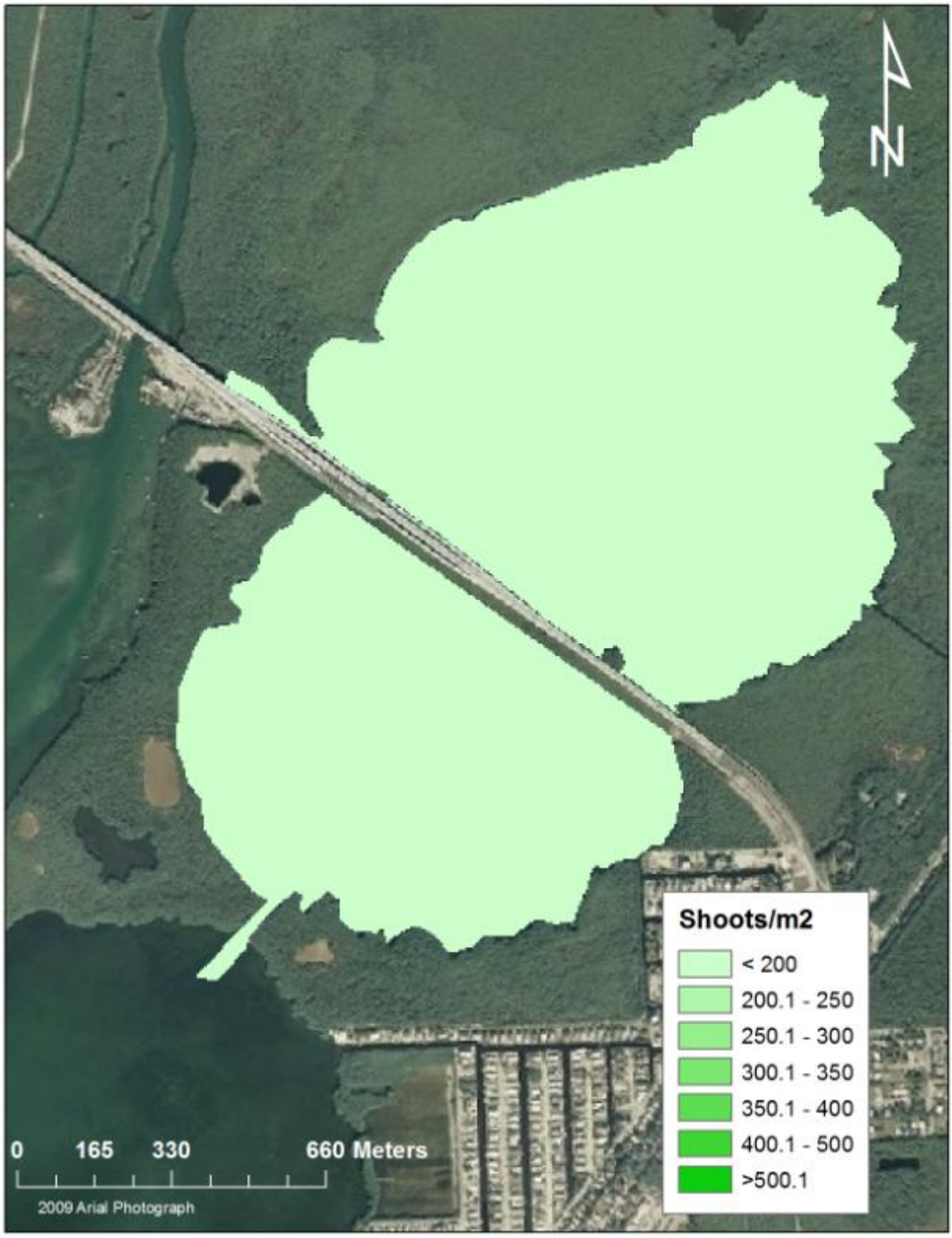

Figure 250. *Halodule wrightii* shoot density (winter 2008)

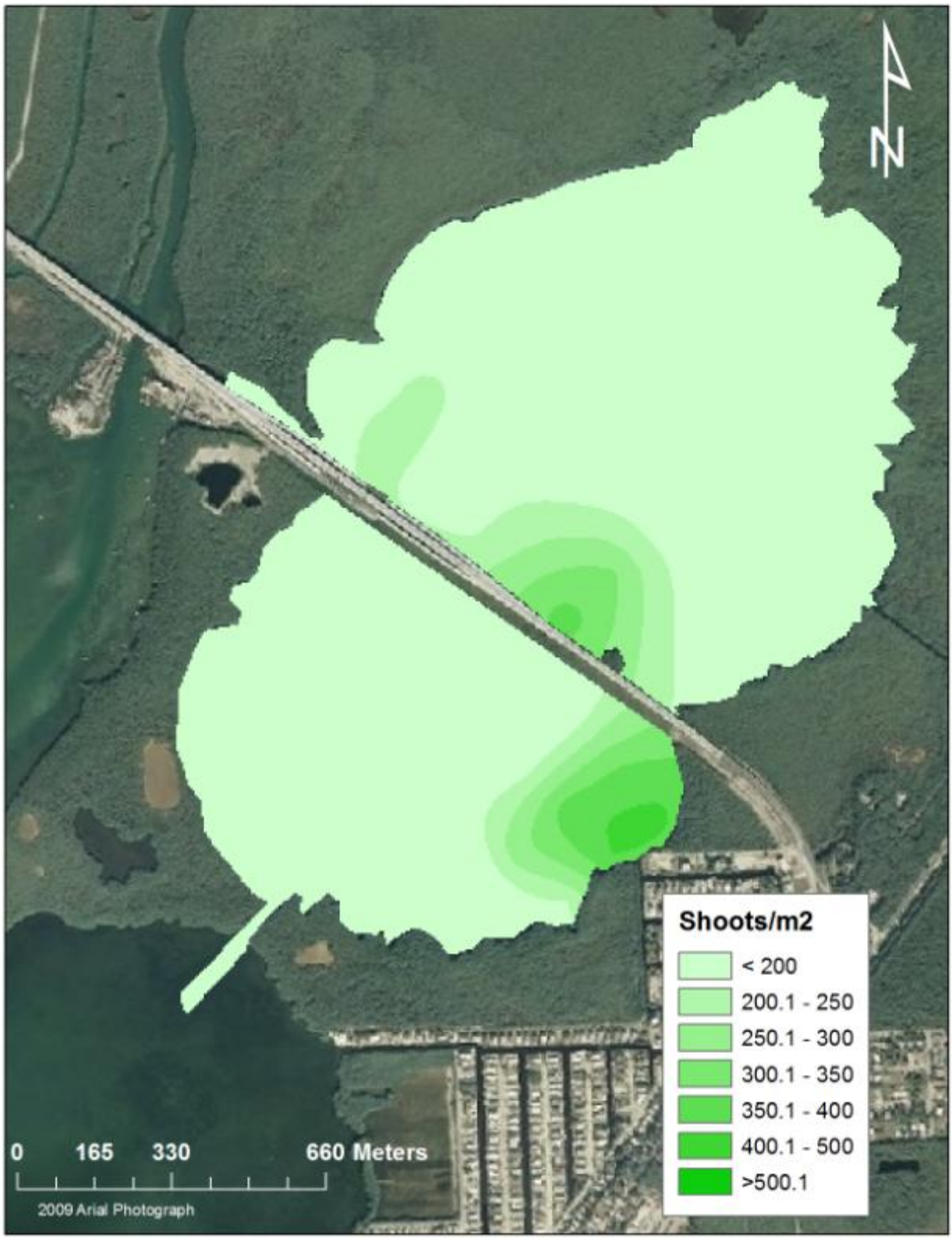

Figure 251. *Halodule wrightii* shoot density (summer 2009).

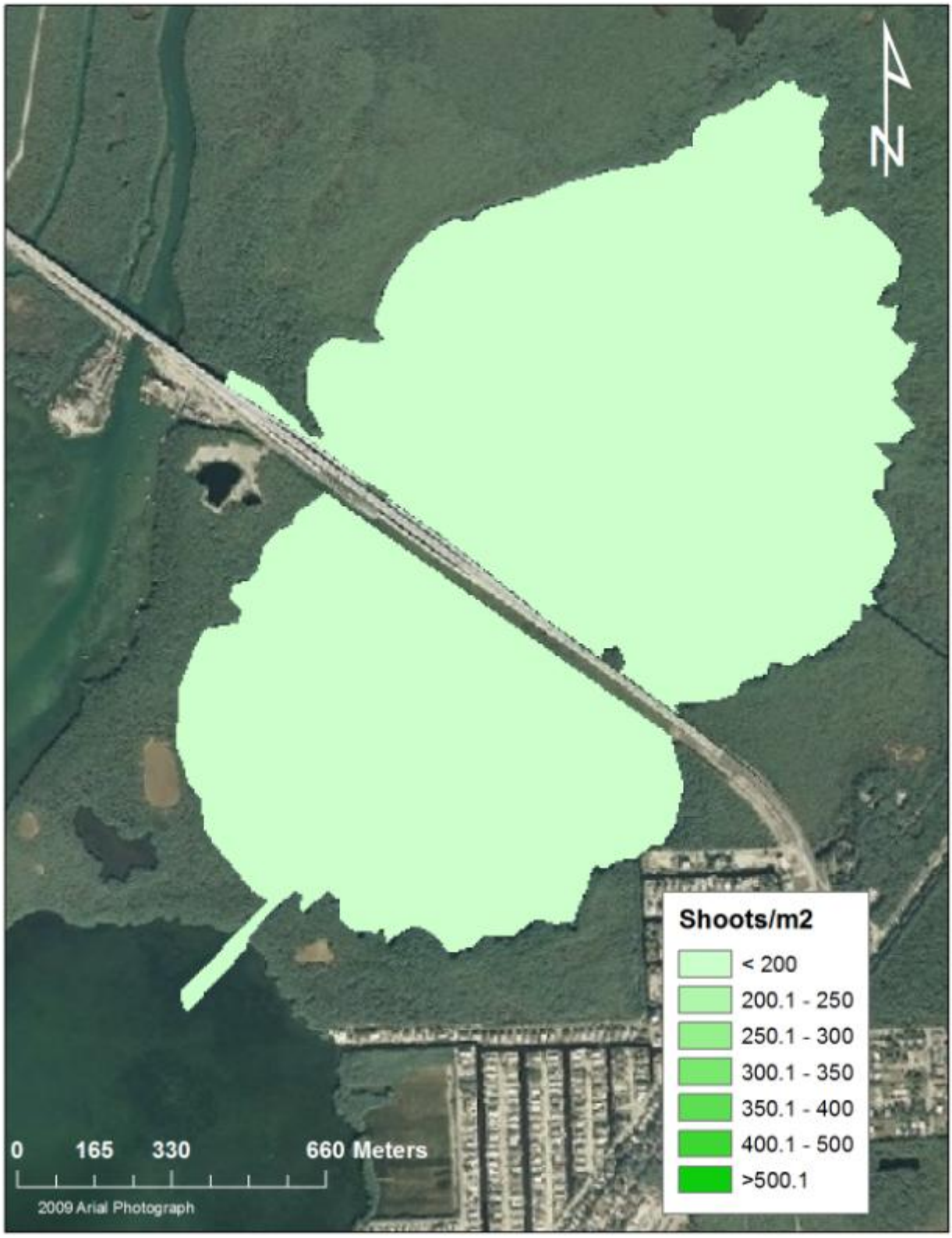

Figure 252. *Halodule wrightii* shoot density (winter 2009)

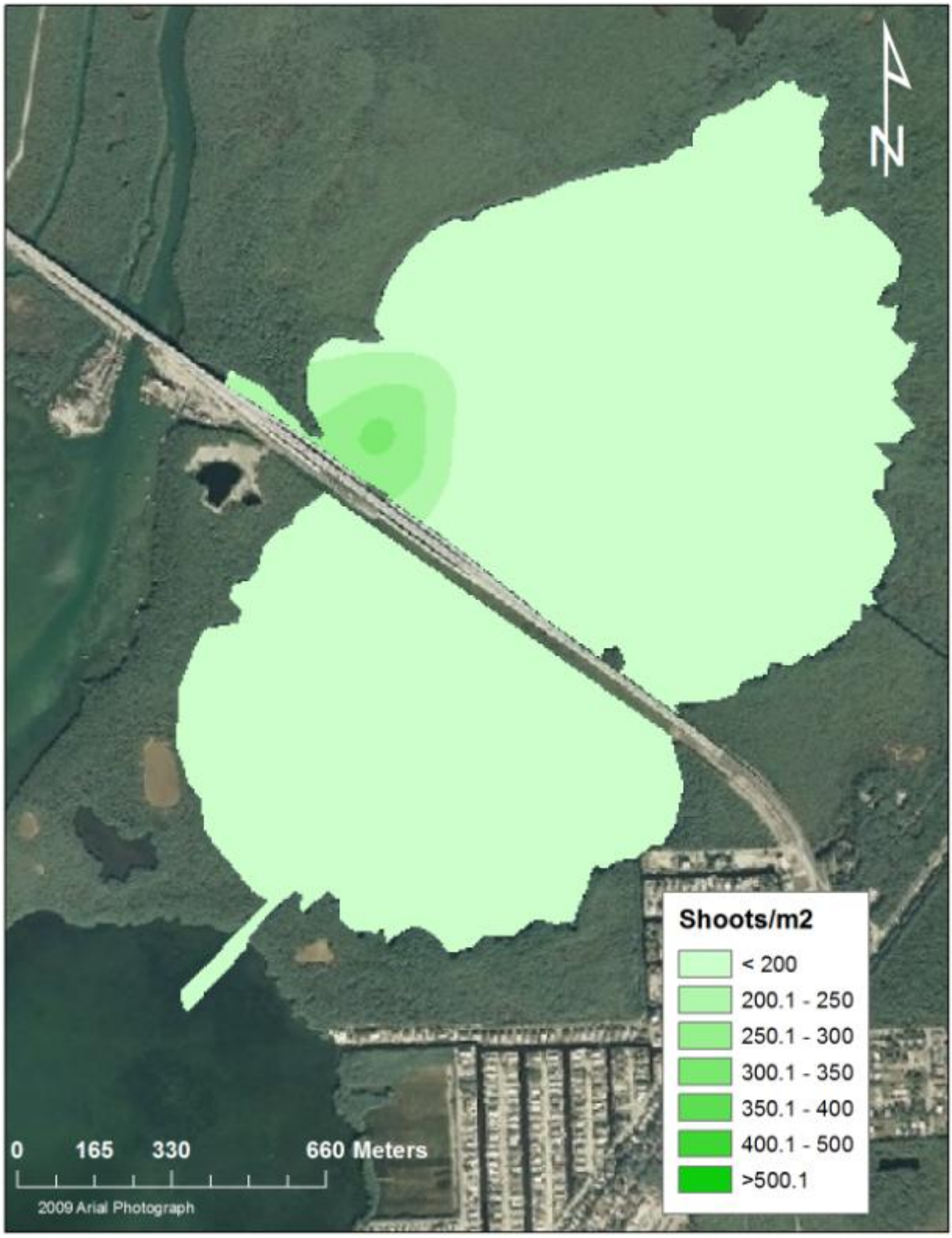

Figure 253. *Halodule wrightii* shoot density (summer 2010).

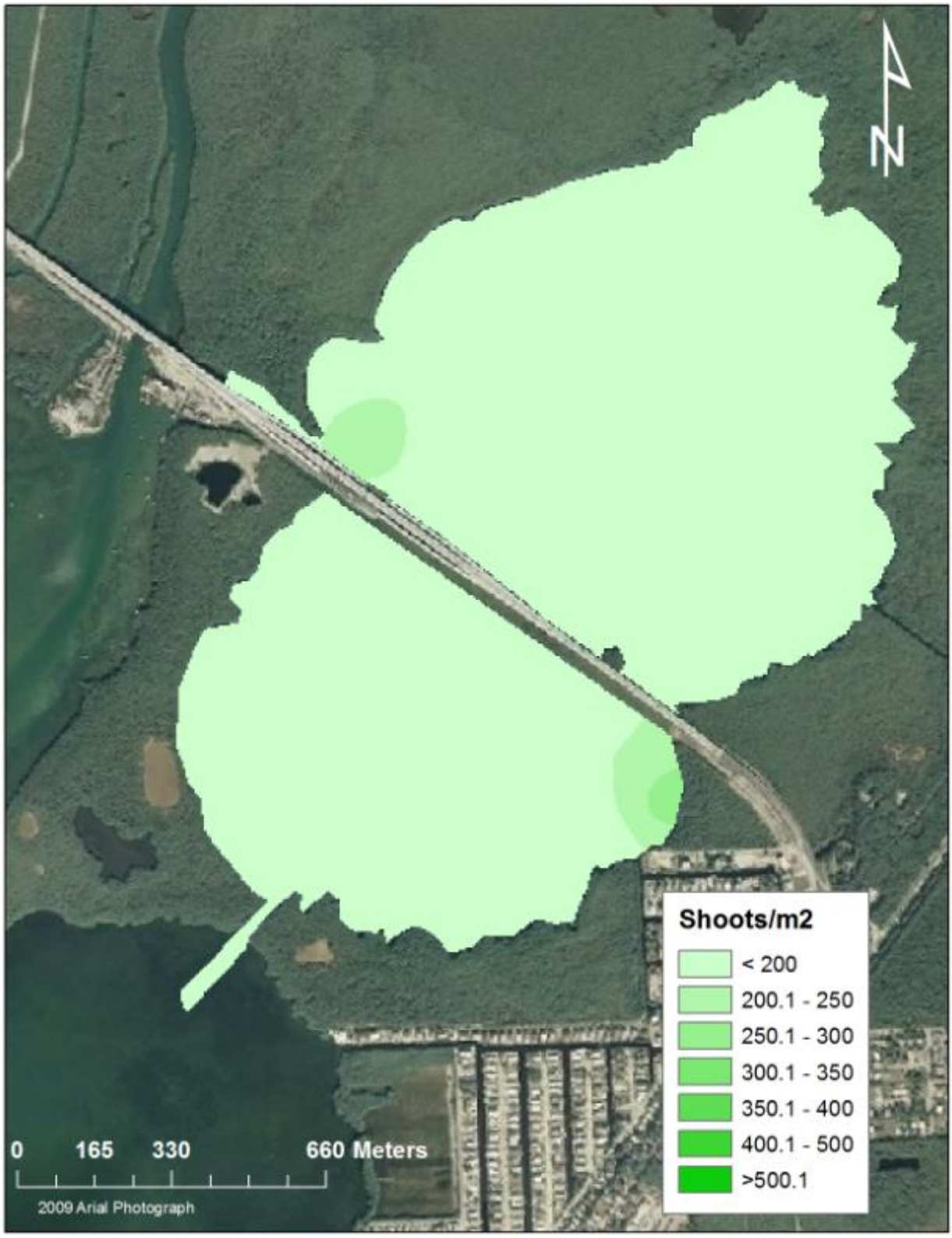

Figure 254. *Halodule wrightii* shoot density (winter 2010).

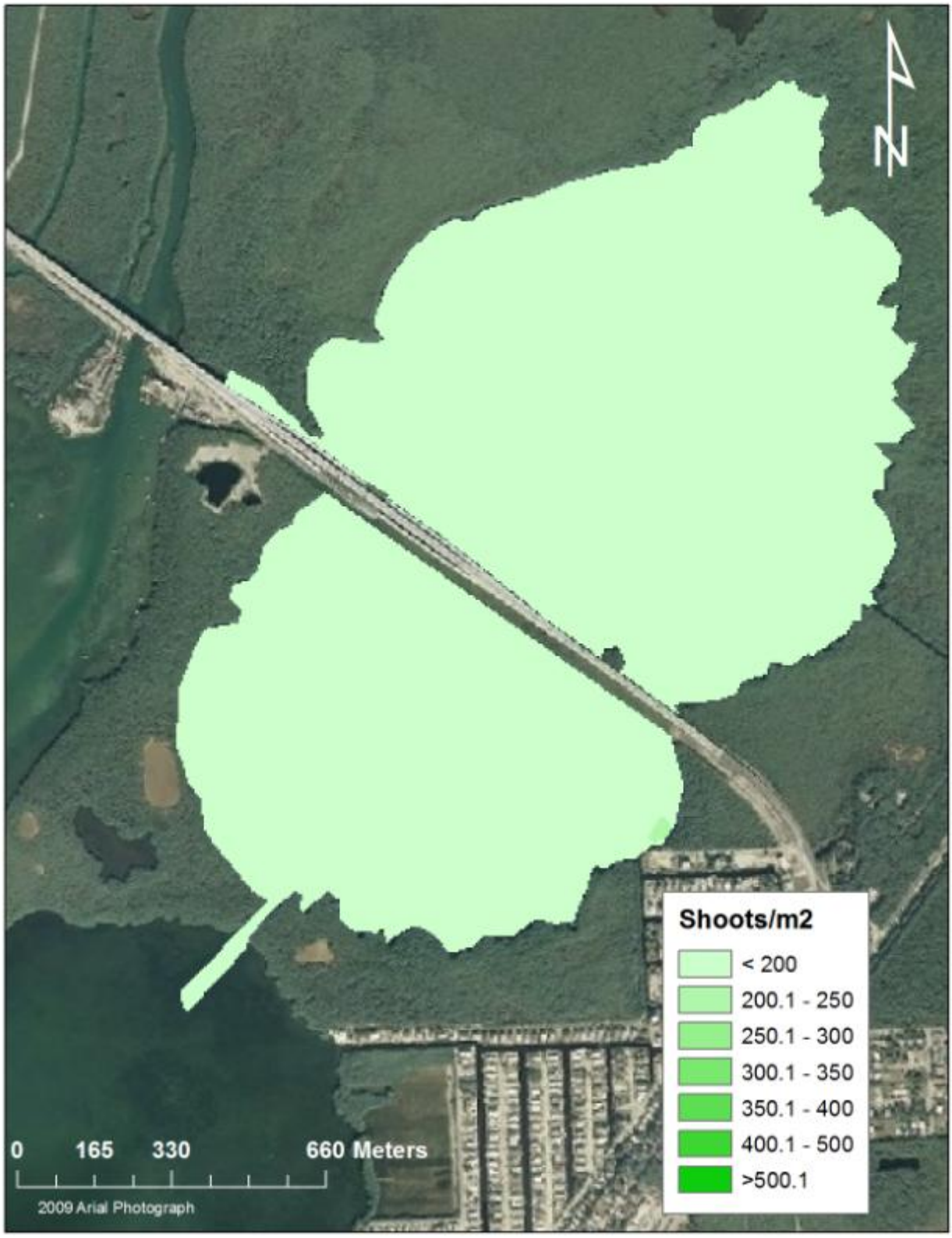

Figure 255. *Halodule wrightii* shoot density (summer 2011.).

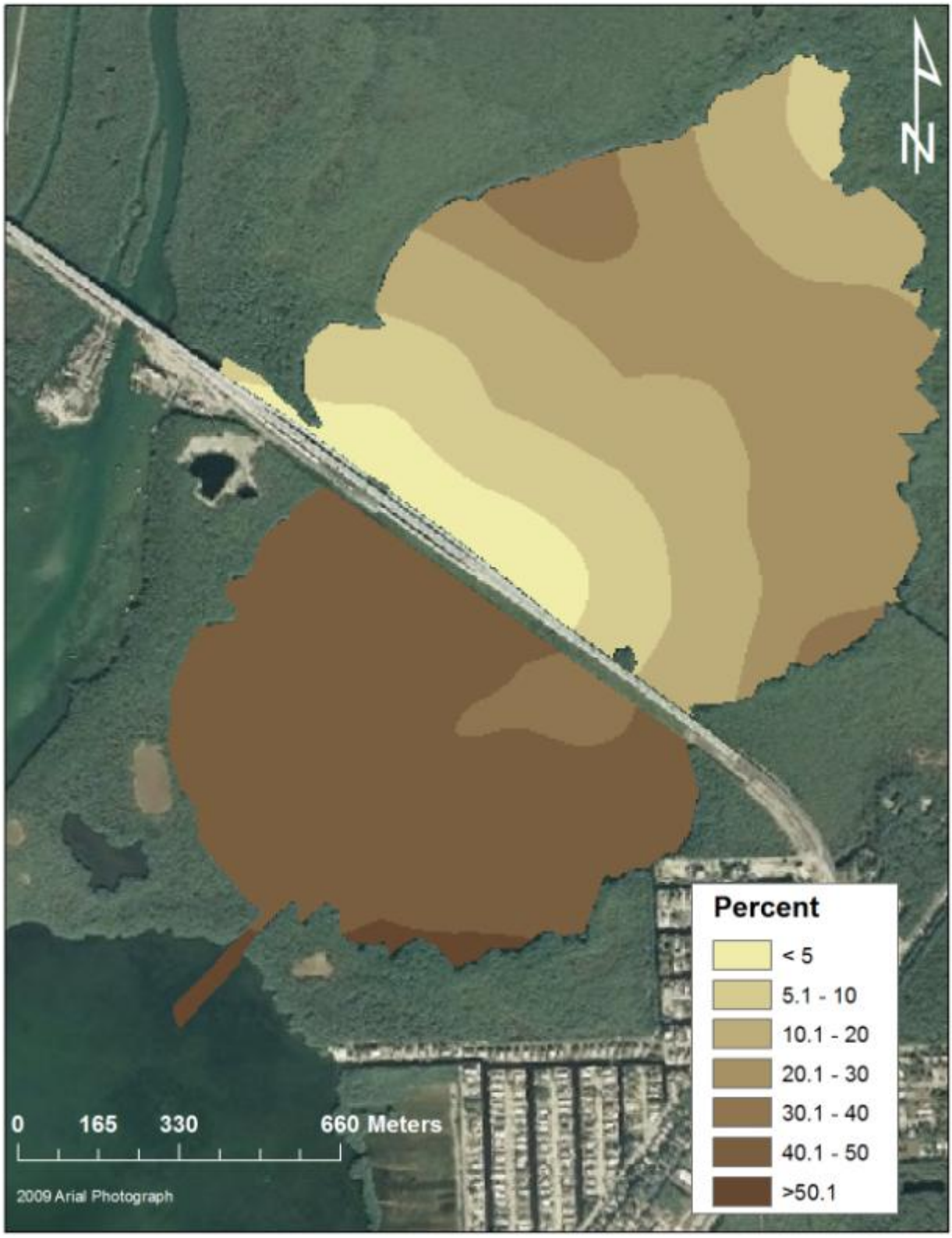

Figure 256. Algae percent cover (winter 2008).

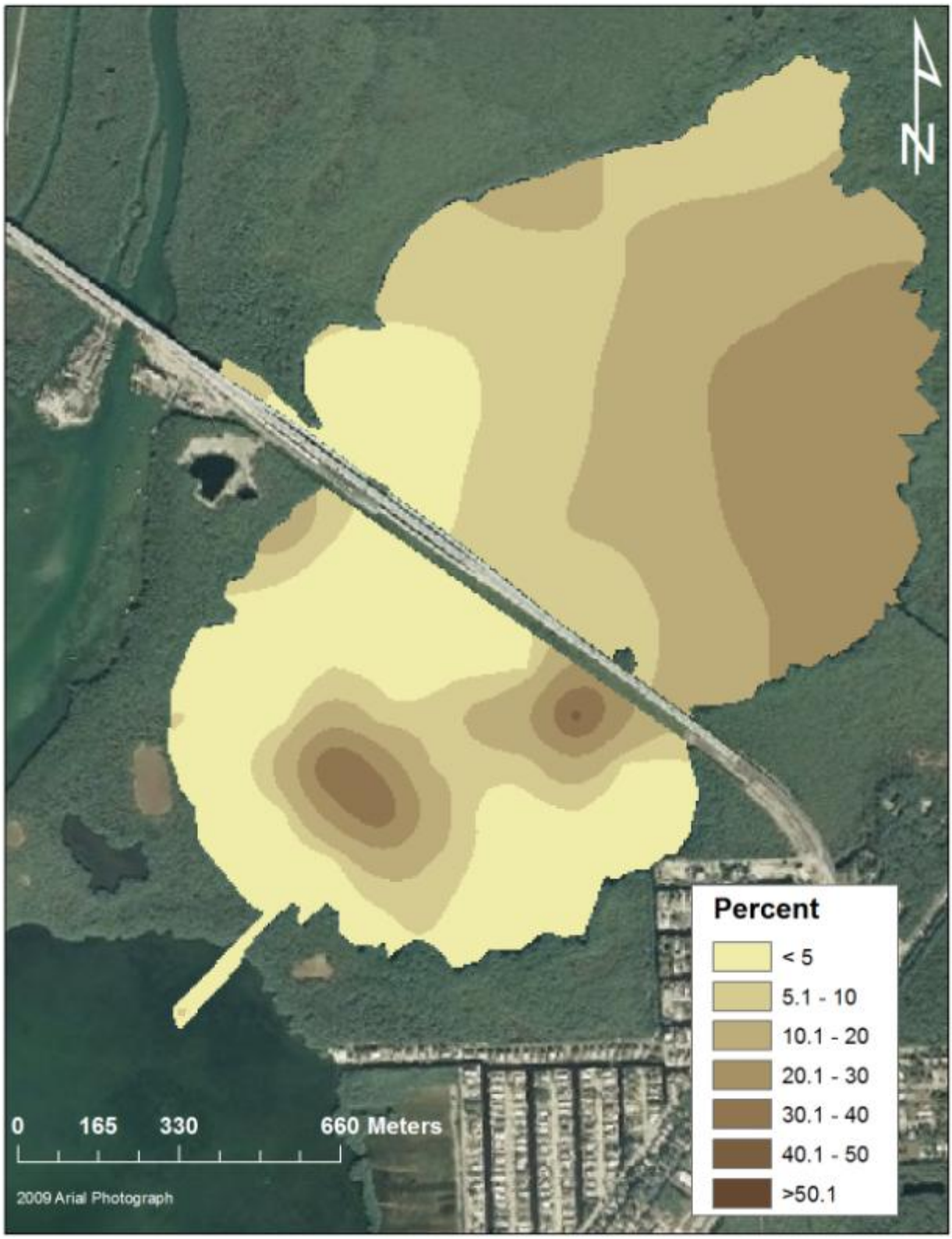

Figure 257. Algae percent cover (summer 2009).

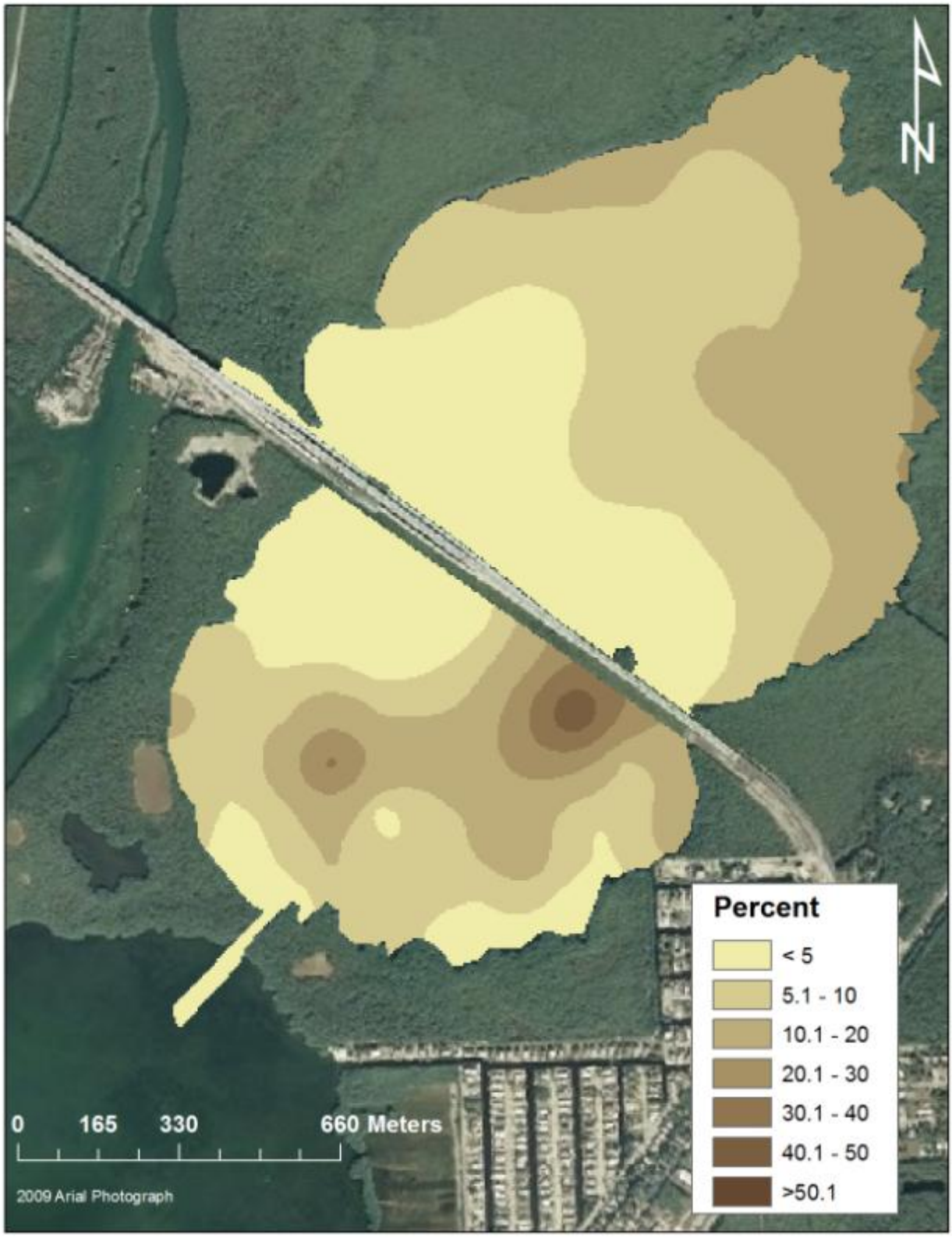
N
Percent
< 5
5.1 - 10
10.1 - 20
20.1 - 30
30.1 - 40
40.1 - 50
>50.1
0
165
330
660 Meters
2009 Arial Photograph

Figure 258. Algae percent cover (winter 2009).

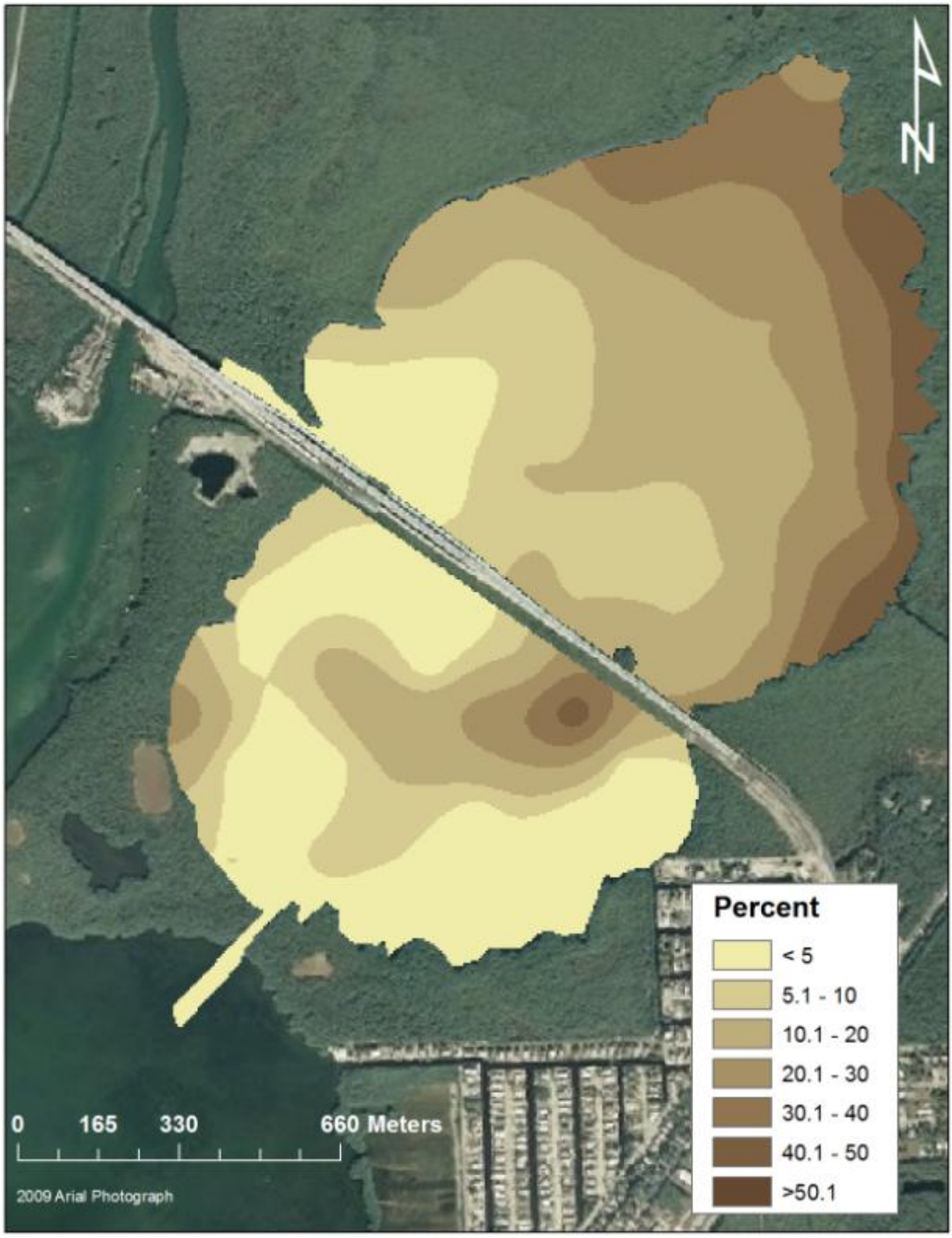
N
Percent
< 5
5.1 - 10
10.1 - 20
20.1 - 30
30.1 - 40
40.1 - 50
>50.1
0
165
330
660 Meters
2009 Arial Photograph

Figure 259. Algae percent cover (summer 2010).

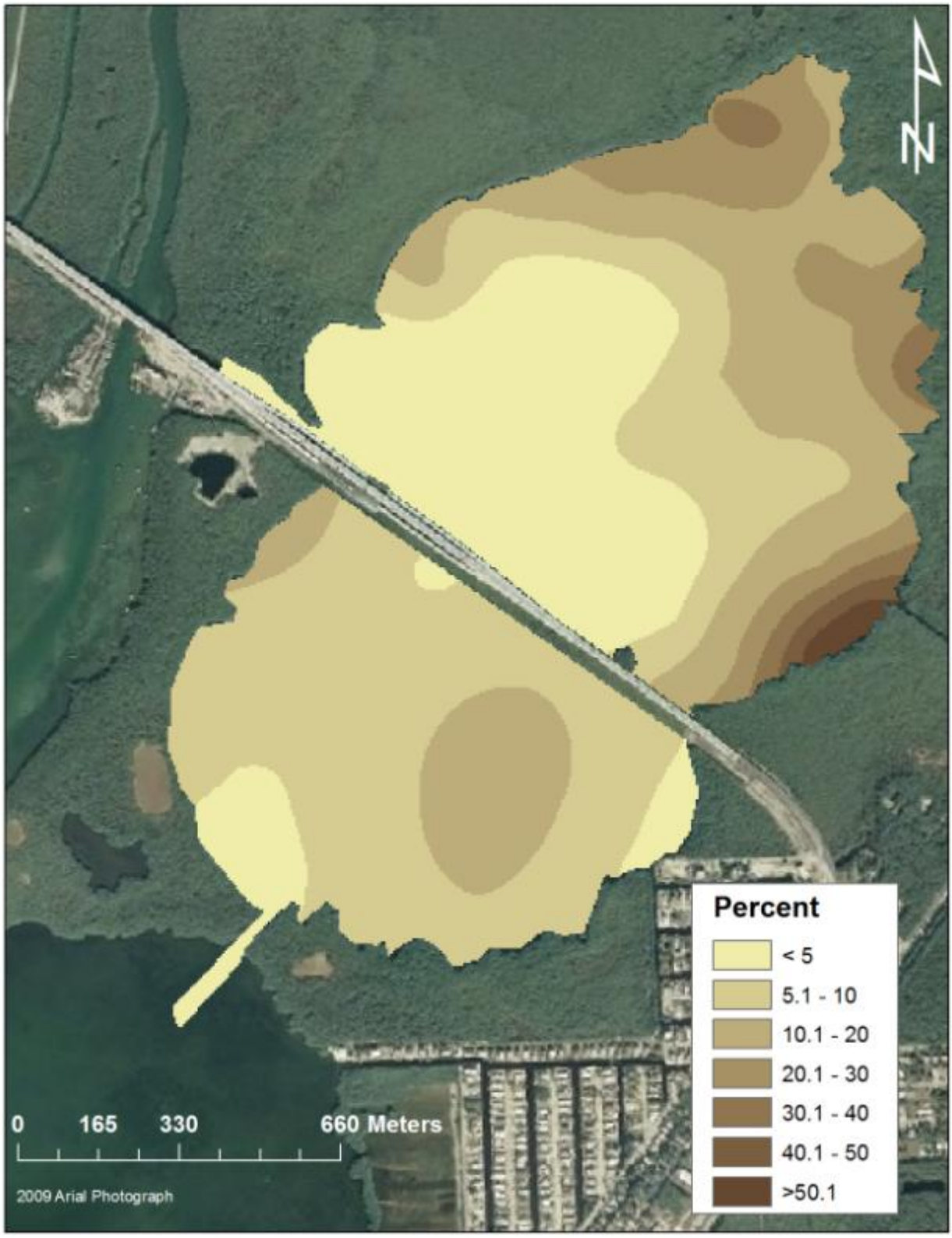
N
Percent
< 5
5.1 - 10
10.1 - 20
20.1 - 30
30.1 - 40
40.1 - 50
>50.1
0
165
330
660 Meters
2009 Arial Photograph

Figure 260. Algae percent cover (winter 2010).

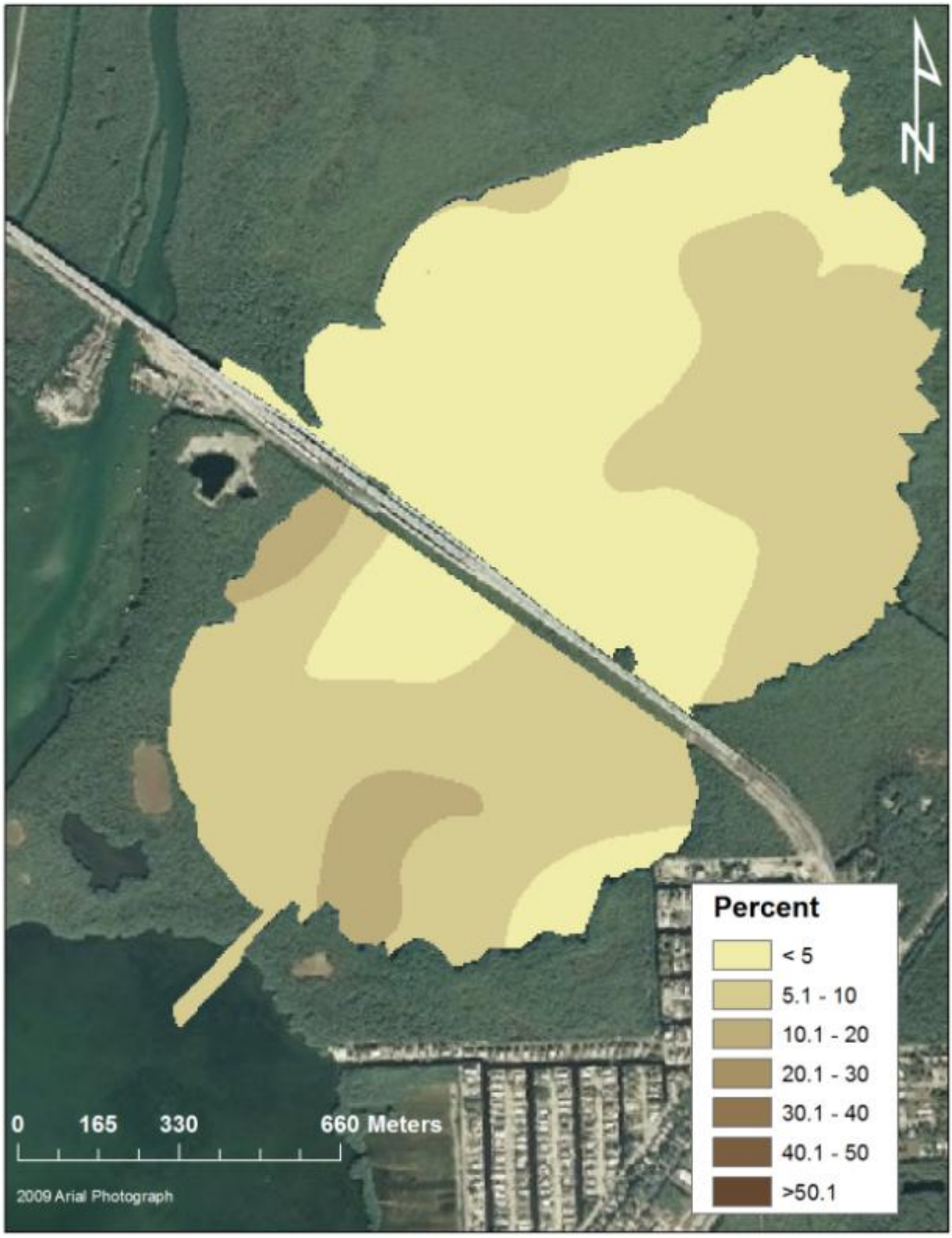
N
Percent
< 5
5.1 - 10
10.1 - 20
20.1 - 30
30.1 - 40
40.1 - 50
>50.1
0
165
330
660 Meters
2009 Arial Photograph

Figure 261. Algae percent cover (winter 2011).

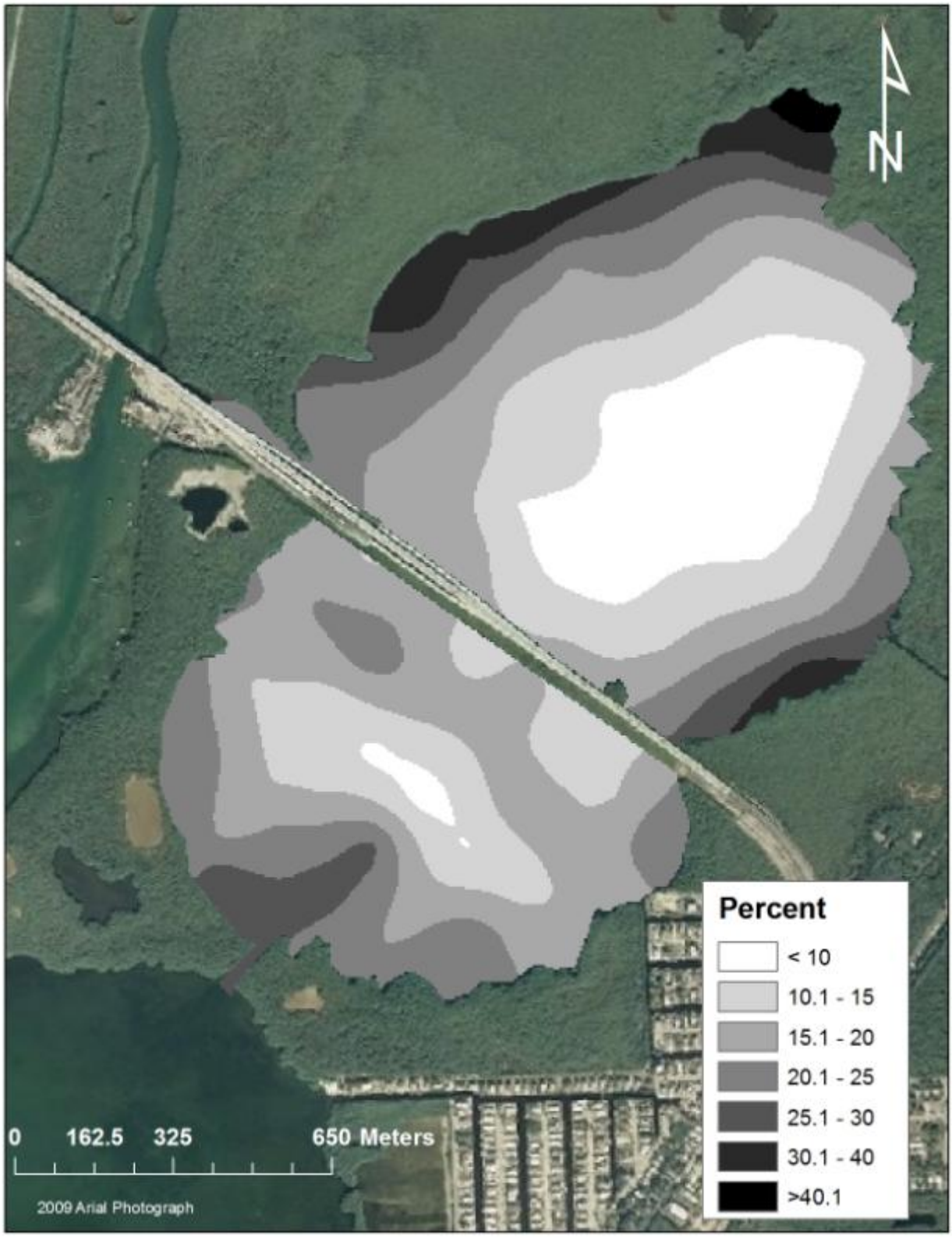
N
Percent
< 10
10.1 - 15
15.1 - 20
20.1 - 25
25.1 - 30
30.1 - 40
>40.1
0
162.5
325
650 Meters
2009 Arial Photograph

Figure 262. Soil organic matter (winter 2008).

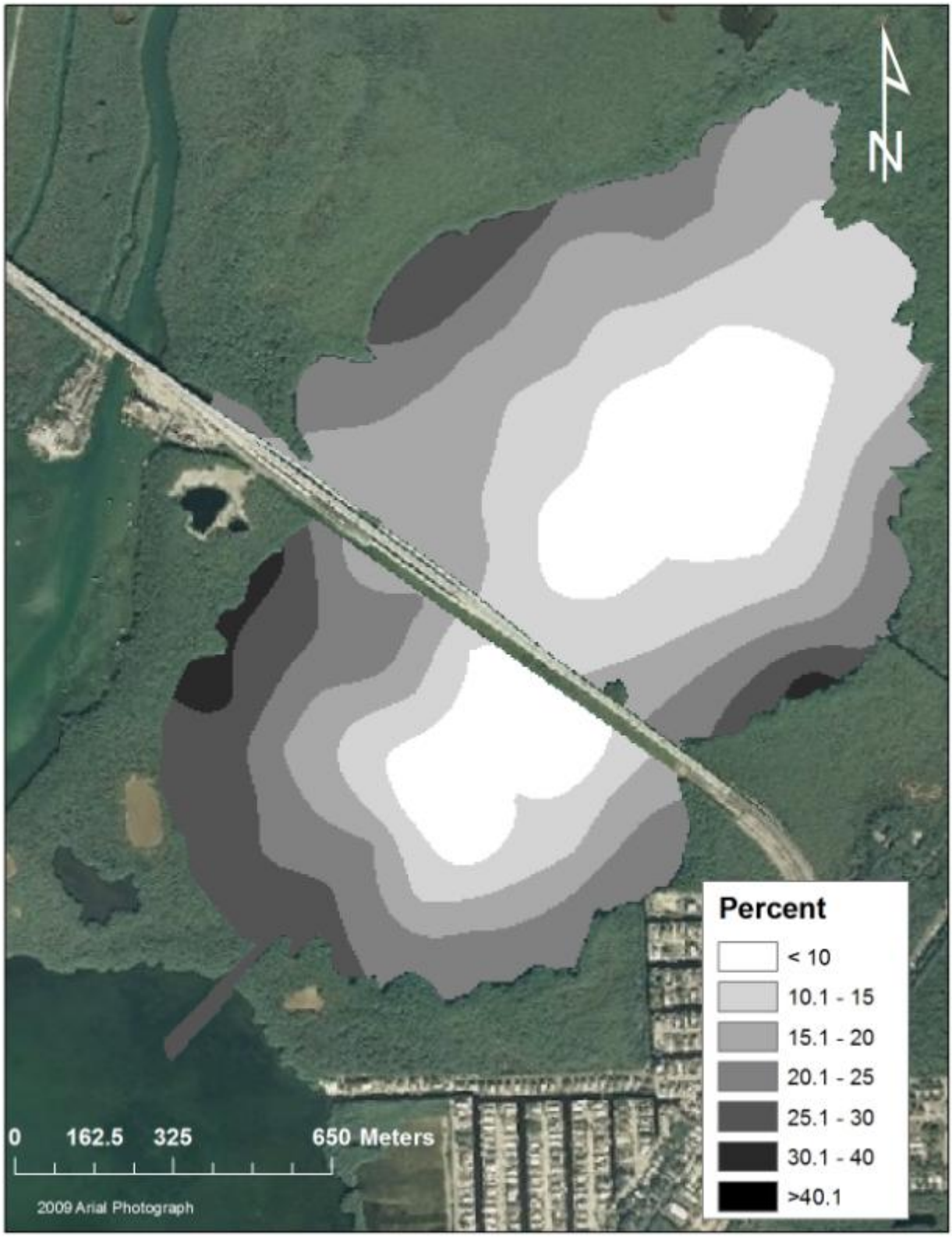
N
Percent
< 10
10.1 - 15
15.1 - 20
20.1 - 25
25.1 - 30
30.1 - 40
>40.1
0
162.5
325
650 Meters
2009 Arial Photograph

Figure 263. Soil organic matter (summer 2009).

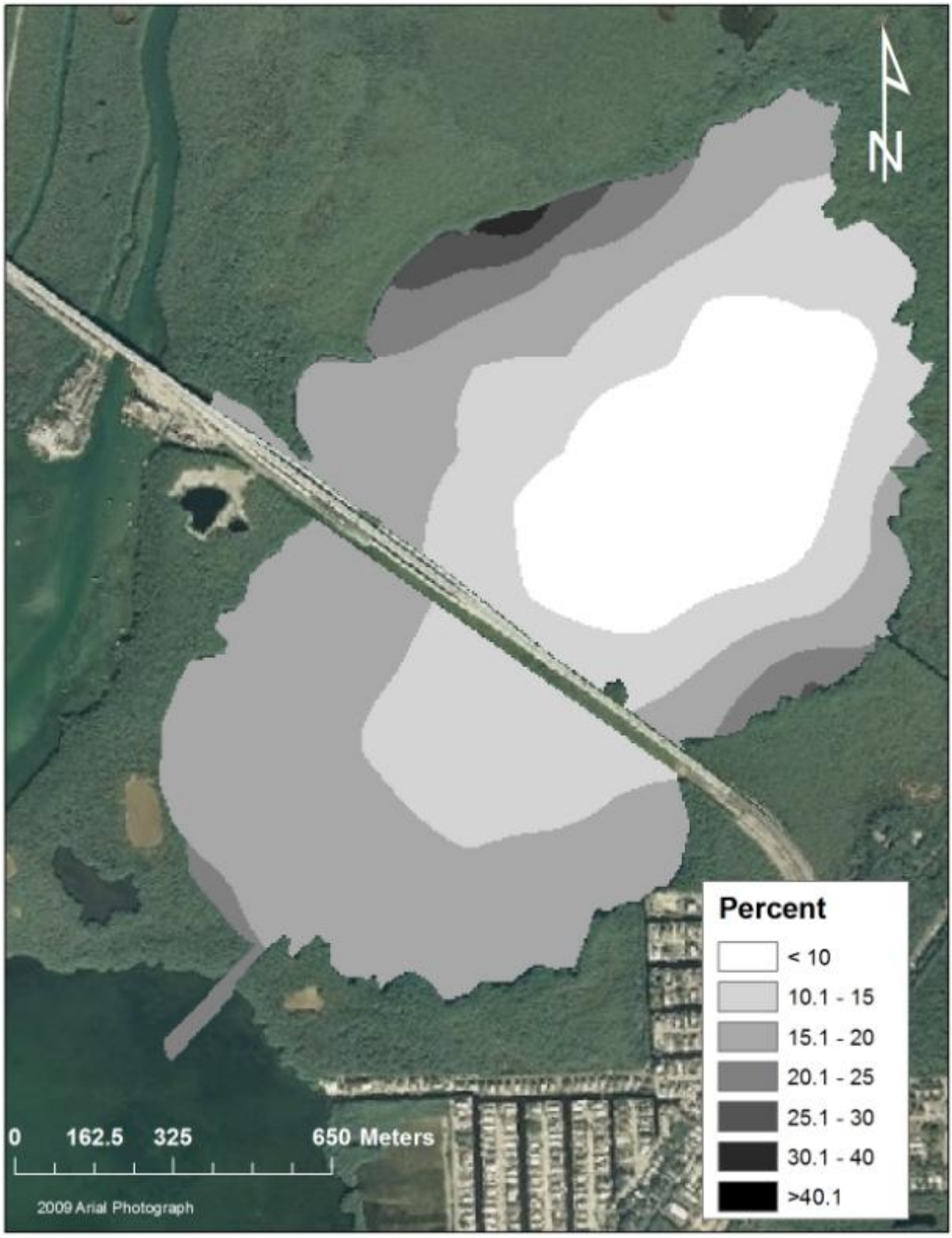
N
Percent
< 10
10.1 - 15
15.1 - 20
20.1 - 25
25.1 - 30
30.1 - 40
>40.1
0
162.5
325
650 Meters
2009 Arial Photograph

Figure 264. Soil organic matter (winter 2009).

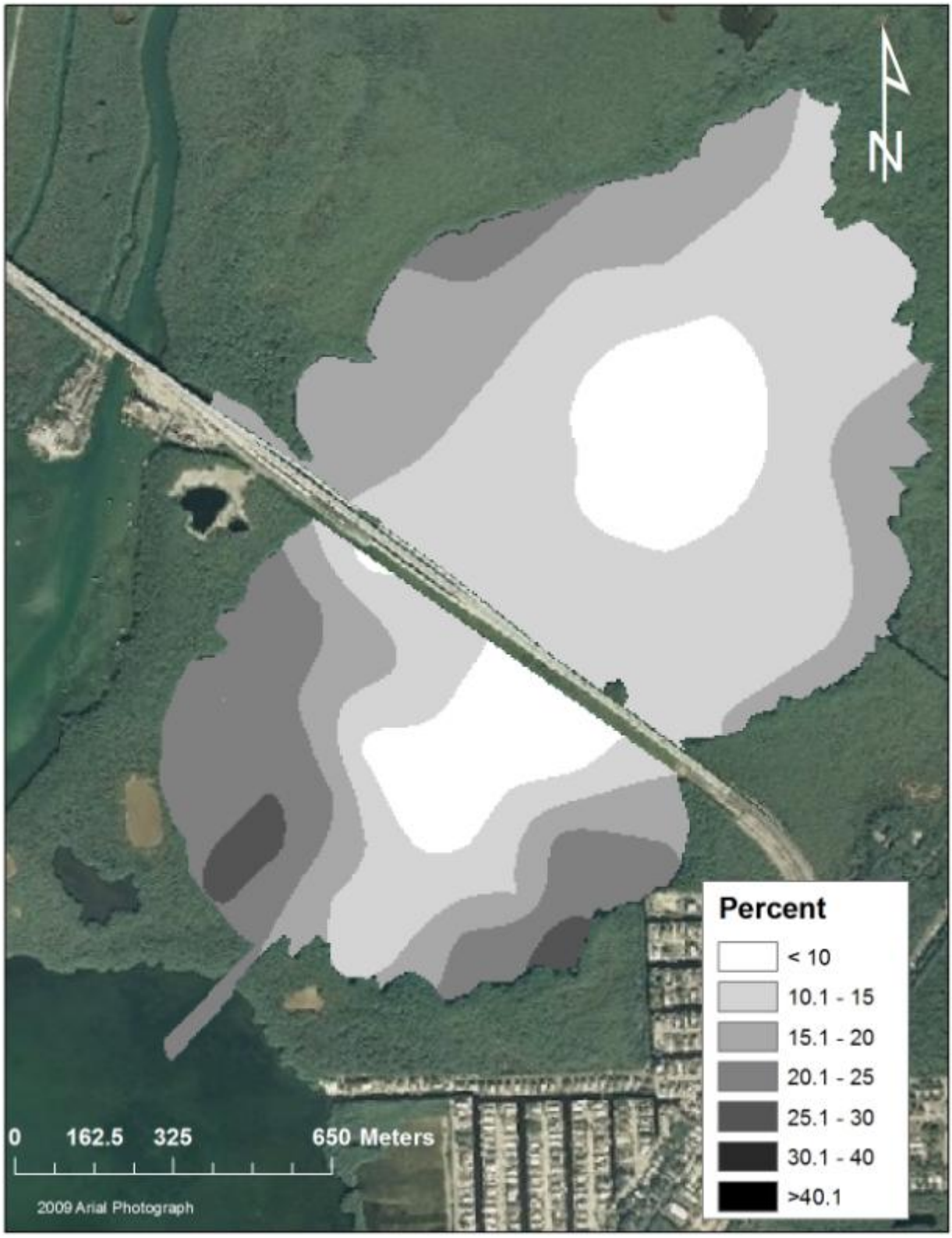
N
Percent
< 10
10.1 - 15
15.1 - 20
20.1 - 25
25.1 - 30
30.1 - 40
>40.1
0
162.5
325
650 Meters
2009 Arial Photograph

Figure 265. Soil organic matter (summer 2010).

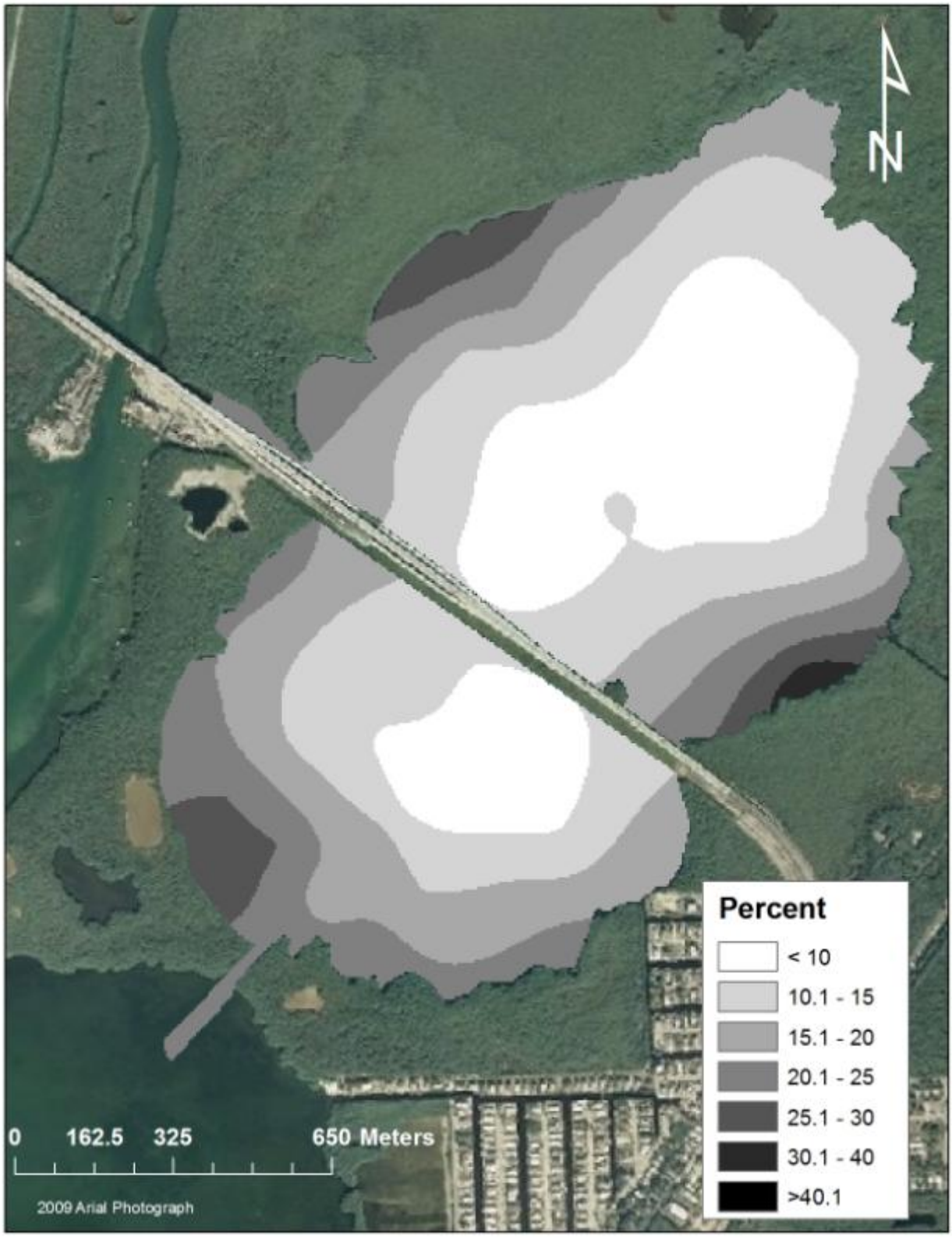
N
Percent
< 10
10.1 - 15
15.1 - 20
20.1 - 25
25.1 - 30
30.1 - 40
>40.1
0
162.5
325
650 Meters
2009 Arial Photograph

Figure 266. Soil organic matter (winter 2010).

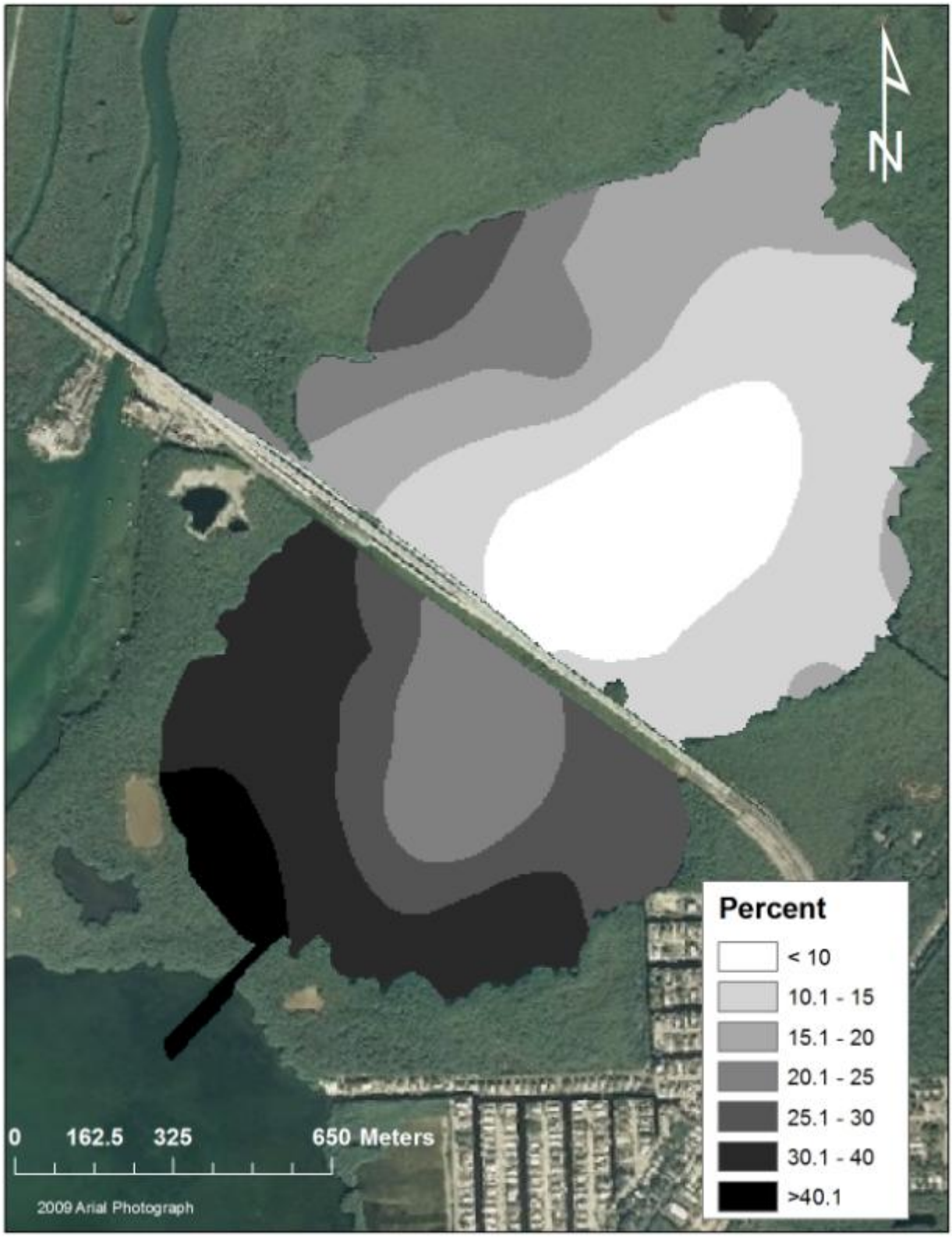
N
Percent
< 10
10.1 - 15
15.1 - 20
20.1 - 25
25.1 - 30
30.1 - 40
>40.1
0
162.5
325
650 Meters
2009 Arial Photograph

Figure 267. Soil organic matter (summer 2011).


N
Percent
< 15
15.1 - 20
20.1 - 25
25.1 - 30
30.1 - 35
35.1 - 40
>40.1
0
165
330
660 Meters
2009 Arial Photograph

Figure 268. Soil clay content (winter 2008).


N
Percent
< 15
15.1 - 20
20.1 - 25
25.1 - 30
30.1 - 35
35.1 - 40
>40.1
0
165
330
660 Meters
2009 Arial Photograph

Figure 269. Soil clay content (summer 2009).


N
Percent
< 15
15.1 - 20
20.1 - 25
25.1 - 30
30.1 - 35
35.1 - 40
>40.1
0
165
330
660 Meters
2009 Arial Photograph

Figure 270. Soil clay content (winter 2009).


N
Percent
<15
15.1 - 20
20.1 - 25
25.1 - 30
30.1 - 35
35.1 - 40
>40.1
0
165
330
660 Meters
2009 Arial Photograph

Figure
271


N
Percent
< 15
15.1 - 20
20.1 - 25
25.1 - 30
30.1 - 35
35.1 - 40
>40.1
0
165
330
660 Meters
2009 Arial Photograph

Figure
272


N
Percent
< 15
15.1 - 20
20.1 - 25
25.1 - 30
30.1 - 35
35.1 - 40
>40.1
0
165
330
660 Meters
2009 Arial Photograph

Figure
273

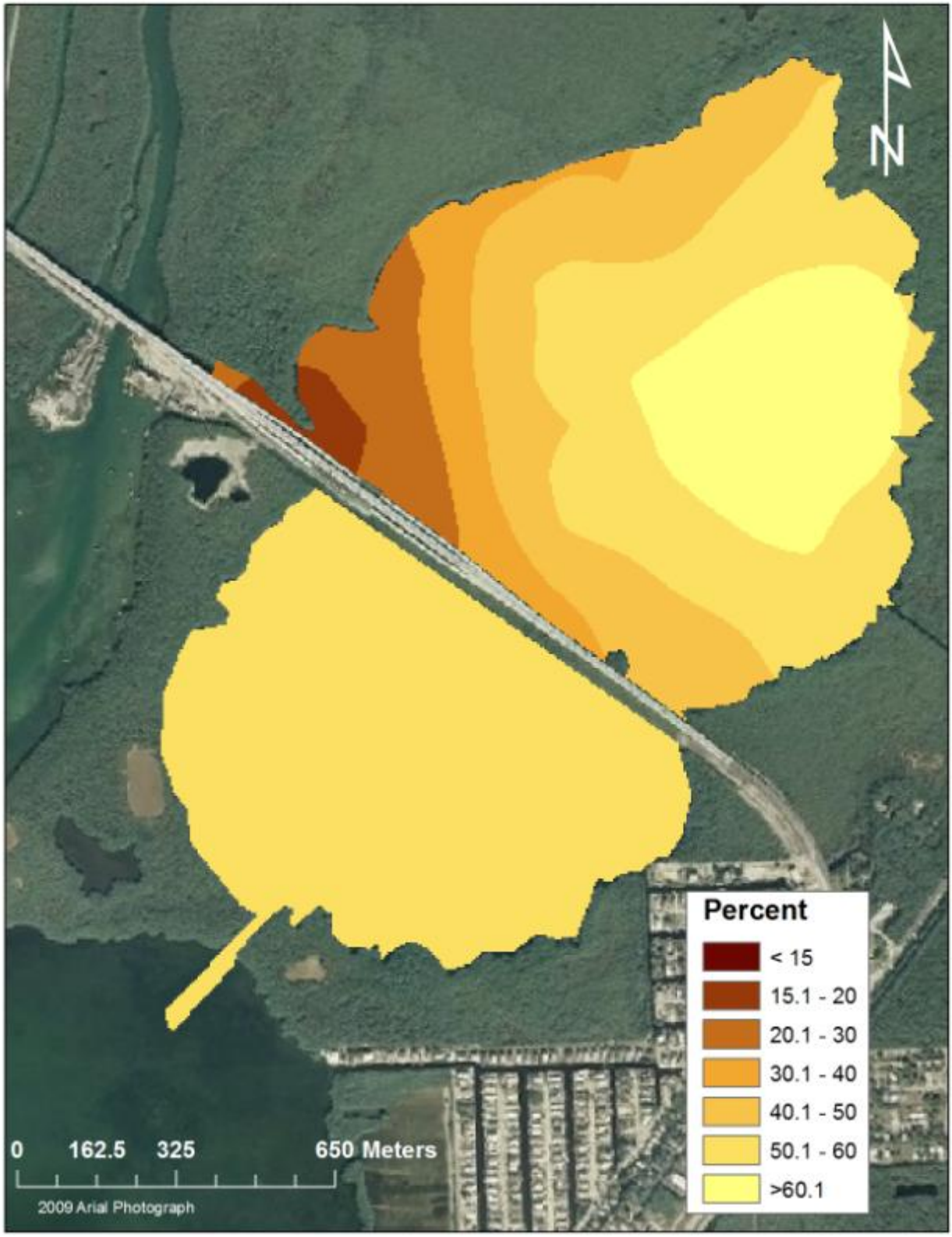
N
Percent
< 15
15.1 - 20
20.1 - 30
30.1 - 40
40.1 - 50
50.1 - 60
>60.1
0
162.5
325
650 Meters
2009 Arial Photograph

Figure
274

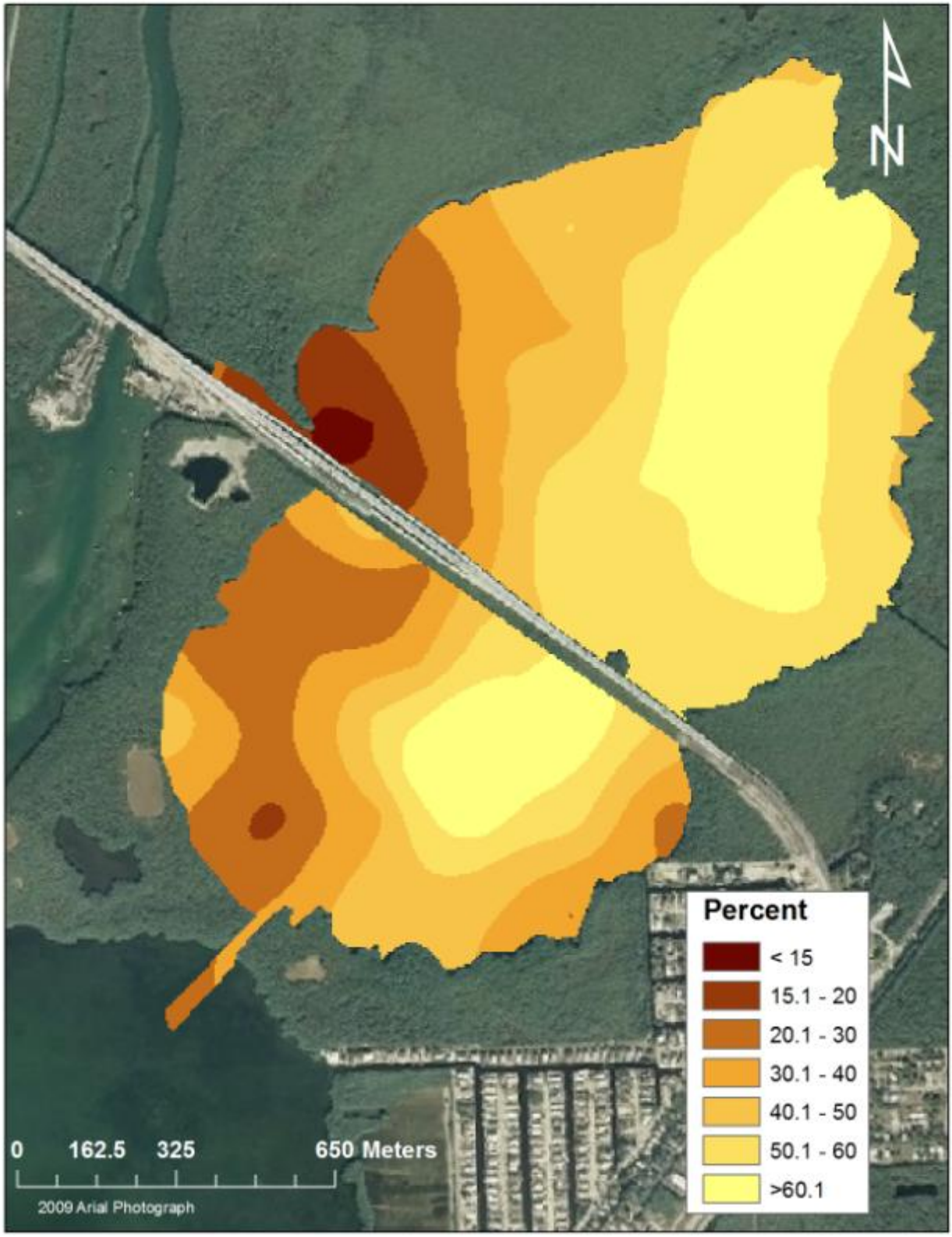
N
Percent
< 15
15.1 - 20
20.1 - 30
30.1 - 40
40.1 - 50
50.1 - 60
>60.1
0
162.5
325
650 Meters
2009 Arial Photograph

Figure
275

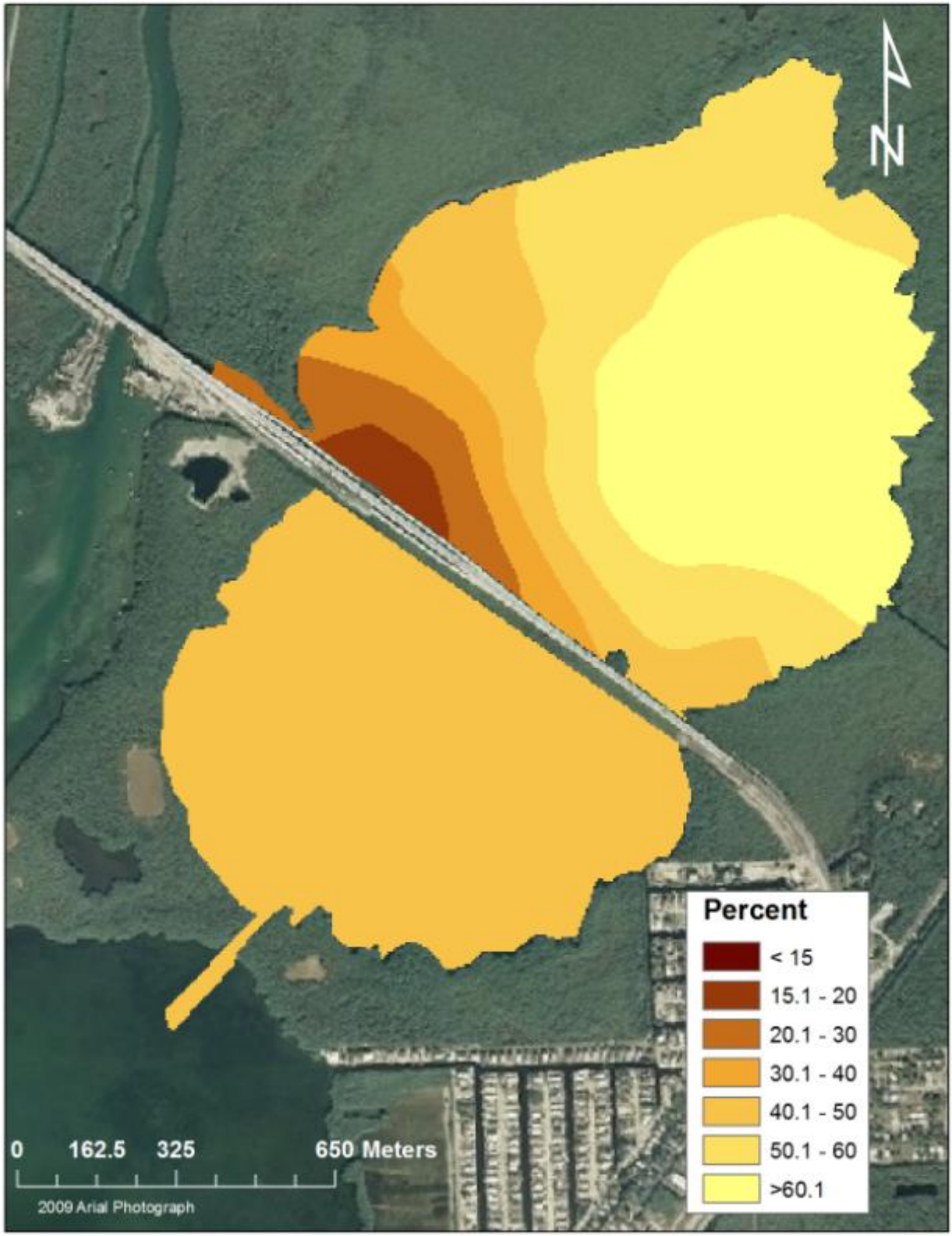
N
Percent
< 15
15.1 - 20
20.1 - 30
30.1 - 40
40.1 - 50
50.1 - 60
>60.1
0
162.5
325
650 Meters
2009 Arial Photograph

Figure
276


N
Percent
< 15
15.1 - 20
20.1 - 30
30.1 - 40
40.1 - 50
50.1 - 60
>60.1
0
162.5
325
650 Meters
2009 Arial Photograph

Figure 277. Soil sand content (summer 2010).

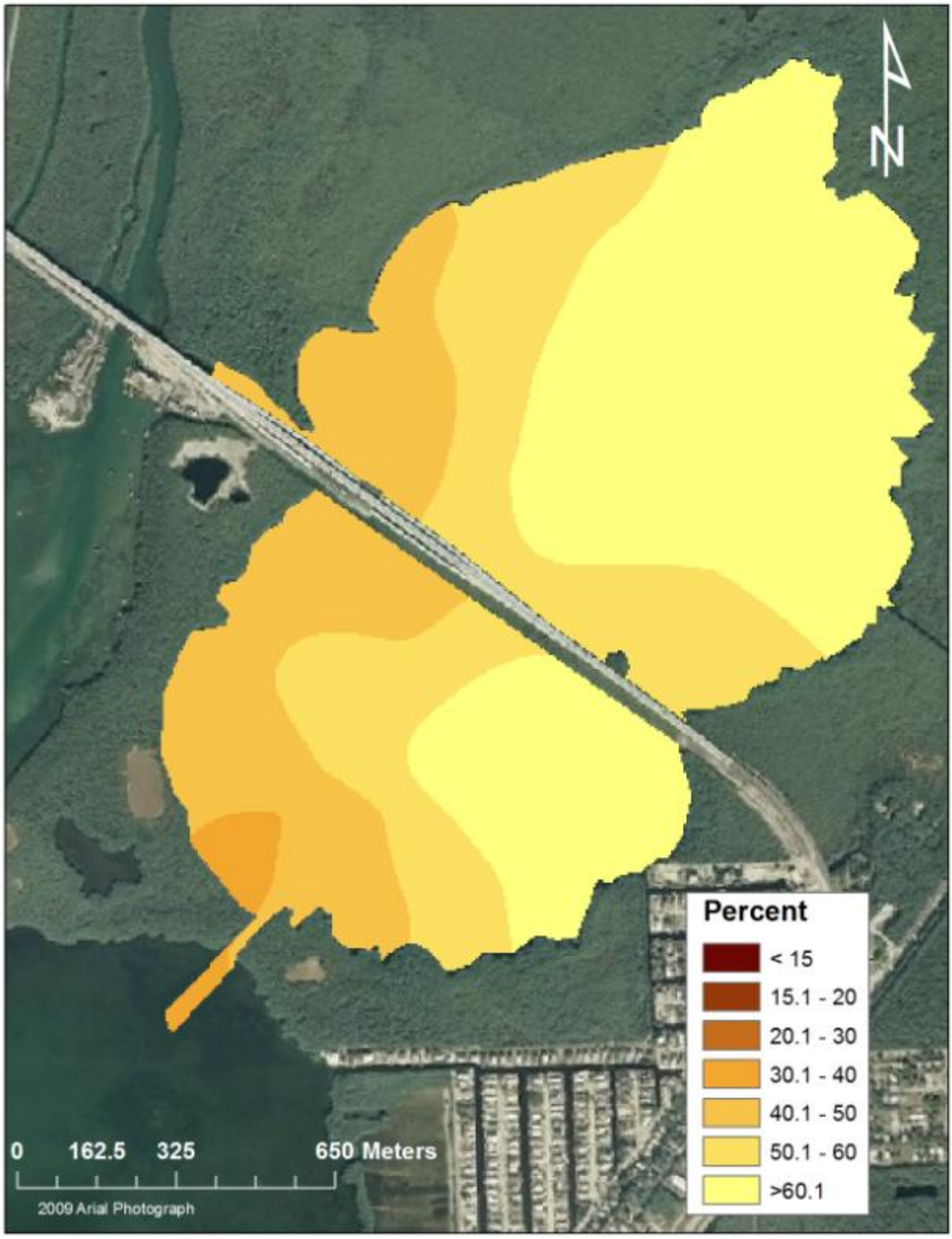

Figure
278


N
Percent
< 15
15.1 - 20
20.1 - 30
30.1 - 40
40.1 - 50
50.1 - 60
>60.1
0
162.5
325
650 Meters
2009 Arial Photograph

Figure
279

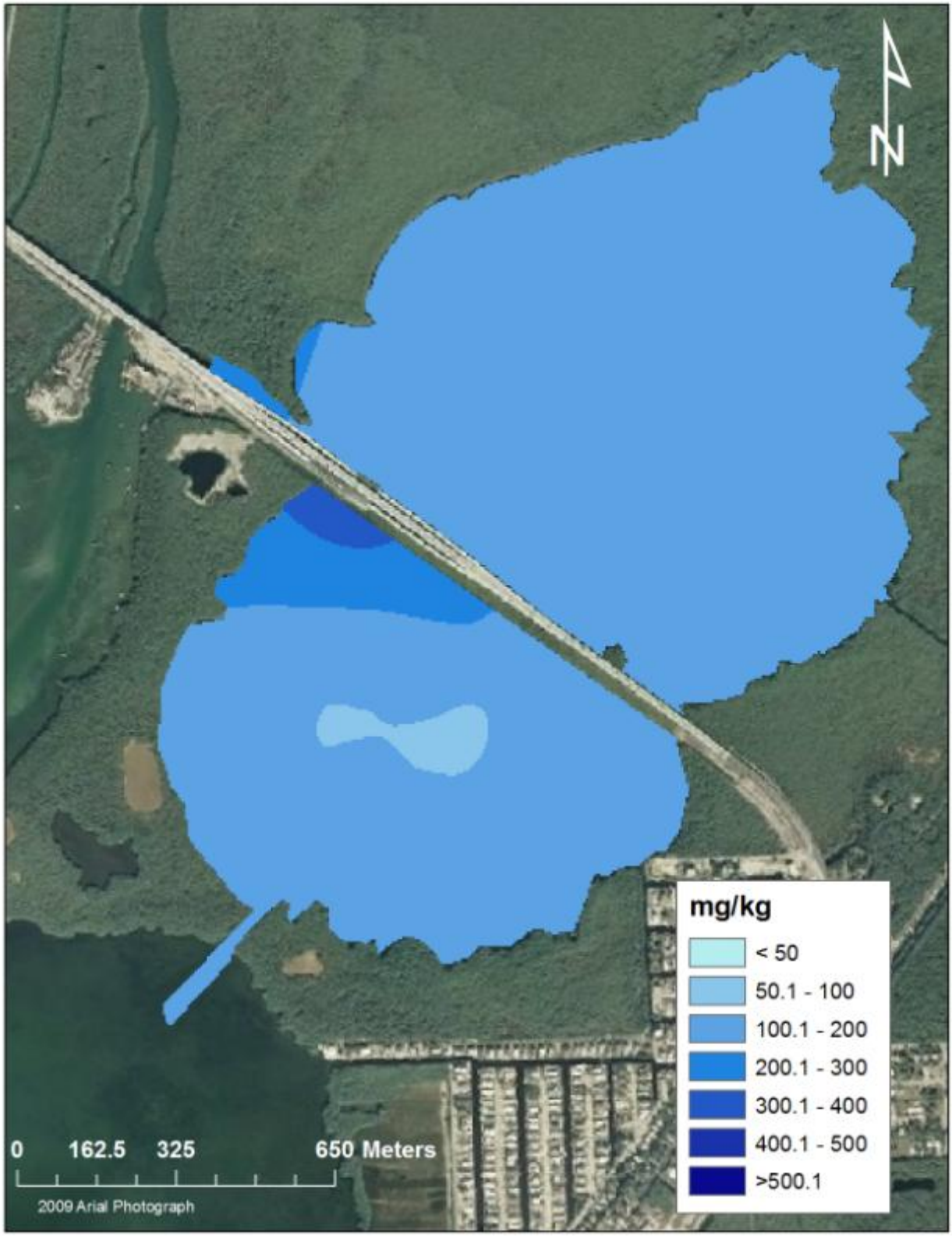
N
mg/kg
< 50
50.1 - 100
100.1 - 200
200.1 - 300
300.1 - 400
400.1 - 500
>500.1
0
162.5
325
650 Meters
2009 Arial Photograph

Figure
280


N
mg/kg
< 50
50.1 - 100
100.1 - 200
200.1 - 300
300.1 - 400
400.1 - 500
>500.1
0
162.5
325
650 Meters
2009 Arial Photograph

Figure
281

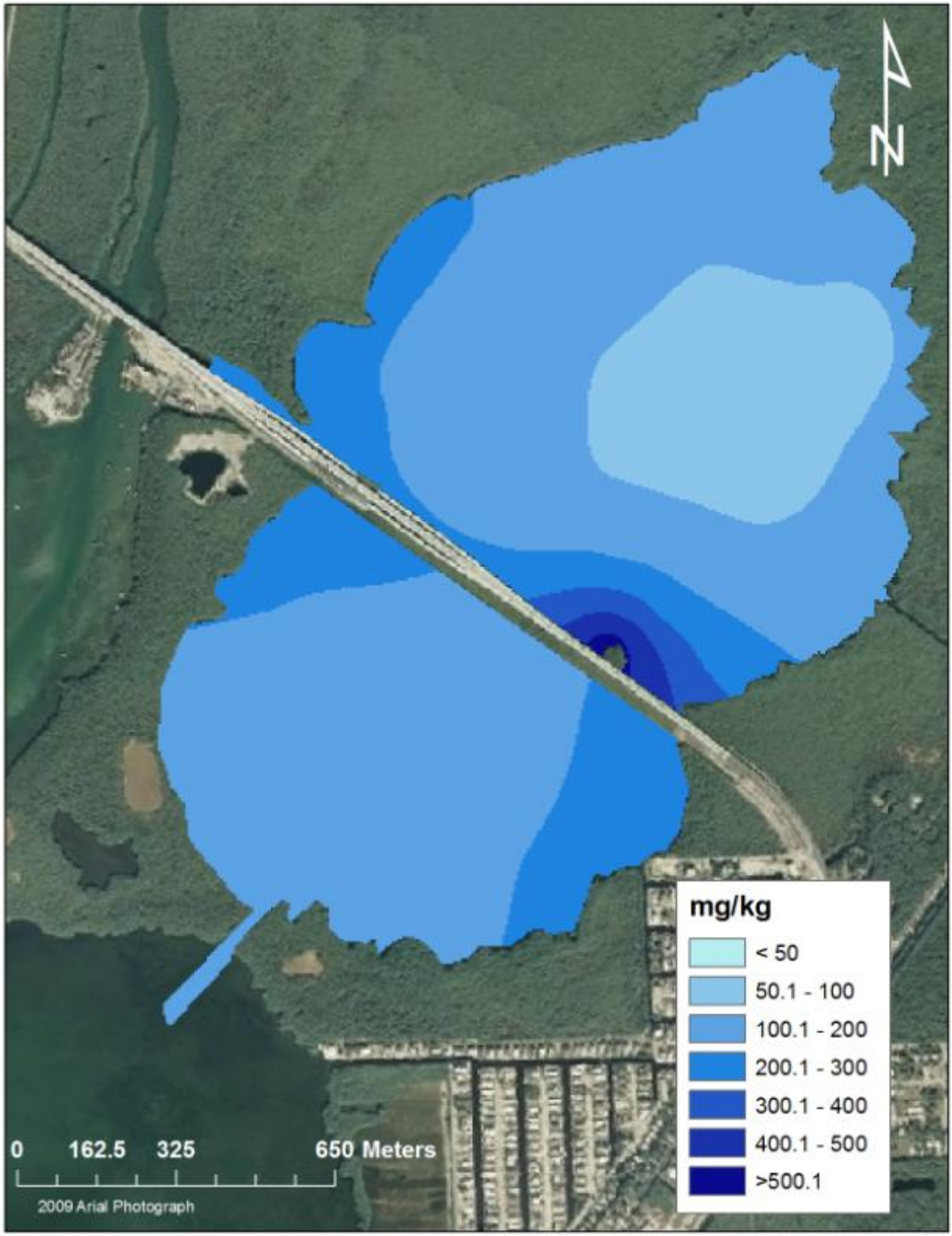
N
mg/kg
< 50
50.1 - 100
100.1 - 200
200.1 - 300
300.1 - 400
400.1 - 500
>500.1
0
162.5
325
650 Meters
2009 Arial Photograph

Figure 282. Soil total phosphorous (winter 2009).

Figure 283. Soil total phosphorous (summer 2010).

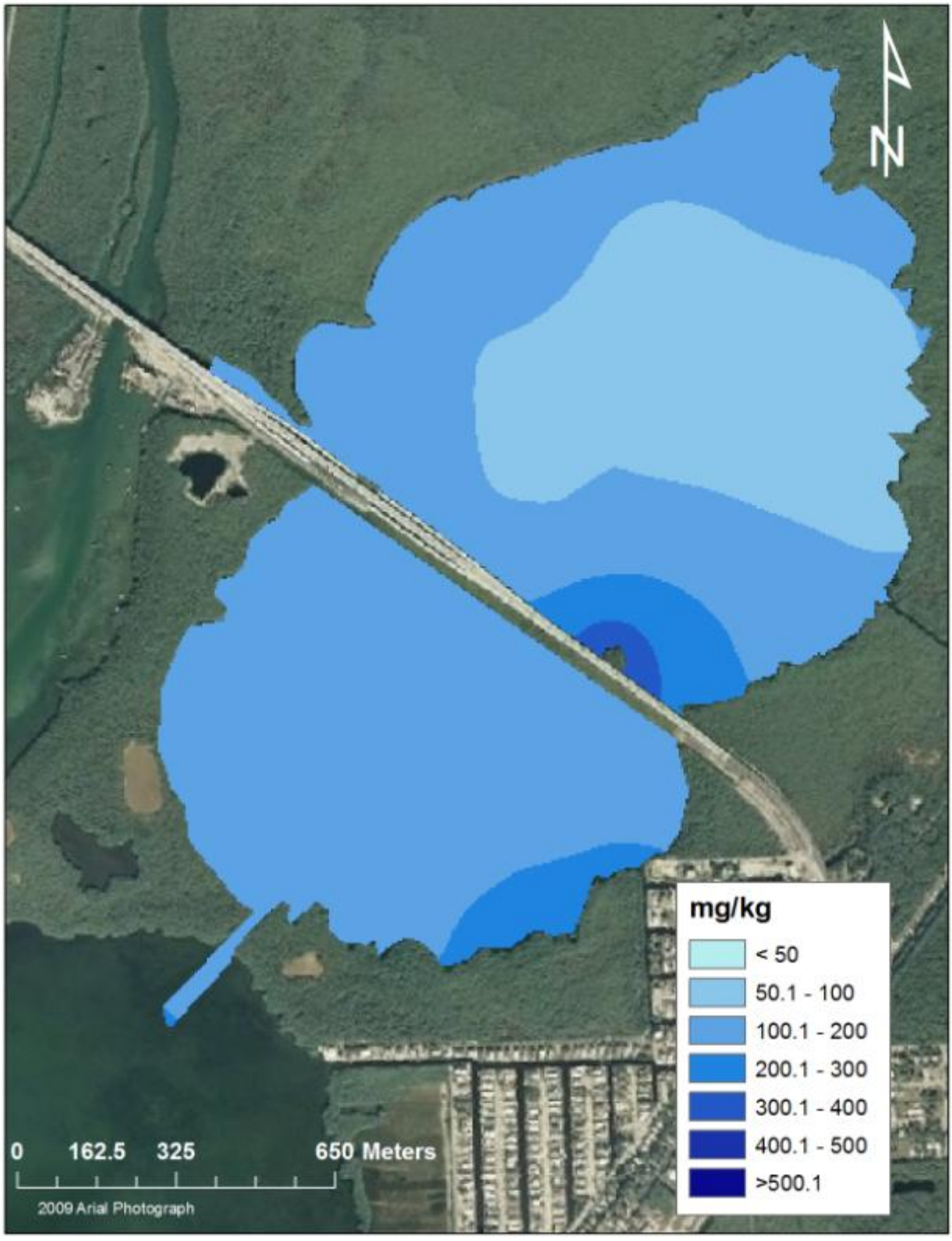
N
mg/kg
< 50
50.1 - 100
100.1 - 200
200.1 - 300
300.1 - 400
400.1 - 500
>500.1
0 162.5 325 650 Meters
2009 Arial Photograph

Figure 284. Soil total phosphorous (winter 2010).

APPENDIX C: CHAPTER 6 STATISTICAL TRANSFORMATIONS

Table 32. How data were transformed to meet the normality assumption prior to running ANOVAs. For parameters, TOC=total organic carbon, TN= total nitrogen, C:N= carbon to nitrogen ratio, ExOC=extractable organic carbon, and MBC=microbial biomass carbon. For transformations, NT= not transformed, Sqrt=square root, and a C (as in X-C) indicates that a constant was subtracted or added to a parameter before it was transformed via square root, log, arcsine, etc.

ANOVA	Depths	Parameter	Transformation
Mangrove factorial	Sediments 0-10	Bulk density	NT
		TOC (conc..)	Log 10 (Arcsin)
		TOC (storage)	Log 10
		TN (conc..)	Log 10 (Arcsin)
		C:N	Sqrt.
		ExOC (conc..)	Log e
		ExOC (storage)	Log e
		MBC (conc..)	Sqrt. (MBC-C)
		MBC (storage)	Log e
		Lability	Log 10
Seagrass factorial	Sediments 1-3	Bulk density	NT
		TOC (conc..)	NT
		TOC (storage)	NT
		TN (conc..)	NT
		C:N	Log e
		ExOC (conc..)	Log e (ExOC-C)
		ExOC (storage)	Log 10
		MBC (conc..)	Sqrt. (MBC-C)
		MBC (storage)	NT
		Lability	Log 10
Mangrove factorial	Surface layers	Bulk density	Sqrt.

		TOC (conc..)	Log 10 (Arcsin)
Table cont.		TOC (storage)	Log 10
		TN (conc..)	Log 10 (Arcsin)
		C:N	Log e (C:N-C)
		ExOC (conc..)	Log e (ExOC-C)
		ExOC (storage)	Log 10
		MBC (conc..)	Sqrt.
		MBC (storage)	Sqrt.
		Lability	Log e
Seagrass factorial	Surface layers	Bulk density	NT
		TOC (conc..)	Arcsin (sqrt)
		TOC (storage)	NT
		TN (conc..)	Sqrt.
		C:N	Sqrt.
		ExOC (conc..)	Sqrt.
		ExOC (storage)	Sqrt.
		MBC (conc..)	Sqrt.
		MBC (storage)	Log e
		Lability	NT
Mangrove repeated	Sediments 0-10	Bulk density	Log e
Measures		TOC (conc..)	Sqrt.
		ExOC (conc..)	NT

Table cont.

ANOVA	Depths	Parameter	Transformation
		TN (conc..)	Sqrt.
		C:N	Log e
		Lability	Sqrt.
	Algal mat	Bulk density	NT
		TOC (conc..)	NT
		ExOC (conc..)	NT
		MBC (conc..)	Sqrt. (MBC-C)
		TN (conc..)	Log e (TN-C)
		C:N	Log e
		Lability	Log e
Seagrass repeated Measures	Sediments 0-10	Bulk density	Log e
		TOC (conc..)	Sqrt.
		ExOC (conc..)	NT
		MBC (conc..)	NT
		TN (conc..)	Sqrt.
		C:N	Sqrt.
		Lability	
	Sediments accreted	Bulk density	NT
		TOC (conc..)	Log e
		ExOC (conc..)	Sqrt. (ExOC-C)
		MBC (conc..)	Sqrt.

			TN (conc..)	Log e
Table			C:N	Log e (C:N-C)
Cont.			Lability	NT
		Floc	Bulk density	Log 10
			TOC (conc..)	NT
			ExOC (conc..)	Sqrt. (ExOC-C)
			MBC (conc..)	NT
			TN (conc..)	NT
			C:N	Log e (C:N-C)
			Lability	NT
	Mangrove factorial	Sediments 0-10	$\Delta^{13}c$	NT
	Seagrass factorial	Sediments 1-3	$\Delta^{13}c$	NT
	Mangrove factorial	Surface layers	$\Delta^{13}c$	NT
	Seagrass factorial	Surface layers	$\Delta^{13}c$	Log 10 ($\delta^{13}C$*-1)
	Mangrove repeated measures	Sediments 0-10	$\Delta^{13}c$	NT
		Algal mat	$\Delta^{13}c$	NT
	Seagrass repeated measures	Sediments 0-10	$\Delta^{13}c$	NT
		Sediments accreted	$\Delta^{13}c$	NT
		Floc	$\Delta^{13}c$	NT

APPENDIX D: CHAPTER 7 SUPPLIMENTAL

GEE AND BOUDER PSDA ANALYSIS

Particles < 2mm (Pipette Method) (3A)

Reagents
Hydrogen peroxide (H_2O_2), 30 to 35 percent.

Sodium hexametaphosphate $(NaPO_3)_6$. Dissolve 35.7 grams of $(NaPO_3)_6$ and 7.94 grams of Na_2CO_3 per liter of water.

Demineralized water.

Procedure
Removing organic matter.--Place about 10 air-dry soil containing no particles larger than 2 mm in a tarred Flask. Add about 50-ml of demineralized water (referred to subsequently as water) and then add 5 ml of H_2O_2. Cover the flask with a watch glass. If a violent reaction occurs, repeat the cold H_2O_2 treatment periodically until no more frothing occurs. Heat the flask to about 90°C on an electric hot plate. Add H_2O_2 in 5-ml quantities at 45-min intervals until the organic matter is destroyed, as determined visually. Continue heating for about 30 min to remove any excess H_2O_2.

Removing cementing agents (optional).--Treat the sample with about 200 ml of 1 *N* sodium acetate buffered at pH 5 to remove carbonates. When CO_2 bubbles are no longer evident, wash free of salts with a filter candle system. Highly calcareous samples may need a second treatment. Remove siliceous cementing agents by soaking the sample overnight in 0.1 *N* NaOH. Iron oxide cementing agents are removed by shaking overnight in sodium dithionite (6C2). Wash free of salts with filter candle system before proceeding.

Removing dissolved mineral and organic components.--After the H_2O_2 treatment, place the flask in a rack and add about 150 ml of water in a jet strong enough a short Pasteur- Chamberlain filter of "F" fineness. Five such washings and filterings are usually enough except for soils containing much coarse gypsum. Remove soil adhering to the filter by gentle back pressure; use finger as policeman. Dry the sample overnight in an oven at 105°C, cool in a desiccator, and weigh to the nearest milligram. Use the weight of the oven dry, H_2O_2-treated sample as the base weight for calculating percentages of the various fractions.

Dispersing the sample.--Add 10 ml of sodium hexametaphosphate dispersing agent to the flask containing oven dry treated sample. Make the volume to approximately 200 ml. Stopper and shake overnight on a horizontal reciprocating shaker at 120 oscillations per minute. *Separating sands from silt*

and clay.--Wash the dispersed sample with water on a 300-mesh sieve. Silt and clay pass through the sieve into a 1-L cylinder. Use a clamp and stand to hold the sieve above the cylinder. Avoid using jets of water in washing the sample. Gently tap the sieve clamp with the side of the hand to facilitate sieving. Continue washing until the suspension volume in the cylinder is about 800 ml. Sand and some coarse silt remain on the sieve. It is important to wash all particles of less than 20µ diameter through the sieve. Remove the sieve from the holder, wash the sands into an evaporating dish with water, and dry at 105 to 110°C. Bring the silt and clay suspension in the cylinder to 1 L with water and cover with a watch glass.

Pipetteting.--First pipette the <20µ fraction at a 10-cm depth. Vary sedimentation times according to temperature. Next, pipette the <2µ fraction after a predetermined setting time (usually 4 1/2 to 6 1/2 hr). Vary depth according to time and temperature. Use a Lowy 25-ml automatic pipette and regulate filling time to about 12 s. Before each pipetting, stir material in the 611 sedimentation cylinder, and stir the suspension for 30 s with a hand stirrer, using an up-and-down motion. Note the time at completion of stirring. About 1 min before sedimentation is complete, lower the tip of the pipette slowly into the suspension to the proper depth with a Shaw pipette rack. At the appropriate time, fill the pipette and empty into a 90-ml, wide-mouth bottle. Rinse the pipette into the bottle once. Dry in an oven overnight at 105°C. Cool in a desiccator containing phosphorus pentoxide (P_2O_5). Weigh.

Sieving and weighing the sand fractions.--Transfer the dried sands to a nest of sieves. Shake for 3 min on a shaker that has 1/2-in vertical and lateral movements and oscillates at 500 strokes per minute. Record the weights of the individual sand fractions.

Calculations

Pipetted fractions:

Percentage of pipetted fractions = (A - B)KD

where
A = Weight (g) of pipetted fraction
B = Weight correction for dispersing agent (g)
K = 1000/(ml in pipette)
D = 100/(g of H_2O_2-treated oven dry total sample)

The <20-µ fraction minus the <2-µ fraction equals fine silt.

Sand fractions:
Percentage of sieved fractions = weight (g) of fraction on sieve times D.

Coarse silt fraction:
Obtain by difference. Subtract the sum of the percentages of sand plus the <20-µ fraction from 100.

References

Kilmer and Alexander (1949), Kilmer and Mullins (1954), Tyner (1939), and Grossman and Millet (1961).

Moist Samples (3A2)

If drying affects dispersion of treated sample, oven drying may be avoided by removal of a pipette sample to estimate the total weight of the sample. Pipette 50 ml at a depth of 20 cm at time zero while the suspension is still turbulent. Use the oven dry weight of the aliquot to calculate the total weight of the <0.05-mm fraction. Add this weight to the total weight of the sands to obtain the total weight of the sample. An optional procedure is to carefully weigh out two identical samples and pretreat to remove organic matter and dissolved mineral matter. The first sample is continued through the standard procedure, excluding oven drying. The second sample is oven dried, weighed, and discarded. The oven dry weight of the second sample is substituted in the calculations for the first sample.