ABSTRACT

HARTIS, BRETT MICHAEL. Mapping, Monitoring and Modeling Submersed Aquatic Vegetation Species and Communities. (Under the direction of Stacy A. C. Nelson).

Aquatic macrophyte communities are critically important habitat species in aquatic systems worldwide. None are more important than those found beneath the water's surface, commonly referred to as submersed aquatic vegetation (SAV). Although vital to such systems, many native submersed plants have shown near irreversible declines in recent decades as water quality impairment, habitat destruction, and encroachment by invasive species have increased. In the past, aquatic plant science has emphasized the restoration and protection of native species and the management of invasive species. Comparatively little emphasis has been directed toward adequately mapping and monitoring these resources to track their viability over time. Modeling the potential intrusion of certain invasive plant species has also been given little attention, likely because aquatic systems in general can be difficult to assess. In recent years, scientists and resource managers alike have begun paying more attention to mapping SAV communities and to address the spread of invasive species across various regions. This research attempts to provide new, cutting-edge techniques to improve SAV mapping and monitoring efforts in coastal regions, at both community and individual species levels, while also providing insights about the establishment potential of *Hydrilla verticillata*, a noxious, highly invasive submersed plant. Technological advances in satellite remote sensing, interpolation and spatial analysis in geographic information systems, and state-of-the-art climate envelope modeling techniques were used to further assess the dynamic nature of SAV on various scales. This work contributes to the growing science of mapping, monitoring, and modeling of SAV.

Mapping, Monitoring and Modeling Submersed Aquatic Vegetation Species and Communities

by
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DEDICATION

To my amazing parents who have supported my scholastic endeavors from day one.

BIOGRAPHY

Brett Hartis was born and raised in Marion, North Carolina the son of Gary and Brenda Hartis. His love for all things outdoors began at a young age and was greatly fostered by his family. Brett found interests in fishing and hunting throughout the beautiful Blue Ridge Mountains of Western North Carolina. After graduating high school in the top of his class, Brett headed east to East Carolina University and quickly began studying the estuarine and marine systems of coastal NC. After having the opportunity to do research with various experts in the field of fisheries science, Brett graduated from the ECU with a 4.0 and an award for outstanding senior in the Department of Biology. He then moved to Raleigh, North Carolina to pursue further interests in natural resources and geographic information systems, obtaining a Master of Science degree in Fisheries, Wildlife, and Conservation Biology and North Carolina State University.

In 2010, Brett began work as a research assistant with Dr. Stacy Nelson of NCSU to utilize various geospatial platforms to assess aquatic plant populations in the Currituck Sound of North Carolina. In 2012, he accepted a position in the Department of Crop Science as an aquatics extension associate, providing extension and outreach to various groups regarding aquatic plant science and management. During this time, Brett realized a passion in life to provide education and knowledge to individuals and groups regarding aquatic ecosystems and their functions. While working as an extension associate and research assistant, Brett worked on his doctorate in Fisheries, Wildlife and Conservation Biology.

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CHAPTER 1

Introduction

1.1 Mapping, Monitoring and Modeling Submersed Aquatic Vegetation

Aquatic systems are as mysterious as they are complex. Unlike terrestrial systems, the monitoring, management and assessment of such environments is greatly hindered by limitations of humans, as organisms foreign to life in aquatic habitats. Extensive monitoring protocols, management handbooks, and assessment manuals have been written for many terrestrial ecosystems. By comparison, the aqueous domain of planet Earth is poorly understood.

Some aspects of aquatic systems, such as the biotic and physical realms, have seen standardization of protocols and methodologies for monitoring and assessment over the past few decades. However, one of the most important aquatic communities in need of understanding still lacks widely accepted methodologies that can provide consistently precise and accurate measures (Systma 2008). Aquatic vascular plant populations found in various water bodies worldwide, while critically important habitat species, generally have been "beyond reach" except for relatively primitive methods for monitoring and assessment. Understanding the dynamic, multifaceted intricacies of aquatic plant populations has been recognized as much more important in more recent years due to global declines in native populations concomitant with invasions of non-native species through global human movement (Madsen and Wersal 2012, Waycott et al. 2009). A subgroup of aquatic plants that poses the greatest challenges to monitoring and assessment are those that grow and

reproduce beneath the water's surface. Submersed aquatic vegetation (SAV) is a diverse group of vascular plants found in various climates throughout the world (Waycott et al 2009, Street et al. 2005). SAV provides food, shelter and critical habitat for many animal species (Larkum et al. 2006). The disappearance of native species or the introduction and dispersal of invasive species can cause severe ecological and economic impacts (Langeland 1996, Charles and Dukes 2007). Ecological impacts are centered on the fact that the entire food web dynamics can be significantly altered by the addition or removal of a single keystone or exotic species (Irlandi et al. 1995, Hovel and Lipcius 2002). Invasive submersed species, in particular, displace native plants and can shift balanced, heterogeneous ecosystems to monocultures with severely altered food web dynamics (Richardson et al. 2012). Invasive SAV can also promote lethal impacts affecting higher trophic levels. For example, an epiphytic cyanobacterium commonly found on invasive *Hydrilla verticillata* has been shown to produce a neurotoxin(s) that causes Avian Vacuolar Myelinopathy (AVM). AVM is an often-lethal neurological disease of waterfowl and their predators in the southeastern United States, from coots (*Fulica americana*) to bald eagles (*Haliaeetus leucocephalus*) (Wilde et al. 2005, Williams et al. 2007). Furthermore, large quantities of invasive SAV biomass are prime habitat for mosquitoes, which can carry a number of diseases (Nichols and Shaw 1986). Major economic loss has been sustained from invasive SAV whose growth inhibits flood control, hydropower generation, irrigation, navigation and recreation in infested water bodies (Pimental et al. 2005). Related loss of *native* SAV species can result in the collapse of fish populations (Wislon et al. 2013). Many have quantified the direct economic losses associated with both native losses and invasive introductions as ranging from tens of

thousands to millions of U.S. dollars (e.g. Larkum et al. 2006, Pimental et al. 2005, Costanza et al. 1997, Anderson 1993). Indirect economic losses, more difficult to define and sometimes of major importance, only add to this deficit (Pimental et al. 2005). Thus, the methods by which to detect, monitor and assess SAV, including evaluation of the efficacy of management actions, have become increasingly important (Blossey 2004). Potentially as important as determining SAV species composition in a given water body can be determining the geographic extent of species distributions, confounded by climate change (Johnston 1986). This is especially the case with invasive species, which can cause great economic hardship and ecological damage in the wake of their expansion.

Various methodologies for detection and monitoring of SAV lack rigorous standardization and explicit protocols. Most commonly, these methods are employed to develop species lists, estimate plant abundance, and determine species distributions within a given water body (Madsen and Wersal 2012). Techniques for monitoring SAV range from low-cost, high-effort point-intercept sampling to high-cost, moderate-effort remote sensing, each of which can provide very different information. For example, point-intercept sampling provides the researcher with the presence or absence of various species, but without intensive sampling, information on the extent and distribution of plants is left to speculation or some degree of interpolation. Remote sensing can provide valuable distribution and extent information but cannot be used to detect individual species presence or absence (Madsen and Bloomfield 1993). Application of certain methodologies can also be confounded by the environmental and physical parameters of a water body (Yin and Kreiling 2011, Middleboe and Markager 1997). These strengths and limitations often determine which method will be

most applicable to a given project. The method that most meets the desired objectives of the study is used; no one-size-fits-all method can be fit across all systems (Spencer and Whitehand 1993).

Detection and monitoring of SAV has become extremely important over the past few decades, especially as non-native species have continued to establish and spread throughout various water bodies in North America as throughout the world (Williams and Meffe 1998). Determining where a species presently exists in a water body is important, yet accurately estimating the extent to which a species may expand beyond its established range can be just as vital (Johnstone 1986). For example, accurate prediction of the potential establishment of an invasive SAV species can greatly aid resource managers in maximizing preventative and precautionary measures.

This dissertation explores various techniques to map, monitor and model native and invasive SAV species. Chapter 2 assesses the use of newly available satellite imagery to develop predictive models of SAV presence/absence in a highly turbid coastal system. In Chapter 3, a geographic information systems (GIS)-based and field-driven mapping technique is developed to identify the existing spatial locations and coverage of individual native and invasive SAV species in a coastal system. In Chapter 4, the notorious invasive SAV species, *Hydrilla verticillata*, is modeled to determine its establishment potential throughout the United States and Canada.

1.2 Primary Research Questions and Rationale

Chapter 2: Large-scale submersed aquatic vegetation (SAV) surveys are rarely done due to logistical difficulties and high costs. This characterization especially fits the shallow, low-salinity, highly diverse coastal aquatic habitats of North Carolina. The primary reason for difficulty in obtaining survey data over large areas is largely due to the expense, intensive labor need and time constraints associated with sampling SAV. In past years, remote sensing coupled with modeling and interpolation techniques has shown the potential to be an important tool to obtain survey information on SAV over large lakes across the country (Nelson et al. 2006, Valley et al., 2005). However, this approach has limited applicability for assessing SAV distributions in shallow, coastal regions where frequently changing, tidal and wind-driven currents cause high, persistent turbidity. Therefore, an approach that can address the need for regional-scale water body mapping and monitoring of SAV in these coastal areas would be valuable. Survey assessments are further complicated in coastal regions covering thousands of hectares. The second chapter of this dissertation examines whether remote sensing can be used to detect SAV presence and/or levels of cover in the Currituck Sound, a shallow, low-salinity water body on the northeastern coast of North Carolina.

Chapter 3: Mapping of Submersed aquatic vegetation (SAV) in highly diverse, coastal aquatic habitats is a top priority of resource managers, especially in more recent years as restoration, mitigation and invasive species management activities have steadily increased (Ailstock et al. 2010, Busch et al. 2010, Gordon 1998) While various mapping and survey techniques are often employed to inform such activities, each has limitations for identifying key information about the spatial extent and coverage of SAV species in large areas with

nearly continual littoral coverage. In such systems, a method to identify key areas for further exploration and survey should be employed to help focus management efforts. Understanding the dynamics of SAV populations in a given water body has become increasingly important, especially with the increasing encroachment of various invasive species that can severely alter community dynamics (Madsen and Wersal 2012). Furthermore, assessing individual species dominance and coverage within large stands of vegetation provides the essential information needed to make sound decisions about optimal management strategies. Here, I developed a geographic information systems (GIS)-based and field driven mapping technique to identify the present spatial locations and coverage of SAV species in the Currituck Sound of North Carolina. The spatial distribution of the invasive species, *Myriophyllum spicatum*, was also targeted.

Chapter 4: Hydrilla (*Hydrilla verticillata*) is a highly invasive submersed aquatic plant species that has caused severe ecological damage and economic hardship (Langeland 1996). After introduction into Florida during the 1950s, the species has continued to spread across the U.S. mainland, establishing as far north as Maine and as far west as Washington (Bailey and Calhoun 2008, Madeira et al. 2000). Hydrilla establishment in colder environments has recently raised questions about the ability of this plant to extend its range throughout North America (Richardson et al. 2012). Despite the issue of hydrilla spreading northward, there has been a paucity of attempts to model its potential expansion (Peterson et al. 2003, Gallardo and Aldridge 2013). Langeland (1996) suggested that the monoecious biotype could spread as far north as southern Canada (1996) based on its range in Europe at the time. However, hydrilla has since established much farther north in other areas of the world than

previously expected (Balciunas and Chen 1993). It has even been found growing in the subarctic climates of Latvia, Russia, and Poland (Brunel 2009, Nesterova 1994, Klosowski 2006). This study modeled the establishment potential of hydrilla using a modified version of the climate envelope method, based on the known global geographic distribution of this species in the Northern Hemisphere, particularly along its northernmost limits.

1.3. References

Ailstock, M. S., D. J. Shafer, and A. D. Magoun. 2010. Effects of planting depth, sediment grain size, and nutrients on Ruppia maritima and Potamogeton perfoliatus seedling emergence and growth. Restoration Ecology 18: 574-583. DOI: 10.1111/j.1526-100X.2010.00697.x

Anderson LWJ. 1993. Aquatic weed problems and management in North America (a) Aquatic weed problems and management in the western United States and Canada. In: AH Pieterse and KJ Murphy (eds.), Aquatic Weeds. Oxford University Press, Oxford. pp. 371-391.

Bailey JE and Calhoun AJK. 2008. Comparison of three physical management techniques for controlling variable-leaf milfoil in maine lakes. J Aquat Plant Manage 46:163-7.

Balciunas J and Chen P. 1993. Distribution of hydrilla in northern china - implications on future spread in north-america. J Aquat Plant Manage 31:105-9.

Blossey B. 2004. Monitoring in weed biological control programs, pp. 95-105. In: EM Coombs, JK Clark, GL Piper and AF Cofrancesco, Jr. (eds.). Biological control of invasive plants in the United States. Oregon State University Press, Corvallis.

Brunel S. 2009. Pathway analysis: Aquatic plants imported in 10 EPPO countries. Bulletin OEPP 39(2):201-13.

Busch, K. E., R. R. Golden, T. A. Parham, L. P. Karrh, M. J. Lewandowski, and M. D. Naylor. 2010. Large-scale *Zostera marina* (eelgrass) restoration in Chesapeake Bay, Maryland, USA. Part I: A comparison of techniques and associated costs. Restoration Ecology 18: 490-500. DOI: 10.1111/j.1526-100X.2010.00690.x

Costanza R, R d'Arge, R de Groote, S Farber, M Grasso, B Hannon, K Limburg, S Naeem, RV O'Neill, J Paruelo, RG Raskin, P Sutton, and M van den Belt. 1997. The value of the world's ecosystem services and natural capital. Nature 387:253-260.

Charles H and JS Dukes. 2007. Impacts of invasive species on ecosystem services. Biol Invas 193:217- 237

Gallardo B and Aldridge DC. 2013. The 'dirty dozen': Socio-economic factors amplify the invasion potential of 12 high-risk aquatic invasive species in Great Britain and Ireland. J Appl Ecol 50(3):757-66.

Gordon, D.R., 1998. Effects of Invasive, Non-indigenous plant species on ecosystem processes: Lessons from Florida. Ecological Applications. 8: 975-989.

Hovel KA, Lipcius RN (2002) Effects of seagrass habitat fragmentation on juvenile blue crab survival and abundance. J Exp Mar Biol Ecol 271:75–98

Irlandi EA, Ambrose WG Jr, Orlando BA (1995) Seascape ecology and the marine environment: how spatial configu-ration of seagrass habitat influences growth and survival of the bay scallop. Oikos 72:307–313

Johnstone IM. 1986. Plant invasion windows: A time-based classification of invasion potential. Biological Reviews 61(4):369-94.

Klosowski S. 2006. The relationships between environmental factors and the submerged potametea associations in lakes of north-eastern poland. Hydrobiologia 560:15-29.

Langeland, K.A. 1996. Hydrilla verticillata (L.F.) Royle (Hydrocharitaceae), "The Perfect Aquatic Weed". Castanea. 61(3): 293-304.

Larkum, A.W.D., Orth, R.J., Duarte, C., 2006. Seagrasses: Biology, Ecology and Conservation. Springer, The Netherlands. 691.

Madeira, P.T., C.C.Jacono, and T.K. Van. 2000. Monitoring hydrilla using two RAPD procedures and the nonindigenous aquatic species database. J. Aquat. Plant Manage. 38:33-40.

Madsen JD and JA Bloomfield. 1993. Aquatic vegetation quantification symposium: an overview. Lake and Reserv. Manage. 7:137-140.

Madsen JD and RM Wersal. 2012. A Review of Aquatic Plant Monitoring and Assessment Methods. Aquatic Ecosystem Restoration Foundation.

Middelboe AL and S Markager. 1997. Depth limits and minimum light requirements for freshwater macrophytes. Freshwater Bio. 37:553-568

Nelson, S.A.C., Cheruvelil,K.S. and Soranno, P.A. 2006. Satellite Remote Sensing of Freshwater Macrophytes and the Influence of Water Clarity. Aquatic Botany 85.4, 289-98.

Nesterova IA. 1994. New records of aquatic and littoral plants in sikhote-alin reserve. Botanicheskii Zhurnal (St.Petersburg) 79(3):116-7.

Nichols C and Shaw B. 1986. Ecological life histories of the 3 aquatic nuisance plants, myriophyllum-spicatum, potamogeton-crispus and elodea-canadensis. Hydrobiologia 131(1):3-21.

Peterson A, Papes M, Kluza D. 2003. Predicting the potential invasive distributions of four alien plant species in north america. Weed Sci 51(6):863-8.

Pimentel D, R Zuniga and D Morrison. 2005. Update on the environmental and economic costs associated with alien-invasive species in the United States. Ecol. Econ. 52: 273-288.

Richardson et al. 2012. Monoecious Hydrilla – A Review of the Literature. Northeast Aquatic Nuisance Species Panel. Accessed Online: http://www.nyis.info/user_uploads/files/Monoecious%20Hydrilla%20Lit%20Review%20-%20Final.pdf

Spencer DF and LC Whitehand. 1993. Experimental design and analysis in field studies of aquatic vegetation. Lake and Reserv. Manage. 7:165-174.

Street, M.W., Deaton, A.S., Chappel, W.S., Mooreside, P.D., 2005. North Carolina Coastal Habitat Protection Plan, North Carolina Department of Environment and Natural Resources, Division of Marine Fisheries, Morehead City, NC. 656 pp. Online at http://www.ncfisheries.net/habitat/chpp2k5/_Complete%20CHPP.pdf

Sytsma MD. 2008. Introduction: workshop on submersed aquatic plant research priorities. J. Aquat. Plant Manage. 46:1-7.

Valley, R.D., Drake, M.T., Anderson, C.S., 2005. Evaluation of alternative interpolation techniques for the mapping of remotely-sensed submersed vegetation abundance. Aquat. Bot. 81, 13–25.

Waycott, M., Duarte, C.M., Carruthers, T.J., Orth, R.J., Dennison, W.C., Olyarnik, S., Calladine, A., Fourqurean, J.W., Heck, K.L., Hughes, A.R., Kendrick, G.A., Kenworthy, W.J., Short, F.T, Williams, S.L., 2009. Accelerating loss of seagrasses across the globe threatens coastal ecosystems. Proceedings of the National Academy of Sciences 106, 12377-12381.

Wilde, S.B., T.M. Murphy, C.P. Hope, S.K. Habrun, J. Kempton, A. Birrenkott, F. Wiley, W.W. Bowerman, and A.J. Lewitus. 2005. Avian vacuolar myelinopathy linked to exotic aquatic plants and a novel cyanobacterial species. Environ. Toxicol. 20:348–353.

Williams, S.K., J. Kempton, S.B. Wilde, and A. Lewitus. 2007. A novel epiphytic cyanobacterium associated with reservoirs affected by avian vacuolar myelinopathy. Harmful Algae. 6:343–353.

Williams, J.D.; G. K. Meffe (1998). "Nonindigenous Species". Status and Trends of the Nation's Biological Resources. Reston, Virginia: United States Department of the Interior, Geological Survey 1.

Wilson CJ, Wilson PS, Greene CA, Dunton KH. 2013. Seagrass meadows provide an acoustic refuge for estuarine fish. Mar Ecol Prog Ser 472:117-27.

Yin Y and RM Kreiling. 2011. The evaluation of a rake method to quantify submersed vegetation in the Upper Mississippi River. Hydrobiologia 675:187-195.

CHAPTER 2

Plants and Pixels: Developing Predictive Models of Submersed Aquatic Vegetation using Satellite Imagery in the Currituck Sound, North Carolina USA

2.1 Introduction

Large-scale submersed aquatic vegetation (SAV) surveys are rarely possible, although effective SAV management depends in part on understanding the coverage and spatial location of SAV. The primary reason for such difficulty in obtaining survey data over large areas is largely due to the expense, intensive labor need and time constraints associated with sampling SAV. Survey assessments are further complicated in coastal regions covering thousands of hectares. SAV plays an extremely important role ecologically, providing critical habitat for fish, shellfish, and other wildlife as well as supporting local fisheries based economies (Larkum et al. 2006). The most extensive SAV communities in North Carolina tend to be found in shallow, low-salinity waters on the leeward side of the state's barrier island network (Davis and Brinson 1983, Ferguson and Wood 1994, Street et al. 2005). Inventorying an area this large becomes cost-, time-, and labor-prohibitive using traditional field sampling techniques such as visual delineation, sampling along transects, or subsampling randomly selected points. Although these techniques have been shown to produce acceptable estimates of SAV coverage for smaller bodies of water (Madsen 1999), they have been logistically impractical when attempts have been made to apply them to large water bodies (Zhang 1998). The extensive Currituck Sound of North Carolina is such a system; nearly all of its large expanse is accessible to SAV establishment (Thayer et al. 1984)

Remote sensing using modeling and interpolation techniques have exhibited the potential to be an important tool to obtain survey information on SAV cover in large lakes (Nelson et al. 2006, Valley et al. 2005). The utility of remote sensing to measure SAV has been demonstrated through the process of mapping general SAV distributions through visually driven delineations (Orth and Moore, 1983; Marshall and Lee, 1994), including North Carolina's coastal waters (Ferguson and Korfmacher 1997). Remote sensing has been successfully applied to assess SAV in clear tropical waters worldwide (Chollett and Mumby 2012, Dierssen et al. 2010, Lyzenga et al. 2006). However, this approach has limited applicability for assessing SAV distributions in shallow, coastal regions where frequently changing, wind-driven currents yield high, persistent turbidity. Therefore, it would be valuable to design an approach that could be used for large-scale water body mapping and monitoring of SAV in these turbid coastal areas.

One approach is to use high-resolution images such as Digital Globe's Quickbird and Worldview-2 satellite imagery. The two sensors currently have the highest commercially available spatial resolution (2.44 m and 2.0 m multispectral respectively) and possess the capability of synoptically capturing large areas within a single image (272 km^2). Although Quickbird and Worldview-2, like Landsat before them, were primarily designed for detecting land features, recent improvements have provided improved spatial and spectral resolutions in aquatic systems (Curran 2011, Dogan 2009). Nevertheless, satellite remote sensing of SAV has remained poorly studied in comparison to terrestrial and emergent vegetation because of the difficulties inherent in interpreting reflectance values within water (Lehmann et al. 1997). For example, terrestrial remote sensing does not require correcting for scatter or

absorption of light in the water column. Additionally, turbid water often contains appreciable suspended sediment and other constituents that also scatter or absorb light. As a result, researchers typically have used remotely sensed data to detect emergent or floating vegetation, rather than SAV (see Baschuk et al. 2012, Albright and Ode 2011, Midwood and Chow-Frasier 2010). Improvements in spatial resolution are providing greater differentiation in the sample scene, whereas advancements in spectral resolution may make it possible to discern spectral properties of submersed plants that previously could not be identified.

Water characteristics and quality may also need to be taken into consideration when attempting to remotely sense SAV. For decades remotely sensed images have been used to measure characteristics such as chlorophyll, water depth, Secchi depth transparency, and suspended sediment concentrations (Nelson et al. 2003, Khorram and Cheshire 1985), all of which may influence SAV detection. Coastal waters like the Currituck Sound can vary widely in several of these water quality characteristics, which in turn can influence how well ~~aquatic~~ SAV can be characterized with remote sensing. Characteristics such as turbidity and water depth may influence the sensor's ability to detect SAV, making it necessary to incorporate such factors into predictive models for SAV detection. In past research focusing on much smaller water bodies, water depth was successfully incorporated into models to detect SAV using sensors such as Landsat (Nelson et al. 2006). Tremendous potential still exists in the capacity of high-resolution satellite imagery to detect SAV over larger coastal regions.

The objectives in this study were: (1) to determine whether different levels of aquatic plant cover could be detected using the commercially available Quickbird and Worldview-2 satellite sensors or free LANDSAT 5 data and (2) to assess whether predictions of SAV abundance and distribution can be improved by considering environmental characteristics (Secchi disk depth, salinity, sediment type, and water depth) and water quality (total nitrogen, total phosphorus, etc) in the models.

2.2 Materials and Methods

2.2.1 Study area

Currituck Sound is located in the northeastern-most corner of North Carolina and forms the northern arm of the Albemarle-Pamlico Estuary System (APES), the second largest estuary on the United States mainland (McKay et al. 2012), thus making it one of the most important wildland habitats in the nation (Figure 1).This shallow embayment has a surface area of 39,600 ha (396 km^2), a mean depth of 1.6 m and maximum depth of approximately 3.96 m (McKay et al. 2012). The sound's shallow depths, low salinity and large expanse make nearly every centimeter of the water body accessible to SAV establishment (Thayer et al. 1984, Street et al. 2005). The Sound stretches approximately 48 km from north to south and 5 to 13 km from east to west. On its northernmost end the Sound extends to Back Bay, Virginia and into the Albemarle-Chesapeake Canal. To the south, it joins the Albemarle Sound and the rest of the APES system. The freshwater inputs to Currituck Sound include North Landing River and Northwest River, both with headwaters in the Great Dismal Swamp of North Carolina. Back Bay also contributes water (both salt and fresh) into the Sound

through shallow water channels. Inputs of brackish water from federal canals also might influence the salinity of Currituck Sound. The sound is separated from the Atlantic Ocean by a narrow strip of barrier islands known as the outer banks which are no more than a mile wide. Unlike many other sounds which are tidally driven, water level fluctuations in Currituck Sound are a product of the constantly changing wind. Thus, water level can fluctuate wildly from week to week, even from day to day during changing weather conditions. The Sound stretches through two counties, Dare and Currituck, with level or slightly sloping terrain.

The SAV survey area spans the mid portion of the Currituck Sound encompassing the Currituck County mainland and Outer Banks as well as the Dare County Outer Banks (Figure 1). The study area is approximately 21 km long by 8 km wide, stretching from just south of the towns of Corolla and Duck, NC on the eastern side and Parker's creek to Webster's creek on the western side.

2.2.2. SAV sampling

Currituck Sound was sampled three times during the growing season of many SAV (June –September 2010). SAV sample sites were sampled using a modification of the point-intercept method (Madsen, 1999). The study area was overlain by a grid matrix wherein the centroids were converted into 174 points for use during each sample period: Survey 1 (June 14th, 2010- July 13th, 2010), Survey 2 (July 24th, 2010-August 7th, 2010) and Survey 3 (September 3rd, 2010-September 6th, 2010). Based on preliminary examinations, survey points with an initial depth of 3 m or greater identified in Survey 1 were considered too deep for plant growth or detection from sensors. Also, grid points found to be terrestrial were

removed as SAV would be unable to establish growth on such features. Therefore, only points in the littoral zone of the Sound remained for survey 2 (N = 117). Survey 3 consisted of 41 points within the littoral zone area. All survey points were located in the field using a Magellan Mobile Mapper CX professional grade GPS unit with sub-meter positional accuracy. At each point, water depth was measured to sediment using a marked depth pole, and plant metrics were assessed by recording plant presence and plant cover at each site. This was accomplished by assigning an associated level of plant coverage for each category. Plant cover was assessed at each point for an area of 10 m x 10 m by using a two-sided sampling rake thrown in four cardinal directions from the point of anchor. A two-sided rake is a widely accepted survey method for assessing plant presence/absence, wherein the rake is thrown and drug along the bottom to retrieve plant material (Madsen 1999). A locational error of +/- 1.52 m was estimated through frequent repositioning.

Plant cover levels were initially separated into 10% field interval categories ranging from 0 (0%) to a level of 10 (91-100%). These levels were then combined in the lab to represent four levels most likely to be discernible by each sensor: 0 (0–20% plant cover), 1 (21–40% plant cover), 2 (41–80% plant cover), and 3 (81–100% plant cover). An additional binomial category of total littoral zone plant cover was developed by combining the four levels recorded for each plant category at each point. This category captured littoral plant presence or absence at each point by assigning each site either a 0 (0–20% plant cover) or a 1 (21–100% plant cover). All values of plant cover less than 20% were considered to be undetectable by the sensors in North Carolina coastal waters (Curran 2011, Nelson et al. 2006), and were therefore assigned a value of “0” or absence. To assess sensor ability to

detect different levels of SAV coverage, a littoral percent plant cover was calculated as the total number of points sampled with any plant category greater than level 0 (i.e., >1% cover at an individual site), divided by the total number of points in the littoral zone. These values were used to estimate the applicability of using literature defined thresholds for each sensor. In turbid North Carolina coastal bodies of water, SAV is unable to typically establish or grow in depths greater than 1.83 m due to inadequate light (Ferguson and Korfmacher 1997, Kenworthy and Haunert 1991). Thus, areas greater than 2 m in depth were deemed "pelagic."

2.2.3. Water Quality Characteristics

Water clarity was estimated using a 20-cm diameter Secchi disk. Secchi depth was determined by averaging two measurements taken over the shady side of the boat during SAV sampling. Pelagic water samples were taken from the deepest area of the study area (sample point 169 = 3.2m) directly adjacent to multiple aggregated sampling areas for comparison. A temperature profile was also established using an onboard thermometer during all SAV sampling. Salt content of the water was measured to investigate salinity in the sound with the use of a handheld refractometer by taking the mean of four readings over the side of the boat. Finally, sediment type estimates were developed by collecting samples during each SAV survey in the littoral zone area of the aggregated sampling areas with a bottom grab from directly under the boat. Bottom sediments were categorized into ten different types that represent identified bottom sediment texture including clay, clay loam, loam, loamy sand, sand, sandy clay loam, sandy loam, silt, silt clay, and silt loam.

For water quality estimations, a representative dataset was developed from which to test water quality. This representative dataset was then interpolated to provide water quality at each SAV sample point. Water quality parameters identified below were tested between SAV sampling runs: Water Quality sampling 1 (July 8th, 2010 – July 23rd, 2010) and Water Quality sampling 2 (August 8th, 2010 – August 14th, 2010). Water quality parameters were estimated with the use of a field spectrophotometer. Measures of water quality included total nitrogen (TN), total phosphorus (TP), ammonia nitrogen, nitrate-N, color, dissolved oxygen (DO), nitrite-N, Phosphate-P, and pH. DO, pH, and color were all derived in the field using procedures designated for testing by LaMotte (2000**)**. All other samples were collected into 950 ml sampling containers, preserved using procedures specified by LaMotte (2000), transported to the lab in complete darkness, packed in ice, and processed the same day as collection.

2.2.4. Satellite imagery

Quickbird satellite imagery (2.44 m) and Worldview-2 imagery (2.0m) were acquired from Digital globe and LANDSAT 5 imagery for the entire Currituck Sound study area, and were matched to the SAV and water quality samples. Image rectification and geoprocessing were conducted using ERDAS Imagine 2010 image processing software (Intergraph 2010). Quickbird and Worldview-2 images were available in a georectified format. However, visual inspection showed that further georeferencing was necessary for some images. Images taken on September 13th, 2010 and August 5th, 2010 were georeferenced to the Worldview-2 image collected on July 22nd, 2010. All imagery was georeferenced using a 1st order polynomial transformation with no less than 10 ground control points. Error was minimized to less than

1 pixel per transformation. All imagery was inspected for atmospheric differences occurring between scenes and histogram matching was completed when necessary. Due to the high occurrence of clouds in most images, a cloud removal masking technique was required to remove all pixels containing clouds or cloud shadows. All land features were masked out using similar techniques. Data points lying within pixels containing clouds, shadows or other interference were subsequently removed from the dataset before statistical analyses were performed. Dark object subtraction was used in an attempt to adjust for further atmospheric correction (Teillet and Fedosejevs 1995).

The spectral pixel values or digital number (DN) values for all single pixels containing the position of each sample point, using the field-recorded GPS coordinates, were extracted using ESRI ArcMap 10.0 (ESRI 2011). Digital Number values were extracted from each individual scene and combined with all SAV sampling data into one database file. Spectral DN values for the pelagic region were also extracted to analyze the relationship between pelagic zone sound characteristics and spectral values. Because the pelagic zone is more homogeneous than the littoral zone, the spectral DN values for all pixels within the pelagic zone of the aggregated sampling areas were averaged resulting in one pelagic spectral value. Spectral and spatial properties of each sensor used are included in Tables 1, 2, and 3.

Because of the high spatial resolution of the Worldview-2 and Quickbird sensors, it was necessary to resample pixel size to more closely match the resolution of SAV survey sites (10m X 10m). Resampling was achieved using a bilinear interpolation in ESRI ArcMap 10.0 (ESRI 2011). LANDSAT 5 DN extraction was based solely on the single pixel containing the GPS sample point, since LANDSAT imagery is of much coarser spatial

resolution (30m) than both Worldview-2 and Quickbird.

2.2.5. Statistical analysis

Survey point extractions were analyzed for outliers and determination of outliers was completed using visual and statistical inspection of each DN value at each point. Any DN value identified as an outlier (> 1.5 x IQR) in SAS Enterprise Guide 4.2 (SAS 2009) was ultimately inspected for atmospheric interference and removed upon confirmation. Points removed were deemed unusable in model development due to the high degree of influence from sources outside of the target. This procedure was completed for each image/ SAV sampling dataset combination. All spectral digital number values were independently and statistically evaluated for interference from atmospheric or sensor defects before attempting to develop a spectral model data set for each sensor.

The satellite imagery DN values and SAV data were analyzed using binomial and multinomial logistic regression (logit models) in SAS Enterprise Guide 4.2 (SAS 2009). Stepwise and best-subset regression techniques were used to fit individual and combined spectral bands to the sample data.

All image/SAV combined sampling data points not eliminated during outlier detection were included in all logit models for each sensor and combined models. The multinomial and binomial categories for plant cover and plant presence absence served as individual response variables for each logit model. The logit model uses the explanatory and interaction covariates to predict the probability that the response variable will take on a given value (SAS Institute Inc., 1995).

For binomial logistic regression, the logit model indicated how the explanatory variable (DN values by band) affects the probability of the event (SAV presence/absence) being observed versus not being observed. For the multinomial logistic regression, probable outcomes of observations were calculated by analyzing a series of binomial sub models that represented the overall ability of the model to predict each of the plant cover response variables. For all logit model analyses, the descending option was used to select the highest plant category level as the response variable reference (level 3 for plant cover and level 1 for littoral plant presence/absence). This selection ensured that the results will be based on the probabilities of modeling an event (SAV present), rather than a non-event (SAV not present).

Model fit was determined by examining the percent concordant values, the Wald test statistic, likelihood ratio, and score test. The percent concordant values provided an indication of overall model quality through the association of predicted probabilities and observed responses. These values were based on the maximum likelihood estimation of the percent of paired observations of which values differed from the response variable (Kleinbaum 1994). Thus, the higher the predicted event probability of the larger response variable (based on the highest plant category level), the greater the percent concordant value. The Chi-square level of significance for the Wald test statistic, Likelihood ratio and score tests were used to test the hypothesis that the coefficients of the independent variables were significantly different from zero. This was done by fitting the model using the intercept terms (Kleinbaum, 1994; Pampel, 2000). Hosmer and Lemeshow Goodness of fit was used to determine the overall model fit and applicability; it tests the null hypothesis that the data are generated by the model fitted by the researcher. The test divides subjects into deciles based

on predicted probabilities, and then computes a chi-square from observed and expected frequencies (Hosmer and Lemeshow 2000). Then a probability (p) value is computed from the chi-square distribution with 8 degrees of freedom to test the fit of the logistic model. If the Hosmer and Lemeshow Goodness-of-Fit test statistic is 0.05 or less, the null hypothesis is accepted that there is no difference between the observed and model-predicted values of the dependent. (This means the model predicts values significantly different from observed values.) If the Hosmer and Lemeshow Goodness-of-Fit test statistic is greater than 0.05, then the null hypothesis cannot be rejected - there is no difference, implying that the model's estimates fit the data at an acceptable level.

To examine whether there were significant differences between data obtained from multiple images of the same sensor, individual-image logit models were developed. The model output and model coefficients from each image were compared using a two sample t-test to test for differences between the means of the model coefficients (log transformed). Resulting p-values for the paired variance and significance were determined at the 0.05 α level. Insignificant results from these tests suggest that the means of the individual image data show no significant difference. The means of the percent concordant values from the individual image data were then compared. In this analysis, the absence of large differences between the data percent concordant values was used to support the validity of creating a sensor specific model across multiple images.

Logit models for individual images were used to examine whether various water quality characteristics helped improve predictions of SAV cover using Quickbird, Worldview-2 and LANDSAT 5 imagery. Ordinary least squares regression was used to regress each of the model coefficients from the individual image logit models against each of the measured water quality characteristics individually: Secchi depth, water depth, salinity, water temperature, sediment type, TN, TP, NH_4^+N, nitrate-N, color, DO, nitrite-N, Phosphate-P, and pH.

2.2.6 LOGIT Model Calibration

Model calibration was accomplished using previously selected points from each SAV sampling run not used in model development. Results derived using these datasets were then compared to results developed using SAV sampling data during model development only. The calibration was made by investigating point specific logit predictions and comparing them to point-specific SAV sampling data of actual groundtruthed data. The logit values represented the cumulative probability of each sample point being each plant cover level (0, 1, 2, and 3) or littoral SAV presence (0 or 1) within each plant category. The cumulative probability value of the logit was used to calculate the actual probability of each sample point being each plant cover level or plant presence/absence category. The actual probabilities were then averaged to determine the overall probability of sample points belonging in each plant cover level and plant category.

2.2.7. Accuracy Assessment of LOGIT models

Typical measures of error in models are “omission” and “commission” errors. These measures are most often used to determine how well a model corresponds with groundtruthed data at the same location. For this study, we compared rake sample results to the sensor derived LOGIT models as an additional means for evaluating model results. We used two classes to develop error estimates of correspondence between field and sensor derived presence/absence category data: ‘SAV’ (for indication of SAV presence) or ‘no SAV’ (for no indication of SAV presence). We used four classes to develop error estimates of correspondence between field and sensor derived percent cover category data: ‘No Cover’ (for indication of <20% coverage), ‘Low Cover’ (for indication of 21-40% coverage), ‘Medium Cover’ (for indication of 41-80% coverage) and ‘High Cover’ (for 81-100% coverage).

The omission error was calculated as the proportion of points where survey data showed SAV presence, yet models predicted no SAV presence. A high omission error suggests that the sensor derived model falsely identified the absence of SAV that was present at that location, or a false negative. The commission error was calculated as the proportion of points where survey data showed SAV absence, yet models predicted SAV presence. A high commission error suggests that the sensor derived model falsely identified the presence of SAV that was absent at that location, or a false positive.

2.3. Results

2.3.1. SAV cover in the Currituck Sound

The average Secchi depth in the study area was 0.42 m throughout the sample period. Water quality and other environmental conditions are summarized in Table 4. During summer sampling, SAV was found to be present on average at 47% of all points sampled, and the majority of all points (53%) were devoid of plants. Although nearly half of all points sampled were vegetated, only 22% of the SAV was in classes designated as "detectable" through remote sensing (> 20% coverage). Almost half of all vegetated points fell below this 20% threshold (24%). The category 1 level (21-40%) comprised 11% of all vegetated points, while levels 2 (41-80%) and 3 (81-100%) represented 8% and 3% of all vegetated points, respectively (Figure 2). Thus, SAV distribution within the Currituck Sound exhibited a highly patchy distribution which further complicated detection with remote sensing. Six SAV species were identified during summer sampling, including five native species as *Ruppia maritima* (widgeon grass), *Najas guadalupensis* (southern naiad), *Stuckenia pectinata* (Sago pondweed), *Vallisneria americana* (wild celery), *Potamogeton perfoliatus* (redhead grass) and the invasive species, *Myriophyllum spicatum* (Eurasian watermilfoil). All six identified species previously have been identified in Currituck Sound (Sincock 1966). Of points with SAV present, *Ruppia maritima* was most widely distributed at 87% of vegetated points, followed by *Stuckenia pectinata* at 61% of points, *Najas guadalupensis* at 43% of points, *Myriophyllum spicatum* at 35% of points, *Potamogeton perfoliatus* at 6% of points and *Vallisneria americana* at 5% of points. The deepest point sampled was 3.2 m, and most plants were found at ~1.31m. No plants were detected at a depth greater than 2.89 m.

2.3.2. LOGIT Model Results

Sensor-derived logistical regression models were developed for SAV presence or absence. However, no models could be developed for the multinomial variable (plant cover) due to a low ratio of events to non-events. In the case of the multinomial variable (plant cover), an event was defined as any plant cover category 1-3 which indicated some type of SAV presence. A non-event was defined as the plant cover category 0 which indicated no plant presence. The automated stepwise selection method led to the final, most reasonable model as decided upon in the best-subset procedure for the regression analysis. For a variable to enter into or remain in the model, a p-value of < 0.01 was necessary. A model was considered fit if the Hosmer and Lemeshow Goodness-of-Fit test yielded an insignificant difference in groups ($p > 0.05$). Sensor specific models were developed for both the Quickbird and Worldview-2 sensors, but LANDSAT 5 models proved to be inconclusive. Odds ratios were used to evaluate relative influence of variables selected in the final models. Prediction maps were developed to display 3 categories: correct prediction (prediction = observation), false positives (prediction = 1, observation = 0), or false negatives (prediction = 0, observation = 1).

Several environmental characteristics were highly correlated with SAV presence/absence, but only Secchi depth and water depth improved actual model predictions. Worldview-2 derived models yielded the highest percent concordant value for correct classification of SAV presence or absence with both the sensor specific average (81.3%) and the image specific (91.6%). Quickbird derived models yielded a percent concordant of 73.1% (Table 5).

2.3.2.1 Worldview-2

LOGIT model results suggested that the Worldview-2 sensor provided the best predictive model of the binary predictor variable (presence/absence), with percent concordant values between 67.9 and 94.7% and Wald, Likelihood and Score values of < 0.0001 each. Three variables were included in the Worldview-2 sensor specific prediction model (Table 6). The most influential predictor variable for the Worldview-2 sensor-specific model was the interaction between band 4 and Secchi depth, followed by the interaction between band 3 and Secchi depth, band 4 alone and band 3 alone. The negative β coefficient for band 4 alone was consistent with knowledge of the reflective properties of submersed plants in wavelengths from 700 to 1100 nm (Klancik et al. 2012). The negative β coefficient associated with the interaction of band 3 and Secchi depth was consistent with both the reflective properties of plants in wavelengths from 600 to 700 nm and the fact that light penetration decreases as Secchi depth increases. The positive associations with band 3 alone and the interaction between band 4 and Secchi depth were also consistent with reflective properties of plants. Regarding the best image provided for the Worldview-2 sensor (August 5th, 2010), model outputs demonstrated only 4 false negatives (observation dataset = presence, prediction dataset = absence) and 3 false positives (observation dataset = absence, prediction dataset = presence).

Due to the poor quality of the Worldview-2 image taken on July 22nd, 2010, an image- specific model was developed for the best Worldview-2 image (August 5th, 2010) to test for any difference. The image-specific model contained only two of the original three variables that were used for prediction in the Worldview-2 sensor specific model.

This image-specific model suggested that only the interaction between band 4 and Secchi depth was the best predictor for the model. This image-specific model yielded a percent concordant value of between 88.5% and 94.7% and a Wald, Likelihood and Score values of <0.001. The positive β coefficient association with the interaction between band 4 and Secchi depth was consistent with knowledge of the reflective properties of SAV from 700 to 1100 nm.

Parameter estimates for each Worldview-2 model are included in Table 7 for the sensor- specific model and in Table 8 for the image-specific model. Prediction output examples for the proposed Worldview-2 sensor derived model and the image-specific model (August 5th, 2010 dataset) are shown in Figure 3 and Figure 4, respectively. For comparison of prediction output to actual groundtruth estimates, SAV percent cover was overlain for each image/model combination.

2.3.2.2. Quickbird

The Quickbird sensor-derived predictive model yielded a percent concordant value of 73.1% with a Wald of 0.04, Score of 0.0097 and a Likelihood ratio of 0.0175. The most influential predictor variable was band 3 alone, followed by the interaction of band 2 and Secchi depth, band 3 and Secchi depth, band 2 alone, and a small influence provided by the interaction between band 4 and depth. The positive β coefficient for band 3 alone was consistent with knowledge of the reflective properties of submersed plants in wavelengths from 600 to 700 nm. The negative β coefficient associated with band 2 alone was consistent with knowledge of the reflective properties of submersed plants in wavelengths from 520 to

600 nm. The positive β coefficient for the interaction between band 2 and Secchi depth was consistent with knowledge of the reflective properties of submersed plants in wavelengths from 520 to 600 nm and the fact that light penetration decreases as Secchi depth increases. The negative β coefficient associated with the interaction of band 3 and Secchi depth was consistent with both the reflective properties of plants in wavelengths from 600 to 700 nm and the fact that light penetration decreases as Secchi depth increases. Lastly, the negative β coefficient associated with the interaction of band 4 and depth was consistent with knowledge of submersed plant reflection in wavelengths from 700 to 1000 nm and the fact that light penetration decreases as depth increases. Parameter estimates for the Quickbird-derived model are included in Table 9. The sensor-specific model prediction output for the Quickbird sensor is shown in Figure 5.

2.3.2.3. LANDSAT5

The LANDSAT-5-derived models yielded inconsistent results, often varying from scene to scene, and suggested various bands that have not historically been associated with plant reflectance. Although various images were available for analysis, many were affected by clouds, atmospheric haze, and sun-glint.

2.3.3. Accuracy Assessment of Models

Accuracy assessments were completed for derived models from each sensor, and are summarized in Tables 10 and 11.

4. Discussion

Remote sensing in Currituck Sound provides some potential for inventorying the distribution of SAV, but a number of limitations still exist and will be discussed in the following section. Overall, the Worldview-2 sensor provided the best predictive model of SAV presence or absence. A percent concordant value greater than 80% has been loosely recognized as an indicator of strong model agreement when developing predictive models that analyze the presence of SAV (Nelson et al 2006).

Models derived from the Worldview-2 sensor produced higher predictive capacity based on the groundtruthed field data in the Sound. Image and sensor specific models when applied to the August 5th, 2010 image provided percent concordant values of 81.3 to 91.6%. However, accuracy assessments of both the sensor specific and image specific Worldview-2 models showed relatively large commission and omission errors. False positives using the Worldview-2 derived models, although few, most often occurred in areas of shallow depth (<0.9 m) and could have been caused by bottom reflectance. However, four out of the seven false positives that occurred across all images with the Worldview-2 sensor took place at points with at least some SAV present (1-20%). The > 20% coverage threshold used to develop our models designated these points as “absent” of SAV, because these data points fell below the imposed definition of the threshold for SAV presence. The higher percent concordant value and lower rates of error suggest that the Worldview-2 image specific model out-performed other sensor models for detecting SAV in Currituck Sound. The sensor-specific model was influenced by the Worldview-2 image captured on July 22, 2010. This

image appeared very pixilated, having been acquired on a day of high wind and wave action. False negatives most often occurred in areas of low to moderate plant density (< 40%) or in areas of deeper water (> 0.9 m). Perhaps spectral contribution of SAV was inhibited by greater distances between plant canopy and water surface or high turbidity. Lower densities of SAV may also not provide an adequate spectral contribution to be differentiated from the water column alone. Patchy distributions of SAV, common in Currituck Sound, may also have contributed to false negatives. For example, SAV coverage in the survey area may not have covered an adequate amount of each pixel area for detection.

The Quickbird sensor provided poor results with a total of 10 false negatives and 1 false positive, and a percent concordant value of 73.1%. Unfortunately, one of the difficulties encountered with development of the Quickbird model was the lag time between image acquisition and field sampling (+ 36 days). This lag time prevented acquisition of near-coincidental satellite and field sampling, which may have contributed to limitations in model development; there were also large amounts of cloud cover in the Quickbird image. Most false negatives in the image were located along a large cloud extending from the southern to northern reach of the study area. Atmospheric interference inherent around clouds can severely alter the spectral signature of the target (Levin and Levina 2007), thus leaving even points not directly within the cloud at risk for atmospheric influences.

Use of the free LANDSAT 5 data proved to be inconclusive. Extreme obscurity from clouds within each scene made virtually all data points unusable. The more coarse spatial resolution of LANDSAT-5 imagery also hindered attempts to contain only plant growth

within individual pixels. Pixels likely contained plant matter but were potentially subject to complicated optical effects in very shallow waters, i.e. substratum reflectance contributed to tainted spectral reflectance. Several bands not historically associated with plant reflectance were potentially important, but were inconsistent. For example, band 5 and 7 (SWIR1 and SWIR2) were often identified as important. Given that these wavelengths do not penetrate water (Poole and Atkins 1926), these observations may have been related to spectral mixing of land or emergent vegetation features not sampled within the images large pixel size (30mx30m).

Although three models were developed, common variables emerged from each Worldview-2 model as well as the Quickbird Model - Band 4 and Secchi Depth. These two predictor variables were used in each model and should be investigated first in future attempts at remote sensing of SAV with the Worldview-2 sensor. The reflective properties of plants in the Near-Infrared portion of the electromagnetic spectrum have been cited in numerous papers regarding aquatic plants (e.g. Klancnik 2012, Nelson 2006), but water readily absorbs light in the NIR. This most likely limits the effective depth of Band 4 in most sensors to less than 1m in depth. Remote sensing of SAV using the NIR band, in this case, most likely detects plant material at shallow sites that are very close to the water surface.

The NIR portion of the spectrum should always be considered when remote sensing of SAVs is desired with the Worldview-2 sensor, and depth of water and height of vegetation canopy should be monitored closely. The spatial resolution of the Worldview-2 sensor also

made it possible to more adequately capture the sample area from which the groundtruth data were based. This became the major limitation to using freely available LANDSAT 5 data. The sensors 30-m spatial resolution most likely allowed spectral contribution from sources other than the target SAV (i.e. water-column constituents, turbidity, bottom reflectance, etc.). Although LANDSAT 5 imagery have been used in past attempts to remote sense SAV, the patchy distribution of SAV in Currituck Sound is likely the reason for the sensor's poorer performance in this attempt to apply it for remote sensing of SAV presence/absence.

Complications arose using all sensors due to the low occurrence of events to non-events in the groundtruthed data making it difficult to satisfy general assumptions for applying logit models. Logistic regressions are improved when features of interest, in this case SAV presence, constitute a third of the total number of sample sites (Harrell 1997). Development of predictive models was for SAV presence, as this feature ("event") comprised at least one-third of all observed points. In the case of SAV percent coverage, the occurrence of events to non-events per class was low (see Figure 2). SAV density was inversely proportional to the number of points per coverage class. The areas most likely to be detected by remote sensing (Class 3: 81-100%) occurred least often. This made modeling of SAV percent coverage difficult, given the lack of dense stands of SAV in the Sound. Thus, increasing the sample size of SAV points within Currituck Sound may actually lead to an increase ratio of non-events to events. Remote sensing of SAV in percent coverage classes with limited coverage in water bodies lacking substantial areas of higher percent coverage classes should be undertaken with caution.

Difficulties in image acquisition from commercial vendors led to less-than-ideal available acquisition dates. Originally, images were scheduled to be acquired as close temporally to the actual dates of field surveys as possible. Inability to do so especially applied to the Quickbird image acquired in mid-September, leaving Surveys 1 and 2 unusable for model development and therefore requiring last-minute data collection in a third survey. Survey 3 was not scheduled during preliminary planning, but was deemed necessary due to late data collection of the Quickbird sensor. Survey 3 was combined with existing Survey 2 data in attempts to develop a model for the Quickbird sensor. Actual acquisition dates and time between Survey 1 and image acquisition also left the utility of survey 1 as a groundtruthed dataset questionable. The first Worldview-2 image was acquired on July 22nd, 2010, 9 days past the last collection day for survey 1 (July 13th, 2010). This image was actually closer in acquisition date to the beginning of Survey 2 (July 24th, 2010 – 2 days). The second Worldview-2 image (August 5th, 2010) was acquired during Survey 2 (July 24th,2010 – August 7th, 2010). Given the highly variable water levels within the Sound, it was extremely important to collect very near incidental imagery alongside groundtruthed data points. In coastal systems like Currituck, water level can change in a matter of days, even hours, resulting in shallower or deeper plants or even semi-emergent plants that had previously been submersed. Future attempts to replicate or advance beyond this study should take special care to schedule and obtain imagery from dates as near-coincidental as possible to actual SAV sampling, and or should obtain water height data at time of acquisition to facilitate adjustment. Other options would be to consider using non-commercial remote sensors such as Sentinel 2 and LANDSAT-8.

SAV in North Carolina coastal waters, and worldwide, have shown sustained declines in recent years (Waycott et al. 2009, Burkholder et al. 2007, Orth et al. 2010), which should be of great concern to resource managers. To further complicate the issue, the decline of SAV has traditionally been difficult to reverse (Fonseca et al. 1998, Burkholder et al. 2007). SAV is a well-known indicator of ecologic state and change (Biber et al. 2004); therefore, new methodologies for mapping and monitoring this vital resource should be of paramount importance to resource managers to ensure the ecologic and economic sustainability of these valuable natural resources. Remote sensing using modeling techniques are not appropriate for all applications (i.e. highly turbid, low optical depth areas) when combined with the sensors described in this study. Nevertheless, this technique offers some capability in filling gaps for shallow coastal areas with submersed vegetation, such as the Currituck Sound.

2.5. References

Albright, T.P., and Ode, D.J., 2011. Monitoring the Dynamics of an Invasive Emergent Macrophyte Community using Operational Remote Sensing Data. Hydrobiologia 661.1, 469-74.

Baschuk, M.S., Ervin, M.D.,Clark, W.R., Armstrong, L.M., Wrubleski, D.A., Goldsborough, G.L.. 2012. Using Satellite Imagery to Assess Macrophyte Response to Water-Level Manipulations in the Saskatchewan River Delta, Manitoba. Wetlands 32.6, 1091-102.

Biber, P.D., Paerl, H.W., Gallegos, C.L., Kenworthy, W.J., Fonseca, M.S. 2004. Evaluating Indicators of Seagrass Stress to Light. in SA Bortone (ed.), Estuarine Indicators. CRC Press, Boca Raton. 193-209.

Burkholder JM, Tomasko DA, Touchette BW. 2007. Seagrasses and eutrophication. J Exp Mar Biol Ecol 350(1-2):46-72.

Chollett I and Mumby PJ. Predicting the distribution of montastraea reefs using wave exposure. Coral Reefs. 2012 JUN;31(2):493-503.

Curran, R.W. 2011. The Utility of Digital Globe's WorldView-2 Satellite Data

in Mapping Seagrass in North Carolina Estuaries. Unpublished results. Master's Thesis, East Carolina University.

Davis, G.J.; and M.M. Brinson. 1983. Trends in submerged macrophyte communities of the Currituck Sound: 1909-1979. Journal of Aquatic Plant Management 21, 83-87.

Dierssen HM, Zimmerman RC, Drake LA, Burdige D. Benthic ecology from space: Optics and net primary production in seagrass and benthic algae across the great bahama bank. Mar Ecol Prog Ser. 2010;411:1-15.

Dogan, O.K., Akyurek, Z., and Beklioglu, M. 2009. Identification and Mapping of Submerged Plants in a Shallow Lake using Quickbird Satellite Data. Journal of environmental management 90.7, 2138-43.

Ferguson, R.L., Korfmacher, K. 1997. Remote sensing and GIS analysis of seagrass meadows in North Carolina, USA. Aquatic Botany. 28, 241-258.

Ferguson, R.L.; and L.L. Wood. 1994. Rooted vascular aquatic beds in the Albemarle-Pamlico estuarine system. NMFS, NOAA, Beaufort, NC, Project No. 94-02 , 103.

Fonseca, M.S., Kenworthy, W.J., Thayer, G.W., 1998. Guidelines for the Conservation and Restoration of Seagrasses in the United States and Adjacent Waters. NOAA, Coastal Ocean Program, Decision Analysis Series No. 12. U.S. Department of Commerce, NOAA, Coastal Ocean Office, Silver Spring, MD. 222.

ESRI. 2011, ArcGIS Desktop: Release 10. Redlands, CA: Environmental Systems Research Institute.

Harrell, F. E. 1997. Predicting outcomes: applied survival analysis and logistic regression. University of Virginia Press. University of Virginia School Medicine, Charlottesville, Virginia.

Hosmer D.W. and Lemeshow S., 2000. Applied logistic regression, 2nd ed. New York: John Wiley &. Sons, Inc,. ISBN 0-471-35632-8.

Intergraph. 2010. ERDAS Imagine Desktop 2010. Madison, AL: Intergraph Geospatial.

Khorram, S., Cheshire, H.M., 1985. Remote sensing of water quality in the Neuse River Estuary North Carolina. Photogramm. Eng. Remote Sens. 51, 329–341.

Klancnik, K., Mlinar, M., Gaberscik, A., 2012. Heterophylly Results in a Variety of "Spectral Signatures" in Aquatic Plant Species. Aquatic Botany. 98.1, 20-6.

Kleinbaum, D.G., 1994. Logistic Regression A Self-learning Text. Springer-Verlag Publishing, New York.

LamottE. 2000. LaMotte Smart Spectro Operator's Manual. V3.0 and higher. 2000-01-MN

Larkum, A.W.D., Orth, R.J., Duarte, C., 2006. Seagrasses: Biology, Ecology and Conservation. Springer, The Netherlands. 691.

Lehmann, A., Jaquet, J.M., and Lachavanne, JB. 1997. A GIS Approach of Aquatic Plant Spatial Heterogeneity in Relation to Sediment and Depth Gradients, Lake Geneva, Switzerland. Aquatic Botany 58.3-4, 347-61.

Levin IM and Levina E. 2007. Effect of atmospheric interference and sensor noise in retrieval off optically active materials in the ocean by hyperspectral remote sensing. Appl Opt 46(28):6896-906.

Lyzenga DR, Malinas NR, Tanis FJ. Multispectral bathymetry using a simple physically based algorithm. IEEE Trans Geosci Remote Sens. 2006 AUG;44(8):2251-9.

Madsen, J.D., 1999. Point intercept and line intercept methods for aquatic plant management. Army Corps of Engineers Waterways Experiment Station Technical Note MI-02.

Marshall, T.R., Lee, P.F., 1994. Mapping aquatic macrophytes through digital image analysis of aerial photographs: assessment. J. Aquat. Plant Manage. 32, 61–66.

McKay, S. K., Wilson, C. R., Piatkowski, D. 2012. Currituck Sound Estuary Restoration: A Case Study in Objective Setting. EMRRP-EBA-17

Midwood, J.D., and Chow-Fraser, P. 2010. Mapping Floating and Emergent Aquatic Vegetation in Coastal Wetlands of Eastern Georgian Bay, Lake Huron, Canada.Wetlands 30.6, 1141-52.

Nelson, S.A.C., Cheruvelil,K.S. and Soranno, P.A. 2006. Satellite Remote Sensing of Freshwater Macrophytes and the Influence of Water Clarity. Aquatic Botany 85.4, 289-98.

Nelson, S.A.C., Soranno, P.A., Cheruvelil, K.S., Batzli, S.A., Skole, D.L., 2003. Regional assessment of lake water clarity using satellite remote sensing. J. Limnol. 62, (Suppl. 1), 27–32.

Orth, R.J. Marion, S.R., Moore, K.A., Wilcox, D.J., 2010. Eelgrass (Zostera marina L.) in the Chesapeake Bay region of mid-Atlantic coast of the USA: challenges in conservation and restoration. Estuaries and Coasts 33, 139-150.

Orth, R.J., Moore, K.A., 1983. Submerged vascular plants: techniques for analyzing their distribution and abundance. Mar. Technol. Soc. 17, 38–52.

Pampel, F.C., 2000. Logistic Regression: A Primer. Sage University Papers Series Quantitative Applications in the Social Sciences, series no. 07-132. Sage Publications, CA.

Poole HH and Atkins WRG. 1926. On the penetration of light into sea water. Jour Marine Biol Assoc 14((1)):177-98.

SAS Institute Inc. 2009. Administering SAS Enterprise Guide 4.2. Cary, NC: SAS Institute Inc.

Sincock, J.L. 1965. Back Bay – Currituck Sound data report. U.S. Fish and Wildlife Service, North Carolina Wildlife Resources Commission, and Virginia Commission of Game and Inland Fisheries. 1600.

Street, M.W., Deaton, A.S., Chappel, W.S., Mooreside, P.D., 2005. North Carolina Coastal Habitat Protection Plan, North Carolina Department of Environment and Natural Resources, Division of Marine Fisheries, Morehead City, NC. 656 pp. Online at http://www.ncfisheries.net/habitat/chpp2k5/_Complete%20CHPP.pdf

Thayer, G.W., W.J. Kenworthy, Fonseca, M.S., 1984. The ecology of eelgrass meadows of the Atlantic coast: a community profile. U.S. Fish and Wildlife Service. FWS/OBS-84/02. 147.

Teillet, PM and G Fedosejevs. 1995. On the dark object approach to atmospheric correction of remotely sensed data. Canadian Journal of Remote Sensing. 21:373-387.

Waycott, M., Duarte, C.M., Carruthers, T.J., Orth, R.J., Dennison, W.C., Olyarnik, S., Calladine, A., Fourqurean, J.W., Heck, K.L., Hughes, A.R., Kendrick, G.A., Kenworthy, W.J., Short, F.T, Williams, S.L., 2009. Accelerating loss of seagrasses across the globe threatens coastal ecosystems. Proceedings of the National Academy of Sciences 106, 12377-12381.

Valley, R.D., Drake, M.T., Anderson, C.S., 2005. Evaluation of alternative interpolation techniques for the mapping of remotely-sensed submersed vegetation abundance. Aquat. Bot. 81, 13–25.

Zhang, X., 1998. On the estimation of biomass of submerged vegetation using Landsat Thematic Mapper (TM) imagery: a case study of the Honghu Lake, PR China. Int. J. Remote Sens. 19, 11–20.

Table 2.1. Sensor specifications for spatial, spectral, radiometric and temporal resolution.

Sensor	Spatial (m)	Spectral (nm)	Radiometric (bits)	Temporal (days)	Bands
Worldview-2	1.8-2.4	450-1040	11	3.7	4*
Quickbird	2.44-2.88	450-900	11	3.5	4
LANDSAT-5	30 (120 Band 6)	450-1250	8	7	7

* Worldview-2 currently contains 4 additional bands that were not assessed during this study

Table 2.2. Multi-Spectral bands of the WorldVeiw-2 satellite sensor

Worldview-2				
Band	1	2	3	4
Name	Blue	Green	Red	NIR
Spectrum Width (nm)	450 - 510	510 - 580	630 - 690	860 - 1040

Table 2.3. Multi-Spectral bands of the Quickbird satellite sensor

Quickbird				
Band	1	2	3	4
Name	Blue	Green	Red	NIR
Spectrum Width (nm)	450 - 520	520 - 600	630 - 690	760 - 900

Table 2.4. Summary statistics of all water quality and condition parameters collected during summer field sampling.

Parameter	mean (SD*)	min	max
depth (m)*	1.73 (0.602)	0.335	3.2
Secchi (m)*	0.42 (0.093)	0.2	0.75
temperature (^{o}C)	29 (2.177)	22.22	32.33
Turbidity (NTU)	0.693 (0.541)	0.02	2
Ammonia-N (mg/L)	0.295 (0.094)	0.12	0.57
Color (cu)	327.28 (41.985)	184	405
DO (mg/L)	7.485 (0.934)	4.9	10.2
Nitrite-N (mg/L)	0.147 (0.042)	0.07	0.27
Nitrate-N (mg/L)	10.075 (1.664)	6	13
pH	7.378 (1.209)	5.7	9.2
Phosphate-P	0.015 (0.0163)	0	0.08
TN (mg/L)	1.075 (1.490)	0	8
TP (mg/L)	0.693 (0.541)	0.02	2
TN : TP	7.519 (19.353)	0	100

*Standard Deviation

Table 2.5. Percent concordant value for each presence/absence model developed. *WVSS = Worldview-II Sensor Specific, WVIS = Worldview-II Image Specific, QB = Quickbird

Model	Percent Concordant	No. of Usable Points
WV SS	81.3	196
WV IS	91.6	80
QB	73.1	90

Table 2.6. Analysis of Maximum Likelihood estimates for the Worldview-II sensor specific model applied to the August 5th, 2010 dataset.

Parameter	DF	Estimate	SE	Wald	Pr > Chi Sq
Intercept	1	-2.2755	0.7678	8.7843	0.003
B3	1	0.5497	2.1038	0.0683	0.7939
B4	1	-4.9392	3.0326	2.6527	0.1034
B3*SD	1	-5.4005	4.6811	1.3309	0.2486
B4*SD	1	21.6477	9.0837	5.6793	0.0172

Table 2.7. Parameter estimates for the Worldview-II sensor specific model applied to the July 22nd, 2010 dataset.

Parameter	DF	Estimate	SE	Wald	Pr > Chi Sq
Intercept	1	-1.8425	0.295	39.0086	< 0.0001
B3	1	19.1947	7.5488	6.4655	0.011
B4	1	-22.3063	7.8728	8.0278	0.0046
B3*SD	1	-46.3251	20.0271	5.3506	0.0207
B4*SD	1	53.8189	20.6317	6.8045	0.0091

Table 2.8. Parameter estimates for the best Worldview-II image specific model applied to the August 5th, 2010 dataset.

Parameter	DF	Estimate	SE	Wald	Pr > Chi Sq
Intercept	1	-2	0.4859	16.94	< 0.0001
B4*SD	1	5.8799	1.3345	19.4146	< 0.0001

Table 2.9. Analysis of Maximum Likelihood estimates for the Quickbird sensor specific model applied to the September 13th, 2010 dataset.

Parameter	DF	Estimate	SE	Wald	Pr > Chi Sq
Intercept	1	1.8684	0.3393	30.3157	<.0001
B2	1	-11.4288	5.4478	4.401	0.0359
B3	1	12.4367	5.373	5.3576	0.0206
B2*SD	1	31.7167	14.3848	4.8615	0.0275
B3*SD	1	-30.4846	14.0404	4.7141	0.0299
B4*D	1	-0.2457	0.1221	4.0476	0.0442

Table 2.10. Worldview-II accuracy assessments for both sensor and image specific models

Image (08/05/10) Specific Model				
		Omission	LOGIT Model	
			SAV	No SAV
Field Data	SAV	31.3%	11	5
	No SAV	4.7%	3	61
	Commission		21.4%	7.6%

Table 2.11. Quickbird accuracy assessments for sensor specific model.

Sensor Specific Model				
		Omission	LOGIT Model	
			SAV	No SAV
Field Data	SAV	62.5%	6	10
	No SAV	1.4%	1	73
	Commission		14.3%	12.0%

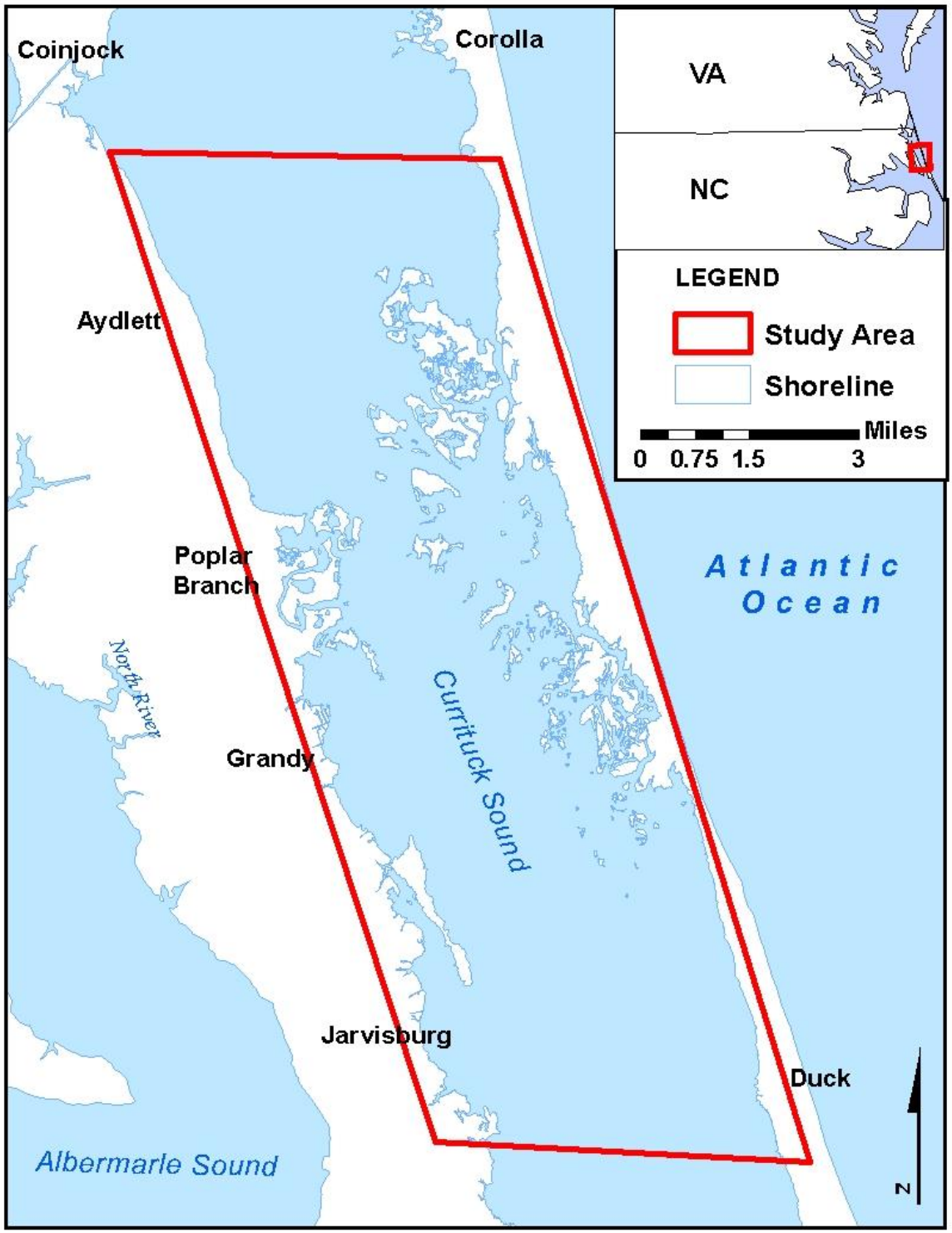

Figure 2.1. Study area for SAV sampling and remote sensing within the Currituck Sound making up the uppermost portion of the Albemarle-Pamlico Estuary System.

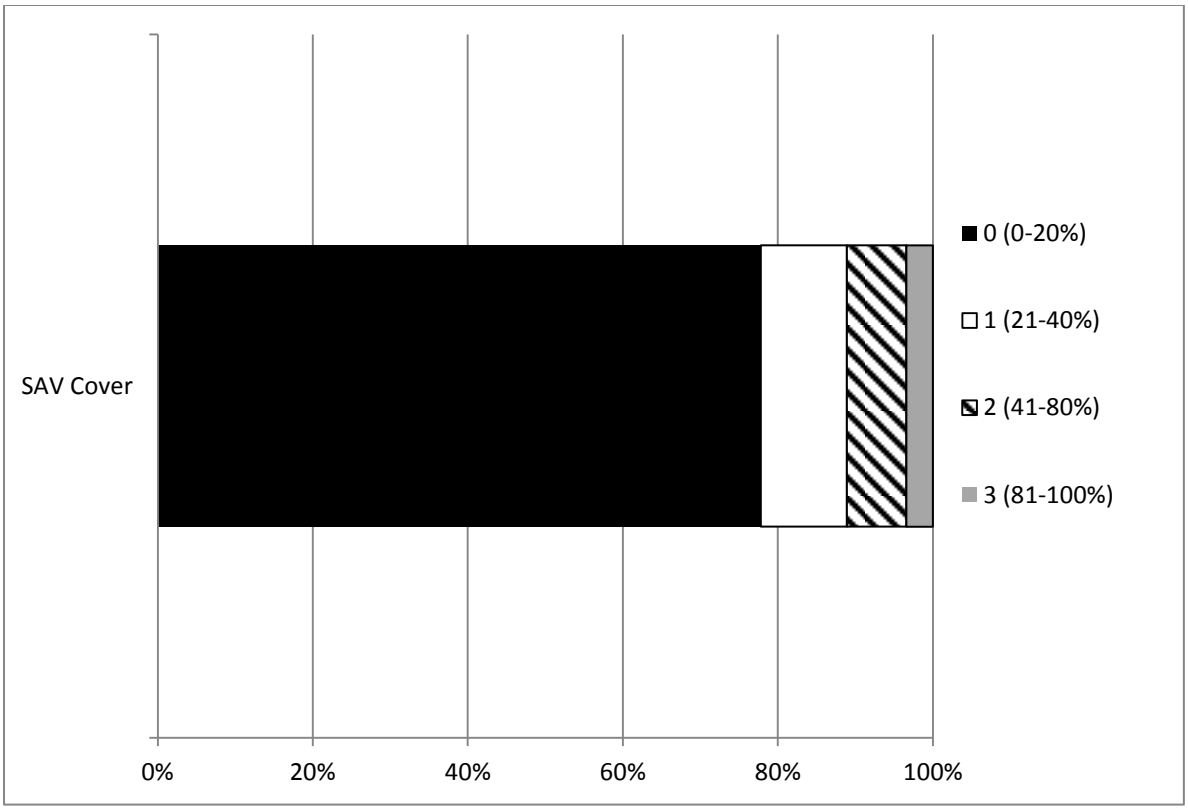

Figure 2.2. The distribution of plant cover levels throughout the Currituck Sound.

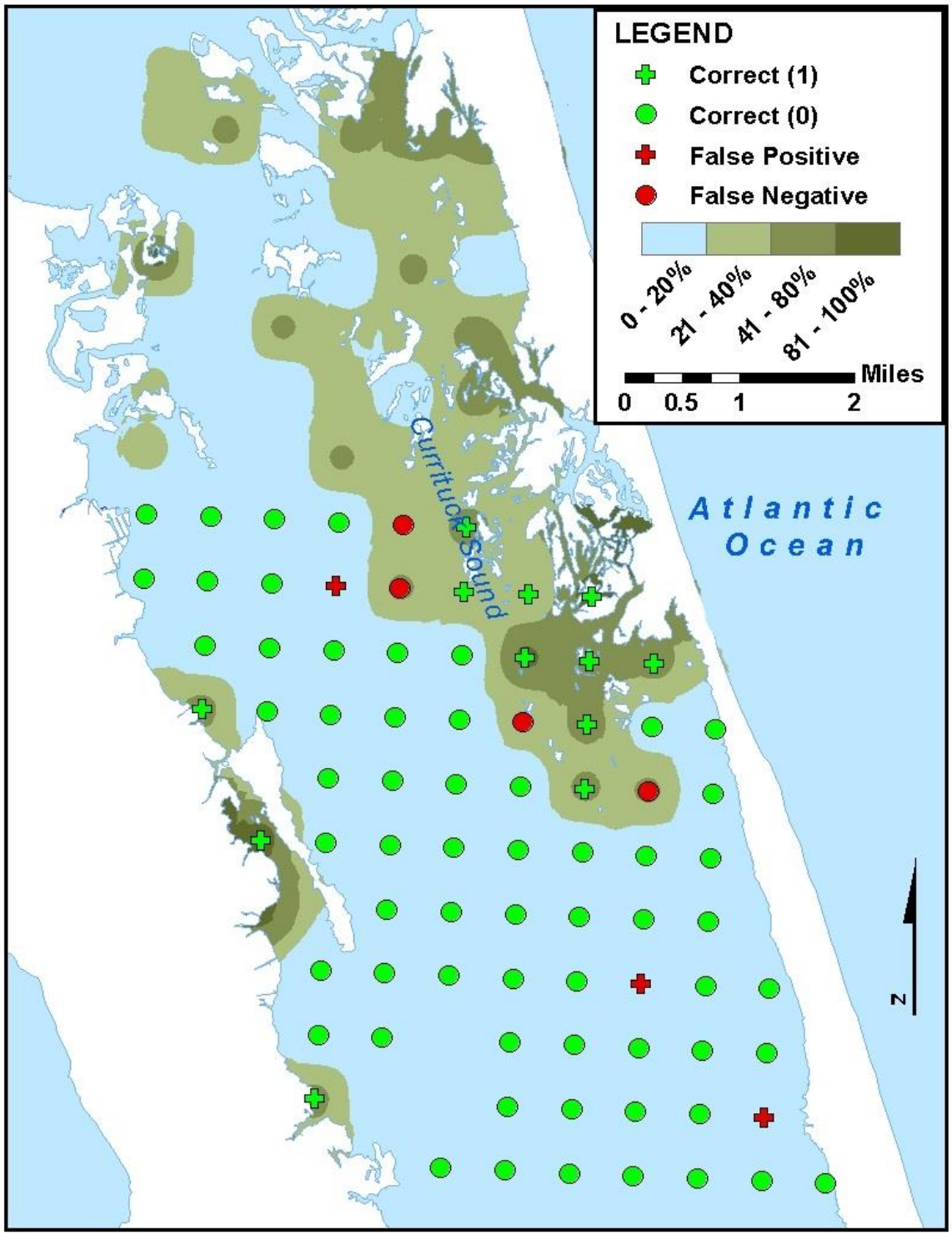

Figure 2.3. Worldview-II sensor specific model predictions applied to image taken on August 5th, 2010 and overlain with SAV percent cover estimations from survey 2.

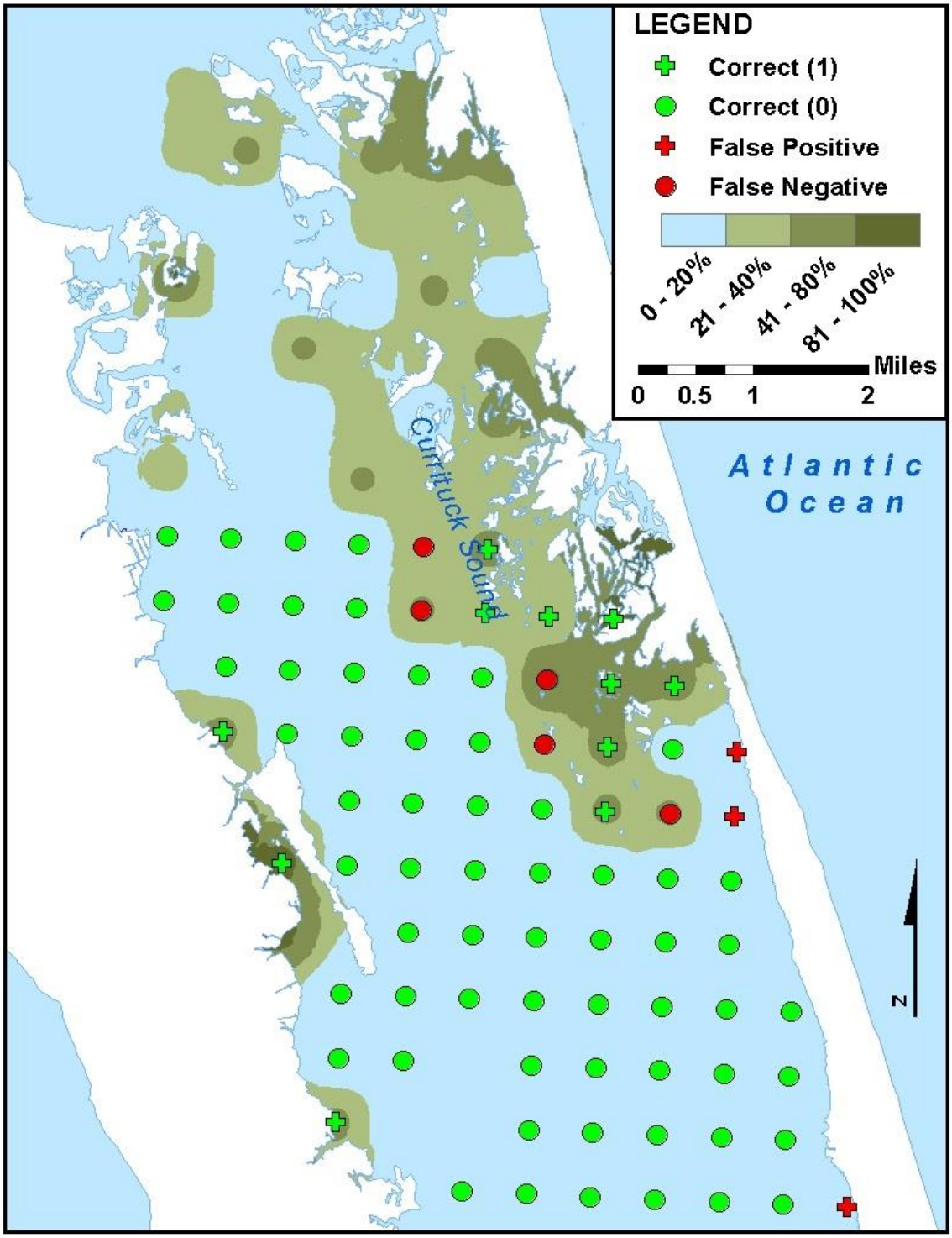

Figure 2.4. Worldview-II image specific model predictions applied to image taken on 08/05/10 and overlain with SAV percent cover predictions from survey 2.

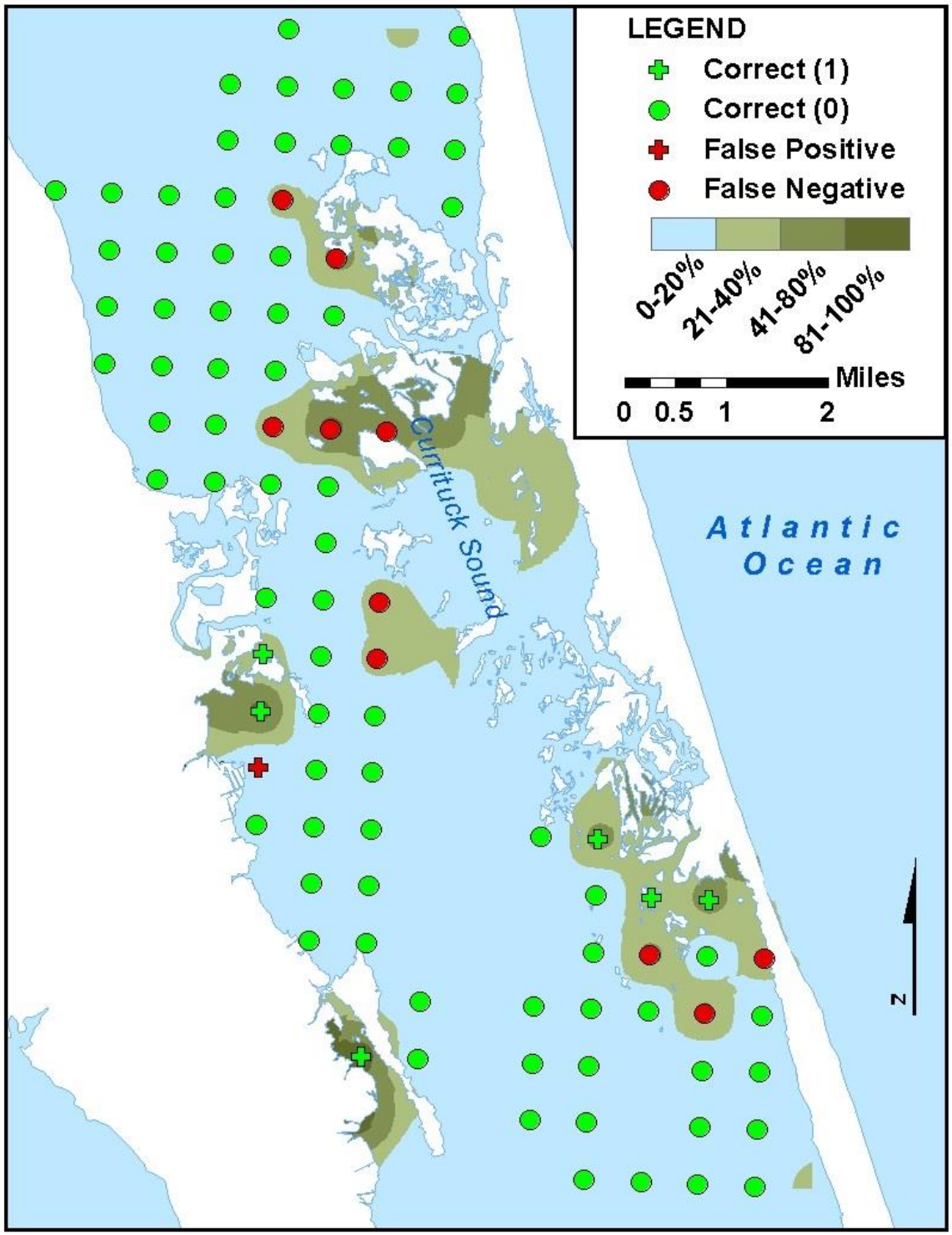

Figure 2.5. Quickbird sensor specific model predictions applied to image taken on July 13th, 2010 and overlain with SAV percent cover predictions from SAV survey 2.

CHAPTER 3

A Novel Technique for Mapping Submersed Aquatic Vegetation Species Dominance and Coverage in Mixed Stands of Large, Shallow Coastal Systems

3.1. Introduction

In recent years, several agencies have developed strategies for protection of existing native submersed aquatic vegetation (SAV) communities through restoration of historical growth areas in large coastal systems (Ailstock et. al 2010, Busch et al. 2010). Along with restoration, efforts to control invasive species are often employed to help restore native populations (Gordon 1998). While various mapping and survey techniques are often employed to aid in these efforts, each has limitations for identifying key information about the spatial extent and coverage of SAV species in large project areas common of coastal systems. For example, aerial photography has commonly been used to address overall SAV coverage in large coastal systems (Orth et al. 2010, Orth et al. 2007, Niedier 2004), but in reality it is often seriously limited by water clarity and depth and does not provide critically needed, species-specific information. Likewise, boat based visual estimations have provided information on the localized extent of SAV but do not provide information on the species present (Madsen and Bloomfield 1993). As in aerial photography, visual estimations can also be confounded by water clarity (Yin and Kreiling 2011). Traditional sampling techniques like random point, line and quadrat sampling can provide excellent information on species presence, but are extremely labor-intensive and only practical for small area assessments (Madsen and Wersal 2012). Issues inherent in all of the above mentioned

techniques are exacerbated in coastal systems with a seemingly endless and dynamic littoral zone. In such systems, a method to identify key areas for further exploration and survey should be employed to help narrow down management efforts. Understanding the dynamics of SAV populations in a water body has become increasingly important, especially with the encroachment of various invasive species that can severely alter community dynamics (Madsen and Wersal 2012). Furthermore, assessing individual species dominance and coverage within large stands of vegetation provides the detrimental information needed to make sound decisions for what types of management should be employed. Historic studies have often reported species dominance on a system-wide basis (McAtee 1919, Kerwin et al 1976, Davis and Carey 1981) rather than expressing quantitative aspects of spatial dominance within the study area. While informative in terms of presence/absence, such an approach provides no spatial reference to particular stand location or individual species dominance, heterogeneity or coverage which can be vital information when assessing plant communities. These features, particularly current species dominance and coverage, play a key role in efforts to restore and protect vital areas while monitoring the spatial location of invasive aquatic plant species (Lirman et al. 2008).

Assessment and identification of individual species dominance and coverage has been commonplace in terrestrial survey science for decades (Bechtold and Patterson 2005). Panistsa et al. showed the importance of assessing multiple aspects of plant communities including heterogeneity, dominance, rarity and threat which they showed more effective in assessing conservation status of various habitat types over simple occurrence and frequency

alone (2011). In more recent years, the combination of field sampling techniques and technological advances like remote sensing have been combined to provide various estimations of tree species coverage, dominance and spatial extent (Hame et al. 2001, Hansen et al. 2002). In particular, Inverse Distance Weighting to map and inventory species has become an accepted method in terrestrial plant surveys (Nelson and Vissage 2006, Robert et al. 2004). As a simple yet commonly used deterministic interpolation method, inverse distance weighting determines the value to be occupied by a location-dependent variable based on the values of known points of the same variable (ESRI 2011). Roberts et al. 2004 showed that through determination of an adequate sampling distance in a systematic sampling method, the IDW method yielded near 85% accuracy using presence/absence data. Other studies have incorporated various metrics developed from field data using interpolation to map plant biodiversity in various terrestrial environments (Dogan and Dogan 2006, Panitsa et al. 2011). Morelli et al. 2013 used plant species heterogeneity on varying spatial scales using the IDW method to assess bird diversity. While various accepted techniques exist for mapping species dominance and coverage in terrestrial systems, there are few accepted procedures for identifying these attributes in regards to SAV.

Currituck Sound in North Carolina is presently being monitored and assessed for SAV restoration while simultaneously addressing a single invasive submersed plant species; Eurasian watermilfoil (*Myriophyllum spicatum*). The Sound once supported large quantities of submersed aquatic vegetation (SAV) across its vast expanse. Historically, Currituck Sound was the principal East Coast wintering ground for waterfowl during the 1800s and early 1900s (Sincock 1965), and had thick growth of diverse native submersed macrophytes

(McAtee 1919, Bourn et al 1932). Such plant communities were characteristic of such low-salinity coastal systems. In the past few decades, various drastic environmental and physical changes to the water body have led to a severe decrease in overall SAV, yielding populations well below historical potential; in fact, many areas of Currituck Sound that used to contain lush SAV meadows are now described as a "desert" for submersed aquatic plants. Shifts in dominance of the few remaining species have also become apparent as beneficial, native species such as wild celery (*V. americana*) and redhead grass (*P. perfoliatus)* have declined more rapidly than other species (Carter and Rybicki 1994). *V. americana* is known as an indicator of good ecological health and its decline or absence suggest a major decline in overall ecosystem function (Kock and Orth 2003). These changes have been well-documented in previous studies throughout the Sound's history (See Sincock 1966, McAtee 1919, Bourn 1932, Davis and Carey 1981, Davis and Brinson 1983, Carter and Rybicki 1994), but there has been little work to address the current status of SAV in Currituck Sound, specifically to identify the spatial distribution and coverage of various native and invasive SAV communities. Chemical treatment to control the known invasive SAV species has been limited to the southernmost portion, due largely to the unknown extent of both *M. spicatum* and important submersed native species. The state environmental agency currently contributes herbicide application funding annually to control of *M. spicatum* in Kitty Hawk Bay (North Carolina Department of Environment and Natural Resources (NCDENR) 2013), found at the southern tip of the Currituck Sound. Identifying such important areas is thought to be important in maintaining existing native stands, identifying areas for restoration and concentrating management efforts while reducing collateral damage of native species during

such activities.

In this study a GIS-based, field-driven mapping technique was developed to identify the existing spatial locations and coverage of SAV species. The objectives were to: 1.) Establish depth contours for all species present within the study area based on field data and literature values, 2.) Determine heterogeneity of SAV stands sound-wide; and 3.) Identify areas of individual species dominance and coverage. This survey and mapping technique will aid in management planning by identifying the spatial extent and location of both existing areas of native SAV and areas of dominant *M. spicatum*. Employing such techniques will help concentrate efforts of protection, mitigation, restoration and management in such a large, unique system.

3.2. Materials and Methods

3.2.1. Study area

Currituck Sound is located in northeastern North Carolina and forms the northern arm of the Albemarle-Pamlico Estuary System (APES), the second largest estuary on the United States mainland (McKay et al 2012), thus making it one of the most important wildland habitats in the nation. The Sound stretches approximately 48 km from north to south and 5 to 13 km from east to west depending on the location. On its northernmost end the Sound extends to Back Bay, Virginia and into the Albemarle-Chesapeake Canal. To the south, it joins the Albemarle Sound and the rest of the APES system. Freshwater inputs to Currituck Sound include North Landing River and Northwest River, both with headwaters in the Great Dismal Swamp of North Carolina. Back Bay also contributes water (both salt and fresh) into

the Sound through shallow water channels. Inputs of brackish water from the Albemarle-Chesapeake Canal also might influence the salinity of Currituck Sound. The sound is separated from the Atlantic Ocean by a narrow strip of barrier islands known as the outer banks which are no more than a mile wide. The Sound has an average depth of 1.52 m and maximum depth of approximately 3.96 m, constituting a nearly continuous littoral zone throughout its entirety. This fact alone would lead one to believe that nearly every centimeter of the Sound has the potential for submersed plant growth. Unlike many other coastal embayments in the State which are tidally driven, water level fluctuations in Currituck Sound are a product of constantly changing wind. Therefore, water level can fluctuate wildly from week to week, even day to day during changing weather conditions. The Sound stretches through two counties; Dare and Currituck, with level or slightly sloping terrain that drains into the Currituck Sound.

The study area spans the mid-Currituck portion of Currituck Sound encompassing the Currituck County mainland and Outer Banks as well as the Dare County Outer Banks (Figure 1). The study area is approximately 13 miles long by 5 miles wide stretching from just south of Corolla to Duck on the eastern side and Parker's Creek to Webster's Creek on the western side.

3.2.2 SAV Field Sampling

Currituck sound was sampled during the summer growing season (June – August 2010). SAV was sampled using a modification of the point intercept method (Madsen 1999). The Sound was gridded into 174 equidistant points that were sampled during time frame of June 14th – July 13th 2010 and encompassed the entire mid-Currituck region. Three

points were found to be terrestrial after the survey was complete and were subsequently eliminated, thus leaving a total of 171 survey points. The sample points were located in the field using a Magellan Mobile Mapper CX professional grade GPS unit. At each point, water depth was measured using a calibrated measuring pole from top of sediment to the water surface. Plant frequency at various depths was examined for all sample sites. Sediment type was also assessed using textural classifications for sand, silt or clay (Link 2006) and plant metrics assessed by recording plant presence and plant cover at each site. This was accomplished by qualitatively assigning a 'plant cover level' for each category. Plant cover was assessed at each point for an area of 10mx10m by utilizing visual interpretation of a two-sided sampling rake with 28 tines thrown in four cardinal directions from the point of anchor. An average estimate of all rake throws was used to determine plant coverage at that sight and to give the point dimensionality for later assessment. A locational error of +/- 1.52 m was estimated to be obtained through frequent repositioning.

Plant cover levels were initially separated into 10% field interval categories ranging from 0 (0%) to a level of 10 (91-100%) based on overall coverage of the rake. These levels were then combined in the lab to represent four levels of SAV coverage (SAV COV) most often reported during ranges of density in the existing literature: 0 (0% plant cover), 1 (1–20% low plant cover), 2 (21–40% mid plant cover), 3 (41–80% mid-high plant cover) and 4 (81-100% high plant cover) (Braun-Blanquet and George 1932). An additional binomial category (SAV PA) of total littoral zone SAV cover was developed by combining the four levels recorded for each plant category at each point. This category captures littoral plant presence or absence at each point by assigning each site either a 0 (0% plant cover) or a

1 (1–100% plant cover). Plant species information was also collected and identified in the field.

A category for dominance was developed and collected based on overall rake coverage (SAV DOM) of each species. A total of all species present at each site was also recorded to determine heterogeneity of sites (TOT SPEC). A plant species was considered dominant if that plant made up more than the majority of all plant material present on the rake divided by 28 tines (51-100%) and thus given a dominance score of 3. A plant species was considered to "share dominance" at a site if that plant made up approximately 25-50% of all plant material present on the rake and was thus given a dominance score of 2. A plant species was considered subdominant if that plant was present on the sampling rake but comprised less than 25% of all plant material on the rake and was given a dominance score of 1. Development of this dominance category is an adaptation of techniques currently employed by the state of Wisconsin (Hauxwell et al. 2010).

3.2.3. Geostatistical Analysis

Investigation of the spatial relationship and distribution of each species was used to determine the applicability of using spatial interpolation techniques to develop continuous mapping of species across the study area. Statistical relationships among plant species were assessed using the Moran's Index (Moran's I) measure of spatial autocorrelation in the Spatial Analyst toolset in the ArcMap 10.1 toolbox. The Moran's I is a measure of spatial autocorrelation capable of taking into account multiple dimensions and directions among observations in space. The tool measures spatial autocorrelation based on feature locations and attribute values using the Global Moran's I statistic. In this study, the Moran's I was

used to investigate and develop appropriate distance thresholds at which species events are influenced by neighboring species events at different distance intervals. Spatial correlation was also utilized to explore relationships across species as well. Given that all points were sampled at equidistant intervals using the point intercept method, there was no need to investigate spatial statistics beyond the above mentioned procedures for applications in this work.

3.2.4. Interpolation: Inverse Distance Weighted

All metrics were combined in order to create a dominance grid of each individual species throughout the study area. At each point, each species dominance score was divided by the total number of species present at that point to give an overall heterogeneity score (SAV HET) per species by point. This metric was designed to represent the ratio of each species at each point relative to the total number of species present.

The Inverse distance weighted (IDW) method was used to develop continuous surfaces for each species based on Depth, SAV HET, SAV COV, and SAV PA. The IDW method is a deterministic method for multivariate interpolation most often used with a scattered set of points. The IDW method is most appropriate with modeling processes where assumptions are made that the closer two events are to one another, the more alike they are, and the further events are from another, the less alike they are. In this study, the IDW was deemed the most appropriate method to interpolate between equidistant points placed along a grid because statistical parameters (i.e. sill, nugget) need not be calculated for intervals of categorical distances. A potential SAV establishment depth profile was developed for each species using existing literature values for North Carolina coastal systems (Ferguson and

Wood 1994) as well as field data collected in this study. Maximum depth values were determined by species and mapped using the IDW method to establish a littoral zone for each species identified in the study. If a certain species was identified at depths greater than that documented in previous literature, then the study depth was used as the maximum depth for species establishment. If depth maxima by species were greater in the literature than those depths identified in this study, then the depths from the literature were used as the maximum depth for species establishment. Finally, values between survey points based on geostatistical parameters were interpolated for SAV HET, SAV COV and SAV PA. IDWs were created for SAV PA based on a 90% likelihood threshold of occurrence, and a 75% likelihood threshold for both SAV COV and SAV HET in an attempt to avoid overestimation of SAV extent.

3.2.5. Development of a Dominance/ Percent Cover Metric

In order to determine individual species contribution to overall SAV coverage at each site, and thus provide mapping of species distribution and coverage throughout the study area, a dominance/percent cover metric (DOMCOV) was developed. Once interpolation maps of both species heterogeneity (SAV HET) and overall SAV coverage (SAV COV) were established, the raster calculator tool found in the map algebra toolset of ArcMap 10.1 was used to develop a map of individual species coverage for the study area. By multiplying the categorical values of overall SAV COV with the categorical values of the SAV HET, a dominance/ percent cover value was derived. For example, if at any site an SAV species was found with 3 other species (species total = 4) and found to be dominant (score = 3) and at a point of overall SAV COV of 3 (41-80%), then that site receives a score of 2.25 for DOM

COV. The minimum score for this metric is 0 (No SAV Present) or 12 (SAV 81-100% cover, species dominant in monoculture). The DOMCOV map was then further refined using the depth contour developed for each species.

3.3. Results

3.3.1. SAV Survey Results

Submersed plants were identified at 44% of all points surveyed (Table 1). Six species were identified during the survey and included five native species as widgeongrass (*Ruppia maritima*), southern naiad (*Najas guadalupensis*), sago pondweed (*Stuckenia pectinata*), Redhead Grass (*Potamogeton perfoliatus*) and Wild Celery (*Vallisneria americana*), and one invasive species, Eurasian watermilfoil (*Myriophyllum spicatum*). The majority of SAV were found to be present at depths of 0.3 to 2.2 m with one instance of plant presence as deep as 3 m (Table 1). *R. maritima* was the most prevalent species in the study area followed by *S. pectinata* and the invasive *M. spicatum.* The maximum depth recorded during the survey was 3.2 m with the majority of survey points between 0.8 and 2.4 m. Depth contour maps for each species are provided in Figure 2.

3.3.2. Species Heterogeneity and Coverage

Along with prevalence, *R. maritima* and *S. pectinata* were found to be the two species most often in homogenous stands throughout the study area with an average dominance score > 2. They were most often found coexisting with only one other species or, in many cases, as a monoculture. *N. guadalupensis* and *M. spicatum* shared dominance with one or more other species on average. *P. perfoliatus* and *V. americana* were most often subdominant coexisting

with two or more species (Table 2). All species were found to be most often identified in a stand of low to mid coverage (21-40%), however *R. maritima*, *N. guadalupensis*, *S. pectinata* and *M. spicatum* were all observed in low (1-20%) to high (81-100%) coverage at one or more sites (Table 2). SAV HET values were derived from SAVDOM metrics and TOTSPEC values. An example output of SAVHET for *M. spicatum* is shown in Figure 3.

3.3.3 Geostatistical Results

A distance threshold of 1,300 m was identified as the maximum distance at which species presence events remained spatially auto-correlated. Autocorrelation substantially decreased as the distance threshold value exceeded that distance. All species demonstrated spatial clustering within the study area using that distance threshold. Spatial autocorrelation estimates were not made with *V. americana* due to low occurrence of that species. Moran's I values for each species are given in Table 3.

3.3.4 Maps of DOMCOV by Species

Maps identifying the existing spatial locations and projected coverage of all SAV by species are included in Figures 4-9. *M. spicatum* grew primarily in the mid-central portion of Currituck Sound, but a large, high-coverage stand of dominant *M. spicatum* was identified in the southwestern corner of the study area. *S. pectinata* was found to be mostly dominant in high coverage in the southeastern portion of the study area with satellite communities on the fringe of other species. *P. perfoliatus* and *V. americana* were identified in very few, spotty areas throughout the study area. *R. maritima* was found in high coverage monocultures in both the northern and mid portions of the study area with vast areas of coverage in between.

3.4. Discussion

The depth contours developed in this study closely resemble those developed by Ferguson and Wood (1994), with one ~~slight~~ difference. The species *Stuckenia pectinata* was identified as growing as deep as 2.2 m in Currituck Sound, which differed greatly from Ferguson and Woods' estimate of up to 0.9 m depth. Depth contours were more consistent with Davis and Brinson's (1983) account of depth contours in Currituck Sound during the late 1970s, except that *Vallisneria americana* was found growing as deep as 3.2 m at that time. The low frequency of *V. americana* in the present study area was probably the main reason why the species was not identified at depths greater than 1.6 m. Similarities in depth distribution of species in other NC coastal water bodies (Ferguson and Wood 1994) suggest that this technique might be applicable in other NC coastal systems, and perhaps also adaptable to other eastern coastal systems.

The species *Ruppia maritima* and *S. pectinata* were identified as the dominant and most widespread taxa present in the study area. One of these two species generally was the most prevalent in the Sound throughout the recorded history for this system (Davis and Brinson 1983). During its introduction, the ~~sole~~ invasive *M. spicatum* was thought to be the dominant species in the Sound and was feared to ~~potentially~~ dominate all SAV after its explosion in the 1960s (Fish 1974). Invasive plants, once established, are known to alter stand structure, recruitment of natives, and resource competition (Gordon 1998). However, *M. spicatum* was not identified as the primary dominant species except in an isolated section in the southwestern portion of the study area. The data indicated that *M. spicatum* was not

the primary dominant species or even the second or third most dominant species. While the species may have had great effects during its introduction and spread in past decades, it is currently less of an issue for resource managers than it has been traditionally, and it may not be a primary threat for restoration and protection of native SAV. More important to resource managers might be the lack of dominant stands of *V. americana* and *P. perfoliatus* throughout much of the Sound. Each of these species was once found throughout the entire study area (Sincock et al. 1966). Various influential factors should be examined in future work.

The DOMCOV maps identify large amounts of *R. maritima* from north to south primarily on the eastern side of the Sound. Areas within the defined littoral zone not occupied by *R. maritima* were most often filled by *S. pectinata* and/or *N. guadalupensis.* The invasive species *M. spicatum* seemed primarily confined to the mid-east section of the study area in low coverage. However, a large section dominated by *M. spicatum* was documented in the southwestern portion of the study area. As further surveys are employed, this study suggests targeting this section of the sound for further exploration and potential management of *M. spicatum* as this area seems to be widely infested. Other than those areas limited by depth, the southern portion of the study area was largely devoid of SAV species. Further investigation of the soil types revealed that these areas consisted primarily of loose sand. Based on the plant species present, the likelihood of establishment in these areas in minimal (Ailstock 2010), suggesting that restoration or protection in these areas pragmatically should not be attempted.

All of the above described methods were used to develop a descriptive map of dominance and coverage for each species. The maps were intended to provide vital information of species dominance, coverage and spatial distribution throughout such a large coastal study area, and to inform efforts of management or restoration based on the sites identified. To retain repeatability and consistency across species and future work, these procedures were completed in a distinct order. Certain limitations of sampling technique may have led to over- or under-estimation of species coverage in the study area. The rake fullness method divides the rake into discrete increments, and when plants are harvested, an abundance ranking is given for each species (Madsen and Wersal 2012). This method, which relies on subjective ratings by an observer, was determined to be the most applicable to the turbid waters of Currituck Sound, as visual sightings were not an option. Here, ratings were made by a single individual to reduce inconsistencies from site to site and photographs were taken of each rake to avoid bias; however, future studies should be careful of this issue in assigning coverage. Perhaps a measure of biomass at each site could be achieved through the use of a spinning rake which is often utilized to make estimates of biomass or coverage at sampling sites (Madsen and Bloomfield 1993).

Use of the rake method for comparisons between species was addressed in this study by comparing coverage values within species, not across species. For example, DOMCOV estimates of *M. spicatum* were not compared to DOMCOV estimates of *V. americana.* Cross-species comparisons are not encouraged unless the efficiency of the rake method has been determined for each species being compared (Yin and Kreilig 2011). This is most apparent when comparing vertically oriented SAV with more basal growing species.

Lastly, usage of these techniques is only recommended in water bodies with a continual littoral zone. Spatial autocorrelation estimates rely on the potential for SAV establishment in several directions without depth as a primary limitation. In Currituck Sound, this assumption was met as depths gradually sloped and were almost always within the range of establishment by SAV species (see Figure 2). On the other hand, these techniques would not be applicable in a system where depth is a great limitation and the littoral zone changes rapidly (i.e. a reservoir system). Use of this procedure in such a water body would require intensive sampling to define a depth contour prior to any interpolation.

3.5. Conclusions

Currituck Sound is a unique system whose large size and vast littoral zone make it a challenge for scientists and resource managers alike. This study provides a framework for large-scale mapping and monitoring of SAV in the Sound and similar coastal water bodies. The techniques described here define general SAV species heterogeneity, coverage, and distribution with minimal field work and help identify areas of particular interest or concern that could improve managers' ability to optimally allocate agency resources, time and funding given the large spatial extent of their project area. These techniques can be employed annually or seasonally track changes in the distribution and general abundance of a fluid and dynamic plant community.

As new technology emerges, the potential for hybridization between traditional techniques and tech-heavy methods becomes greater. For example, hybridized point intercept and hydroacoustics is being used to map various inland water bodies throughout the

United States (Valley 2013). While hydroacoustic sampling may not be feasible to map the entirety of such large coastal systems as the Currituck Sound, the methods described in this dissertation can aid in selection of sites for further surveying with such technology. Although this technique may not provide exact estimates of SAV coverage, it does provide a means of exploration in systems not traditionally and continuously sampled. The hope of such a methodology is to narrow down efforts to allocate future work in a timely fashion. The methods to map and/or monitor a system for SAV are many**;** therefore, it is important to choose an appropriate technique that meets the needs of each program. In coastal systems, these programs can vary from restoration of native SAV to preservation of existing SAV and even management of invasive species which threaten such a unique ecosystem. The methodology described herein provides a promising means to identify the spatial extent and location of various SAV species in a traditionally under-sampled water body.

3.6. References

Ailstock, M. S., D. J. Shafer, and A. D. Magoun. 2010. Effects of planting depth, sediment grain size, and nutrients on *Ruppia maritima* and *Potamogeton perfoliatus* seedling emergence and growth. Restoration Ecology 18: 574-583. DOI: 10.1111/j.1526-100X.2010.00697.x

Bechtold, W.A.; Patterson, P.L., eds. 2005. The enhanced Forest Inventory and Analysis program—national sampling design and estimation procedures. Gen. Tech. Rep. SRS-80. Asheville, NC:U.S. Department of Agriculture, Forest Service. 85 pp.

Bourn, W.S. 1932. Ecological and physiological Studies on certain aquatic angiosperms. Contribution. Boyce Thompson Inst. 4: 425-496.

Braun-Blanquet J and George D. Fuller. 1932. Plant sociology: The study of plant communities.

Busch, K. E., R. R. Golden, T. A. Parham, L. P. Karrh, M. J. Lewandowski, and M. D. Naylor. 2010. Large-Scale *Zostera marina* (eelgrass) restoration in Chesapeake Bay, Maryland, USA. Part I: A comparison of techniques and associated costs. Restoration Ecology 18: 490-500. DOI: 10.1111/j.1526-100X.2010.00690.x

Carter, V and N.B. Rybicki. 1994. Invasions and declines of submersed macrophytes in the tidal Potomac River and estuary, the Currituck Sound-Back Bay System, and the Pamlico River estuary. Report to the Chesapeake Bay SAV Restoration program. Annapolis, MD

Davis, G.J.; and M.M. Brinson. 1983. Trends in submersed macrophyte communities of the Currituck Sound: 1909-1979. Journal of Aquatic Plant Management 21: 83-87.

ESRI. 2012. ArcGIS Desktop Help 10.1 – About Inverse Distance Weighted. ONLINE. Available: http://help.arcgis.com/en/arcgisdesktop/10.0/help/index.html#//00310000002m000000.

Ferguson, R.L. and L.L. Wood. 1994. Rooted Vascular aquatic plant beds in the Albemarle-Pamlico estuarine system. NMFS, NOAA, Beaufort, NC, 103p.

Gordon, D.R. 1998. Effects of Invasive, Non-indigenous plant species on ecosystem processes: Lessons from Florida. Ecological Applications. 8: 975-989.

Häme, T.; Stenberg, P.; Andersson, K.; Rauste, Y.; Kennedy,P.; Folving, S.; Sarkeala, J. 2001. AVHRR-based forest proportion map of the Pan-European area. Remote Sensing of Environment 77: 76-91.

Hansen, M.C., R.S. DeFries, J.R.G. Townshend, R. Sohlberg, C. Dimiceli, and M. Carroll. 2002. Towards an operational MODIS continuous field of percent tree cover algorithm: examples using AVHRR and MODIS data. Remote Sensing of Environment. 83: 303-319.

Hauxwell, J. S. Knight, K. Wagner, A. Mikulyuk, M. Nault, M. Porzky, and S. Chase. 2010. Recommended baseline monitoring of aquatic plants in Wisconsin: sampling design, field and laboratory procedures, data entry and analysis, and applications. Wisconsin Department of Natural Resources Bureau of Science Services, PUB-SS-1068 2010. Madison, WI.

Kerwin, J.A., R.E. Munro and W.W.A. Peterson. 1976. Distribution abundance of aquatic vegetation in the upper Chesapeake bay, 1971-1974. In: J. Davis (coord.) the effects of tropical storm Agnes on the Chesapeake Bay Estuarine System. Chesapeake Research Consortium Publ. No. 54. The John Hopkins University Press, Baltimore, MD, 639 pp.

Koch, E. W. and R. J. Orth. 2003. The seagrasses of the Mid-Atlantic coast of the United States. pp. 234-243. In: E.P. Green and F.T. Short (eds.). World Atlas of Seagrasses. University of California Press, Berkley, CA.

Link, A.G. 1966. Textural classifications of sediment. Sedimentology 7: 249-254.

Lirman, D., G. Deangelo, J. Serafy, A. Hazra, D. Smith Hazra, J. Herlan, J. Luo, S. Bellmund, J. Wang, R. Clausing. 2008. Seasonal changes in the abundance and distribution of submerged aquatic vegetation in a highly managed coastal lagoon. Hydrobiologia 596: 105-120.

Madsen, J.D. 1999. Point and line intercept methods for aquatic plant management. APCRP Technical Notes Collection (TN APCRP-M1-02), U.S. Army Engineer Research and Development Center, Vicksburg, MS, 16 pp.

Madsen JD and JA Bloomfield. 1993. Aquatic vegetation quantification symposium: an overview. Lake and Reserv. Manage. 7: 137-140.

Madsen JD and RM Wersal. 2012. A Review of Aquatic Plant Monitoring and Assessment Methods. A note to the Aquatic Ecosystem Restoration Foundation.

McAtee, W.L. 1919. Notes on the flora of Churches Island, North Carolina. J. Elisha Mitchell Sci. Society 35:61-75.

McKay, S. K., Wilson, C. R., Piatkowski, D. 2012. Currituck Sound Estuary Restoration: A Case Study in Objective Setting. EMRRP-EBA-17

Morelli F., Pruscini, F., Santolini, R., Perna, P. and Y Benedetti. Landscape heterogeneity metrics as indicators of bird diversity: Determining the optimal spatial scales in different landscapes. Ecological Indicators V.34 pp. 372-379.

NCDENR. 2013. North Carolina Aquatic Weed Control Program 2013 Work Plan. North Carolina Department of Environment and Natural Resources. Available ONLINE: http://www.ncwater.org/Education_and_Technical_Assistance/Aquatic_Weed_Control/2013_work_plan.pdf, last accessed in May 2013.

Nelson M.D. and J. Viassage. 2006. Mapping Forest Inventory and Analysis of Forest Land Use: Timberland, Reserved Forest Land and Other Forest Land. Proceedings of the Seventh Annual Forest Inventory and Analysis Symposium 185-191.

Nieder, W.C., E. Barnaba, S.E.G. Findlay, S. Hoskins, N. Holochuck, E.A. Blair. 2004. Distribution and abundance of submerged aquatic vegetation and *Trapa natans* in the Hudson River Estuary. J. Coast. Res. Sci. 45: 150–161

Orth, R.J., D. J. Wilcox, J. R. Whiting, L. Nagey, A. L. Owens, and A. K. Kenne. 2011. Distribution of Submerged Aquatic Vegetation in Chesapeake and Coastal Bays. VIMS Special Scientific Report Number 153. Final report to EPA. Grant No. CB97392801-0, http://www.vims.edu/bio/sav/sav10, last accessed in June 2011.

Orth, R. J., D. J. Wilcox, L. S. Nagey, A. L. Owens, J. R. Whiting, and A. Kenne. 2007. Distribution of submerged aquatic vegetation in the Chesapeake Bay. VIMS Special Scientific Report Number 150. Final Report to NOAA, Washington, DC. Available at: http://www.vims.edu/bio/sav/sav06, last accessed in July 2011.

Roberts, E.A., Sheley, R.L. and R.L. Lawrence. 2004. Using Sampling and Inverse Distance Weighted Modeling for Mapping Invasive Plants. Western North American Naturalist V. 64: 3 312-323.

Sincock, J.L. 1966. Back Bay-Currituck Sound data report. Patuxent Wildlife Research Center. Laurel, MD, 1600 pp.

Yin Y and RM Kreiling. 2011. The evaluation of a rake method to quantify submersed vegetation in the Upper Mississippi River. Hydrobiologia 675:187-195.

Table 3.1. Plant frequency-depth relationships for Currituck Sound study area.

Depth	No. of Sites	RuMar	% freq	NaGuad	% freq	StPect	% freq	MySpic	% freq	PoPerf	% freq	VaAmer	% freq	any species	% freq
0.3-0.6	2	2	100%	1	50%	0	0%	2	100%	0	0%	1	50%	2	100%
0.6-0.8	5	3	60%	1	20%	0	0%	1	20%	0	0%	0	0%	4	80%
0.8-1.0	16	12	75%	6	38%	5	31%	3	19%	1	6%	0	0%	12	75%
1.0-1.2	13	11	85%	5	38%	6	46%	5	38%	1	8%	0	0%	11	85%
1.2-1.4	18	10	56%	8	44%	12	67%	9	50%	3	17%	0	0%	16	89%
1.4-1.6	19	11	58%	4	21%	8	42%	4	21%	0	0%	1	5%	14	74%
1.6-1.8	19	7	37%	5	26%	4	21%	4	21%	0	0%	0	0%	8	42%
1.8-2.0	17	4	24%	2	12%	3	18%	1	6%	0	0%	0	0%	4	24%
2.0-2.2	23	1	4%	1	4%	2	9%	2	9%	0	0%	0	0%	3	13%
2.2-2.4	13	0	0%	0	0%	0	0%	0	0%	0	0%	0	0%	0	0%
2.4-2.6	8	0	0%	0	0%	0	0%	0	0%	0	0%	0	0%	0	0%
2.6-2.8	10	0	0%	0	0%	0	0%	0	0%	0	0%	0	0%	0	0%
2.8-3.0	7	1	14%	1	14%	0	0%	1	14%	0	0%	0	0%	1	14%
3.0-3.2	1	0	0%	0	0%	0	0%	0	0%	0	0%	0	0%	0	0%
Total	171	62	36%	34	20%	40	23%	32	19%	5	3%	2	1%	75	44%

Table 3.2. Average values (Min and Max) by species for SAVDOM, TOTSPEC and SAVCOV.

Species	DOM	TOT SPEC	COV	N
RuMar	2.34 (1-3)	2.45 (1-4)	1.90 (1-4)	62
NaGuad	1.2 (1-3)	3.21 (2-4)	2.06 (1-4)	34
StPect	2.24 (1-3)	2.63 (1-4)	2.09 (1-4)	40
MySpic	1.31 (1-3)	3.19 (2-4)	2.13 (1-4)	32
PoPerf	1 (1)	3.6 (2-4)	2 (1-3)	5
VaAmer	1.5 (1-2)	3.5 (3-4)	3 (3)	2

Table 3.3. Moran's-I spatial autocorrelation parameters for each species.

Species	Moran's I	Expected I	Variance	Z-score	p-value
RuMar	0.3056	-0.0059	0.0018	7.33	0.0001
NaGuad	0.2066	-0.0059	0.0018	5.03	0.0001
StPect	0.1851	-0.0059	0.0018	4.51	0.0001
MySpic	0.3399	-0.0059	0.0019	8.2	0.0001
PoPerf	0.126	-0.0059	0.0015	3.44	0.0005
VaAmer	n/a	n/a	n/a	n/a	n/a

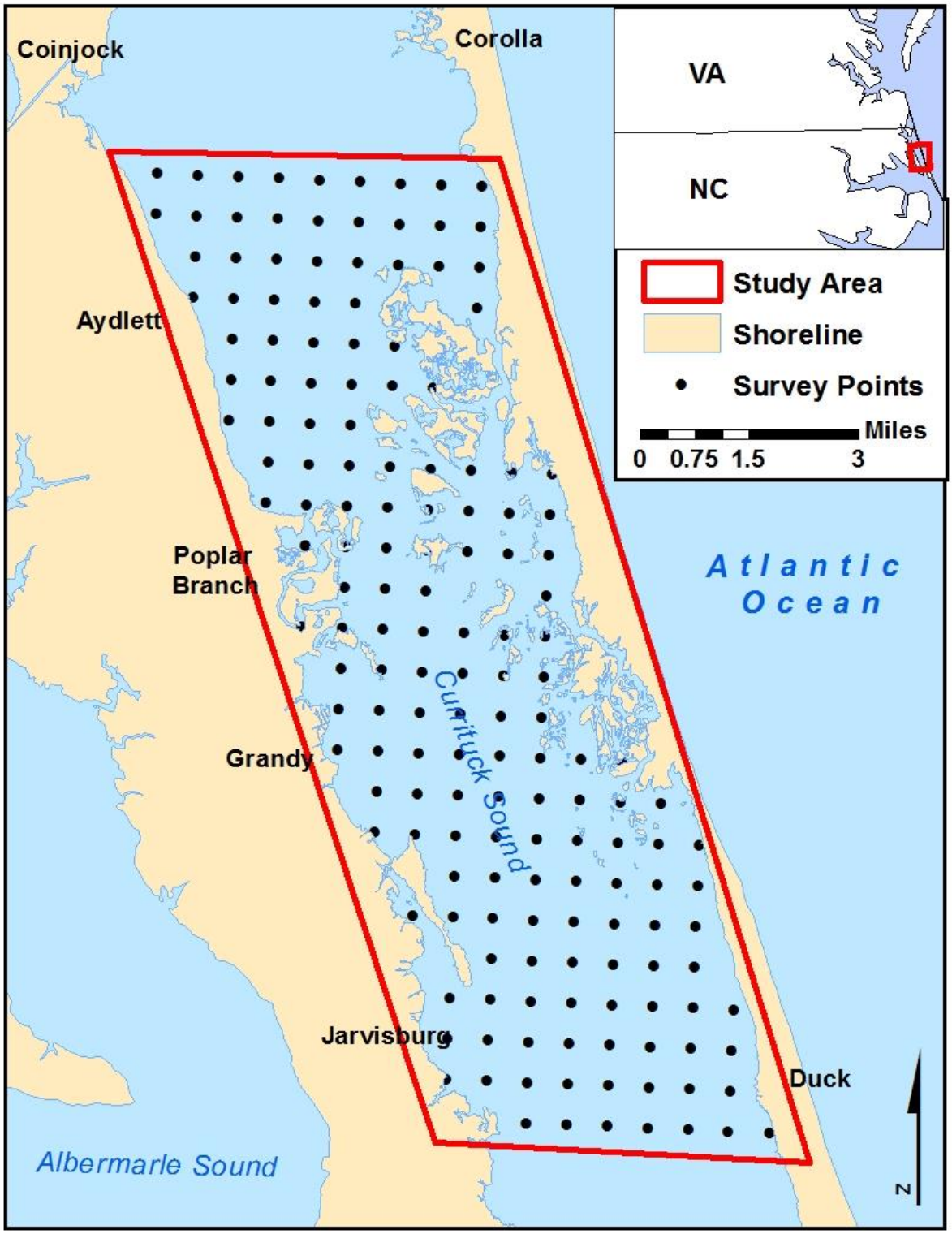

Figure 3.1. Study area for SAV sampling.

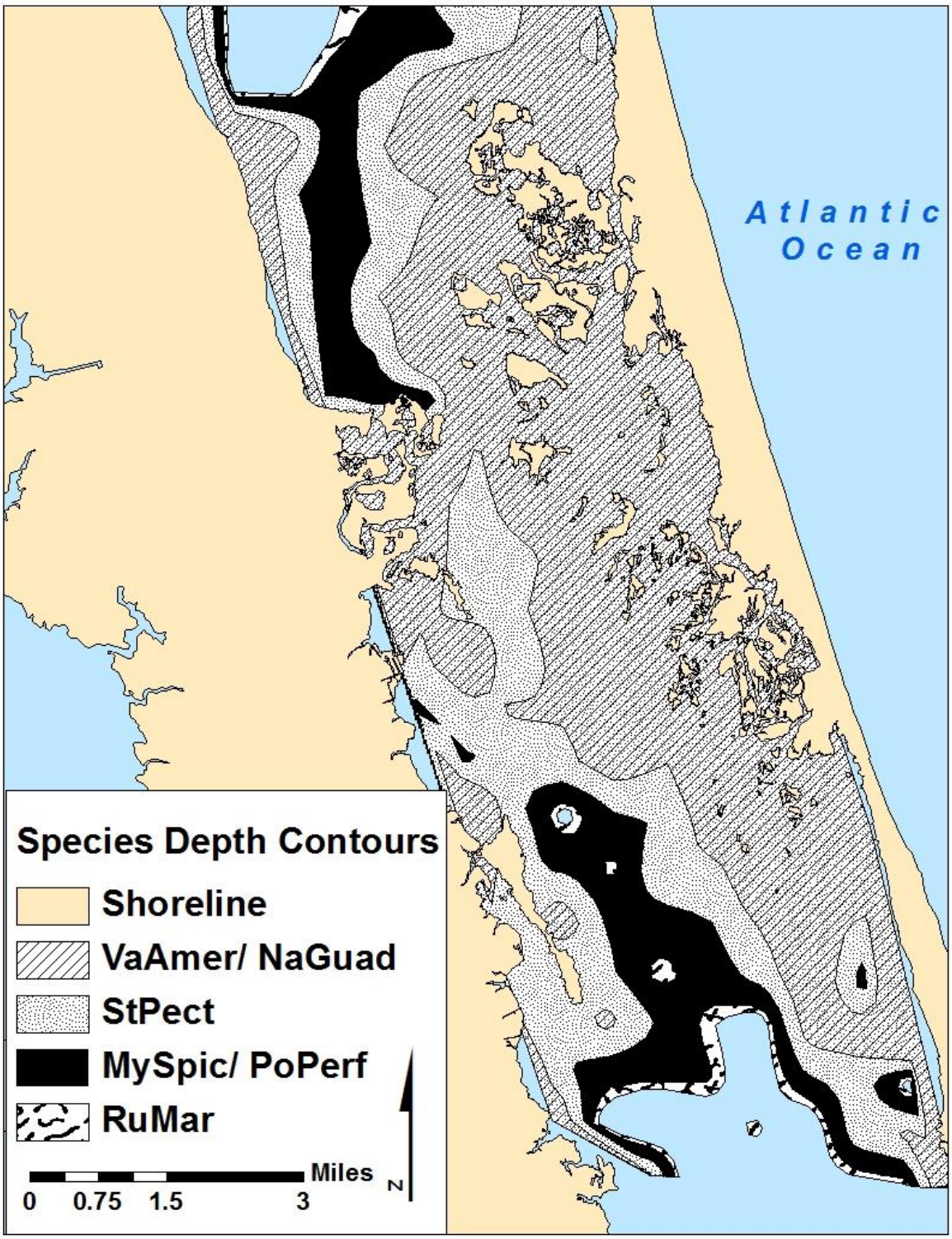

Figure 3.2. Derived depth contours of each SAV species based on field observations.

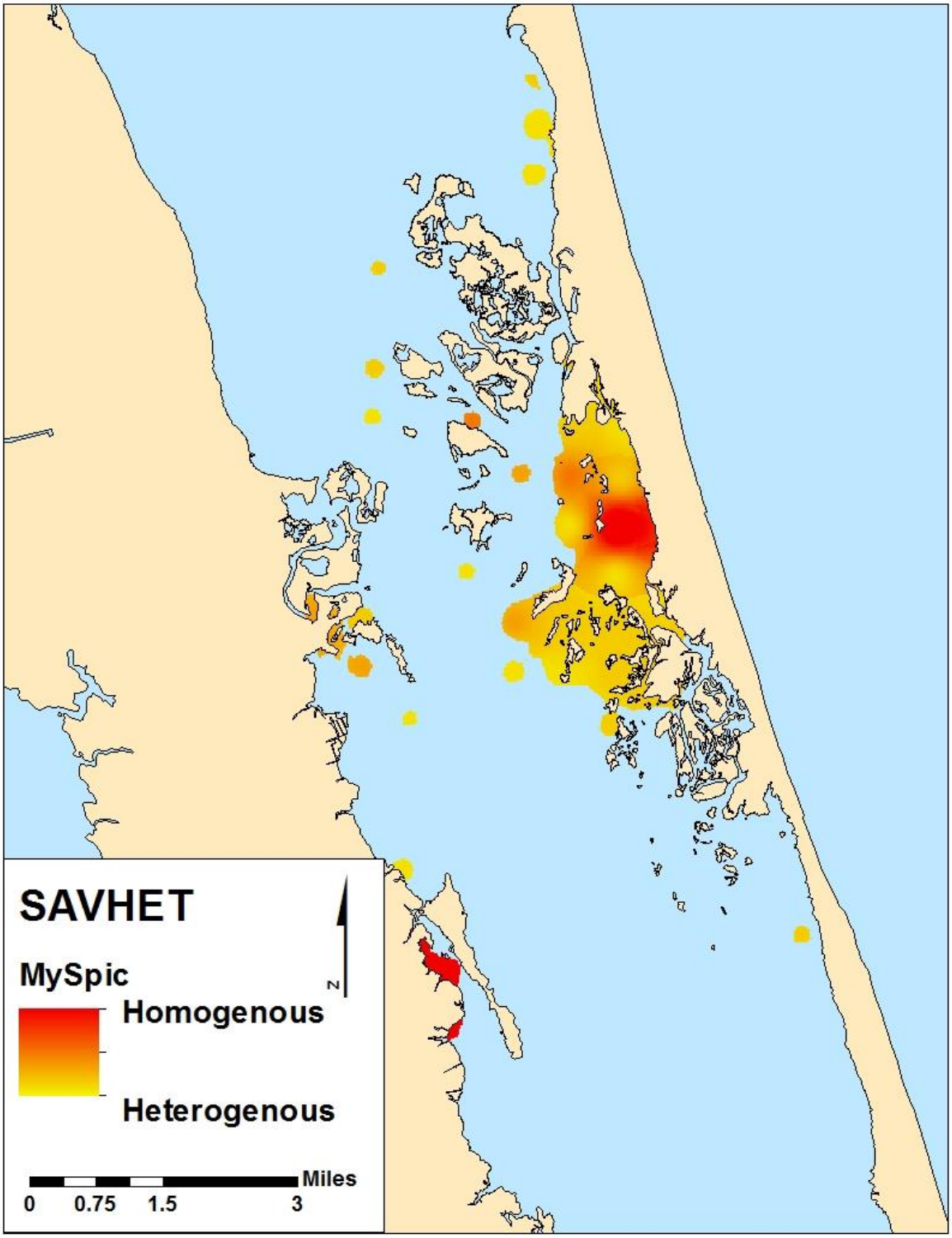

Figure 3.3. Example SAVHET output displaying spatial heterogeneity of *M. spicatum* throughout the study area.

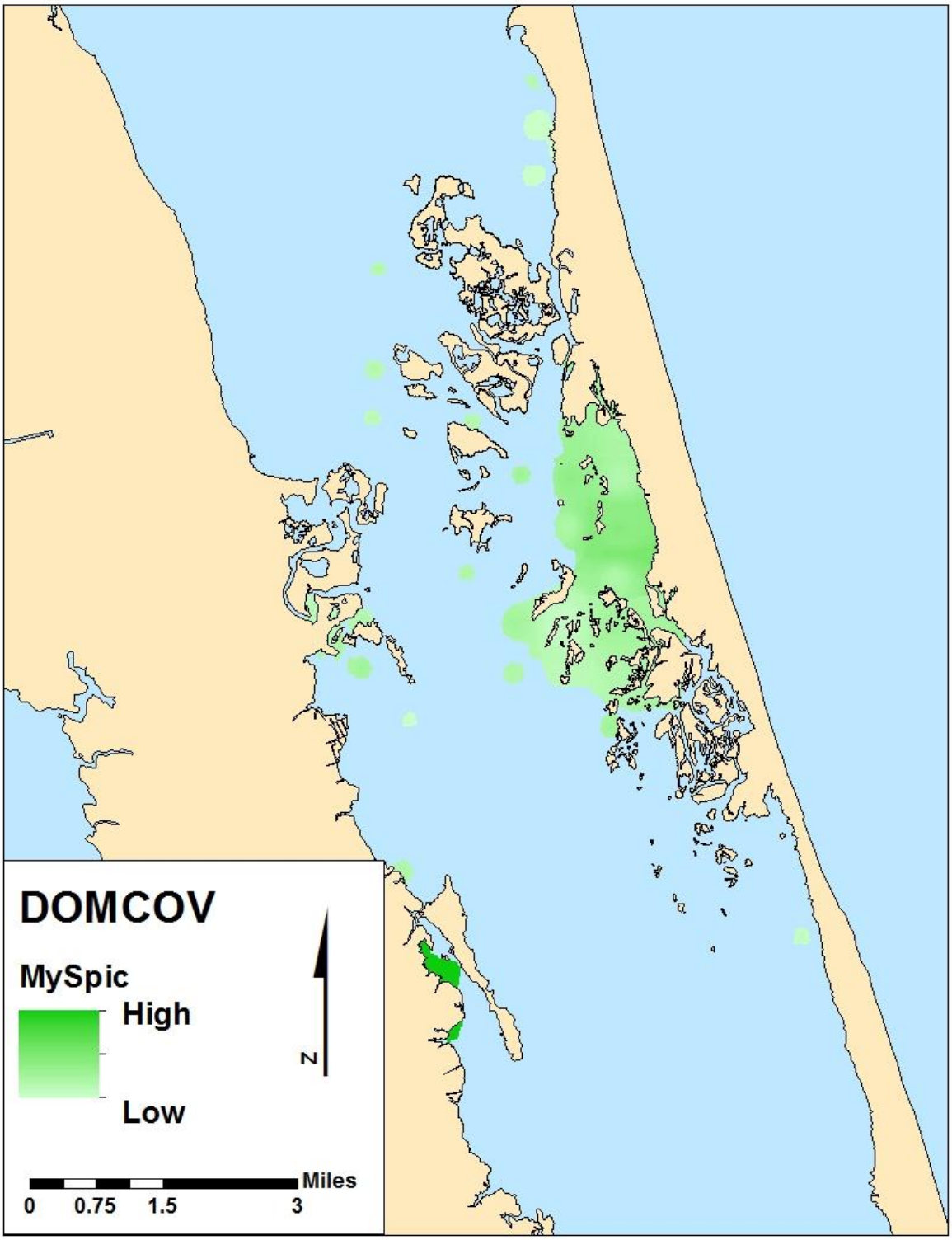

Figure 3.4. DOMCOV output displaying estimated coverage distribution of *M. spicatum* throughout the study area.

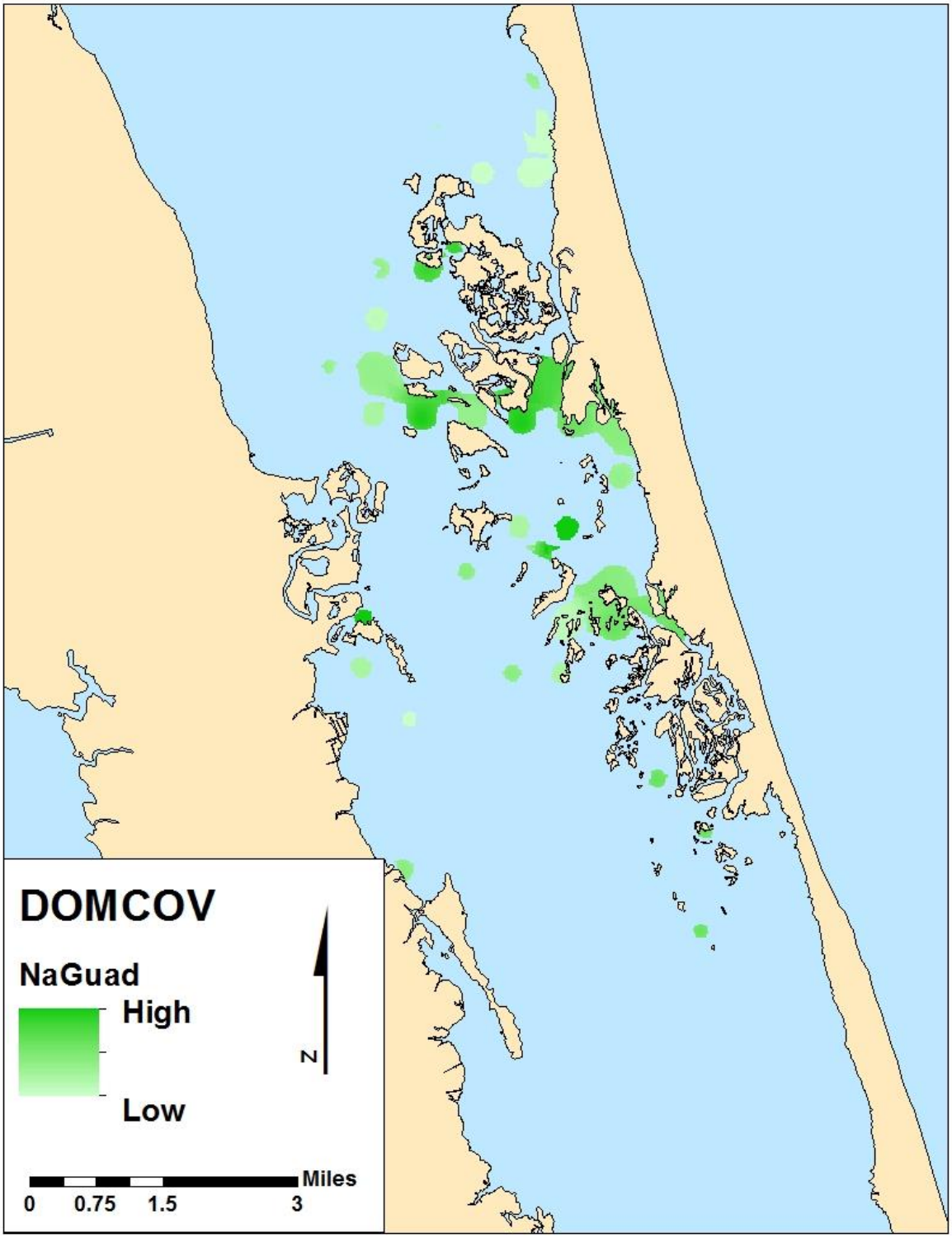

Figure 3.5. DOMCOV output displaying estimated coverage distribution of *N. guadalupensis* throughout the study area.

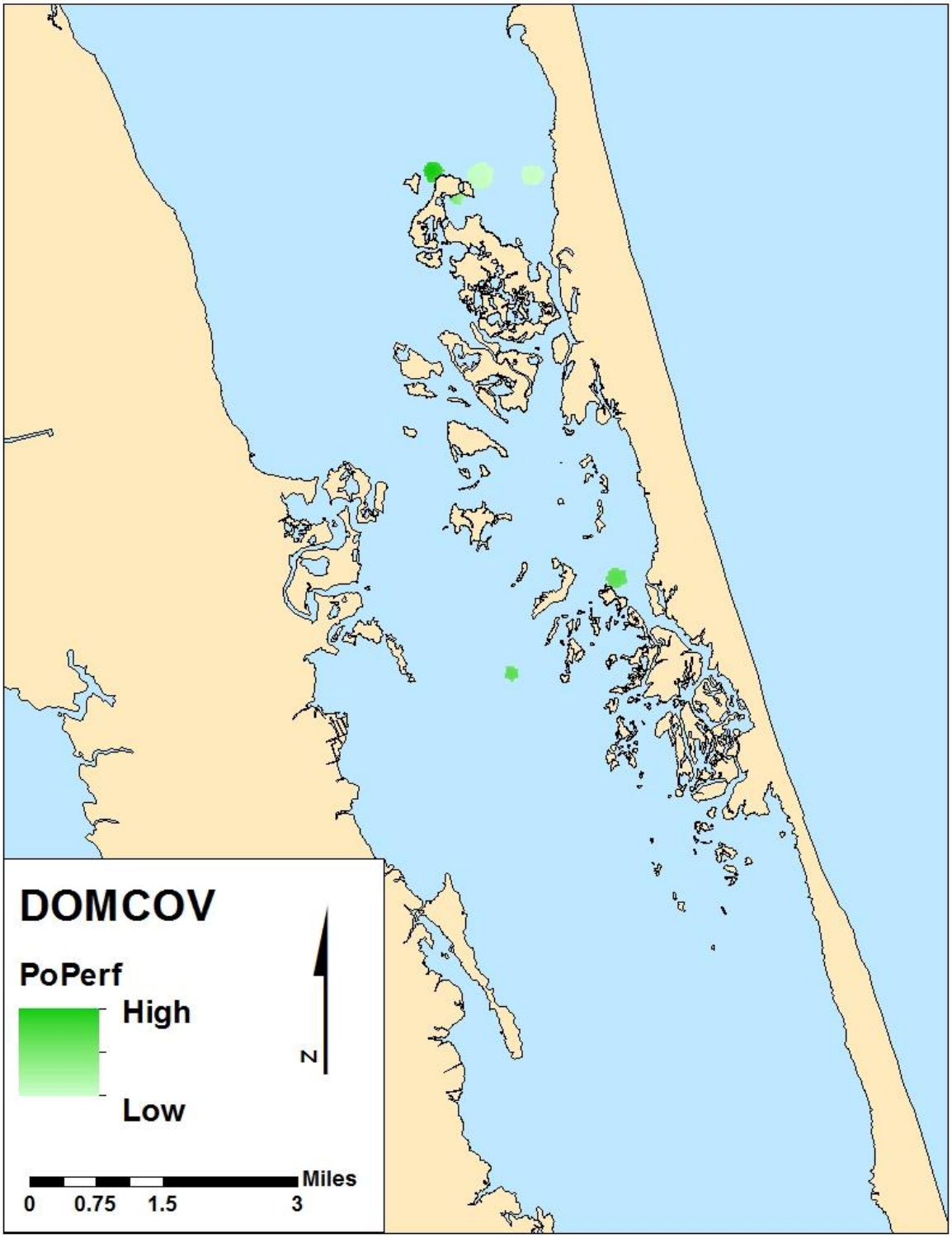

Figure 3.6. DOMCOV output displaying estimated coverage distribution of *P. perfoliatus* throughout the study area.

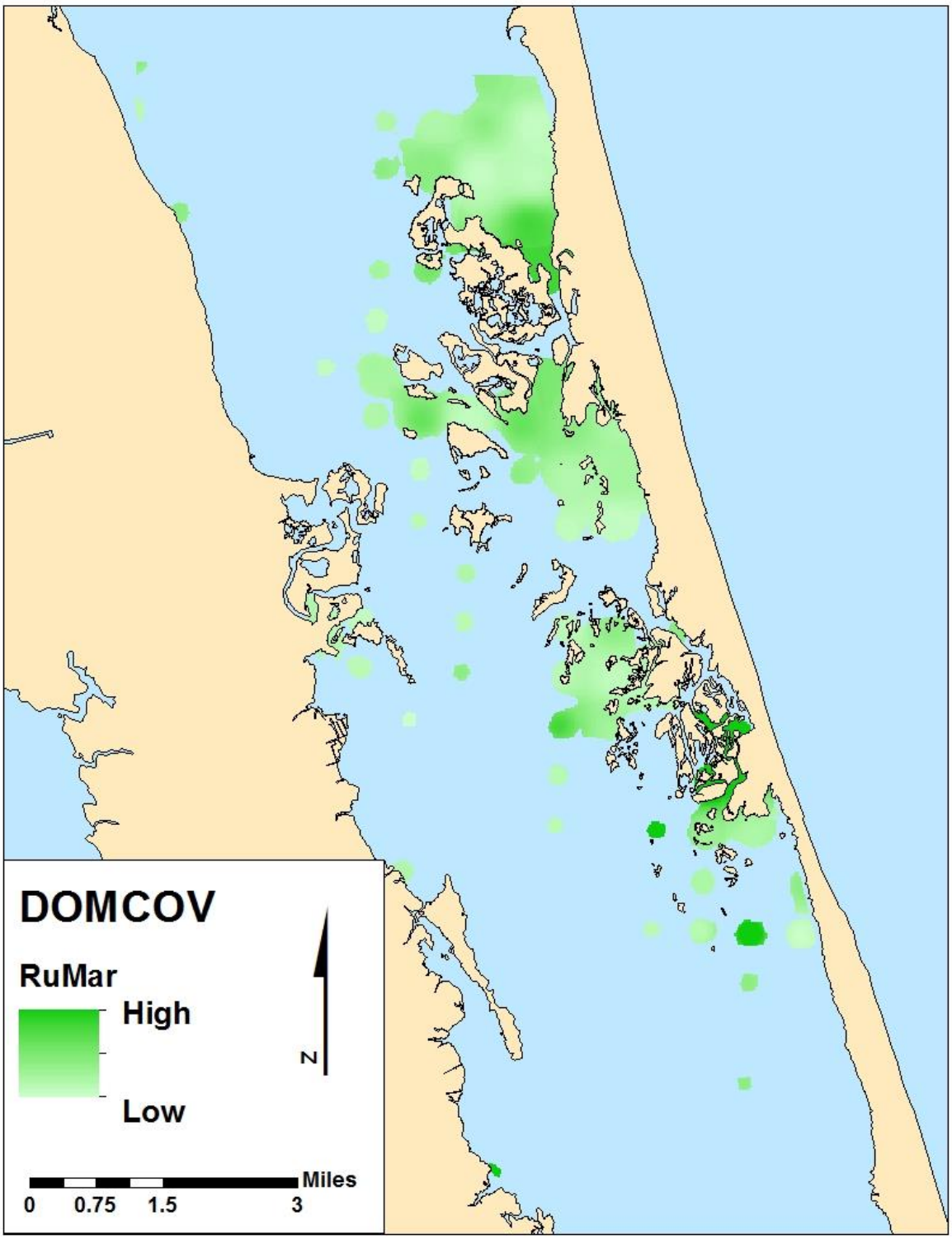

Figure 3.7. DOMCOV output displaying estimated coverage distribution of *R. maritima* throughout the study area.

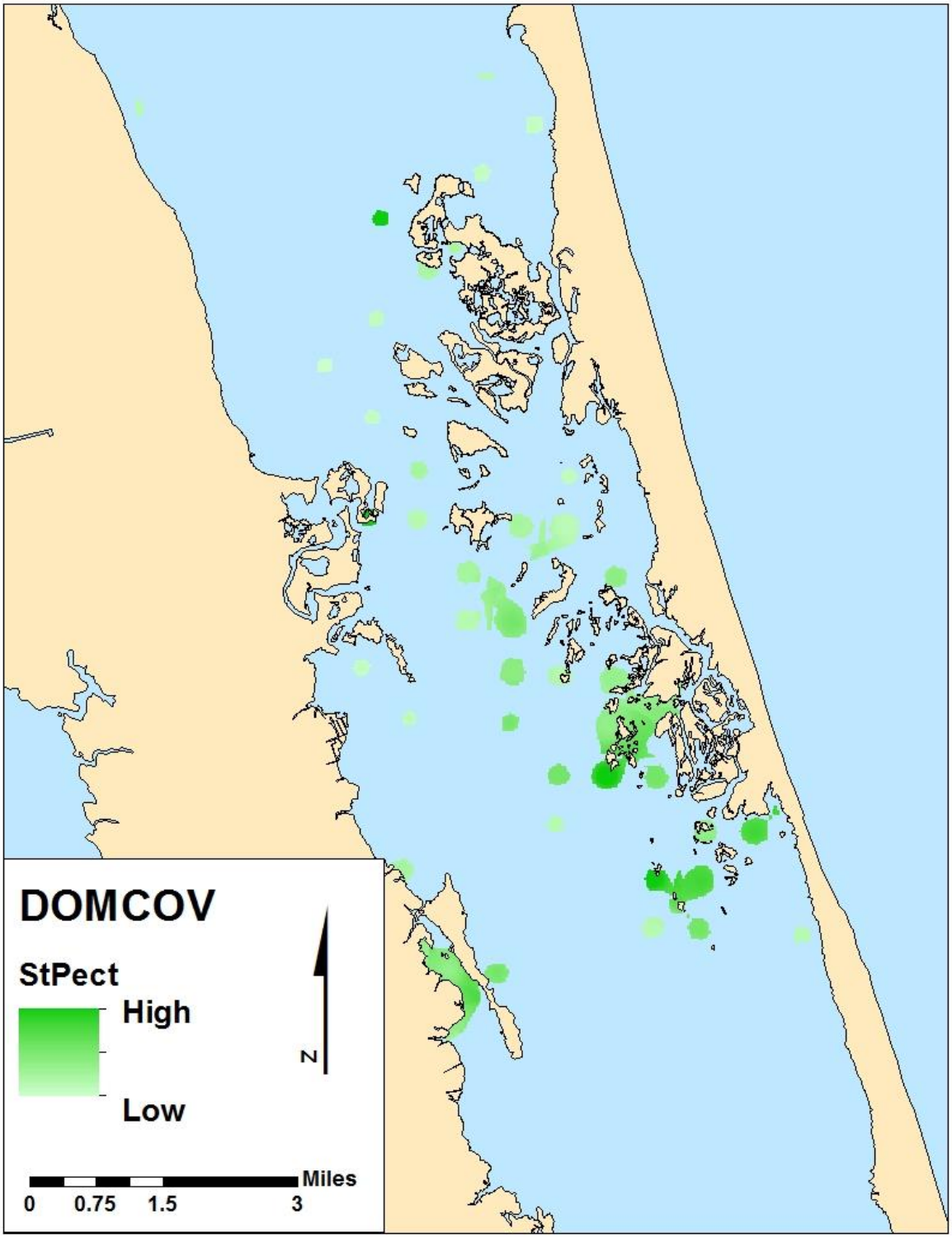

Figure 3.8. DOMCOV output displaying estimated coverage distribution of *S. pectinata* throughout the study area.

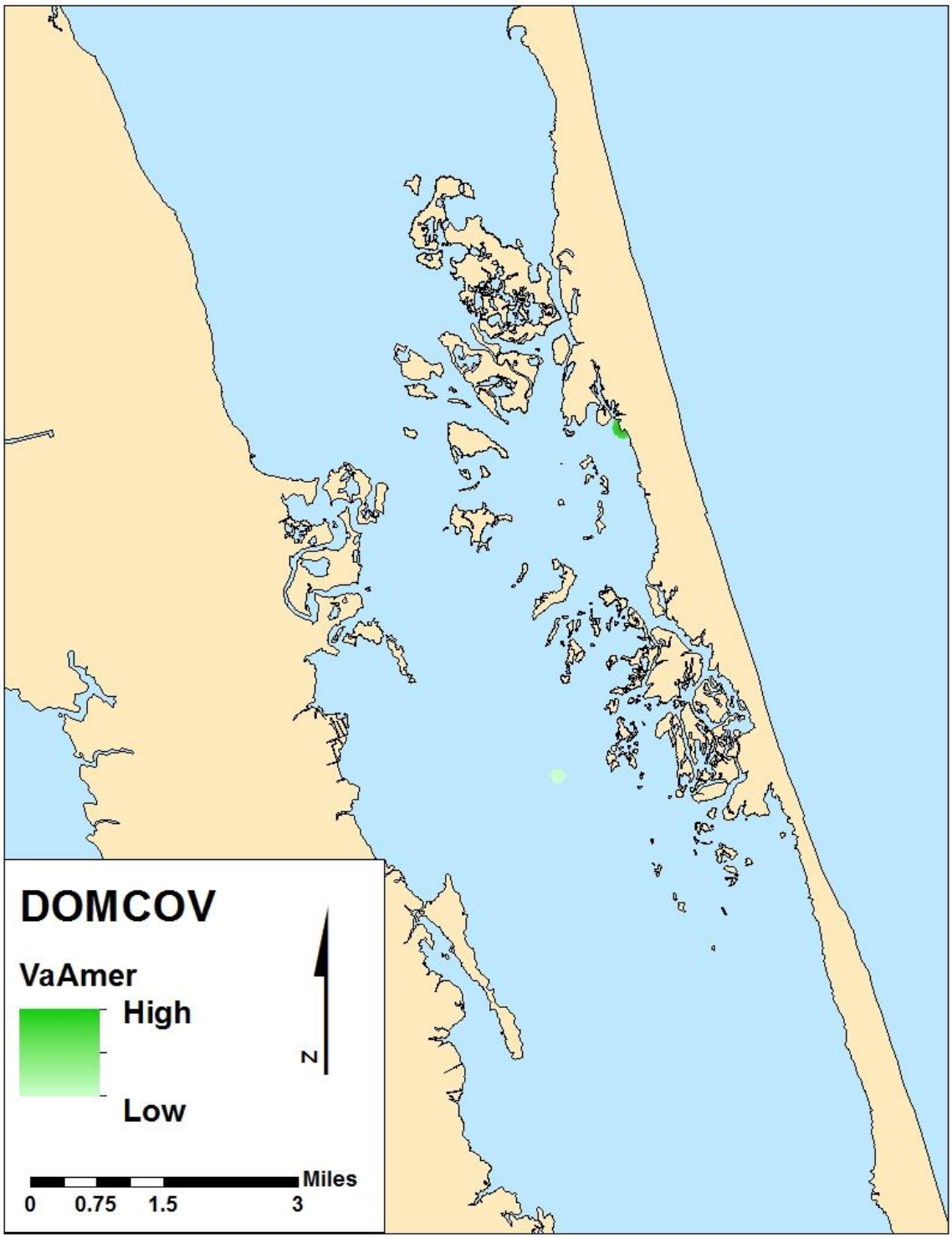

Figure 3.9. DOMCOV output displaying estimated coverage distribution of *V. americana* throughout the study area.

CHAPTER 4

Modeling the Establishment Potential of Hydrilla (*Hydrilla verticillata)* in North America

4.1. Introduction

Hydrilla (*Hydrilla verticillata*) is a submersed aquatic plant species found to be highly invasive and known to cause severe ecological damage and economic hardship after establishment (Langeland 1996). After introduction into Florida during the 1950s, the species has continued to spread, establishing as far North as Maine and as far west as Washington (Bailey and Calhoun 2008, Madeira et al. 2000). Hydrilla establishment in colder environments has recently raised questions about whether this species can extend its range further throughout North America (Richardson et al. 2012). Recent discoveries of the monoecious biotype in Lake Cayuga, NY, the Erie Canal and the Ohio River have further highlighted the need for addressing the potential establishment of this submersed macrophyte. This is especially true of the monoecious biotype which is much more suited for the colder climates prevalent throughout the vast, northern portions of North America (Maki 2008).

The monoecious biotype of hydrilla, while not as extensively studied as its dioecious counterpart, may be found from Georgia and Alabama in the southeast, northward to Maine, and occur as far west as California and Washington (Langeland 1996, Madeira 2000). Unlike dioecious hydrilla, monoecious hydrilla acts as an herbaceous perennial (Harlan et al. 1985) and can survive a much shorter growing season and cooler climates (Madeira et al.

2000, Netherland 1997). This is possible through the prolific formation of carbohydrate-rich reproductive structures, subterranean turions (known as tubers), which can remain dormant for long periods (Owens and Madsen 1998). Monoecious hydrilla in particular only needs roughly four to six weeks of growing conditions during which time the plant can sprout from existing tubers, grow, and generate more tubers before going back into dormancy until conditions are suitable again for regrowth (Spencer and Anderson 1986). Maki and Galatowitsch (2008) reported that monoecious hydrilla tubers survived temperatures as cold as 4 degrees C from 67% (63 day exposure) to 42% (105 day exposure), suggesting successful overwintering in northern latitudes. Furthermore, the monoecious biotype produces more tubers than the dioecious biotype in the field (Harlan et al. 1985, Miller et al. 1976). Steward (1987) showed that tuber production is higher in the monoecious biotype, thus exacerbating the threat of potential spread at the northern borders of expansion in the United States and Canada. This attribute of monoecious hydrilla should be of great concern to natural resource managers in the vast northern reaches of North America, where water bodies number in the hundreds of thousands.

Despite the issue of hydrilla encroaching establishment northward, few have attempted to model the potential spread of the invasive plant into these regions. Langeland (1996) suggested that the monoecious biotype could spread as far north as southern Canada (1996) based on its range in Europe at the time. However, this noxious species has since established much farther north in other areas of the world than previously thought (Balciunas and Chen 1993). It has even been found growing in the subarctic climates of Latvia, Russia,

and Poland. Peterson et al. (2003) attempted to model the invasion potential of hydrilla along with three terrestrial species, using species occurrence and an ecological niche model. While informative, this ecological niche model used various habitat parameters that may not be appropriate for modeling a submersed aquatic plant species and may not be quantifiable on such a large geographic scale. Others have modeled the risk of infestation of hydrilla but only on local scales that do not take into account global establishment (Gallardo and Aldridge 2013). The Peterson et al. (2003) model used 15 layers of varying spatial resolution including slope of landscape, annual frost days and precipitation. While applicable to terrestrial species on which many of these variables play a limiting role, these variables may not functionally limit an obligate aquatic species. In fact, there are very few variables that can be quantified on a global scale that limit a submersed aquatic plant like hydrilla. The majority of climate models currently used are conducted in terrestrial systems, mainly because important aquatic predictor variables are scarce or unavailable for most regions in the world (Barbosa et al. 2012). Therefore, ecological niche and habitat suitability models may be geographically limiting the potential establishment capability of hydrilla and other submersed aquatic plants. Given these constraints, a less robust modeling technique may be actually more appropriate.

Climate envelope models are a simple, yet statistically informative tool used to identify areas of vulnerability to invasive species on a global scale, especially in response to climate change (Araujo and Peterson 2012). These models identify locations of a native species and established invasive ranges and then map the potential geographic range of establishment based on various aspects of climate (Pearson and Dawson. 2003, Broennimann

et al. 2007, Villemant et al. 2011). Traditionally, climate envelope models use variables associated with climate (temperature, rainfall, etc.) to compare current range values to similar values around the world. In the case of hydrilla, few variables are likely limiting other than soil, salinity, light and temperature. Of those, temperature is most easily quantifiable on a global scale given widely available climate data measured on comparable spatial scales. This type of modeling is traditionally used for assessing worldwide species invasions to identify at-risk areas (Leroux et al. 2013). These models can be designed on a global scale and applied to sub-regions where other variables may be collected at finer scale (Sheppard 2013). Climate envelope models are extremely useful when modeling an invasive species potential establishment on such a large scale, as many analyses have indicated that particularly invasive species follow similar distribution patterns even across very different geographic areas (Peterson et al. 2003).

In this study I attempt to model the establishment potential of hydrilla using a modified version of the climate envelope model, based on the plants known global geographic distribution in the Northern Hemisphere, particularly along its northernmost limits. The objectives of this study were to develop a risk scale based on a regression of climate and occurrence frequencies, model the potential establishment of hydrilla in the United States and Canada based on the developed risk scale; determine state or province specific establishment potential based on existing water bodies; and determine if risk within a state/province differs based on different water body sizes. The purpose for such work is to inform researchers and resource managers alike of the establishment potential of hydrilla along northern tier states and provinces in North America. Identifying such areas would

inform resource managers on how best to allocate prevention, monitoring and management efforts for this invasive species prior to its spread into non-hydrilla water bodies.

4.2. Materials and Methods

4.2.1. Data Sources

Hydrilla occurrence points were accumulated from various sources (see appendix X) within the species native and invasive ranges in the Northern Hemisphere. These points were compiled from existing herbarium specimens, field identifications and literature identifying the exact location of discovery. Nearly 10,000 (n = 9,707) data points were identified after the data was georeferenced and examined for duplicates. Of particular interest were occurrence sites identified at high elevations and northernmost latitudes. When values were identified through the literature, ESRI's ArcGIS (Version 10.1, ESRI 2012) was used to create point values at the exact location specified in the literature. All data were then combined into a spatial database for further analysis.

Global climate data representing mean monthly minimum temperature were obtained from the WorldClim datatset (Hijmans et al. 2005) for the months of June, July and August during the years 1950-2000 to represent the minimum growing season of hydrilla required for tuber sprouting, growth and tuber regeneration in the Northern Hemisphere (Spencer et al. 1986). When occurrence sites were identified in the literature by state or province, sites were extracted only if the specified site could be identified at smaller geographic units (i.e. county) and that unit displayed relatively low variation in climate across its entirety (less than .01 degree Celsius). Values were extracted on a point grid of the smallest unit identified. If

occurrence sites showed greater than 0.01^{o}C variability within the smallest measurable geographic unit, they were subsequently eliminated from analysis.

4.2.2. Risk Scale Development

Mean monthly minimum temperature was obtained at each occurrence site using a point extraction algorithm in ArcMAP 10.1 (ESRI 2012) for the months of June, July and August and a frequency table of northern hemisphere hydrilla occurrence by month was developed. A hydrilla frequency raster was created for each month and plotted using a scatterplot to identify minimum temperature values for known hydrilla occurrence sites during those months (Figure 1). Minimum occurrence values were then extrapolated across the United States and Canada for each month using a reclassification technique (ESRI 2012). Values within the WorldClim dataset that fell at or above the minimum hydrilla occurrence values were classified as “1” and values below were classified as “0”. A GIS was then used to identify potential hydrilla establishment throughout the northern hemisphere for each month. This was achieved by recoding values at or above the minimum potential hydrilla value as “1” and values below the minimum as “0”.

Next, data from the three months were combined to represent the minimum growing season required for tuber sprouting, growth and tuber regeneration. This was achieved through the use of various reclassifications and raster combinations. All months were combined and an average computed for minimum monthly temperature (Figure 2). No less than two months of hydrilla occurrence climate values were required for inclusion into the final potential establishment model to represent the minimum required growing period in the

Northern Hemisphere (Spencer et al. 1986). For example, if an individual cell of each raster did not exceed at least two months of the minimum required temperature for hydrilla occurrence, then that area was reclassified as having no potential for establishment. Furthermore, ice and glacial data was acquired and utilized to identify areas of permanent ice formation. This layer was then used to create an overlay with each raster dataset and areas within permanent ice formation were reclassified as having no potential for establishment.

Finally, low and high values from the final combined dataset of all three months were joined to identify the threshold at which hydrilla currently or previously existed during 1950-2010 time period. The minimum temperature and lowest occurrence at which hydrilla had previously been found was designated as having the lowest potential for establishment and the first peak of highest occurrence and highest temperature designated as the highest potential for hydrilla establishment (Figure 2). A linear regression model was subsequently developed based on occurrence data falling between the low and high temperature and lowest and highest worldwide occurrence values. Risk was subsequently split into 10 categories (1-lowest risk, 10-highest risk) based on the derived linear regression equation. A stretched continuous raster mapping was symbolized to display the potential establishment of hydrilla based on a stretch of the combined dataset histogram of minimum temperature (low occurrence value) and first peak (first highest occurrence value). This was completed in an attempt to most accurately represent hydrilla potential establishment categories based on the linear regression of existing and verifiable hydrilla occurrence data points to various temperature ranges.

4.2.3. State/ Province Risk Assessment

GIS layers for water bodies in both the United States and Canada were obtained from the National Hydrography Dataset (NHD) and ESRI North American Water Bodies (NAWB) dataset. Each dataset includes all inland water features in both Canada and the United States, therefore only Framework Classification Code (FCC) water bodies from the H (hydrography dataset) of codes H1 and H2 were used to extract only lakes and reservoirs (Canals, Streams, Pits, and gulf/bay features were eliminated) and surface area of each calculated in a GIS. The GIS was then used to reconstitute the boundaries of continuous water bodies and ensure they were not spatially represented as subsections of the same water body. This served to more adequately represent a count feature per State/ Province. A point feature representative of each water body was created for extraction of values representative of hydrilla potential establishment using a point extraction technique (ESRI 2012). Climate values were averaged across the entire water body and extracted to each point. The final dataset was examined thoroughly for outliers, duplicates and accuracy for quality assurance.

High and low values for potential establishment identified during risk development were used to identify states or provinces at ten different levels from low (1) potential to high (10) potential of hydrilla establishment. Potential establishment categories were separated using an equal interval assignment from the lowest temperature/ occurrence (lowest establishment potential) to the first peak of high temperature/ high occurrence (highest establishment potential) based on the worldwide risk scale (figure 2). An upper limit was not defined in this study as establishment potential is only limited by cold temperatures in North

America. Risk per state was assessed using the MEANS procedure in SAS Enterprise Guide 4.2 (SAS 2009). Those states/ provinces falling below the maximum establishment value of 10 (highest potential establishment) were subsequently investigated to determine the number of water bodies present within each category.

4.2.4. Risk Based on Water Body Size

States displaying a maximum potential establishment value (10) were not included in water body size assessment as potential establishment did not vary within the state's boundary. The NHD water bodies dataset was separated into three classes (small, medium and large) based on a normal distribution of the size in hectares (acres) per water body. Water bodies less than 40 ha (100 acres) in size were categorized in class 1. Water bodies greater than or equal to 40 ha (100 acres) but less than 4,050 ha (10,000 acres) were in class 2 and water bodies greater than 4,050 ha (10,000 acres) were in class 3. Risk of hydrilla establishment per water body was compared within each state by size using the mixed models ANOVA (proc mixed) procedure in SAS Enterprise Guide 4.2. (SAS 2009). A type-3 test of fixed effects was used to determine if either state, size class or their interaction were significant. A test of effect slices was used to investigate the interaction of state and size on a state by state basis (SAS 2009).

4.3. Results

4.3.1. Risk Scale

Locations at which Hydrilla has been recorded as established were identified at sites with a mean monthly low temperature of -8.5^{o}C for June through August, but these occurrences were few (< 5). The most frequent climate at which hydrilla has established occurs at equal to or greater than 6.7^{o}C mean monthly low temperature for the months of June-August. A gradient between -8.5 ^{o}C and 6.7^{o}C was therefore designated to represent the potential risk of hydrilla establishment from low to high (Figure 2). The dataset showed a linear trend between the high and low temperature values of occurrence with an R^2= 0.96 and a p-value = <0.0001 (figure 3). The ten risk categories divided among lowest occurrence (-8.5^{o}C and first highest occurrence (6.7^{o}C) represent a 136% increase in frequency per risk class on average. For example, risk class 2 represents a 136% increase in frequency of hydrilla occurrence compared to risk class 1.

4.3.2. State/ Province Risk Assessment

This risk gradient was applied to all values across the 67 states/provinces of the United States and Canada, and was mapped (figure 4). Risk was considered at absolute maximum (10) throughout the entirety of the United States mainland, excluding certain portions of the Rocky Mountains. The lower Canadian Provinces also displayed absolute highest risk of establishment with a gradient developing in the upper portions of Manitoba, Saskatchewan and Yukon Territories. Therefore, states and provinces along this gradient were given the most emphasis in our results.

4.3.3. Risk Based on Water Body Size

A total of 661,171 distinct water bodies were identified from the combined NHD and NAWB datasets that included various lakes, reservoirs and ponds across the United States and Canada. Water bodies ranged in size from 18,615,540 ha (46 million acres) (Great Bear Lake and all constituents – Northwest Territories) to less than 0.004 hecatres (1/100th of an acre) (Pond-Pennsylvania). The Northwest Territories contained the largest amount of total acreage (61,916,903 ha or 153 million acres). The District of Columbia contained the least acreage (219 ha or 541 acres) (Table 1). The state of Texas contained the greatest number of water bodies with just over 88,000 whereas the District of Columbia contained the least (10) (Table 1).

Twenty two states were identified as having water bodies which fell below the absolute highest establishment potential of 10 (Table 2). While there were no significant differences between size classes pooled across these 22 states, there were differences for size classes within individual states and size classes (Table 3). Of states/ provinces not identified at the absolute highest potential for hydrilla establishment, Ontario, Saskatchewan, Arizona and New Mexico were identified as being the next highest potential for infestation (Table 2). The Nanavut Territories and Idaho displayed the lowest potential for infestation of all 67 states/provinces assessed (Table 2). In terms of individual water bodies, none of the 22 states or provinces contained a water body with a risk level of less than 3 (Table 2).

Eleven states/ provinces showed significant differences in mean risk based on water body size classes within those states/provinces. Newfoundland and Labrador, Alaska,

Quebec and Idaho all had distinctly different risk means across all three size classes (Table 2). Alberta and Montana showed differences in means among classes 1 and 2, with class 3 being not significantly different from 1 or 2. California, Utah, British Columbia, Wyoming and the Nanavut Territories each had one class that was significantly different than the other two classes (Table 2).

4.4. Discussion

The expansion of *Hydrilla verticillata* in the United States and Canada is a real risk to both the ecologic and economic stability of North America. The model developed here provides insight on the potential establishment of *H. verticillata* in this region based on readily available and statistically relevant datasets of the troublesome invader from other parts of the world. It shows that hydrilla can potentially establish much farther north than what had previously been modeled or inferred (see Peterson et al. 2003, Langeland 1996, Araujo and Peterson 2012). An apparent gradient exists along the northern tier provinces of Canada, through parts of Alaska and along the Rocky Mountains in the United States. Several states and provinces lie on this gradient including parts of Ontario, Saskatchewan, Arizona and New Mexico. More importantly, various states previously thought to be unlikely for establishment, and without current established populations of *H. verticillata*, fell well within the highest level of risk for such establishment. These include the northern states of Minnesota, Michigan, North Dakota, and Wisconsin as well as the Southern Canadian Provinces of Prince Edward Island, Nova Scotia, and New Brunswick. Water resource managers and researchers alike should take note of potential hydrilla establishment in these

states/ provinces, potentially allocating resources and funding for hydrilla prevention and monitoring statewide. This is especially true in states bordering areas with *H. verticillata* already established (i.e. Minnesota being north of Iowa).

At the state and federal regulatory levels, limited budgets exist for aquatic invasive plant species management, especially in terms of prevention and monitoring. Our model attempts aid in available resource flexibility and management prioritizations by identifying states/provinces which fall below the maximum risk level of 10, and determine if there are any differences in risk based on the size of water body within those states/provinces. By determining if risk differences exist between water body size classes within a state/province, resource managers and personnel can better allocate preventative actions with a limited budget. For example, Newfoundland and Labrador, Alaska, Quebec and Idaho each had significantly different risk values across the three size classes (table 2). Newfoundland and Labrador, Alaska and Quebec all saw greater risk in smaller size class 1 water bodies, followed by size class 2 whereas Idaho saw greater risk in larger size class 3 water bodies followed by size class 2 water bodies. On the contrary, California and Utah saw size class 1 water bodies as being of least risk throughout the state (table 2). Whereas the Nunavut Territory saw class 3 water bodies as being of least risk (table 2). This information can be extremely beneficial to resource managers in charge of water bodies within these states and others.

Although not included in our initial objectives, the potential for climate change may affect the risk gradient of *H. verticillata* establishment potential throughout the United States

and Canada. The Intergovernmental Panel on Climate Change (IPCC 2007) suggests an average temperature increase of 0.2^{o}C per decade in the next century as a conservative estimate. Should this change be realized, a 1°C increase will be seen in the next 50 years, a 3 degree C increase in 150 years and a 5°C increase in 250 years. Assuming no inter-latitudinal shift in regional climates, the line of maximum establishment for *H. verticillata* would shift much farther north with each unit of increase in temperature. Changes at 1°C, 3°C, and 5°C were mapped and are included in Appendix C.

4.5. Conclusions

Climate envelope models are an extremely useful tool to model the establishment potential of invasive species like *H. verticillata*. In this study, the climate envelope model provides insight on the potential establishment of *H. verticillata* throughout the United States and Canada. Of course, these models can only be improved by an increase in documented reports of established populations worldwide. This is especially true in developing countries in Africa where the literature contains vague locations of *H. verticillata*, but few spatial datasets currently exist to determine exact locations of the plant. Climate envelope models have various limitations which have been the topic of speculation in various papers over the years (Hampe 2004, Dormann 2007, Sinclair et al. 2010). These models have been particularly criticized for making simplified assumptions of a species potential range without taking into account dispersal, biotic interactions and limitations such as barriers. Dispersal mechanisms obviously play an integral role in the expansion of *H. verticillata,* but these variables are difficult to measure on such a global or even regional scale. Such is the case of

modeling the potential establishment range of hydrilla, in which inclusion of these variables is not relevant as the model only attempts to outline the potential spatial distribution of hydrilla based on suitable conditions across a landscape rather than taking into account processes such as dispersal. This may not be true in other applications, such as modeling extinction risk of a given species, in which there is no quantifiable variable that can be represented in the input data to represent the extinction or decline of that species (Araujo and Peterson 2012).

Furthermore, there are few factors which limit submersed aquatic plant growth including light, soil, salinity, nutrients and temperature (Wetzel 2001), the latter of which was modeled in this paper. For hydrilla in particular, light is hardly constraining as the plant can grow and produce subterranean turions (tubers), arguably its main dispersal mechanism, under relatively low light. Some have shown that all races of hydrilla worldwide produced tubers (91-7182) under short light periods which would certainly be exceeded during the long-lighted days of June, July and August used in this model (Steward 1987, Van and Steward 1990). Salinity, with the exception of Great Salt Lake in Utah, was subsequently eliminated in our assessment as only land-locked, freshwater lakes and reservoirs were modeled. Lastly, soil types are rarely collected for aquatic systems, especially worldwide and were therefore not included in our model. Each of these variables can be accounted for on smaller spatial scales, but was not deemed appropriate for inclusion in the models based on the initial study objectives. Future studies regarding the northern range of *H. verticillata* in North America should investigate these variables at smaller scales to refine model results.

Future studies should also attempt to address the means of dispersal and expansion used by hydrilla. Unlike various fauna species which can disperse by means of immigration and emigration across a landscape, a rooted aquatic plant is limited to expansion within a waterbody. Once established, an aquatic plants spreads within a water body based on its own survival requirements but cannot extend beyond that barrier unless assisted (Madsen and Owens 2000). Our model only attempts to assess establishment potential based on unlimited and unrestrained dispersal through an assisted vehicle (i.e. human-aided introduction). Obviously, the proximity to existing invasive populations of hydrilla, proximity to human interaction, etc. play a large role in the ability of hydrilla to move from water body to water body. Four U.S. states (Washington, California, Colorado, and Idaho) identified in our model as having water bodies of less than the maximum risk of 10 currently have existing *H. verticillata* populations within the state. Six others (Arizona, Nevada, New Mexico, Utah, and Wyoming) share the border of a state with *H. verticillata* currently established. These states may be of higher risk due to the proximity to water bodies where Hydrilla is currently established. Hydrilla has often been introduced through aquarium dumps, and through boat movement from one water body to the next (Barnes 2013, de Winton 2009, Coetzee 2009), but discovery of hydrilla in remote farm ponds and other locations may implicate other modes of transport. Little research has been done on the potential for birds, particularly waterfowl as a vehicle for movement. Waterfowl cover hundreds of thousands of kilometers annually, inhabiting various water bodies during annual migrations. New research is needed to investigate the role of waterfowl and other animals in dispersing invasive aquatic plants like hydrilla to determine what role dispersal mechanisms truly play in driving hydrilla

spread. This model also only attempts to identify areas at which hydrilla has the potential to establish and makes no attempt to address the infestation or impact potential once established. This can certainly vary widely based on a number of conditions. For example, Madsen and Owens (2000) showed that hydrilla can spread through a wide range of temperatures based on average annual air temperature, but the rate of spread greatly increases at 12°C. Some areas identified within this model may not reach that optimum threshold and establishment would theoretically not lead to infestation.

The climate envelope model developed in this study, although simplistic, provides a present-day interpretation of the risk of potential hydrilla establishment based on a large, comprehensive dataset. The regression analysis of this dataset provides a way to assess risk quantitatively based on the increase in global occurrence as temperature increases. The model also provides valuable information about hydrilla establishment potential within a state or province while additionally assessing potential differences in risk among differences in the size of water bodies. This modeling technique offers a promising new source of inferences with predictive connotations regarding the potential establishment of non-native, aquatic plant species. The modeling approach has the advantage of using readily available data to predict the potential establishment of such species on a global or regional scale. This information can provide researchers and resource managers ~~alike~~ with a general prediction of such invasive species establishment, therefore allowing them to better combat the possibility of colonization in large water bodies.

4.6. References

Araujo MB and Townsend Peterson A. 2012. Uses and misuses of bioclimatic envelope modeling. Ecology 93: 1527-39.

BALCIUNAS J and CHEN P. 1993. Distribution of hydrilla in northern China - implications on future spread in North America. J Aquat. Plant Manage. 31:105-109.

Barbosa FG, Schneck F, Melo AS. 2012. Use of ecological niche models to predict the distribution of invasive species: A scientometric analysis. Braz. J. Biol. 72: 821-829.

Barnes MA, Jerde CL, Keller D, Chadderton WL, Howeth JG, Lodge DM. 2013. Viability of aquatic plant fragments following desiccation. Invasive Plant Science and Management abbrev. 6: 320-5.

Bailey JE and Calhoun AJK. 2008. Comparison of three physical management techniques for controlling variable-leaf milfoil in Maine lakes. J. Aquat. Plant Manage. 46: 163-167.

Broennimann, O., U. A. Treier, H. Muller-Scharer, W. Thuiller, A. T. Peterson, and A. Guisan. 2007. Evidence of climatic niche shift during biological invasion. Ecology Letters 10:701–709.

Coetzee JA, Hill MP, Schlange D. 2009. Potential spread of the invasive plant hydrilla verticillata in South Africa based on anthropogenic spread and climate suitability. Biol. Invasions 11: 801-812.

de Winton MD, Champion PD, Clayton JS, Wells RDS. 2009. Spread and status of seven submerged pest plants in New Zealand lakes. N Z J Mar Freshwat. Res. 43: 547-561.

Dormann, C. F. 2007. Promising the future? Global change projections of species distributions. Basic and Applied Ecology abbrev. 8: 387–397.

ESRI 2012. ArcGIS Desktop: Release 10.1. Environmental Systems Research Institute, Redlands, CA.

ESRI. 2012. ArcGIS Desktop Help 10.1 – About Inverse Distance Weighted. ONLINE. Available: http://help.arcgis.com/en/arcgisdesktop/10.0/help/index.html#//00310000002m000000.

Gallardo B and Aldridge DC. 2013. The 'dirty dozen': Socio-economic factors amplify the invasion potential of 12 high-risk aquatic invasive species in Great Britain and Ireland. J. Appl. Ecol. 50: 757-766.

Hampe, A. 2004. Bioclimate envelope models: what they detect and what they hide. Global Ecology and Biogeography 13: 469–471.

Harlan, S. M., G. J. Davis, and G. J. Pesacreta. 1985. Hydrilla in three North Carolina lakes. J. Aquat. Plant Manage. 23:68-71.

Hijmans, R.J., S.E. Cameron, J.L. Parra, P.G. Jones and A. Jarvis, 2005. Very high resolution interpolated climate surfaces for global land areas. International Journal of Climatology 25: 1965-1978.

IPCC (Intergovernmental Panel on Climate Change). 2007. Climate change 2007: The physical science basis. www.ipcc.ch.

Langeland, K.A. 1996. *Hydrilla verticillata* (L.F.) Royle (Hydrocharitaceae), "The Perfect Aquatic Weed".
Castanea. 61(3): 293-304.

Leroux, S.J., M. Larrivee, V. Boucher-Lalonde, A. Hurford J. Zuloaga, J.T. Kerr, and F. Lutscher. 2013. Mechanistic models for the spatial spread of species under climate change. Ecol. Appl. 23: 815-828.

Madeira, P.T., C.C. Jacono, and T.K. Van. 2000. Monitoring hydrilla using two RAPD procedures and the nonindigenous aquatic species database. J. Aquat. Plant Manage. 38:33-40.

Madsen, J. D., and Owens, C. S. (2000). "Factors Contributing to the Dispersal of Hydrilla in Lakes and Reservoirs," Aquatic Plant Control Technical Notes Collection (ERDC TN-APCRP-EA-01), U.S. Army Engineer Research and Development Center, Vicksburg, MS. www.wes.army.mil/ellaqua/aqtn.html, last accessed in May 2013.

Maki, K.C. and S.M. Galatowitsch. 2008. Cold tolerance of two biotypes of hydrilla and northern watermilfoil. J. Aquat. Plant Manage. 46:42-50.

Miller, R.W. 1998. The First State's experiences controlling the northern monoecious form of hydrilla. Aquatics 10: 16-23.

Netherland, M. D. 1997. Turion ecology of hydrilla. J. Aquat. Plant Manage. 35: 1-10.

Owens, C. S. & J. D. Madsen, 1998. Phenological studies of carbohydrate allocation in hydrilla. J. Aquat. Plant Manage. 36: 40–44.

Pearson R. G., Dawson T. P. (2003). "Predicting the impacts of climate change on the distribution of species: are bioclimate envelope models useful?" Global Ecology and Biogeography 12: 361-371.

Peterson A, Papes M, Kluza D. 2003. Predicting the potential invasive distributions of four alien plant species in North America. Weed Sci. 51: 863-868.

Richardson et al. 2012. Monoecious Hydrilla – A Review of the Literature. Northeast Aquatic Nuisance Species Panel. Accessed Online: http://www.nyis.info/user_uploads/files/Monoecious%20Hydrilla%20Lit%20Review%20-%20Final.pdf, last accessed in May 2013.

SAS Institute Inc. 2009. Administering SAS Enterprise Guide 4.2. Cary, NC: SAS Institute Inc., Cary, NC.

Sheppard, C.S. 2013. Potential spread of recently naturalised plants in New Zealand under climate change. Clim Change 117: 919-31.

Sinclair, S. J., M. D. White, and G. R. Newell. 2010. How useful are species distribution models for managing biodiversity under future climates? Ecology and Society 15(1):8. http://www.ecologyandsociety.org/vol15/iss1/art8, last accessed in ____.

Spencer, D.F. and L.W.J. Anderson. 1986. Photoperiod responses in monoecious and dioecious *Hydrilla verticillata*. Weed Sci. 34: 551-557.

Steward, K.K. and T.K. Van. 1987. Comparative studies of monoecious and dioecious hydrilla (*Hydrilla verticillata*) biotypes. Weed Sci. 35: 204-210.

Van, T. K. and K. K. Steward. 1990. Longevity of monoecious hydrilla propagules. J. Aquat. Plant Manage. 28: 74-76.

Villemant, C., M. Barbet-Massin, A. Perrard, F. Muller, O. Gargominy, F. Jiguet, and Q. Rome. 2011. Predicting the invasion risk by the alien bee-hawking yellow-legged hornet *Vespa velutina nigrithorax* across Europe and other continents with niche models. Biological Conservation 144: 2142–2150.

Wetzel, R.G. 2001. Limnology, 3rd edition. Academic Press, New York.

Table 4.1. Summary statistics of all U.S. states/ Canadian provinces analyzed in model development.

State/ Province	Mean Acreage	Min Acres	Max Acres	Total Acreage	N	State/ Province	Mean Acreage	Min Acres	Max Acres	Total Acreage	N
AB	7964.84	0.10	7431460	15260630	1916	NE	59.06	0.02	50001	370823	6279
AK	5990.61	0.84	2453960	19080097	3185	NH	108.92	0.05	84906	308785	2835
AL	38.54	0.03	76736	638376	16565	NJ	15.46	0.04	5119	111779	7228
AR	64.00	0.01	67982	747599	11681	NL	17855.46	0.04	5026510	14498630	812
AZ	67.22	0.02	47063	210267	3128	NM	48.25	0.11	38678	314284	6514
BC	4858.94	0.20	1031970	9669295	1990	NS	158.51	0.04	76934	625006	3943
CA	180.36	0.02	224218	2272685	12601	NT	125999.32	17.75	46015300	153089178	1215
CO	43.67	0.01	25365	420675	9633	NU	73219.39	0.06	6452700	125864137	1719
CT	24.14	0.07	7059	116021	4807	NV	801.60	0.04	225252	965128	1204
DC	54.11	0.44	176	541	10	NY	75.38	0.02	96873	1236831	16408
DE	20.62	0.10	1148	13509	655	OH	1130.05	0.03	11391400	11711854	10364
FL	62.52	0.02	441931	1966800	31461	OK	71.93	0.02	150075	970971	13498
GA	25.56	0.01	98029	772259	30212	ON	15193.01	0.03	8799680	46156351	3038
HI	18.16	0.27	902	6810	375	OR	253.08	0.11	106630	1072559	4238
IA	31.39	0.02	25440	231367	7371	PA	11.75	0.00	28280	316031	26895
ID	437.88	0.23	197175	994435	2271	PE	18.79	0.23	710	9807	522
IL	26.00	0.03	38822	455184	17505	PR	25.92	0.06	883	9981	385
IN	22.90	0.04	18578	248702	10860	QC	23004.33	0.06	4761480	62801834	2730
KS	30.28	0.03	26278	430910	14230	RI	33.46	0.11	6413	42564	1272
KY	60.18	0.02	69174	336960	5599	SC	57.62	0.02	132102	495937	8607
LA	92.70	0.04	78360	1389773	14992	SD	68.83	0.05	554033	1778386	25836
MA	33.04	0.02	43232	272846	8258	SK	38430.37	0.07	5778700	31628197	823
MB	67232.61	0.75	15833200	53718857	799	TN	17.96	0.01	39202	302725	16853
MD	15.25	0.09	6744	60140	3943	TX	31.25	0.01	236258	2764158	88460
ME	536.07	0.15	157758	2005454	3741	UT	1014.05	0.09	1827850	2871800	2832
MI	4216.42	0.03	40763500	89059267	21122	VA	24.16	0.03	67796	244681	10128
MN	261.07	0.05	2256330	8229360	31522	VT	552.44	0.07	492489	600498	1087
MO	22.20	0.02	91110	550245	24791	WA	163.50	0.04	73255	718263	4393
MS	37.20	0.06	77218	474812	12765	WI	251.36	0.05	1263020	3015858	11998
MT	116.76	0.04	537344	1835530	15721	WV	28.96	0.10	4188	35847	1238
NB	209.49	0.07	37470	230440	1100	WY	90.26	0.07	165667	767207	8500
NC	40.25	0.02	61760	477372	11861	YT	41376.86	47.28	406491	4261817	103
ND	73.74	0.15	366413	2399661	32544	Total	6663.08	1.09	2568504	684538786	661171

Table 4.2. Mean establishment risk of states/ provinces falling along the maximum risk gradient and mean establishment risk between water bodies within states/provinces.

State/ Province	Mean Risk	Size Class 1	Class 2	Class 3	Risk Min	Max	Pr > F
ON	9.9997	10	10	9.9979	9	10	0.9994
SK	9.9721	10	9.997	9.9481	9	10	0.8489
AZ	9.9658	9.9668	9.9369	10	8	10	0.9675
NM	9.9065	9.9051	9.9407	10	6	10	0.8976
AB	9.8711	**9.8171**	**9.9641**	9.8841	7	10	**0.0549**
MB	9.8135	9.9821	9.9195	9.7407	8	10	0.0769
NV	9.7716	9.7741	9.7423	10	6	10	0.7446
WA	9.5941	9.5842	9.6852	10	6	10	0.1571
NL	9.5333	**9.973**	**9.689**	**8.9124**	7	10	**<0.0001**
MT	9.4592	**9.4669**	**9.2755**	9.8333	3	10	**0.0003**
AK	9.3859	**9.7814**	**9.3118**	**8.9692**	5	10	**<0.0001**
BC	9.3844	**9.5674**	9.2966	9.2973	6	10	**<0.0001**
CA	9.3728	**9.3662**	9.4632	9.833	4	10	**0.0154**
OR	9.2209	9.2126	9.3014	9.35	4	10	0.3799
QC	9.2165	**9.9543**	**9.7187**	**8.5522**	6	10	**<0.0001**
CO	9.1915	9.1933	9.1457	9	4	10	0.7426
UT	9.0357	**8.9729**	9.6781	9.9286	5	10	**<0.0001**
NT	8.7893	8.3333	8.6296	8.7953	5	10	0.5243
WY	8.7027	8.7315	**8.0714**	9.0714	3	10	**<0.0001**
YT	8.2233	8	8.1	8.2391	6	10	0.9292
ID	7.668	**7.5443**	**8.9956**	**9.6154**	3	10	**<0.0001**
NU	6.6998	7.25	7.06	**6.6809**	4	9	**0.0093**

*Bold denotes significant difference p<0.05.

Table 4.3. Type III test of mixed effects for state and size class.

Effect	Num DF	Den DF	F Value	Pr > F
State	21	89364	64.46	**<0.0001**
Size Class	2	89364	0.22	0.802
State*Size Class	42	89364	24.76	**<0.0001**

*Bold denotes significant difference p<0.05

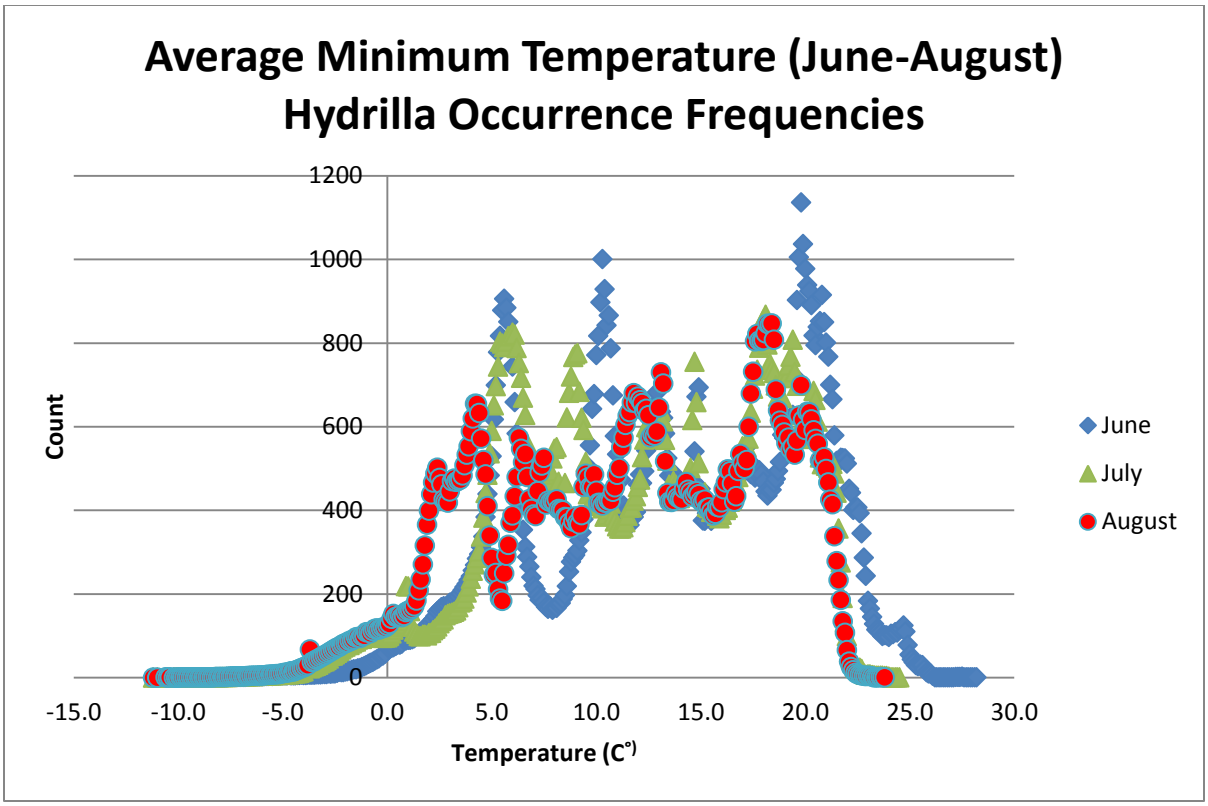

Figure 4.1. Average minimum monthly temperature (June, July and August) of *H. verticillata* occurrences.

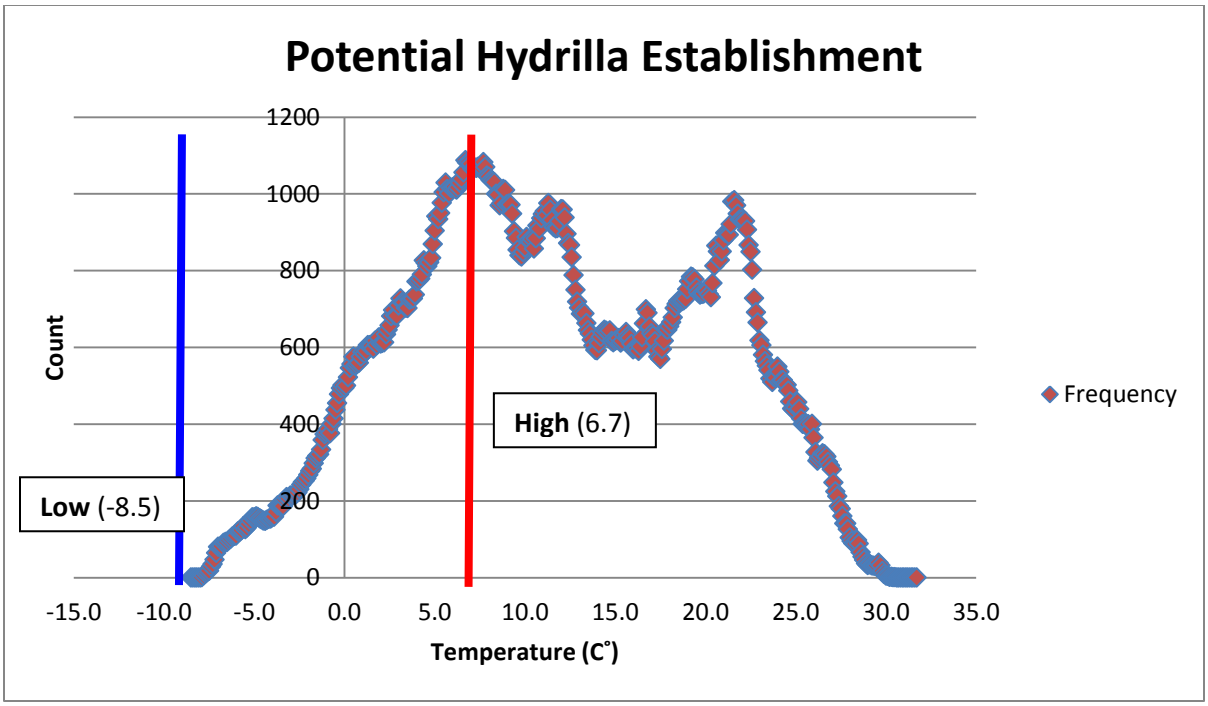

Figure 4.2. Potential *H. verticillata* establishment based on average of all growing season months.

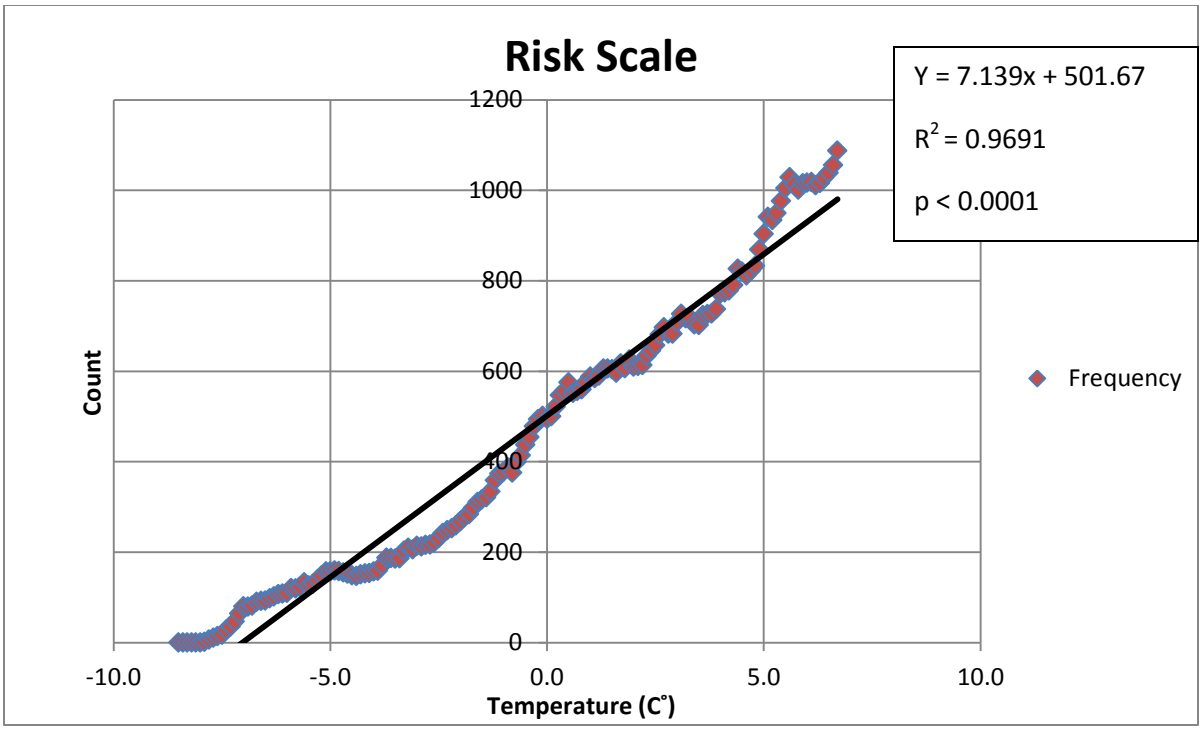

Figure 4.3. Risk scale of *H. verticillata* and linear regression of minimum (low occurrence, low temperature) and maximum (high occurrence, first peak) risk.

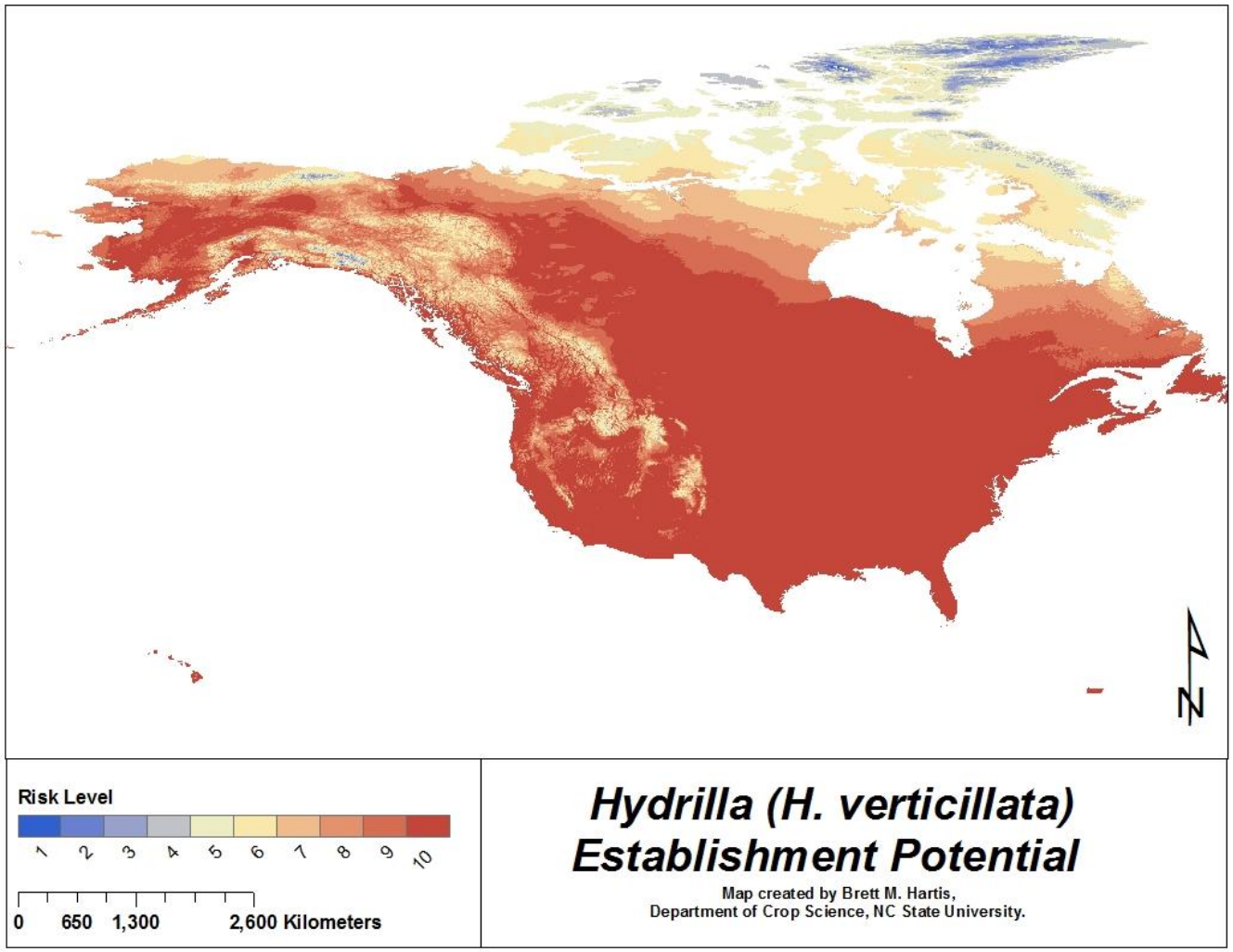

Figure 4.4. Model representation of *H. verticillata* establishment potential in the United States and Canada

CHAPTER 5

5.1. CONCLUSIONS

This research uses various technological advancements including but not limited to remote sensing, GIS, and geospatial modeling in an attempt to better map, monitor and model SAV species and communities. While certain aspects of techniques described in this work have been used for various applications in the past, the contribution of this work can improve existing knowledge in the discipline of aquatic plant science. The techniques described in chapters 2 and 3 present the framework to map and monitor SAV in shallow, coastal systems, arguably one of the most diverse and ecologically important aquatic systems. The modeling procedure described in chapter 4 provides a new outlook of *H. verticillata* establishment not previously thought possible. Efforts in each chapter, although limited in some respects, provide an encompassing look at mapping, monitoring and modeling submersed plants on both local and global scales.

5.2. Chapter 2: Does remote sending provide a viable alternative to traditional survey techniques of SAV in shallow, coastal systems?

This study showed the utility of remote sensing under less than ideal conditions. The purpose of this study was to test the viability of using newly available sensors to detect SAV in conditions not suitable for traditional approaches. The Worldview-2 sensor provides a useful model for detecting SAV but is limited in its applicability at deeper depths in which band 4, identified as the key predictor, begins to lose efficiency. The efficacy of the Quickbird sensor still remains somewhat untested as external variables such as clouds and

issues with acquisition hindered model development. LANDSAT5, although free has a number of limitations making it a less than ideal candidate for use in detecting SAV in such patchy distributions as those found in Currituck Sound, NC.

The models developed with Worldview-2 highlighted the need for further study of this sensor as an alternative for mapping SAV. Future work should certainly take into account certain measures of plant structure in the "Z" dimension including height of canopy and biomass. The Quickbird sensor may provide similar results to those of the Worldview-2 given the comparable spatial and spectral properties. External influences created problems in developing models for this particular sensor.

5.3. Chapter 3: Do spatial interpolations offer an accurate picture of SAV species and community dynamics?

This effort utilized interpolation based on spatial relationships to provide a picture of individual species distribution across a relatively large study area. While the maps developed in this study provide a sound-wide view of SAV species extent, they do not present an accurate estimation of exact space and size encompassed by various stands of vegetation. This limitation is not new to submersed aquatic plant science, as measuring what we cannot see in its entirety without some degree of interpolation is always required. Traditionally, the only way to overcome such obstacles was to collect data at geographic locations as close to one another as possible; however this can be inversely related to the time and cost required for such surveys. Advancements in measuring the underwater continuity of SAV including hydroacoustics and underwater videography continue to tighten gaps between sample points;

however, even these techniques require interpolation and complex algorithms.

This interpolation technique, while simple, does give resource personnel and scientists a tool to determine management efforts in such large systems. The means of measuring SAV species dominance described in this work provides more insight into the mixed SAV communities of our coastal systems. Currently, work is being completed to hybridize methods like hydroacoustics with dominance metrics to provide a better estimation of underwater vegetation. Non species-specific SAV biomass and coverage estimates as determined by such technology are being combined with similar dominance metrics described in this study, to inform management across the nation. Future work should build on the dominance metrics developed in this study to continue to provide precision and accuracy in mapping between measured points.

5.4. Chapters 2 and 3: Implications for impairment of SAV in the Currituck Sound

The ecologic history of the Currituck Sound is only briefly touched upon in chapters 2 and 3 of this work; however, a very rich past and somewhat disheartening current state of the Sound are highlighted in the literature cited. The Currituck Sound has a very rich and ever changing history. Pre-colonial (Mulford 2002) and colonial times (Brimley 1949) took advantage of an extremely productive and lucrative ecosystem that offered rich waterfowl hunting and fishing opportunities. The area was estimated to have hosted more waterfowl than any other state along the Atlantic flyway during colonial times in part, due to the large expanses of SAV (Conoley 1982). Waterfowl were so abundant in the Sound that by the mid-1800s, locals began market hunting the birds to be shipped worldwide, sometimes

harvesting more than a thousand birds in one day (Johnson and Coppedge 1991, Lichon 1998). Inlet closures in the 18th century led to a nearly fresh water system, giving rise to more diverse communities of SAV as well as various fresh to brackish water fauna including blue crab, largemouth bass, oysters (Stick 1958). Up to sixteen different species were identified within the Sound during this era (Sincock 1966, Davis and Brinson 1989).

In the early 1900s, the dominant species of vegetation in the Currituck Sound was sago pondweed followed by bushy pondweed and wild celery. In the 1960s, the Sound saw a shift towards widgeongrass as the dominant species. Up until the 1960s there had been no written report of invasive species having been found, however, a bioinvader known as Eurasian Watermilfoil (*Myriphyllum spicatum*) began to show up in the mid-1960s (Kearson 1976) and had fully colonized the Sound by 1968 (Borawa et al. 1978). The noxious weed remained the dominant species until the late 1970s (Fish 1974) at which time both the invasive *M. spicatum* and various native species began to decline rapidly (Davis and Brinson 1983). Very few sound-wide assessments of SAV were completed during the following decades however speculation and pilot studies suggested a collapse in overall SAV coverage and distribution (Baker and Valentine 2007) followed by a decrease in waterfowl and freshwater fish species (Swihart and Moss 2003, Borawa et al. 1978).

The cause of such a collapse in SAV and other species is as mysterious as the disappearance of those species themselves. Speculations are many ranging from poor water quality, fluctuations in salinity and even indirect effects from increased development but few concrete data sources and published literature are available to confirm such claims. Sincock

suggested impairments to water quality as early as 1960 (1966) whereas Riggs et al suggests increased development of the surrounding landscape as a contributor to nutrients and increased turbidity (2008). Despite the cause, only 35% of SAV distribution seen 25 years ago is estimated to exist today (USACE 2006).

Chapters 2 and 3 shed further light on the troubling current state of a system which once flourished with SAV. The dynamic nature of the Currituck Sound must be more fully understood before irreversible damage takes place. As development in the Currituck watershed continues at near break-neck pace, new management strategies must be developed to help protect and perhaps even enhance such a vital resource. Future work targeting the mapping and monitoring of system-wide SAV, such as that suggested in chapters 2 and 3, should provide important information about the current state of this system and suggest avenues for improvement. Future work in the Sound should also explore the relationships between the decline in SAV and simultaneous declines in waterfowl and fish species. Without adequate study of this system and its intricate processes, assessments cannot be accurately conducted to determine best management strategies. With increased research effort, public support and funding, the area may one day be restored to its historical benevolence.

5.5. Chapter 4: Does *H. verticllata* really have the potential to become invasive across the northern ranges of North America?

This study evaluated the establishment potential of *H. verticillata* across the United States and Canada based on worldwide climate and currently existing populations. The picture painted by this model suggests that *H. verticillata* can establish well beyond extents previously thought, into the sub-arctic ranges of Canada and the state of Alaska. This model, while useful for planning and prevention, does not address the invasive potential of *H. verticillata* once established in any of these water bodies. The short growing window (less than 6 weeks) in some locations suggests that *H. verticillata* could establish but may never reach extreme nuisance potentials as seen in water bodies currently infested farther to the south. This model should only be used as a tool for assessing the potential for this particular invasive to be introduced and complete it's lifecycle within a particular water body.

The expansion and dispersal of *H. verticillata* would likely play a very large role in the true introduction of the plant into water not previously inhabited. The relationship of climate and establishment potential identified in this study is simply a correlation and does not represent causation. Future work should attempt to address such means of introduction, whether a product of human interaction (i.e. boat, aquarium, etc) or wildlife (i.e. waterfowl movement). Proximity to currently infested water bodies should also be included in any further assessment of *H. verticillata* spread. It is recommended that relationships between currently existing populations of *H. verticillata* and potential contributors to spread be investigated further.

5.6. REFERENCES

Baker MD and SC Valentine. 2007. Historical Populations and Long-term Trends of Waterfowl, Fish, and Threatened/ Endangered Species within Back Bay, VA and Currituck Sound, NC. Back Bay, Mackay Island, and Currituck National Wildlife Refuges' Consolidated Report. Available ONLINE: http://www.fws.gov/northeast/planning/back%20bay/pdf/draft_ccp/18w_Entire_Document%285131KB%29.pdf. last accessed in March2011.

Borawa, J, Kerby, J, Huish, M and A Mullis. 1978. "Currituck Sound fish populations before and after infestation by Eurasian water-milfoil," Proceedings of the Southeastern Association of Fish and Wildlife Agencies 32: 520-528.

Brimley, H. 1949. A North Carolina Naturalist, edit. By Eugene Odom, University of North Carolina Press, Chapel Hill.

Conoley, W Jr. 1982. Waterfowl Heritage: North Carolina Decoys and Gunning Lore. Webfoot Inc., Wendell

Davis, G and M Brinson. 1989. A Survey of Submerged Aquatic Vegetation of the Currituck Sound and Western Albemarle-Pamlico Estuarine System. Albemarle-Pamlico Estuarine Study Project No. 89-100.

Johnson, A and B Coppedge. 1991. Gun Clubs & Decoys of Back Bay and Currituck Sound. Johnson and Coppedge, Virginia Beach

Kearson, L. 1976. Observations on the flora of Currituck Sound. Mimeo. Report, North Carolina Wildlife Resources Commission, Raleigh NC.

Lichon, C. 1998. Waterfowling Boats, Blinds, and Related Gear. Outdoor Publications

Mulford, C. 2002. Early American Writings. New York: Oxford University Press.

Riggs, S., Culver, S., Ames, D., Mallison, D. Corbett, D.,and J. Walsh. 2008. North Carolina's Coasts in Crisis: A Vision for the Future. North Carolina Coastal Geology Cooperative Research Program.

Sincock, J.L. 1965. Back Bay – Currituck Sound data report. U.S. Fish and Wildlife Service, North Carolina Wildlife Resources Commission, and Virginia Commission of Game and Inland Fisheries. 1600.

Stick, D. 1958. The Outer Banks of North Carolina. Raleigh: University of North Carolina Press,

Swihart, G. and L. Moss. 2003. Assessment of Recreational Fishery Resources for Enhanced Public Use at Horn Point, Back Bay National Wildlife Refuge. Unpublished report by the U.S. Fish & Wildlife Service, Gloucester Fishery Resources Office, 6669 Short Lane, Gloucester, VA 23061.

United States Army Corps of Engineers – Wilmington District. 2006. Currituck Sound: General Investigation Feasibility Study, Project Management Plan.

APPENDICES

Appendix A – Chapter 2 Supplement

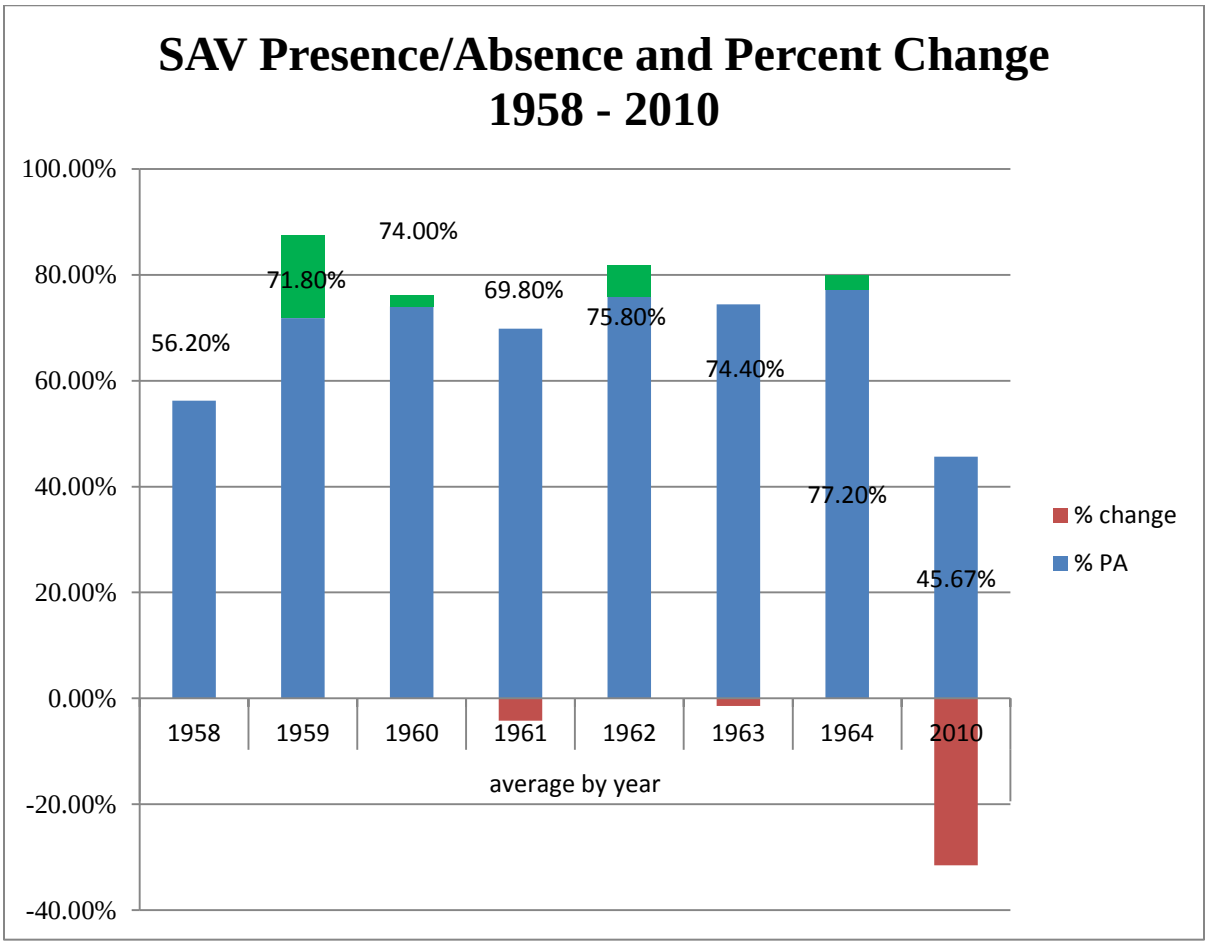

Figure A.1. SAV presence/ absence and percent change over time in Currituck Sound

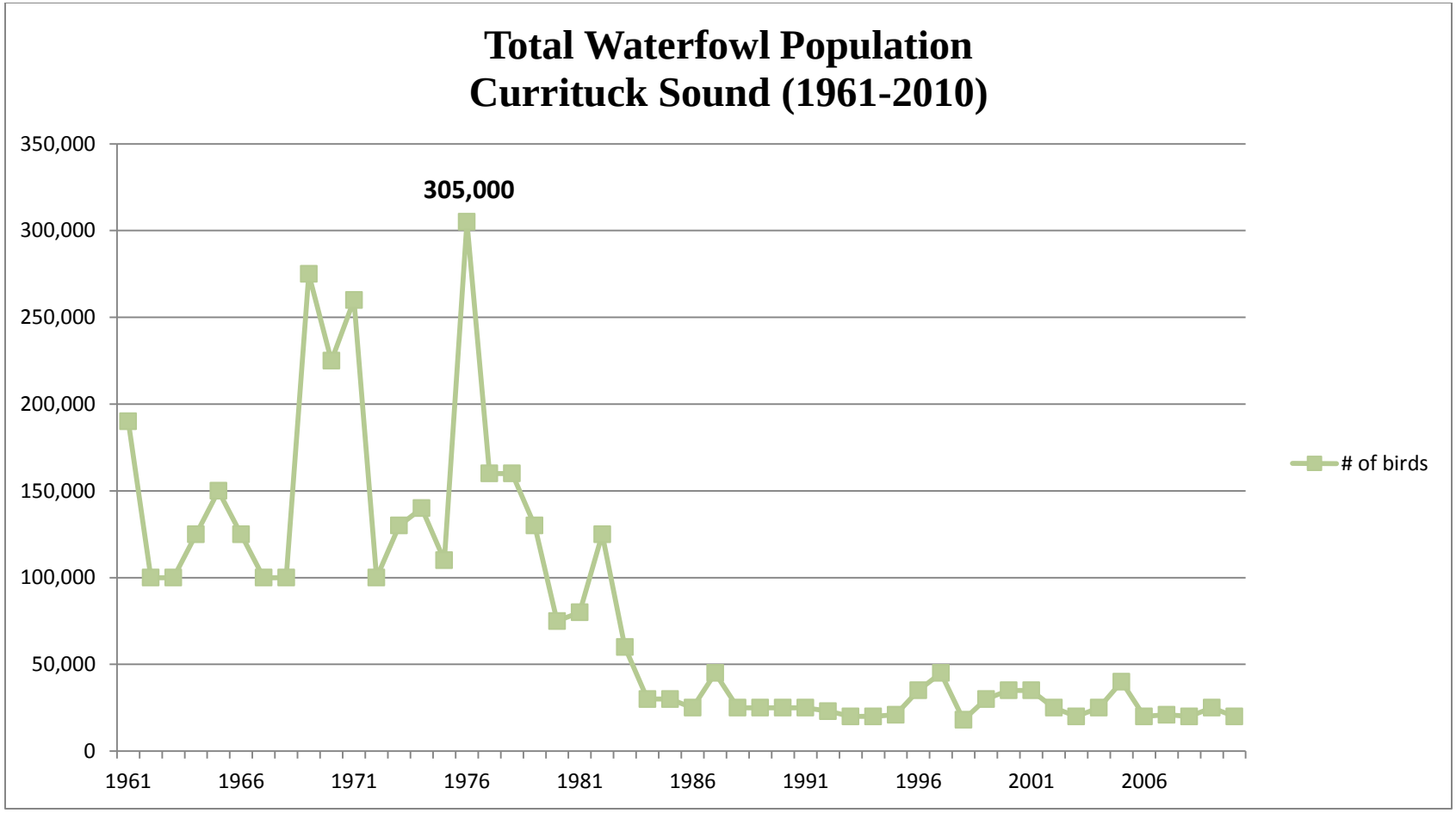

Figure A.2. Total waterfowl population estimates within the Currituck Sound (as adapted from Baker and Valentine 2007).

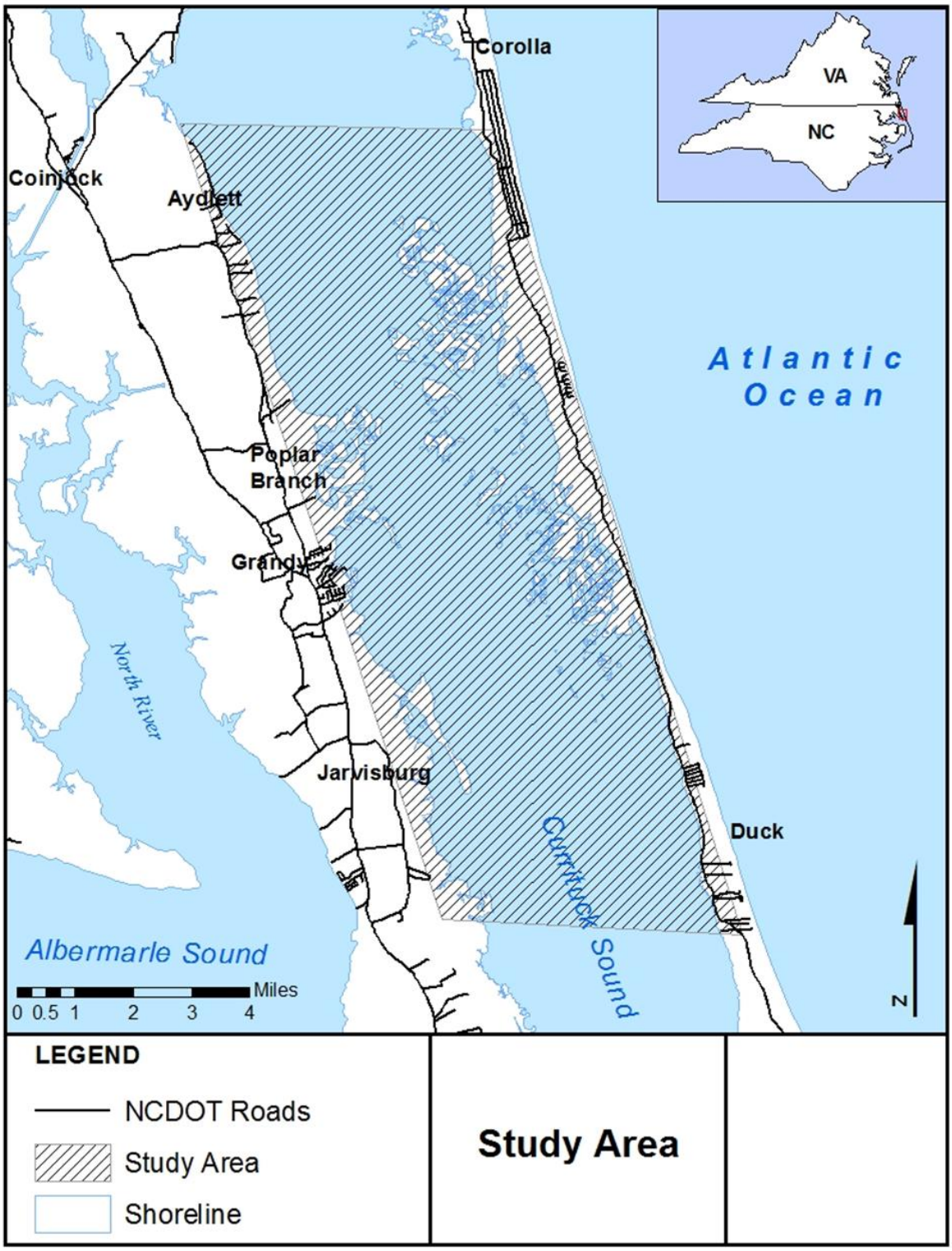

Figure A.3. Image acquisition area.

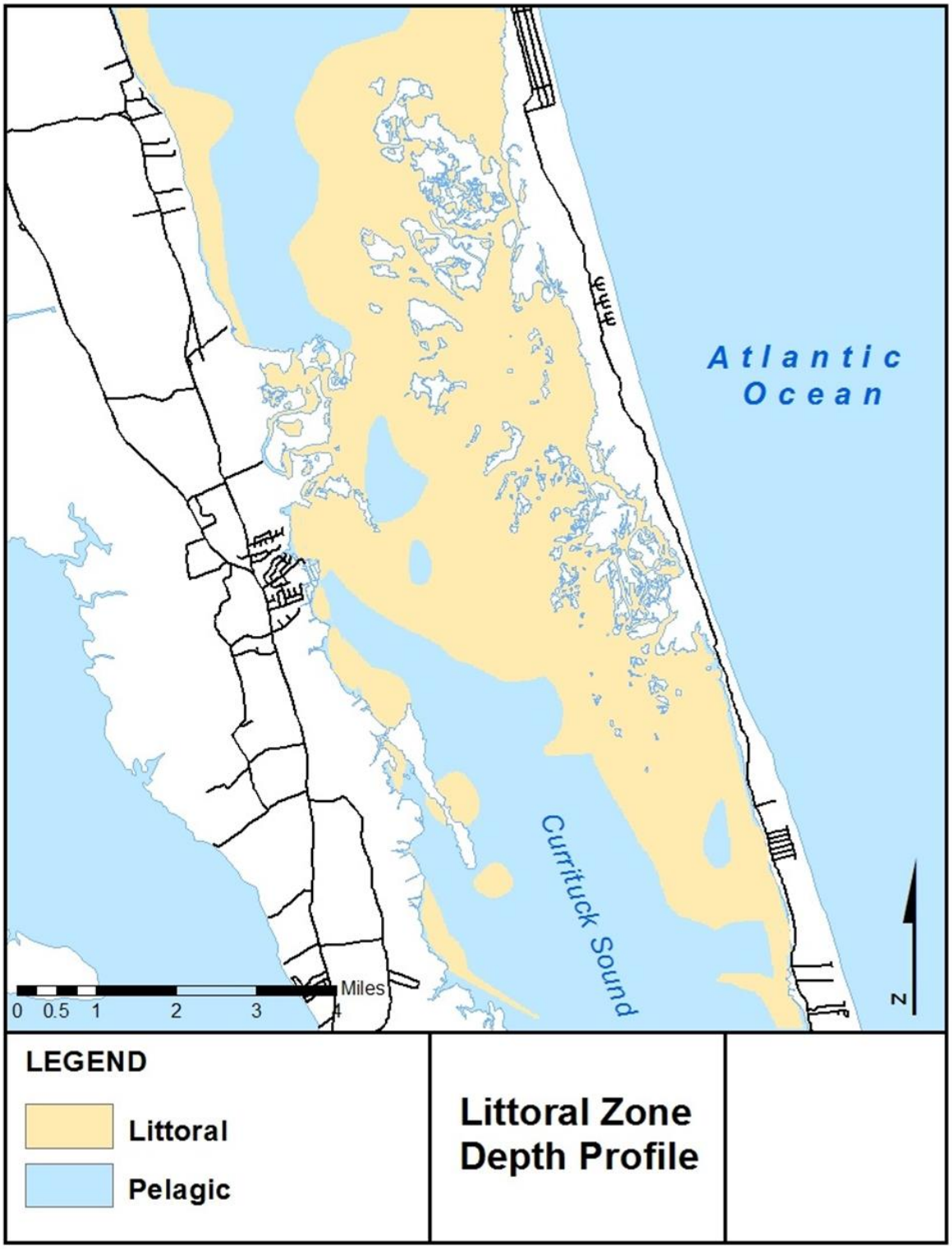

Figure A.4. Littoral zone defined for remote sensing purposes

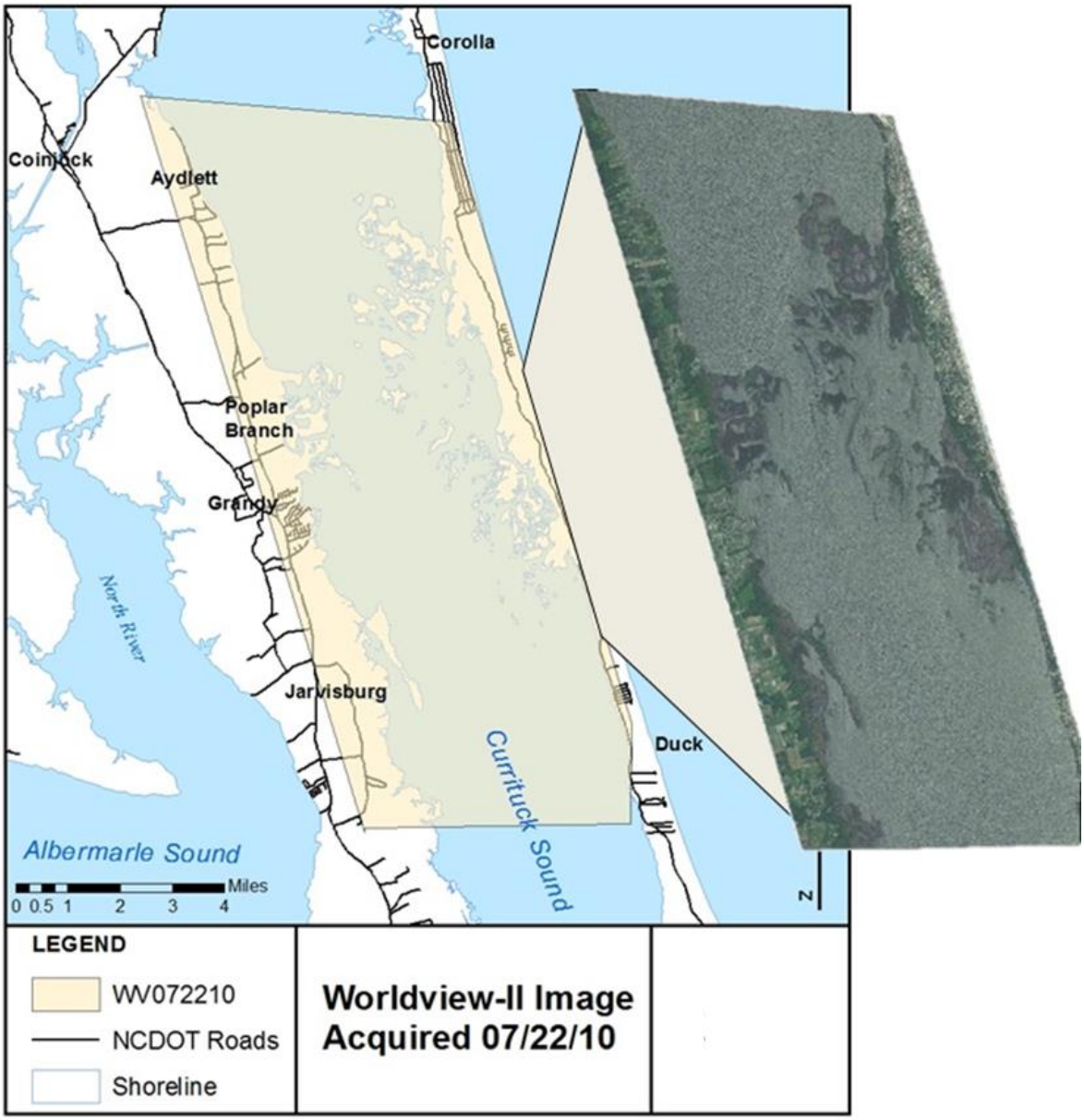

Figure A.5. Worlview-2 image acquired on 07/22/10

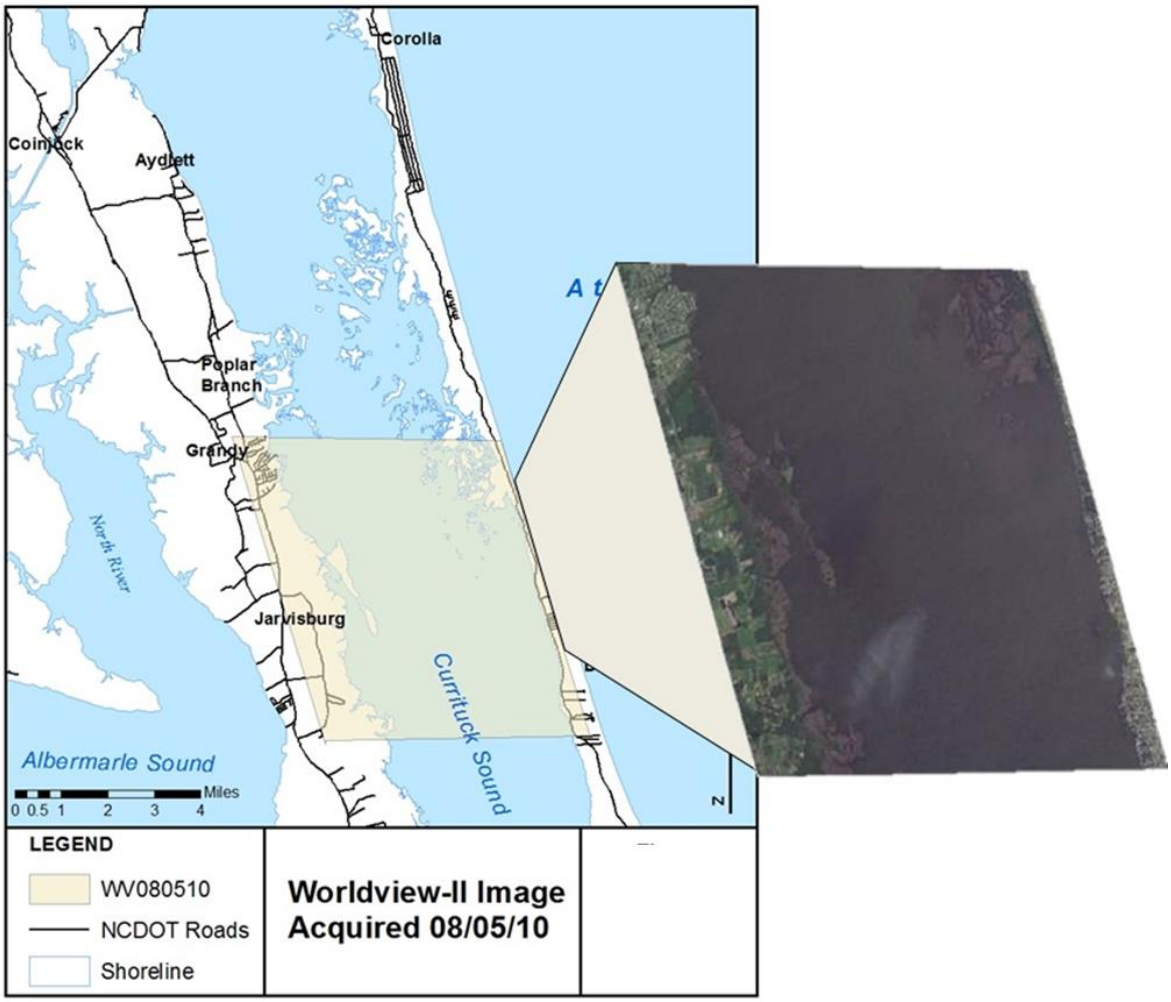

Figure A.6. Worldview-2 Image acquired on 08/05/10

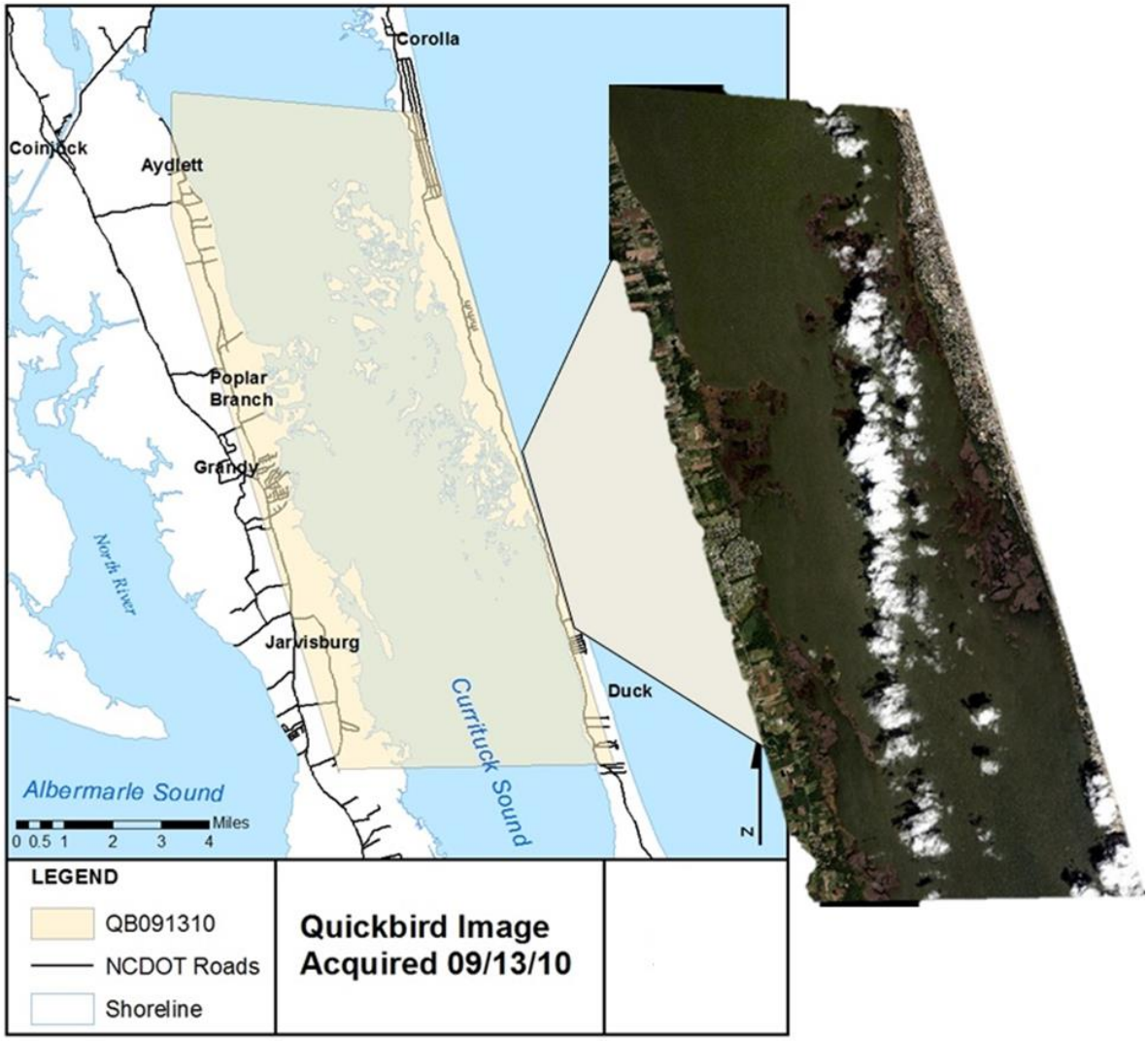

Figure A.7. Quickbird image acquired on 09/13/10

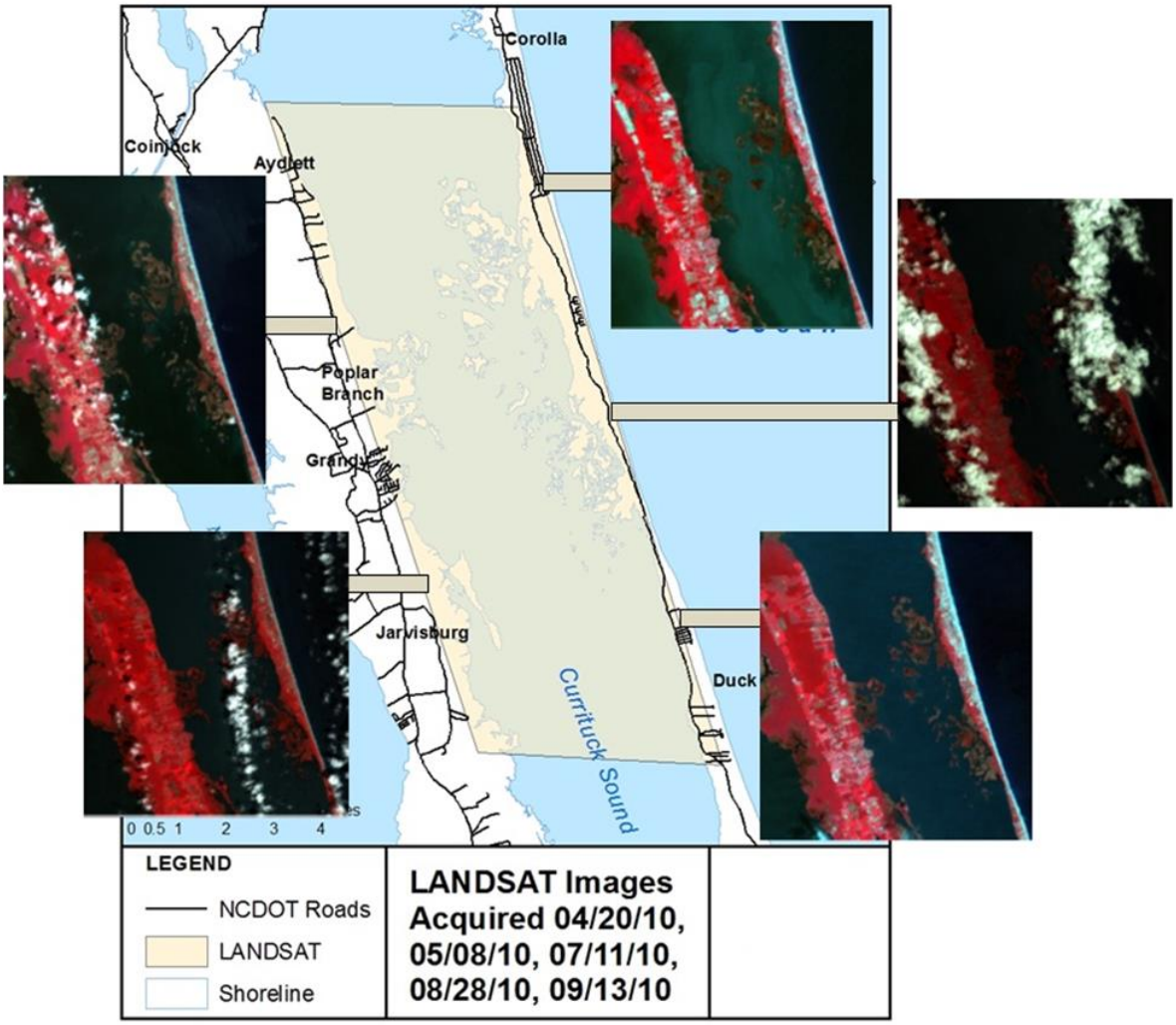

Figure A.8. LANDSAT5 images acquired on multiple dates

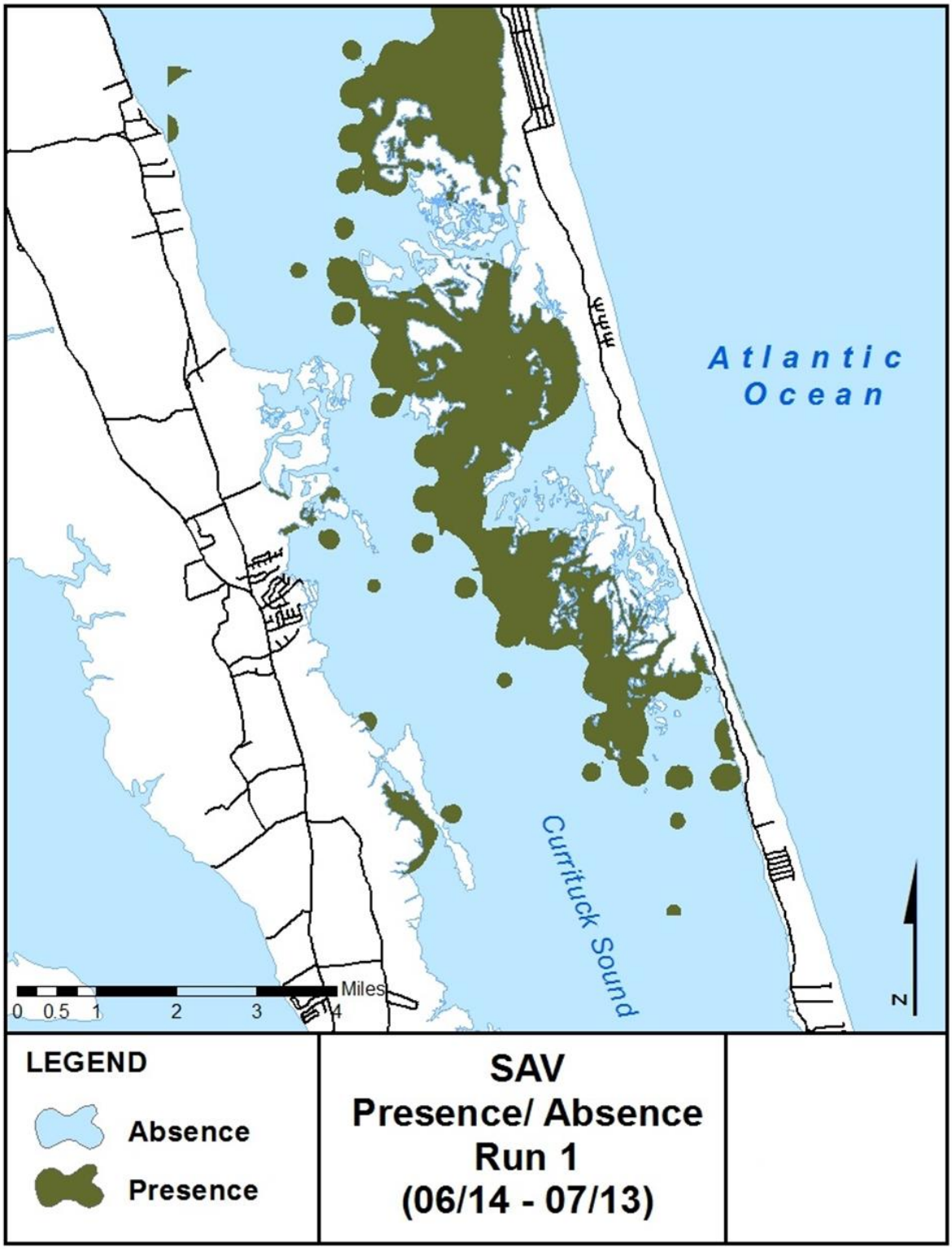

Figure A.9. SAV presence/ absence run 1

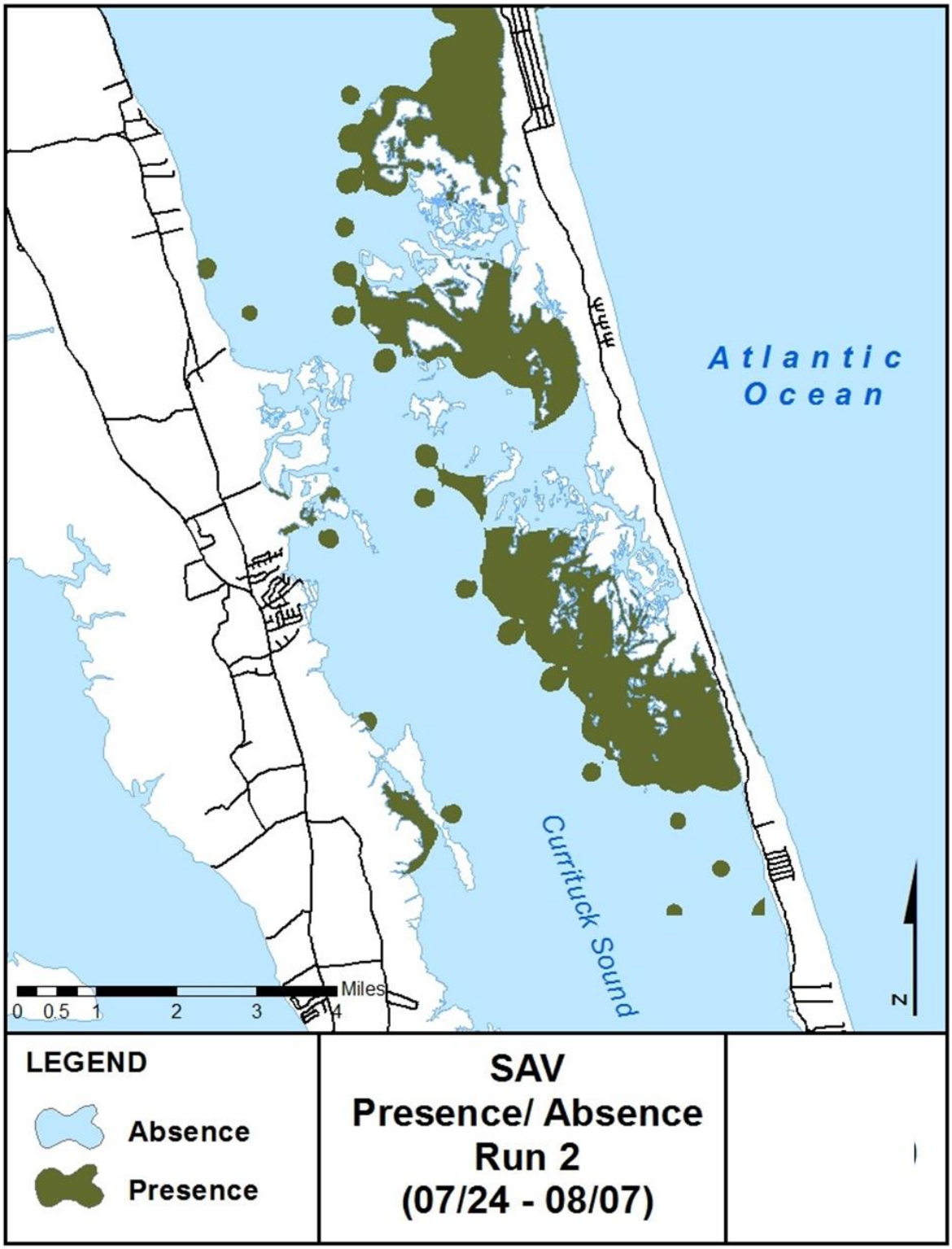

Figure A.10. SAV presence/ absence run 2

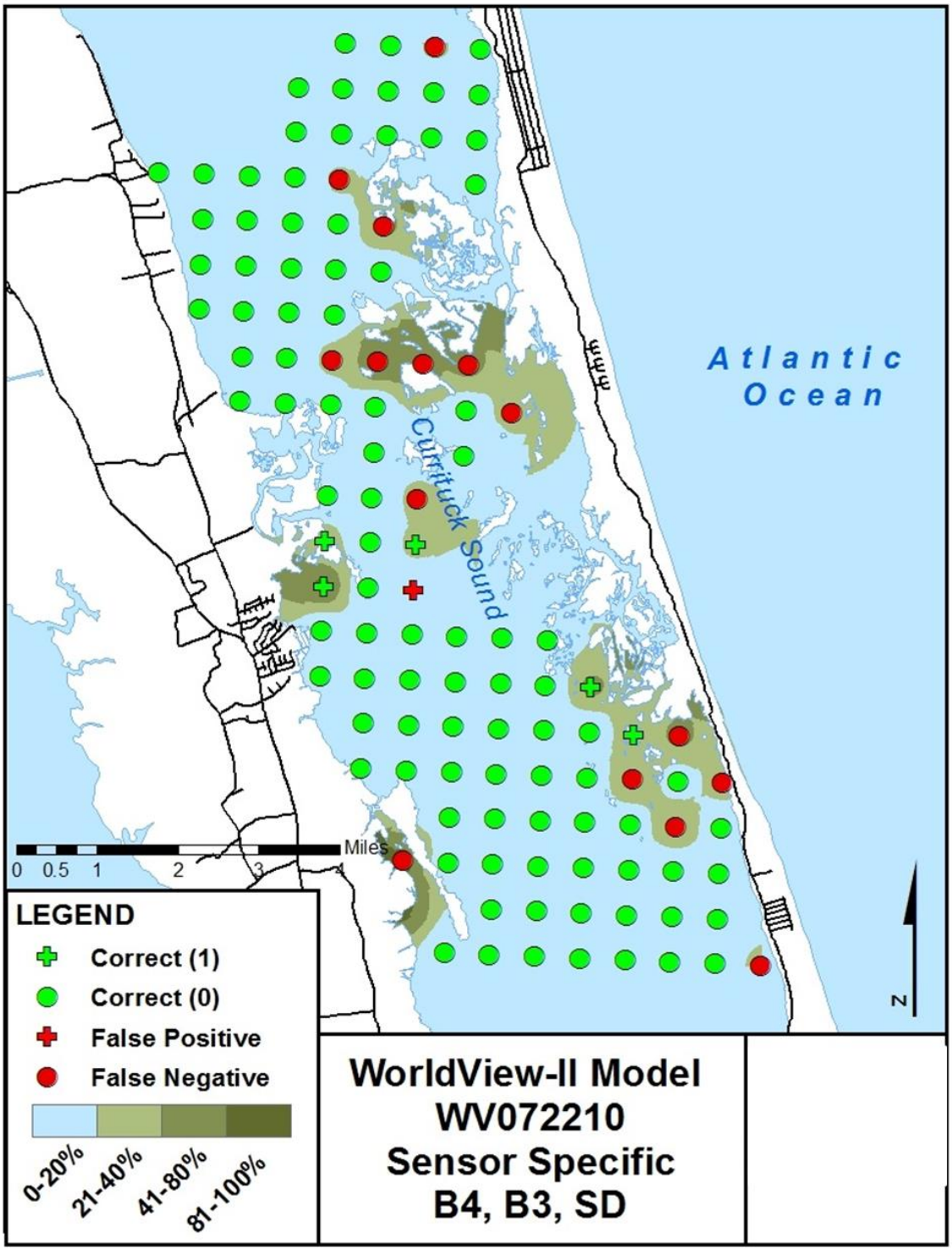

Figure A.11. Worldview-2 sensor specific model overlain with SAV percent cover

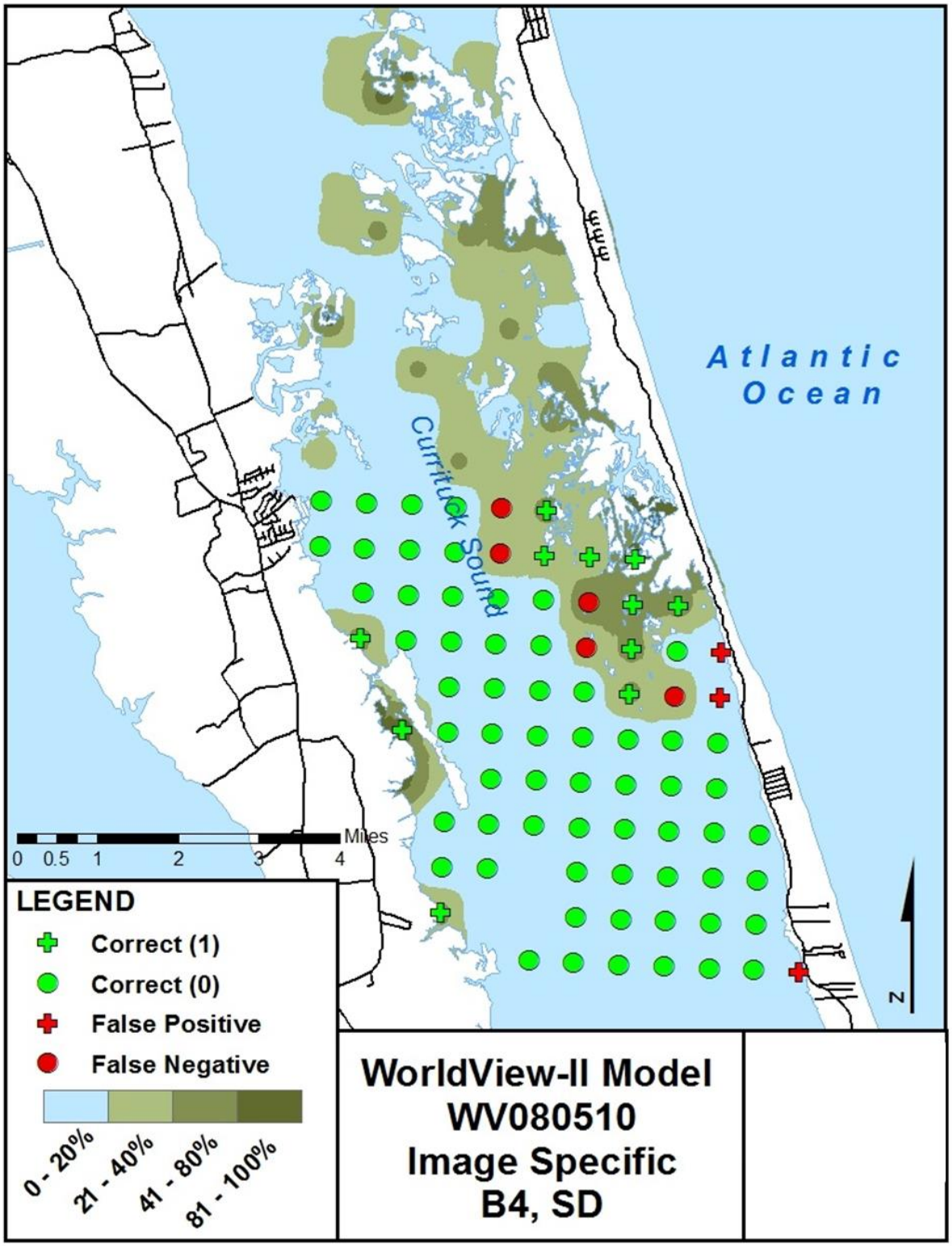

Figure A.12. Worldview-2 image specific model overlain with SAV percent cover

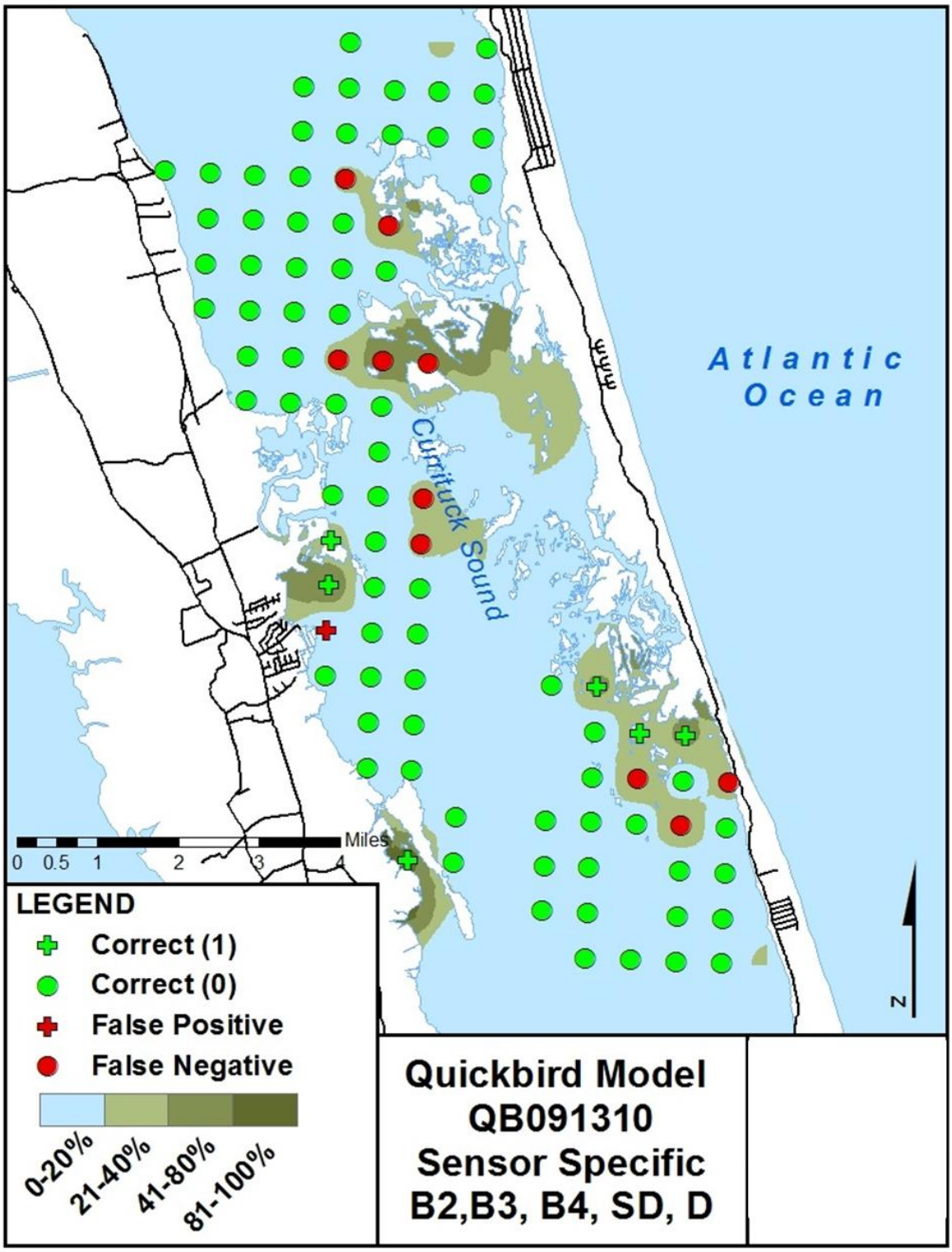

Figure A.13. Quickbird image specific model overlain with SAV percent cover

Figure A.14. Depth gradient along defined littoral zone**.**

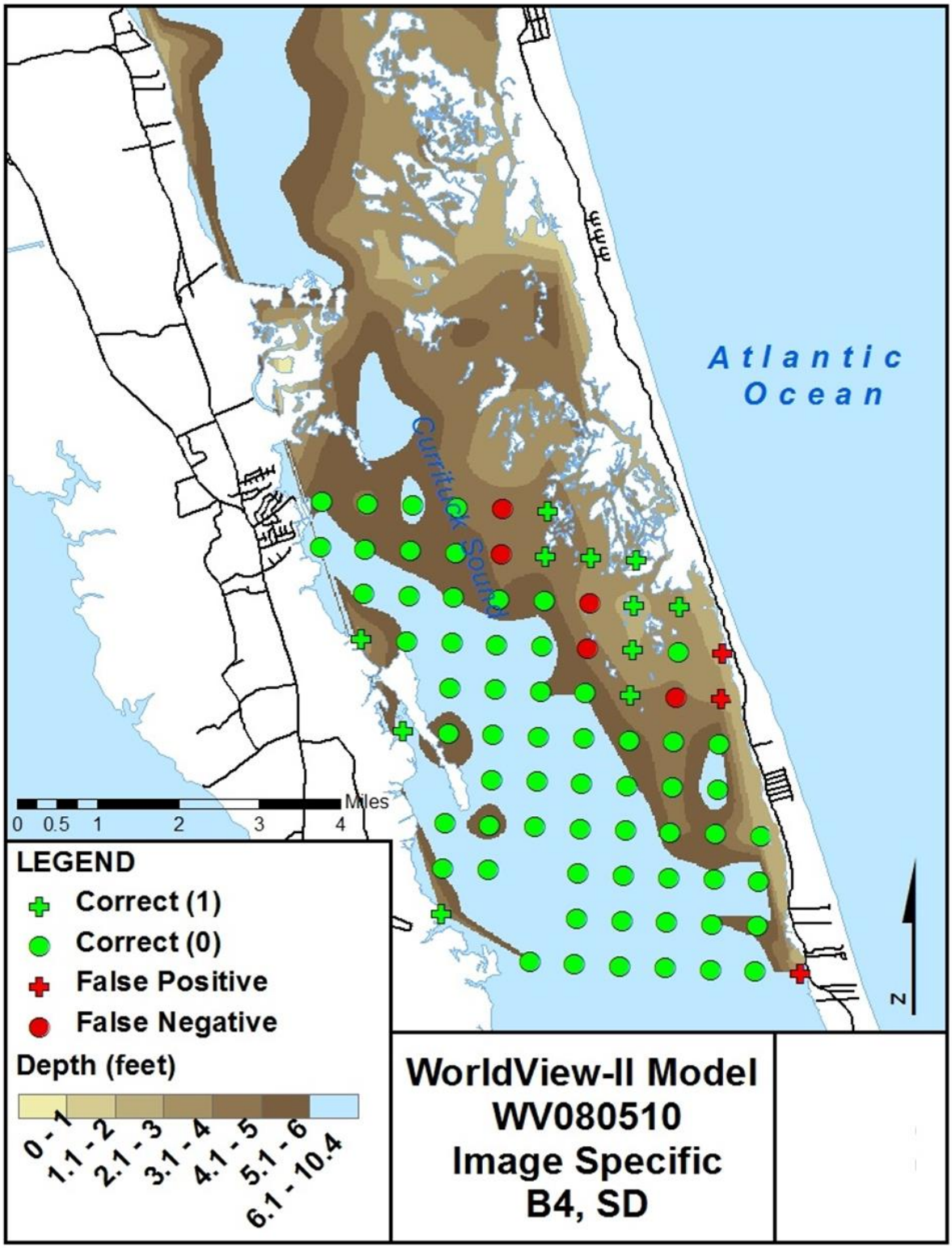

Figure A.15. Worldview-2 image specific model output overlain with depth profile**.**

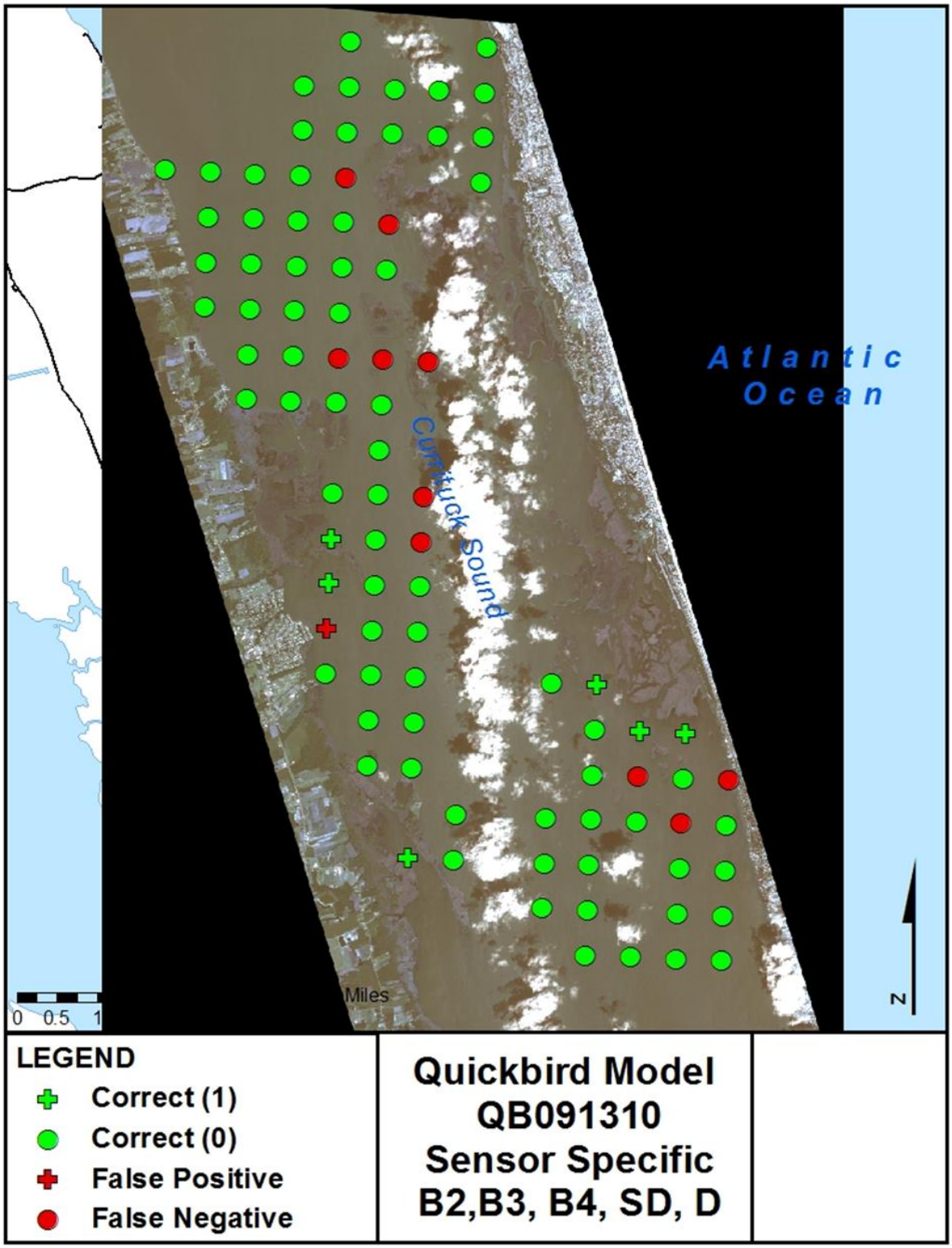

Figure A.16. Quickbird derived model overlain with original image.

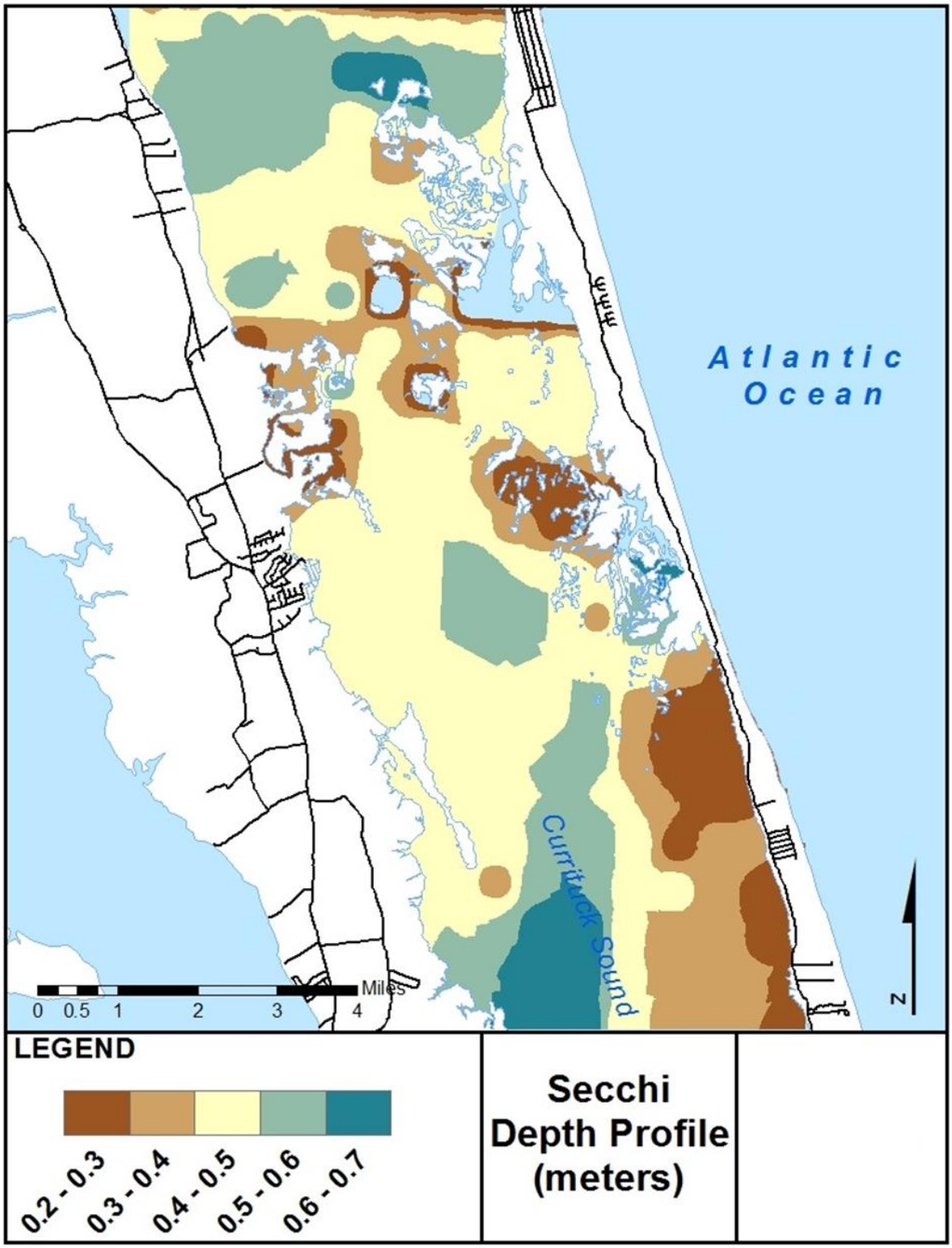

Figure A.17. Secchi depth averaging across entire study area

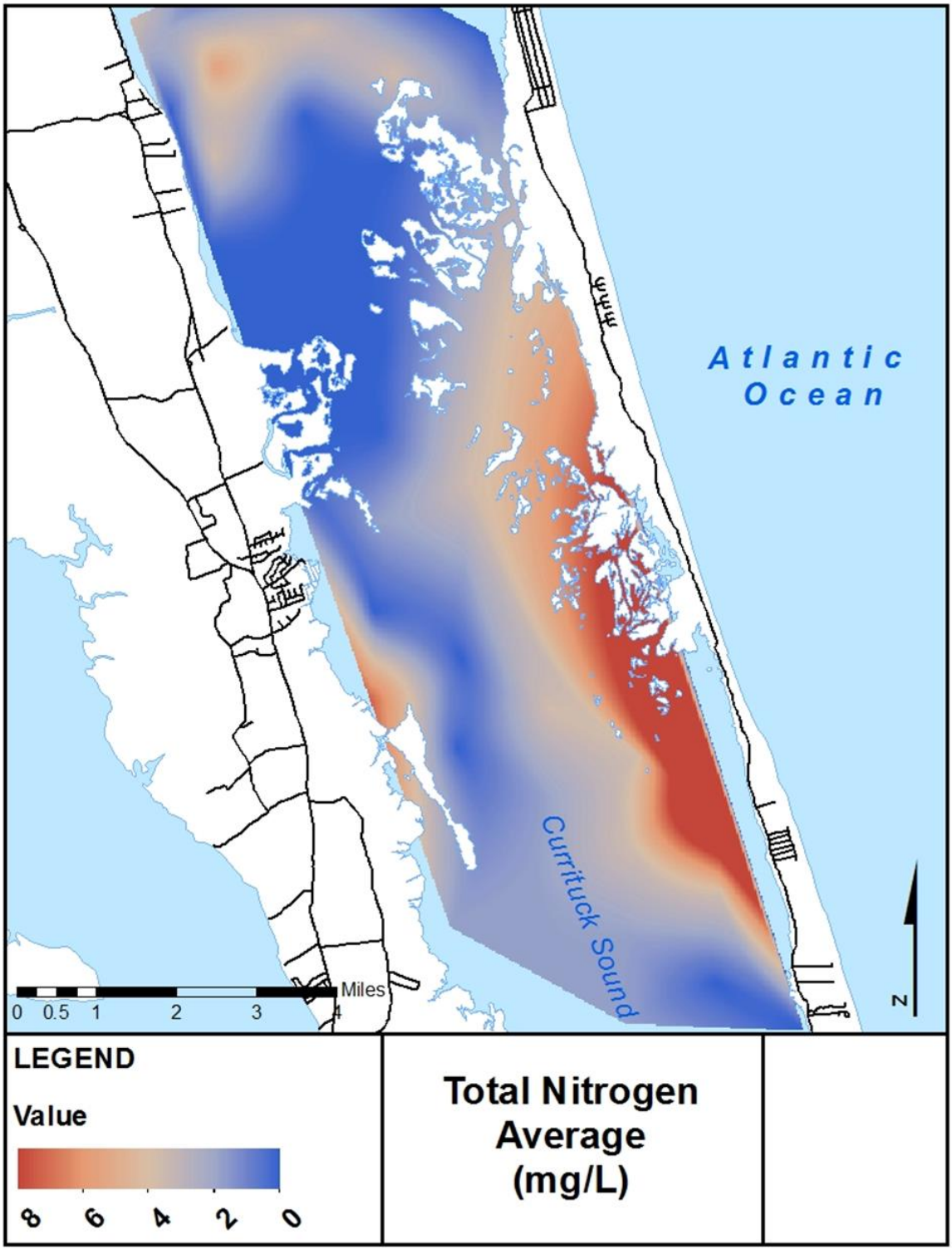

Figure A.18. Total nitrogen profile across study area.

Appendix B – Chapter 3 Supplement

Table B.1. Correlation matrix of SAV across species.

Species	Rumar	NaGuad	MySpic	StPect	PoPerf	VaAmer
Rumar	1	**0.533** ***(0.0001)***	**0.423** ***(0.0001)***	**0.416** ***(0.0001)***	**0.197** ***(0.0324)***	0.113 *(0.2236)*
Naguad	**0.533** ***(0.0001)***	1	0.487 ***(0.0001)***	**0.340** ***(0.0002)***	**0.185** ***(0.0453)***	-0.047 *(0.6136)*
MySpic	**0.423** ***(0.0001)***	**0.487** ***(0.0001)***	1	**0.311** ***(0.0006)***	0.080 *(0.3871)*	-0.039 *(0.6717)*
StPect	**0.416** ***(0.0001)***	**0.340** ***(0.0002)***	**0.311** ***(0.0006)***	1	0.018 *(0.8432)*	0.148 *(0.111)*
PoPerf	0.197 *(0.0324)*	0.185 *(0.0453)*	0.080 *(0.3871)*	0.018 *(0.8432)*	1	-0.015 *(0.872)*
VaAmer	0.113 *(0.2236)*	-0.047 *(0.6136)*	-0.039 *(0.6717)*	0.148 *(0.111)*	-0.015 *(0.872)*	1

*Bold denotes significant difference $p<0.05$.

Table B.2. Sediment type distribution of the littoral zone and number of vegetated points in each.

Sediment Type	Frequency	Percent of total	Vegetated	Percent Vegetated
clay	1	1.09%	1	100%
clay loam	2	2.17%	1	50.00%
loam	13	14.13%	7	53.85%
loamy sand	19	20.65%	11	57.89%
sand	25	27.17%	2	8.00%
sandy clay loam	2	2.17%	0	0.00%
sandy loam	17	18.48%	13	76.47%
silt	3	3.26%	2	66.67%
silt clay	1	1.09%	0	0.00%
silt loam	9	9.78%	5	55.56%

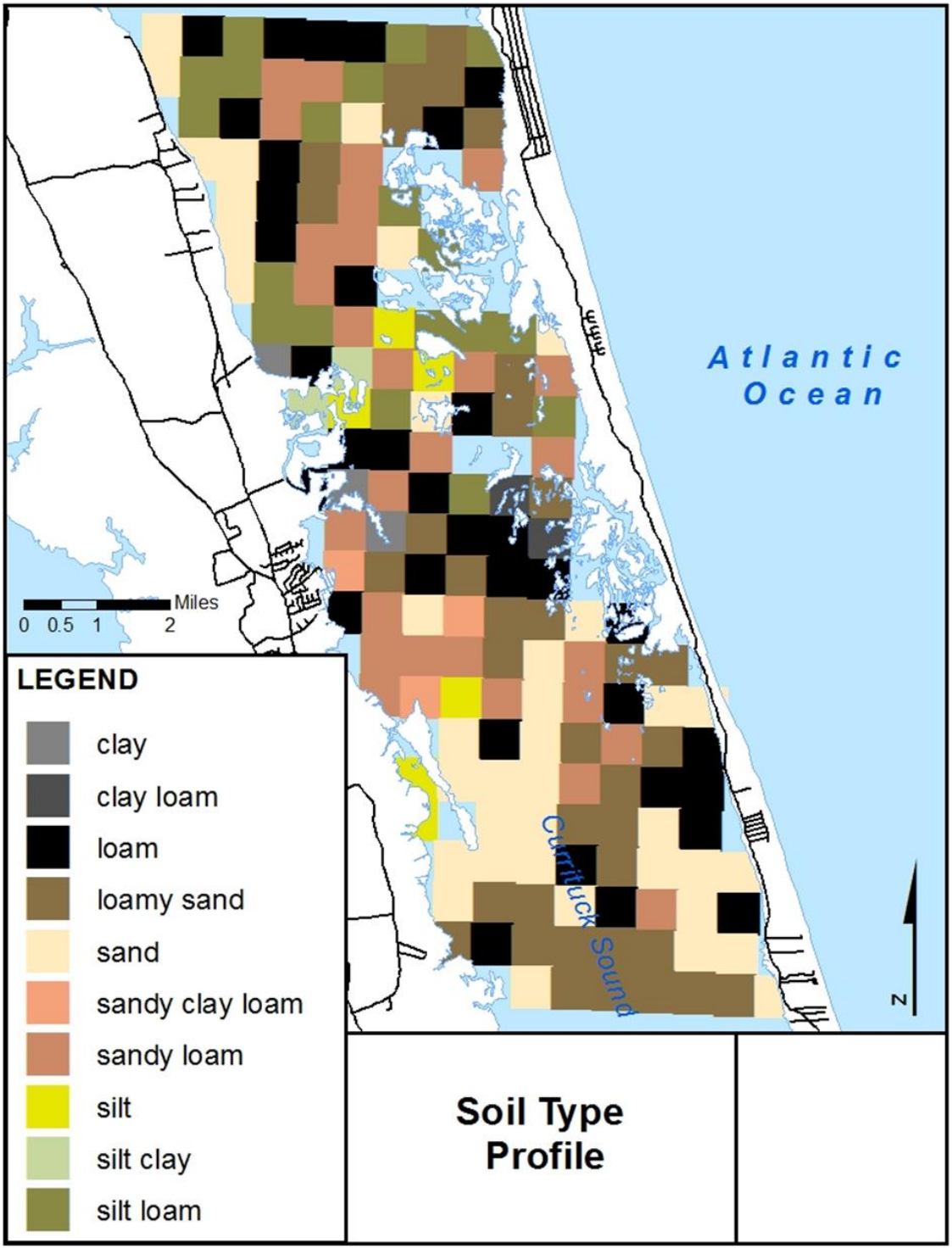

Figure B.1. Distribution of soil type throughout the study area as estimated during SAV sampling

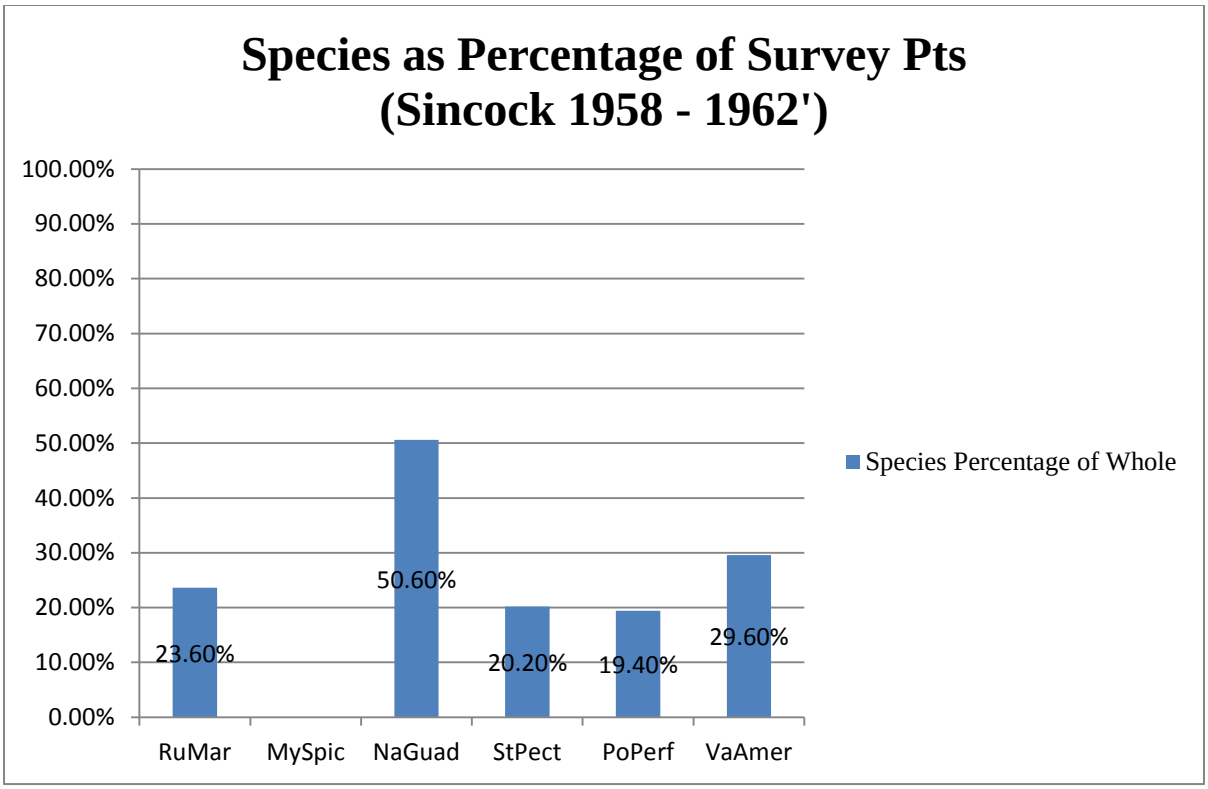

Figure B.2. Species as a percentage of all vegetated points as estimated using Sincock et al. 1965.

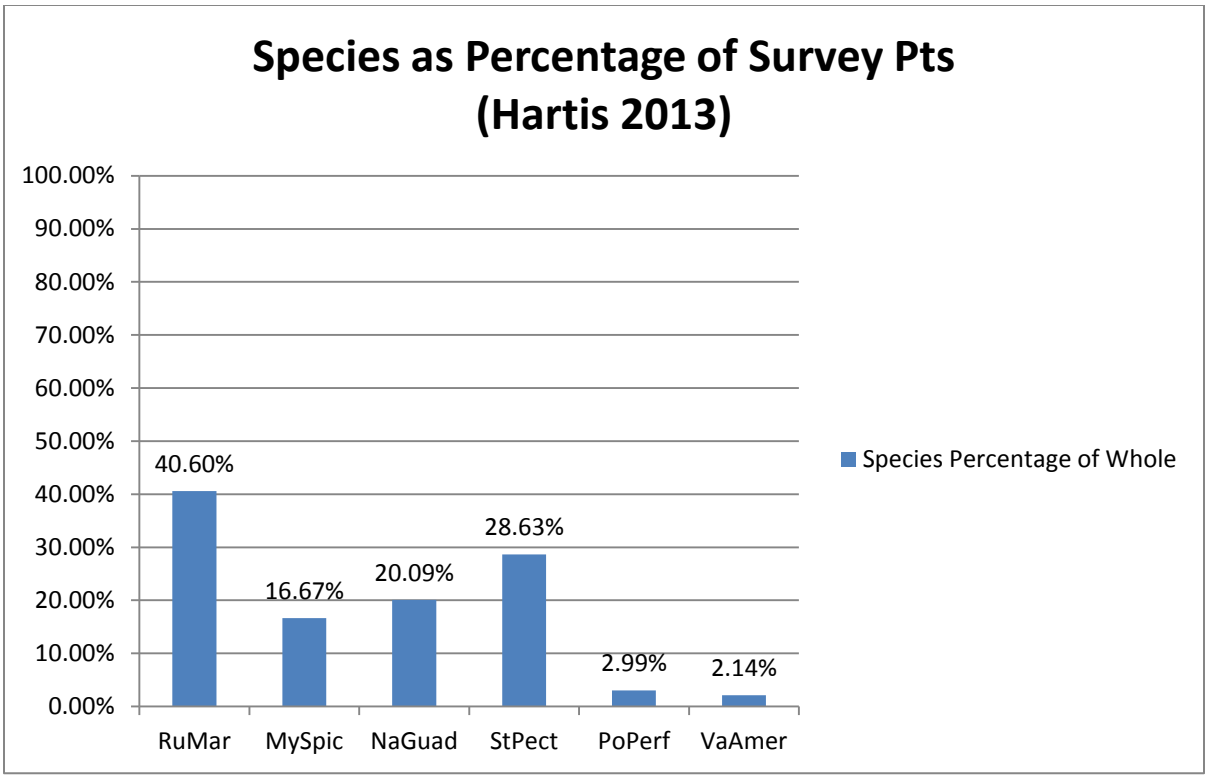

Figure B.3. Species as a percentage of all vegetated points as estimated in this study.

Appendix C – Chapter 4 Supplement

Table C.1. Risk for hydrilla establishment by category and occurrence

Risk Category	Value Range	Increase in Occurrence	Percent Increase
1	-8.5 to -6.8	n/a	n/a
2	-6.7 to -5.1	1.44	144
3	-5.0 to -3.4	1.47	147
4	-3.3 to -1.7	1.68	168
5	-1.6 to 0	1.43	143
6	0 to 1.6	1.16	116
7	1.7 -to 3.3	1.18	118
8	3.4 to 5.0	1.27	127
9	5.1 to 6.7	1.26	126
10	>6.7	1.2	120
	Average	1.34	134.33

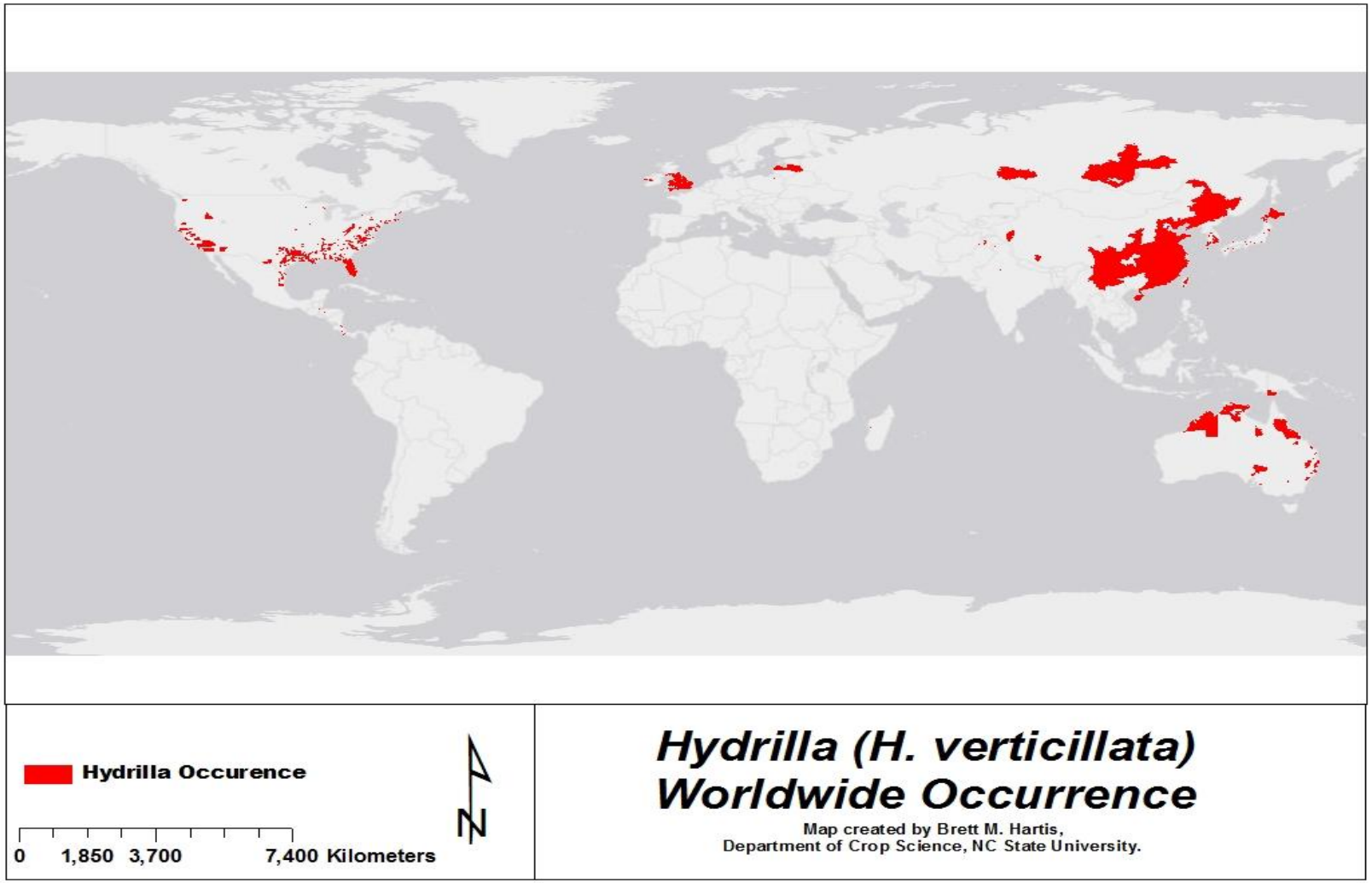

Figure C.1. Hydrilla occurrence worldwide as designated by master dataset

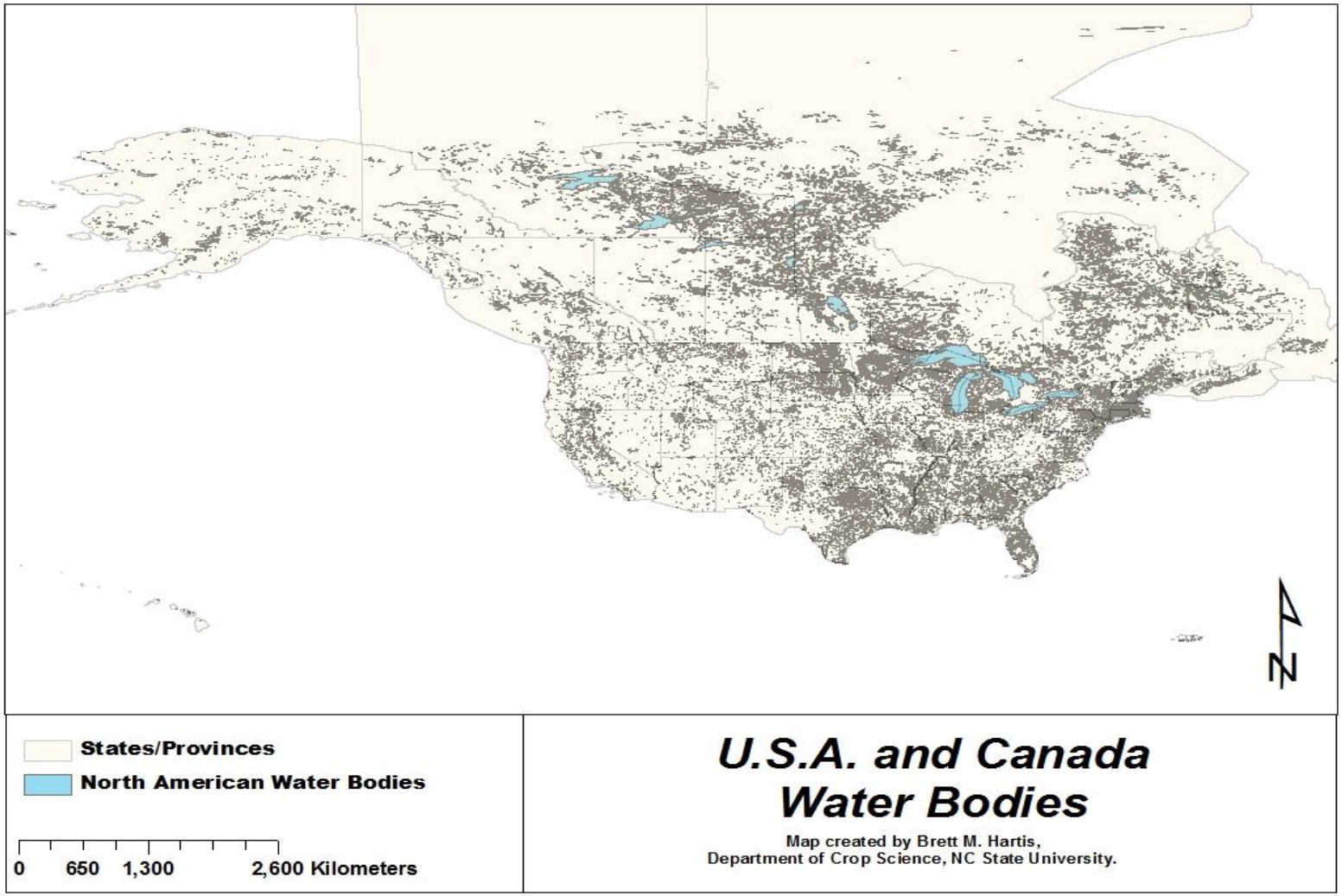

Figure C.2. United States and Canada water bodies

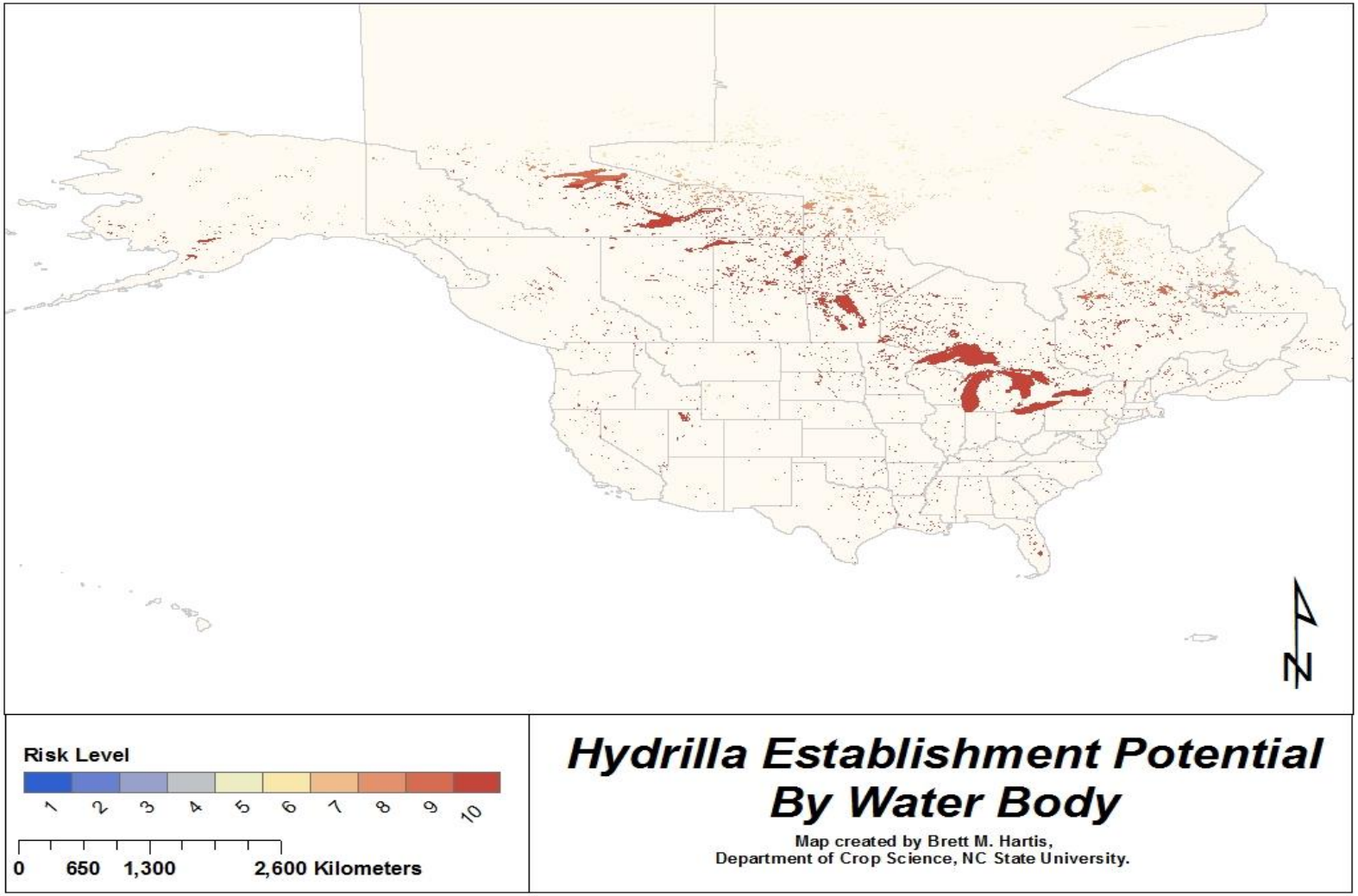

Figure C.3. Hydrilla establishment potential by water body

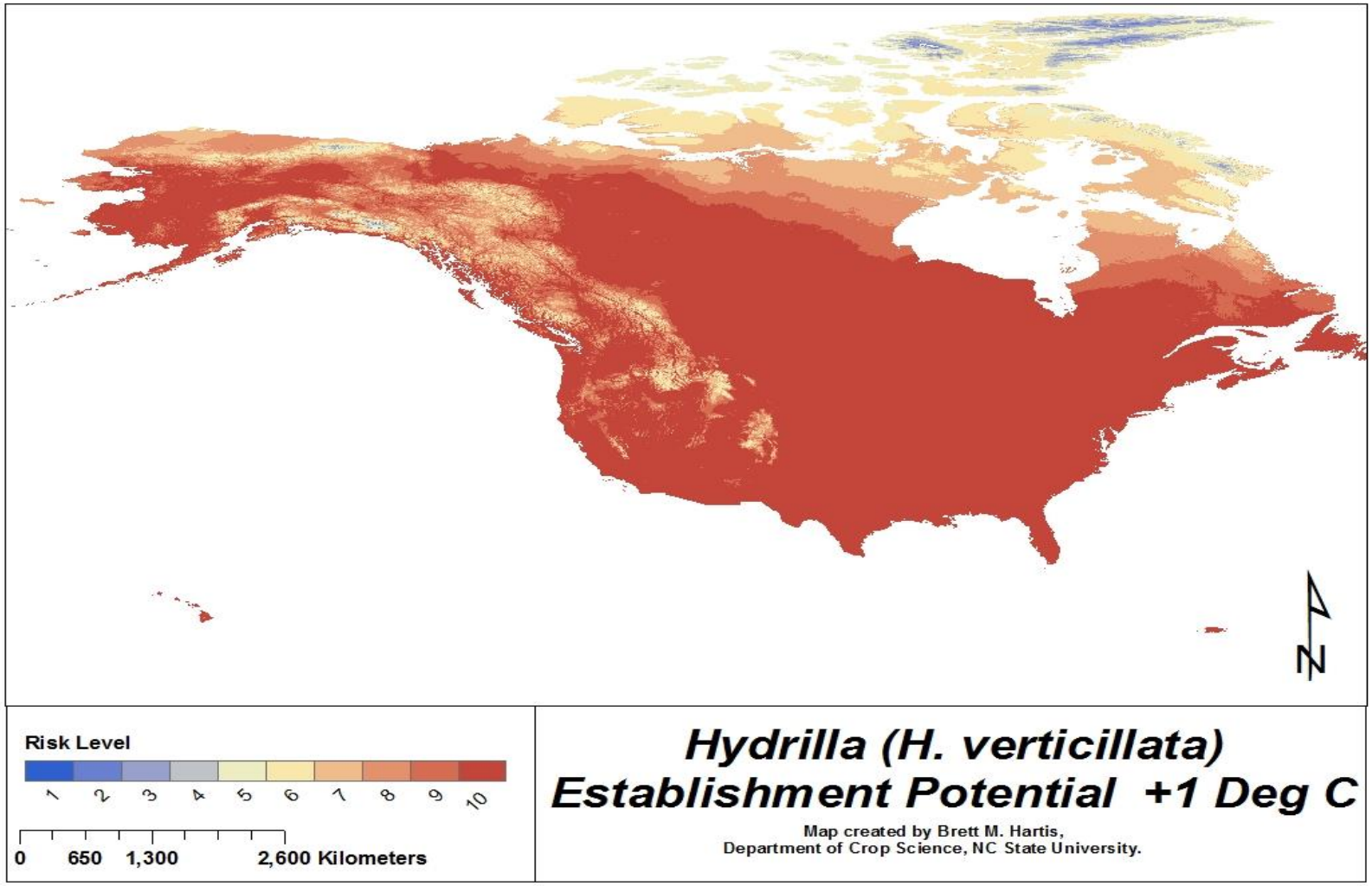

Figure C.4. Projected establishment potential model +1 degree C

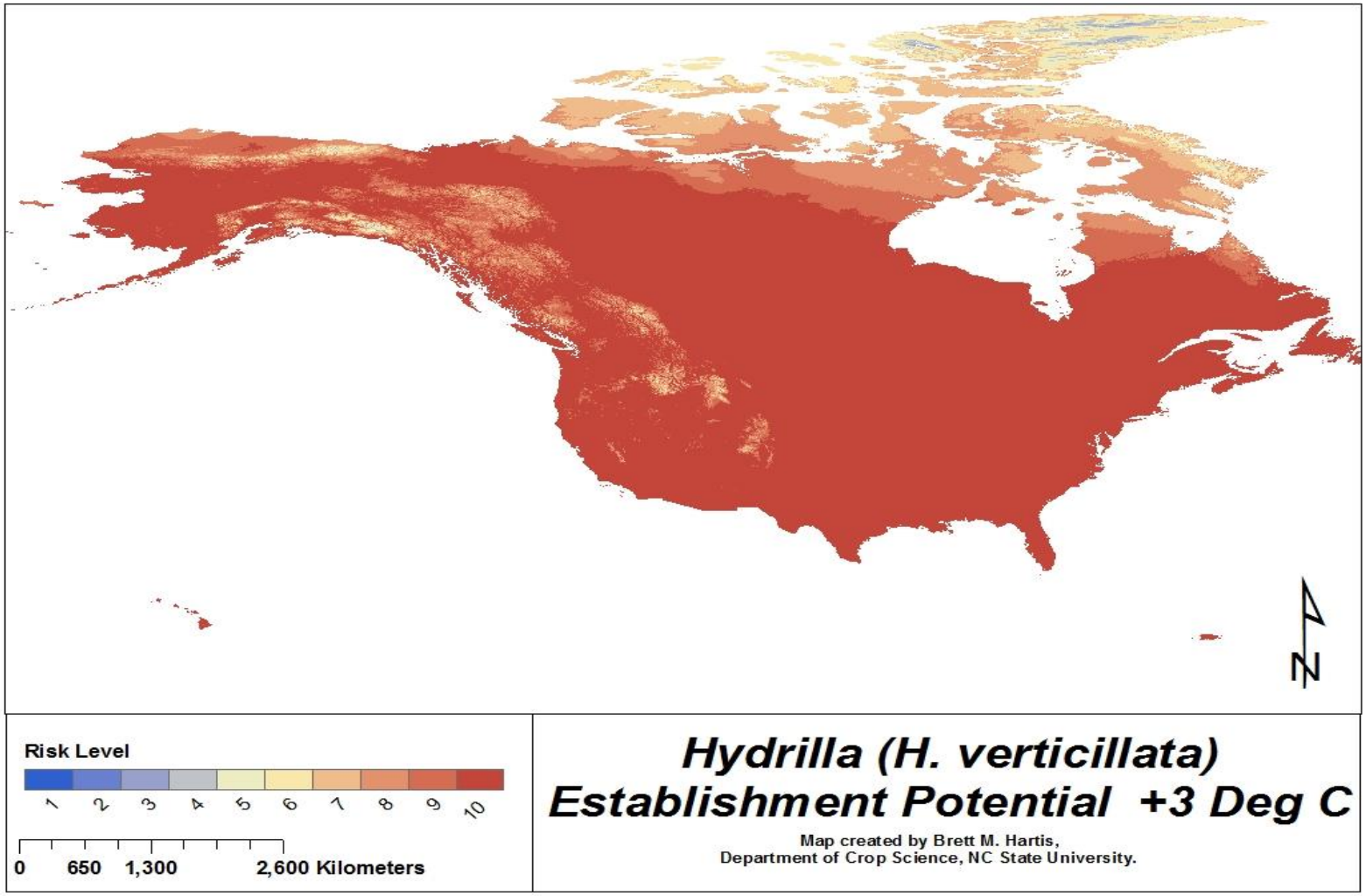

Figure C.5. Projected establishment potential model +3 degree C

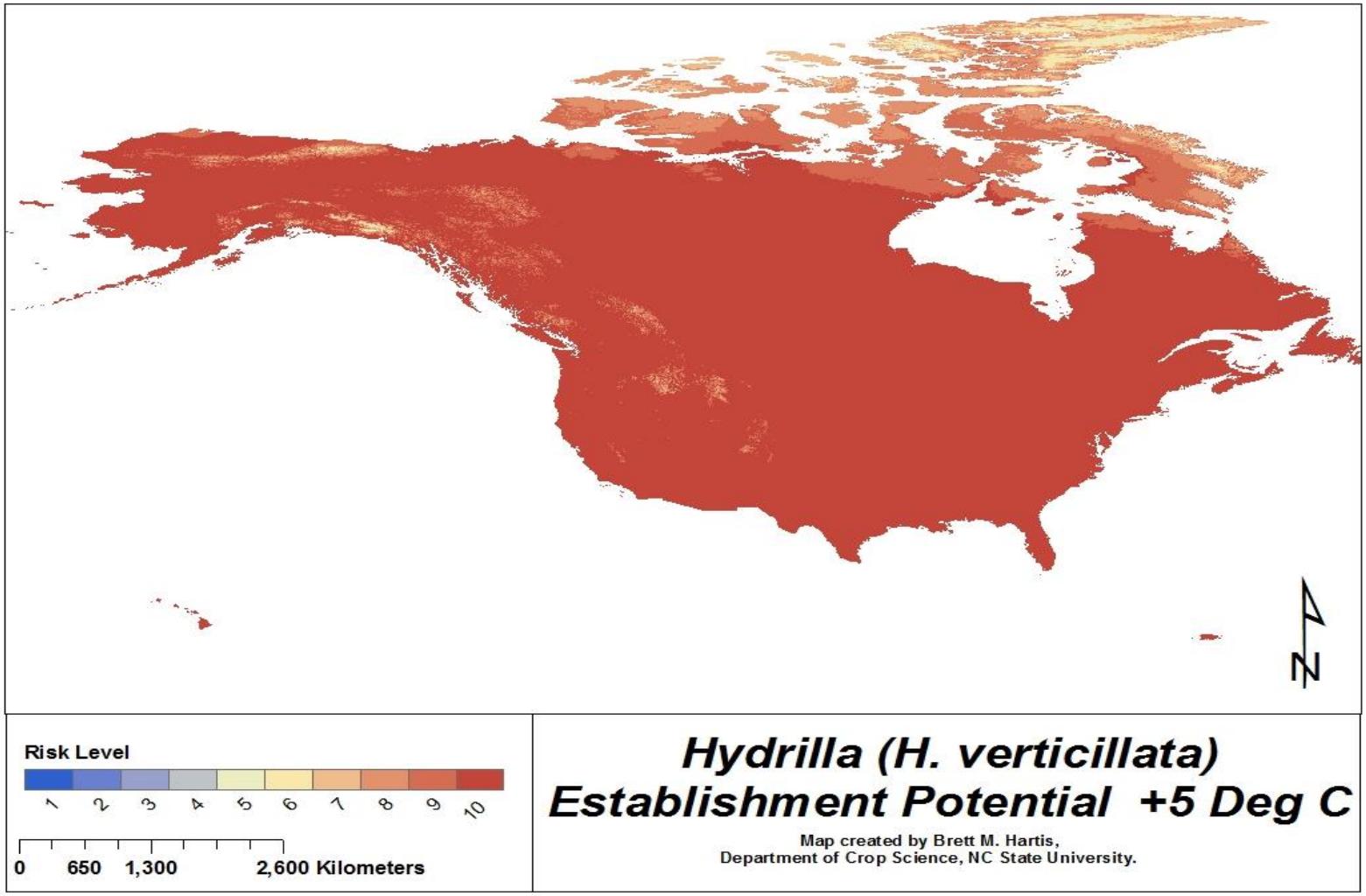

Figure C.6. Projected establishment potential model +5 degree C

Appendix D: SAS Code for Chapter 3

D.1. Summary Statistics of Size Type, Acres and Risk Potential

```
/* ---------------------------------------------------------------------
   Code generated by SAS Task

   Generated on: Thursday, August 08, 2013 at 9:14:09 AM
   By task: Summary Statistics

   Input Data: SASUSER.FINALDATABYLAKES
   Server:  Local
   --------------------------------------------------------------------- */

%_eg_conditional_dropds(WORK.SORTTempTableSorted,
            SASUSER.MEANSUMMARYSTATS_0005);
/* ---------------------------------------------------------------------
   Sort data set SASUSER.FINALDATABYLAKES
   --------------------------------------------------------------------- */
PROC SORT
      DATA=SASUSER.FINALDATABYLAKES(KEEP=size_type acres risk_poten
name_1)
      OUT=WORK.SORTTempTableSorted
      ;
      BY name_1;
RUN;
/* ---------------------------------------------------------------------
   Run the Means Procedure
   --------------------------------------------------------------------- */
TITLE;
TITLE1 "Summary Statistics";
TITLE2 "Results";
FOOTNOTE;
FOOTNOTE1 "Generated by the SAS System (&_SASSERVERNAME, &SYSSCPL) on
%TRIM(%QSYSFUNC(DATE(), NLDATE20.)) at %TRIM(%SYSFUNC(TIME(),
NLTIMAP20.))";
PROC MEANS DATA=WORK.SORTTempTableSorted
      FW=12
      PRINTALLTYPES
      CHARTYPE
      VARDEF=DF
            MEAN
            STD
            MIN
            MAX
            RANGE
            SUM
            N     ;
      VAR size_type acres risk_poten;
      BY name_1;

OUTPUT      OUT=SASUSER.MEANSUMMARYSTATS_0005(LABEL="Summary Statistics
for SASUSER.FINALDATABYLAKES")
```

```
            MEAN()=
            STD()=
            MIN()=
            MAX()=
            RANGE()=
            SUM()=
            N()=

      / AUTONAME AUTOLABEL INHERIT
      ;
RUN;
ODS GRAPHICS ON;
TITLE;
TITLE1 "Summary Statistics";
TITLE2 "Box and Whisker Plots";
PROC SGPLOT DATA=WORK.SORTTempTableSorted ;
BY name_1 ;
      VBOX size_type;
RUN;QUIT;
PROC SGPLOT DATA=WORK.SORTTempTableSorted ;
BY name_1 ;
      VBOX acres;
RUN;QUIT;
PROC SGPLOT DATA=WORK.SORTTempTableSorted ;
BY name_1 ;
      VBOX risk_poten;
RUN;QUIT;
ODS GRAPHICS OFF;
/* ---------------------------------------------------------------------
   End of task code.
   --------------------------------------------------------------------- */
RUN; QUIT;
%_eg_conditional_dropds(WORK.SORTTempTableSorted);
TITLE; FOOTNOTE;
```

D.2. Summary Statistics of Risk Frequency

```
/* ---------------------------------------------------------------------
   Code generated by SAS Task

   Generated on: Tuesday, October 08, 2013 at 9:24:05 AM
   By task: Summary Statistics5

   Input Data: SASUSER.HYDRILLA_MODELS4
   Server:  Local
   --------------------------------------------------------------------- */

%_eg_conditional_dropds(WORK.SORTTempTableSorted,
            SASUSER.MEANSUMMARYSTATS_000B);
/* ---------------------------------------------------------------------
   Sort data set SASUSER.HYDRILLA_MODELS4
   --------------------------------------------------------------------- */

PROC SQL;
      CREATE VIEW WORK.SORTTempTableSorted AS
            SELECT T.Frequency, T.Risk
      FROM SASUSER.HYDRILLA_MODELS4 as T
;
QUIT;
/* ---------------------------------------------------------------------
   Run the Means Procedure
   --------------------------------------------------------------------- */
TITLE;
TITLE1 "Summary Statistics";
TITLE2 "Results";
FOOTNOTE;
FOOTNOTE1 "Generated by the SAS System (&_SASSERVERNAME, &SYSSCPL) on
%TRIM(%QSYSFUNC(DATE(), NLDATE20.)) at %TRIM(%SYSFUNC(TIME(),
NLTIMAP20.))";
PROC MEANS DATA=WORK.SORTTempTableSorted
      FW=12
      PRINTALLTYPES
      CHARTYPE
      NWAY
      VARDEF=DF
            MEAN
            STD
            MIN
            MAX
            MODE
            RANGE
            N     ;
      VAR Frequency;
      CLASS Risk /      ORDER=UNFORMATTED ASCENDING;

OUTPUT      OUT=SASUSER.MEANSUMMARYSTATS_000B(LABEL="Summary Statistics
for SASUSER.HYDRILLA_MODELS4")
```

```
            MEAN()=
            STD()=
            MIN()=
            MAX()=
            MODE()=
            RANGE()=
            N()=

      / AUTONAME AUTOLABEL  WAYS INHERIT
      ;
RUN;
/* ---------------------------------------------------------------------
   End of task code.
   --------------------------------------------------------------------- */
RUN; QUIT;
%_eg_conditional_dropds(WORK.SORTTempTableSorted);
TITLE; FOOTNOTE;
```

D.3. Distribution Analysis of Acreage

```
/* ---------------------------------------------------------------------
   Code generated by SAS Task

   Generated on: Tuesday, September 24, 2013 at 11:46:17 AM
   By task: Distribution Analysis

   Input Data: SASUSER.FINALDATABYLAKES
   Server:  Local
   --------------------------------------------------------------------- */

%_eg_conditional_dropds(WORK.SORTTempTableSorted);
/* ---------------------------------------------------------------------
   PROC SHEWHART does not support DEVICE=ACTIVEX. Switching to PNG.
   --------------------------------------------------------------------- */
OPTIONS DEV=PNG;
/* ---------------------------------------------------------------------
   Sort data set SASUSER.FINALDATABYLAKES
   --------------------------------------------------------------------- */

PROC SQL;
      CREATE VIEW WORK.SORTTempTableSorted AS
            SELECT T.acres
      FROM SASUSER.FINALDATABYLAKES as T
;
QUIT;
TITLE;
TITLE1 "Distribution analysis of: acres";
FOOTNOTE;
FOOTNOTE1 "Generated by the SAS System (&_SASSERVERNAME, &SYSSCPL) on
%TRIM(%QSYSFUNC(DATE(), NLDATE20.)) at %TRIM(%SYSFUNC(TIME(),
NLTIMAP20.))";
      ODS EXCLUDE EXTREMEOBS MODES MOMENTS QUANTILES;

      GOPTIONS htext=1 cells;
      SYMBOL v=SQUARE c=BLUE h=1 cells;
      PATTERN v=SOLID
      ;
PROC UNIVARIATE DATA = WORK.SORTTempTableSorted
            CIBASIC(TYPE=TWOSIDED ALPHA=0.05)
            MU0=0
;
      VAR acres;
      HISTOGRAM   acres / NORMAL    (     W=1   L=1   COLOR=YELLOW  MU=EST
SIGMA=EST)

            CFRAME=GRAY CAXES=BLACK WAXIS=1  CBARLINE=BLACK CFILL=BLUE
PFILL=SOLID ;
      ;
            QQPLOT   acres / NORMAL(            W=1   L=1   COLOR=YELLOW
MU=EST  SIGMA=EST         )
                  CFRAME=GRAY CAXES=BLACK WAXIS=1;
```

```
/* ---------------------------------------------------------------------
   End of task code.
   --------------------------------------------------------------------- */
RUN; QUIT;
%_eg_conditional_dropds(WORK.SORTTempTableSorted);
TITLE; FOOTNOTE;
/* ---------------------------------------------------------------------
   Restoring original device type setting.
   --------------------------------------------------------------------- */
OPTIONS DEV=ACTIVEX;
```

D.4. Linear Regression of Hydrilla Establishment Risk and Scatter Plot

```
/* ---------------------------------------------------------------------
   Code generated by SAS Task

   Generated on: Friday, October 04, 2013 at 1:02:48 PM
   By task: Linear Regression

   Input Data: SASUSER.HYDRILLA_MODELS2
   Server:  Local
   --------------------------------------------------------------------- */
ODS GRAPHICS ON;

%_eg_conditional_dropds(WORK.SORTTempTableSorted,
            WORK.TMP1TempTableForPlots);
/* ---------------------------------------------------------------------
   Determine the data set's type attribute (if one is defined)
   and prepare it for addition to the data set/view which is
   generated in the following step.
   --------------------------------------------------------------------- */
DATA _NULL_;
      dsid = OPEN("SASUSER.HYDRILLA_MODELS2", "I");
      dstype = ATTRC(DSID, "TYPE");
      IF TRIM(dstype) = " " THEN
            DO;
            CALL SYMPUT("_EG_DSTYPE_", "");
            CALL SYMPUT("_DSTYPE_VARS_", "");
            END;
      ELSE
            DO;
            CALL SYMPUT("_EG_DSTYPE_", "(TYPE=""" || TRIM(dstype) ||
""")");
            IF VARNUM(dsid, "_NAME_") NE 0 AND VARNUM(dsid, "_TYPE_") NE 0
THEN
                  CALL SYMPUT("_DSTYPE_VARS_", "_TYPE_ _NAME_");
            ELSE IF VARNUM(dsid, "_TYPE_") NE 0 THEN
                  CALL SYMPUT("_DSTYPE_VARS_", "_TYPE_");
            ELSE IF VARNUM(dsid, "_NAME_") NE 0 THEN
                  CALL SYMPUT("_DSTYPE_VARS_", "_NAME_");
            ELSE
                  CALL SYMPUT("_DSTYPE_VARS_", "");
            END;
      rc = CLOSE(dsid);
      STOP;
RUN;

/* ---------------------------------------------------------------------
   Data set SASUSER.HYDRILLA_MODELS2 does not need to be sorted.
   --------------------------------------------------------------------- */
DATA WORK.SORTTempTableSorted &_EG_DSTYPE_ /
VIEW=WORK.SORTTempTableSorted;
```

```
      SET SASUSER.HYDRILLA_MODELS2(KEEP=Frequency Value_0001
&_DSTYPE_VARS_);
RUN;
TITLE;
TITLE1 "Linear Regression Results";
FOOTNOTE;
FOOTNOTE1 "Generated by the SAS System (&_SASSERVERNAME, &SYSSCPL) on
%TRIM(%QSYSFUNC(DATE(), NLDATE20.)) at %TRIM(%SYSFUNC(TIME(),
NLTIMAP20.))";
PROC REG DATA=WORK.SORTTempTableSorted
            PLOTS(ONLY)=ALL
      ;
      Linear_Regression_Model: MODEL Frequency = Value_0001
            /           SELECTION=NONE
      ;
RUN;
QUIT;

/* ---------------------------------------------------------------------
   End of task code.
   --------------------------------------------------------------------- */
RUN; QUIT;
%_eg_conditional_dropds(WORK.SORTTempTableSorted,
            WORK.TMP1TempTableForPlots);
TITLE; FOOTNOTE;
ODS GRAPHICS OFF;
/* ---------------------------------------------------------------------
   Code generated by SAS Task

   Generated on: Friday, October 04, 2013 at 1:13:06 PM
   By task: Scatter Plot

   Input Data: SASUSER.HYDRILLA_MODELS2
   Server:  Local
   --------------------------------------------------------------------- */

%_eg_conditional_dropds(WORK.SORTTempTableSorted);
/* ---------------------------------------------------------------------
   Sort data set SASUSER.HYDRILLA_MODELS2
   --------------------------------------------------------------------- */

PROC SQL;
      CREATE VIEW WORK.SORTTempTableSorted AS
            SELECT T.Value_0001, T.Frequency
      FROM SASUSER.HYDRILLA_MODELS2 as T
;
QUIT;
      SYMBOL1
      INTERPOL=NONE
      HEIGHT=10pt
      VALUE=CIRCLE
      LINE=1
      WIDTH=2
```

```
CV = _STYLE_
;
Axis1
      STYLE=1
      WIDTH=1
      MINOR=NONE


;
Axis2
      STYLE=1
      WIDTH=1
      MINOR=NONE


;
TITLE;
TITLE1 "Scatter Plot";
FOOTNOTE;
FOOTNOTE1 "Generated by the SAS System (&_SASSERVERNAME, &SYSSCPL) on
%TRIM(%QSYSFUNC(DATE(), NLDATE20.)) at %TRIM(%SYSFUNC(TIME(),
NLTIMAP20.))";
PROC GPLOT DATA=WORK.SORTTempTableSorted
;
PLOT Frequency * Value_0001 /
      VAXIS=AXIS1

      HAXIS=AXIS2

FRAME ;
/* ---------------------------------------------------------------------
   End of task code.
   --------------------------------------------------------------------- */
RUN; QUIT;
%_eg_conditional_dropds(WORK.SORTTempTableSorted);
TITLE; FOOTNOTE;
GOPTIONS RESET = SYMBOL;
```

D.5. Mixed Models ANOVA of risk by water body size type

```
**pgm.sas;


%let
inc='AK','AB','AZ','BC','CA','CO','ID','MB','MT','NV','NM','NL','NU','NT',
'ON','OR','QC','SK','UT','WA','WY','YT';
Title "The following were included in analysis-'AK',
'AB','AZ','BC','CA','CO','ID','MB','MT','NV','NM','NL','NU',
'NT','ON','OR','QC','SK','UT','WA','WY,'YT'";
libname in 'C:\Users\bmhartis.CROPSCI-AD\Desktop\Extension
Position\Extension\Hydrilla Extent';
%include "C:\Users\bmhartis.CROPSCI-AD\Desktop\Extension
Position\Extension\Hydrilla Extent\danda.sas";

data a; set in.lakedata;
Where state in(&inc);
if acres > 10000 then acres=10000;
run;

ods listing close;
ods pdf file="C:\Users\bmhartis.CROPSCI-AD\Desktop\Extension
Position\Extension\Hydrilla Extentpgm1.pdf";

ods graphics on;

ods exclude pearsonpanel residualpanel lsmeans diffs;
proc mixed data =a plots=studentpanel;
      class state size_type;
      model risk_poten= state|size_type/ddfm=kr residual;
      *lsmeans state size_type;
    lsmeans state*size_type/ pdiff slice=state;
      ods output diffs=ppp lsmeans=mmm;
run;

%pdmix800(ppp,mmm,alpha=.05,sort=yes, slice=state);
proc sort data=a; by state;
proc sgplot; by state;
      scatter y=risk_poten x=acres;
run;


proc reg data=a;
      model risk_poten=acres;
run;

proc sort data=a; by risk_poten;
proc means data=a maxdec=2 min mean median range; class risk_poten;
var acres;
run;
```

```
proc freq data=a nlevels;
      tables state*size_type  risk_poten*size_type/norow nocol nopercent;
      tables state*risk_poten / nocol nopercent;
      tables risk_poten;
run;

ods graphics off;
ods pdf close;
ods listing;
title;
```

D.6. Tukey-Kramer Analysis of risk by water body size type

```
                                        The SAS System        11:56
Wednesday, October 16, 2013

1          ;*';*";*/;quit;run;
2          OPTIONS PAGENO=MIN;
3          %LET _CLIENTTASKLABEL='pgm1-2';
4          %LET _CLIENTPROJECTPATH='C:\Users\bmhartis.CROPSCI-
AD\Desktop\Extension
4        ! Position\Extension\Hydrilla
Extent\Water_All\Final_Data\processing2.egp';
5          %LET _CLIENTPROJECTNAME='processing2.egp';
6          %LET _SASPROGRAMFILE='C:\Users\bmhartis.CROPSCI-
AD\AppData\Local\Temp\pgm1-2.sas';
7
8          ODS _ALL_ CLOSE;
9          OPTIONS DEV=ACTIVEX;
NOTE: Procedures may not support all options or statements for all
devices. For details, see
      the documentation for each procedure.
10         GOPTIONS XPIXELS=0 YPIXELS=0;
11         FILENAME EGSR TEMP;
12         ODS tagsets.sasreport12(ID=EGSR) FILE=EGSR STYLE=Analysis
12       !
STYLESHEET=(URL="file:///C:/Program%20Files/SAS/SharedFiles(32)/BIClientSt
yles/4.2/An
12       ! alysis.css") NOGTITLE NOGFOOTNOTE GPATH=&sasworklocation
ENCODING=UTF8
12       ! options(rolap="on");
NOTE: Writing TAGSETS.SASREPORT12(EGSR) Body file: EGSR
13
14         GOPTIONS ACCESSIBLE;
15         **pgm.sas;
16         %include "C:\Users\bmhartis.CROPSCI-AD\Desktop\Extension
Position\Extension\Hydrilla
16       ! Extent\danda.sas";
5997
5998       %let
5998     !
inc='AK','AB','AZ','BC','CA','CO','ID','MB','MT','NV','NM','NL','NU','NT',
'ON','OR','
5998     ! QC','SK','UT','WA','WY','YT';
5999       Title "The following were included in analysis-'AK',
5999     ! 'AB','AZ','BC','CA','CO','ID','MB','MT','NV','NM','NL','NU',
5999     ! 'NT','ON','OR','QC','SK','UT','WA','WY,'YT'";
6000       libname in 'C:\Users\bmhartis.CROPSCI-AD\Desktop\Extension
6000     ! Position\Extension\Hydrilla Extent';
NOTE: Libref IN was successfully assigned as follows:
      Engine:        V9
      Physical Name: C:\Users\bmhartis.CROPSCI-AD\Desktop\Extension
Position\Extension\Hydrilla
      Extent
```

```
6001       %include "C:\Users\bmhartis.CROPSCI-AD\Desktop\Extension
Position\Extension\Hydrilla
6001     ! Extent\danda.sas";
11982


11983      data a; set in.lakedata;
11984      Where state in(&inc);
11985      if acres > 10000 then acres=10000;
11986      run;

NOTE: There were 89365 observations read from the data set IN.LAKEDATA.
      WHERE state in ('AB', 'AK', 'AZ', 'BC', 'CA', 'CO', 'ID', 'MB',
'MT', 'NL', 'NM', 'NT',
      'NU', 'NV', 'ON', 'OR', 'QC', 'SK', 'UT', 'WA', 'WY', 'YT');
NOTE: The data set WORK.A has 89365 observations and 15 variables.
NOTE: DATA statement used (Total process time):
      real time           0.11 seconds

      cpu time            0.09 seconds


11987      ods listing close;
11988      ods pdf file="Pgm1 Tukey.pdf";
WARNING: Unsupported device 'ACTIVEX' for PDF destination. Using device
'ACTXIMG'.
NOTE: Writing ODS PDF output to DISK destination "C:\Windows\system32\Pgm1
Tukey.pdf",
      printer "PDF".
11989
11990      ods graphics on;
11991
11992      ods exclude pearsonpanel residualpanel lsmeans diffs;
11993      proc mixed data =a plots=studentpanel;
11994       class state size_type;
11995       model risk_poten= state|size_type/ddfm=kr residual;
11996       *lsmeans state size;
11997          lsmeans state*size_type / pdiff slice=state adjust=tukey;
11998       ods output diffs=ppp lsmeans=mmm;
11999      run;

NOTE: The data set WORK.MMM has 66 observations and 8 variables.
NOTE: The data set WORK.PPP has 2145 observations and 12 variables.
NOTE: PROCEDURE MIXED used (Total process time):
      real time           42.16 seconds
      cpu time            3.27 seconds


12000
12001      %pdmix800(ppp,mmm,alpha=.05,sort=yes, slice=state);
PDMIX800 1.29 processing
```

```
state*size_type AB Tukey-Kramer(P<.05) average value=0.22565, min=0.14327,
max=0.2722
  state*size_type AK Tukey-Kramer(P<.05) average value=0.18408,
min=0.12755, max=0.22285
  state*size_type AZ Tukey-Kramer(P<.05) average value=0.96824,
min=0.28034, max=1.3261
  state*size_type BC Tukey-Kramer(P<.05) average value=0.2196,
min=0.14168, max=0.26438
  state*size_type CA Tukey-Kramer(P<.05) average value=0.394, min=0.11126,
max=0.54049
  state*size_type CO Tukey-Kramer(P<.05) average value=1.02239,
min=0.15794, max=1.45859
  state*size_type ID Tukey-Kramer(P<.05) average value=0.62181,
min=0.22537, max=0.83304
  state*size_type MB Tukey-Kramer(P<.05) average value=0.3028,
min=0.25447, max=0.35139
  state*size_type MT Tukey-Kramer(P<.05) average value=0.59941,
min=0.11561, max=0.84496
  state*size_type NL Tukey-Kramer(P<.05) average value=0.27477,
min=0.23999, max=0.30923
3                                       The SAS System        11:56
Wednesday, October 16, 2013

  state*size_type NM Tukey-Kramer(P<.05) average value=0.93365,
min=0.19233, max=1.31088
  state*size_type NT Tukey-Kramer(P<.05) average value=1.02031,
min=0.56457, max=1.30916
  state*size_type NU Tukey-Kramer(P<.05) average value=0.5777, min=0.4164,
max=0.72031
  state*size_type NV Tukey-Kramer(P<.05) average value=0.60367,
min=0.22763, max=0.80272
  state*size_type ON Tukey-Kramer(P<.05) average value=0.14236,
min=0.11525, max=0.15815
  state*size_type OR Tukey-Kramer(P<.05) average value=0.49176,
min=0.15886, max=0.66614
  state*size_type QC Tukey-Kramer(P<.05) average value=0.1573,
min=0.11905, max=0.17737
  state*size_type SK Tukey-Kramer(P<.05) average value=0.32595,
min=0.21275, max=0.3862
  state*size_type UT Tukey-Kramer(P<.05) average value=0.5913,
min=0.19841, max=0.79818
  state*size_type WA Tukey-Kramer(P<.05) average value=0.59336,
min=0.15608, max=0.81821
  state*size_type WY Tukey-Kramer(P<.05) average value=0.57266,
min=0.15263, max=0.78946
  state*size_type YT Tukey-Kramer(P<.05) average value=2.30814,
min=0.96583, max=3.04222
NOTE: PROCEDURE OPTLOAD used (Total process time):
      real time           0.02 seconds
      cpu time            0.00 seconds


12002
```

```
12003      proc sort data=lsdvalavgzz out=lsd nodupkey; by state;
NOTE: There were 22 observations read from the data set WORK.LSDVALAVGZZ.
NOTE: 0 observations with duplicate key values were deleted.
NOTE: The data set WORK.LSD has 22 observations and 7 variables.
NOTE: PROCEDURE SORT used (Total process time):
      real time           0.00 seconds
      cpu time            0.00 seconds

12004      proc print data=lsd;

12005      title2 "Print LSD Values from Danda Macro";
12006      run;

NOTE: There were 22 observations read from the data set WORK.LSD.
NOTE: PROCEDURE PRINT used (Total process time):
      real time           0.05 seconds
      cpu time            0.01 seconds


12007
12008
12009      proc sort data=a; by risk_poten;

NOTE: There were 89365 observations read from the data set WORK.A.
NOTE: The data set WORK.A has 89365 observations and 15 variables.
NOTE: PROCEDURE SORT used (Total process time):
      real time           0.06 seconds
      cpu time            0.04 seconds

12010      proc means data=a maxdec=2 min mean median range; class
risk_poten;

12011      var acres;
12012      run;
```



```
NOTE: There were 89365 observations read from the data set WORK.A.
NOTE: PROCEDURE MEANS used (Total process time):
      real time           0.08 seconds
      cpu time            0.09 seconds


12013
12014
12015      proc freq data=a nlevels;
12016       tables state*size_type state*risk_poten risk_poten*size_type /
nocol nopercent;
12017       tables risk_poten;
12018      run;
NOTE: There were 89365 observations read from the data set WORK.A.
NOTE: PROCEDURE FREQ used (Total process time):
```

```
real time           7.90 seconds
      cpu time            1.85 seconds


12019
12020      ods graphics off;
12021      ods pdf close;
12022      ods listing;
12023      title;
12024
12025
12026      GOPTIONS NOACCESSIBLE;
12027      %LET _CLIENTTASKLABEL=;
12028      %LET _CLIENTPROJECTPATH=;
12029      %LET _CLIENTPROJECTNAME=;
12030      %LET _SASPROGRAMFILE=;
12031
12032      ;*';*";*/;quit;run;
12033      ODS _ALL_ CLOSE;
12034
12035
12036      QUIT; RUN;
12037
```