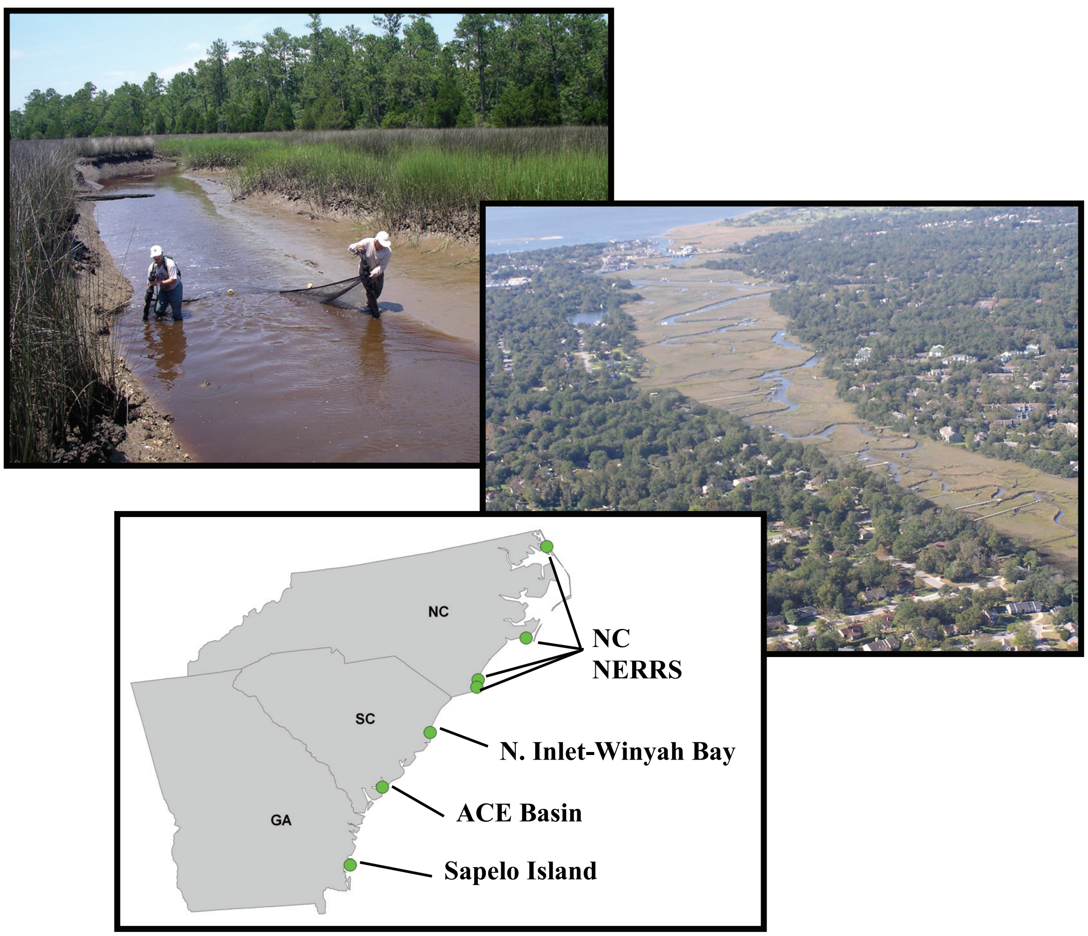
Support for Integrated Ecosystem Assessments of NOAA's National Estuarine Research Reserves System (NERRS), Volume I:

The Impacts of Coastal Development on the Ecology and Human Well-being of Tidal Creek Ecosystems of the US Southeast



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Abstract

A study was conducted, in association with the Sapelo Island and North Carolina National Estuarine Research Reserves (NERRs), to evaluate the impacts of coastal development on sentinel habitats (e.g., tidal creek ecosystems), including potential impacts to human health and well-being. Uplands associated with southeastern tidal creeks and the salt marshes they drain are popular locations for building homes, resorts, and recreational facilities because of the high quality of life and mild climate associated with these environments. Tidal creeks form part of the estuarine ecosystem characterized by high biological productivity, great ecological value, complex environmental gradients, and numerous interconnected processes. This research combined a watershed-level study integrating ecological, public health and human dimension attributes with watershed-level land use data. The approach used for this research was based upon a comparative watershed and ecosystem approach that sampled tidal creek networks draining developed watersheds (e.g., suburban, urban, and industrial) as well as undeveloped sites. The primary objective of this work was to clearly define the relationships between coastal development with its concomitant land use changes and non-point source pollution loading and the ecological and human health and well-being status of tidal creek ecosystems.

Nineteen tidal creek systems, located along the southeastern United States coast from southern North Carolina to southern Georgia, were sampled during summer (June-August), 2005 and 2006. Within each system, creeks were divided into two primary segments based upon tidal zoning: intertidal (i.e., shallow, narrow headwater sections) and subtidal (i.e., deeper and wider sections), and watersheds were delineated for each segment. In total, we report findings on 24 intertidal and 19 subtidal creeks. Indicators sampled throughout each creek included water quality (e.g., dissolved oxygen concentration, salinity, nutrients, chlorophyll-*a* levels), sediment quality (e.g., characteristics, contaminants levels including emerging contaminants), pathogen and viral indicators, and abundance and genetic responses of biological resources (e.g., macrobenthic and nektonic communities, shellfish tissue contaminants, oyster microarray responses).

For many indicators, the intertidally-dominated or headwater portions of tidal creeks were found to respond differently than the subtidally-dominated or larger and deeper portions of tidal creeks. Study results indicate that the integrity and productivity of headwater tidal creeks were impaired by land use changes and associated non-point source pollution, suggesting these habitats are valuable early warning sentinels of ensuing ecological impacts and potential public health threats. For these headwater creeks, this research has assisted the validation of a previously developed conceptual model for the southeastern US region. This conceptual model identified adverse changes that generally occurred in the physical and chemical environment (e.g., water quality indicators such as indicator bacteria for sewage pollution or sediment chemical contamination) when impervious cover levels in the watershed reach 10-20%. Ecological characteristics responded and were generally impaired when impervious cover levels exceed 20-30%. Estimates of impervious cover levels defining where human uses are impaired are currently being determined, but it appears that shellfish bed closures and the flooding vulnerability of headwater regions become a concern when impervious cover values exceed 10-30%. This information can be used to forecast the impacts of changing land use patterns on tidal creek environmental quality as well as associated human health and well-being. In addition, this

study applied tools and technologies that are adaptable, transferable, and repeatable among the high quality NERRS sites as comparable reference entities to other nearby developed coastal watersheds. The findings herein will be of value in addressing local, regional and national needs for understanding multiple stressor (anthropogenic and human impacts) effects upon estuarine ecosystems and response trends in ecosystem condition with changing coastal impacts (i.e., development, climate change).

Key Words: National Estuarine Research Reserve System (NERRS), North Carolina NERR, Sapelo Island NERR, tidal creek, sentinel habitat, conceptual model, impervious cover, land use, urbanization, sediment and tissue contaminants, water quality, pathogens, nekton, oysters, macrobenthos, physical and chemical environment.

1. Introduction

This project, "Support for Integrated Ecosystem Assessments of NOAA's National Estuarine Research Reserves System (NERRS)", initiated in 2006, was completed as part of an emerging research partnership between the National Centers for Coastal Ocean Science (NCCOS) and the National Estuarine Research Reserve System (NERRS) to develop approaches for characterizing the ecosystem condition and public-health status of NERRS sites as compared to nearby, similar, more developed watersheds. This collaboration also provides a framework for continued monitoring (including the transfer of techniques and technologies) and prediction of future conditions in these important protected estuarine systems.

There are two components of the overall study: (1) a sentinel habitat component conducted in tidal creeks at the NERRS sites in Georgia (Sapelo Island) and North Carolina (Masonboro Island); and (2) a subtidal probabilistic-sampling component conducted at all four North Carolina NERR sites (Currituck Banks, Rachel Carson, Masonboro Island, Zeke's Island). This effort involves four NCCOS Centers (Hollings Marine Laboratory, Center for Coastal Environmental Health and Biomolecular Research, Center for Coastal Monitoring and Assessment, Center for Coastal Fisheries and Habitat Research) working in close collaboration with the NERRS program to address common research and coastal management goals. NCCOS's mission is to provide coastal managers with scientific information and tools needed to balance society's environmental, social, and economic goals (NCCOS 2004). The mission of NERRS is to practice and promote coastal and estuarine stewardship through innovative research and education activities (NERRS 2005). Through this collaboration, a number of high priority U.S. coastal management issues are being addressed such as assessing impacts of changing watershed land use, coastal habitat change, nutrient runoff on coastal rivers and bays, and providing tools and information for modeling the effects of sea level change with coastal watersheds (Pew Ocean Commission 2003, Coastal States Organization 2004).

This NERRS-NCCOS partnership for this project resulted in solid contributions by the NERRS in terms of planning, field support and logistics, and data interpretation. The NERRS local information assisted greatly in the identification of tidal creek sampling sites and any nearby watershed influences on water quality (i.e., land use change and development) that supported improved watershed and historical data interpretation. Reserve access, both on land and on the water, was facilitated through NERRS and enabled improved access to sampling locations.

Together, the two project components demonstrate the utility of two complementary assessment tools, (1) a sentinel habitats study designed to evaluate the impacts of development on tidal creek ecosystems, including potential impacts to human health and well-being; and (2) a probabilistic study providing a means for assessing the spatial extent of condition throughout a targeted resource category (i.e., sub-tidal estuarine waters of a reserve) and how the relative proportions of healthy vs. degraded areas may be changing with time. An associated objective of the project is to provide a prototype framework of assessment strategies (including tools and technology transfer) that can be applied systematically across other reserves, to support national and regional comparisons. This collaboration is a crucial step towards determining if healthy coastal ecosystems are associated with healthy people and healthy economies as well as assessing trends in ecosystem characteristics over time.

The sentinel habitats portion of this effort was also conducted as part of the Hollings Marine Laboratory's (HML) Center of Excellence in Oceans and Human Health (OHH). The Center brings basic, applied, and medical researchers together to identify and understand factors that affect the health of coastal ecosystems and the humans who live in or visit the coastal zone. The science focus of the HML Center is to develop biotechnology that identifies and evaluates linkages between coastal development, the condition of the marine ecosystems, and public health and well-being.

As human land use in coastal areas has shifted from industry to residential development, point source industrial discharge has been replaced by non-point source pollution as the primary threat to estuarine ecological health. The amount, timing, and quality of stormwater runoff from rapid, relatively unplanned development directly affects the introduction of freshwater, sediments, chemicals, bacteria, viruses, and other pollutants into tidal creeks and salt marshes. These pollutants often come from non-point sources (NPS), which can be difficult to identify, and can substantially degrade environmental quality. Pathogens and chemical contaminants can accumulate to such high levels in the water, sediments, and organisms that seafood products become unsafe to eat and water becomes unsafe for contact recreation. Thus, human activities, depending on the extent, may impair these valuable resources and result in negative feedback to human health.

Tidal creeks provide a useful sentinel habitat for coastal systems. Tidal creeks form part of the estuarine ecosystem characterized by high biological productivity, great ecological value, complex environmental gradients, and numerous interconnected processes. River and tidal creek networks form the primary hydrologic link between estuaries and land based activities. Finally, these networks are critical feeding grounds, spawning areas, and nursery habitats for many species of fish, shellfish, birds, waterfowl, and mammals. Thus, the significant economic and ecological value of tidal creek habitats and the degree of human interaction with these habitats are disproportionate to their spatial area. Recent research has demonstrated that the ecological condition of intertidal headwater areas of southeastern tidal creek ecosystems is a sensitive indicator of land use changes within the local watershed (Holland et al. 2004, Sanger et al. 2004, DiDonato et al.in press). These headwater reaches are the first zone of impact for non-point source pollution runoff, and as a result, the levels of microbial and chemical contamination in headwaters are frequently an order of magnitude greater than levels reported for their contiguous deeper open-water environments (Holland and Sanger Unpublished, DiDonato et al. in press). As such, tidal creek ecosystems serve as sentinel habitats for assessing the impact of watershed development on ecosystem condition and public health risk. They also provide a reliable tool for identifying pollution sources and serve as sentinels for assessment of the effectiveness of corrective actions aimed at recovering ecosystem-scaled health.

A conceptual model linking watershed development (stressors), the associated physical and chemical exposures, and ecological responses has been developed for South Carolina tidal creeks (Holland et al. 2004, Figure 1). Changes in the rate and volume of stormwater runoff resulting from increases in impervious cover were a predominant factor driving ecological impairment. Adverse changes in the physical and chemical environment occurred when impervious cover exceeded 10-20%. Ecological processes responded when impervious cover exceeded 20-30%.

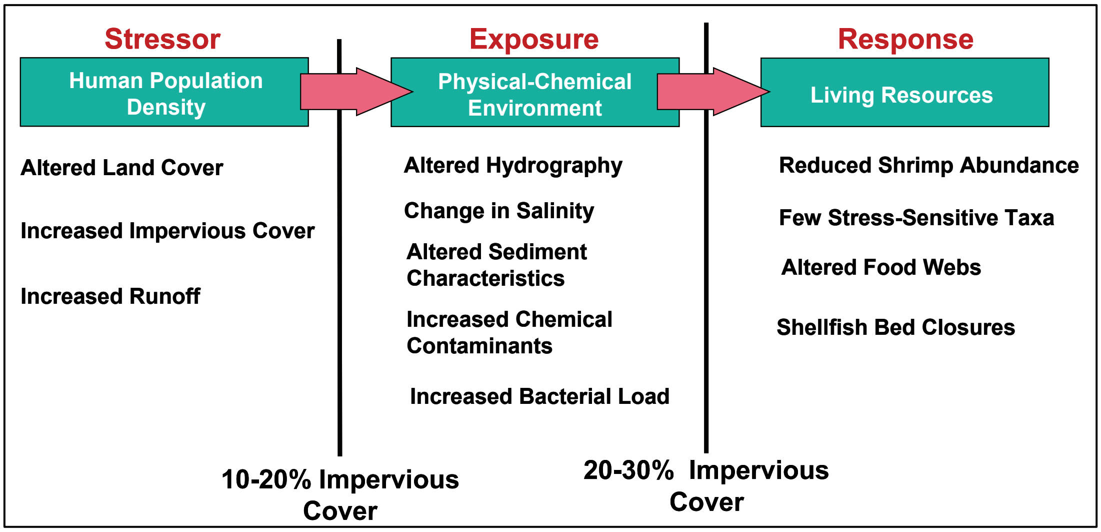

Figure 1. Conceptual model developed by Holland et al. (2004) identifying linkages between development of the upland and the ecological response of South Carolina tidal creeks. Ranges of impervious cover percent denoting transition from one model phase to the next are shown at the bottom of the model.

The overall goals of the sentinel habitat study were: 1) to evaluate the applicability of the current tidal creek classification framework and conceptual model linking urban and suburban growth to tidal creek ecological condition and potential impacts on ecosystem function, including human health and well-being, in the southeast region through collaborations with Sapelo Island NERR and North Carolina NERR; and 2) to support the transfer of this information/data to appropriate coastal managers to improve decision making related to land use/development issues and human health concerns in the Southeast. Results of this study are presented here in Volume I of a two-volume report for the overall NERRS project. Results of the probabilistic survey of ecological condition throughout subtidal waters of the four North Carolina NERR sites are presented in the companion Volume II.

2. Methods

2.1 Study Sites

Nineteen tidal creek networks located from New Hanover County, NC, to Glynn County, GA, were sampled during summer (June-August) periods in 2005 and 2006 (Figure 2). Twelve networks were sampled in SC in 2005, and four networks were resampled in 2006. In 2006, four creek networks were sampled in GA, and three networks were sampled in NC. The GA and NC sites were sampled in association with the NERRS sites (Table 1).

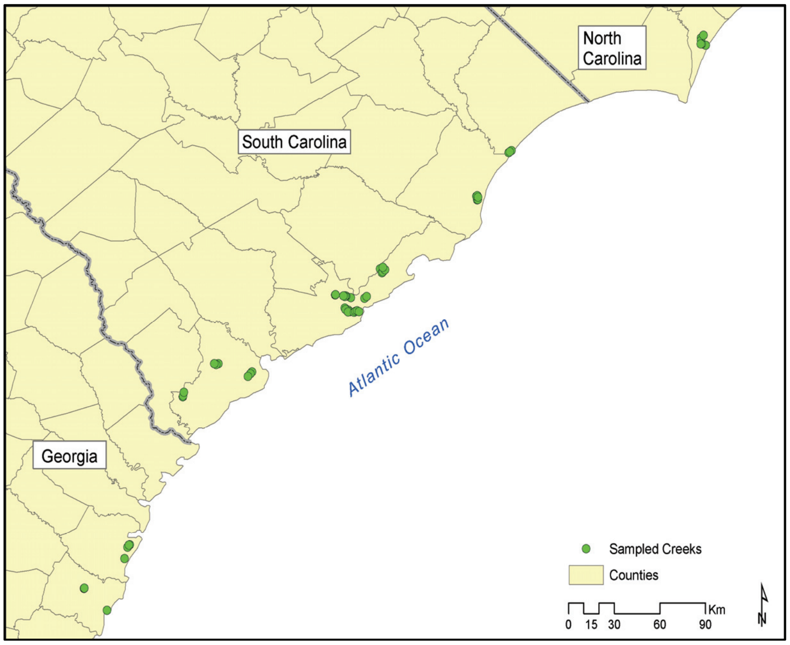

Figure 2. North Carolina, South Carolina, and Georgia sampling sites.

A longitudinal gradient was identified by applying a freshwater stream classification model (Horton, 1945; Strahler, 1957) to tidal creek systems. The first order, or headwater, of each creek directly drained coastal uplands or salt marsh habitat and was characterized by its narrow width and predominately intertidal habitat. These first order sections will be referred to as intertidal in the remaining text. The second order of each creek was formed by the confluence of two or more first order creeks. Second order systems were wider and had subtidally-dominated habitats. The third order of each creek was formed by the confluence of two or more second order creeks. Third order systems were large creeks with a proportionally small amount of intertidal habitat and substantial subtidal habitat. For simplicity, the second and third order systems will be collectively referred to as subtidal throughout the remaining text. While combining second and third orders resulted in the loss of some information about the tidal creek longitudinal gradient, two points support their pooling in this study: (1) in SC, the differences between second and third orders were small, and (2) only one third order creek could be sampled outside of SC, making regional evaluation of that order impossible. Each order was divided into three equidistant reaches using ArcGIS 9 (ESRI, Redlands, CA); by convention, the first reach within an order was the furthest upstream, while the second reach was the middle, and the third reach was the furthest downstream section sampled. Within any reach of any creek order, stations were randomly located for sample collection. The specific sampling activities within each reach are detailed below.

2.2 Land Use and Watershed Determinations

Watersheds and subwatersheds were identified using ArcGIS 9 to evaluate the land use and impervious cover of each creek and order. Watersheds and their sub-watersheds were delineated based on elevation contours. Elevation Derivatives for National Applications (EDNA) data were downloaded from the United States Geological Survey (USGS, http://edna.usgs.gov/). The EDNA watersheds have a resolution of 30 meters that corresponds to a scale of 1:24,000. Single watershed boundaries were identified for the respective research area and overlaid with digital elevation model (DEM) and USGS topographic data. Visual confirmation of the delineation of the EDNA watersheds using the topographic maps was conducted, and subwatersheds were delineated for each creek order. In general, the EDNA data were found to represent the expected watershed boundaries with only slight modifications needed to reflect specific elevation gradients or other attributes such as roads that might impede surface runoff. These changes were made by hand digitization.

Table 1. Orders sampled, date sampled, latitude, and longitude for each study creek network by state. NOC = North Carolina NERR, NIWB = North Inlet-Winyah Bay NERR, ACE = Ashepoo, Combahee and Edisto NERR, SAP=Sapelo Island NERR.

Creek System	Orders Sampled	Date Sampled 2005	Date Sampled 2006	Latitude	Longitude
North Carolina					
Hewlitts	1, 1, 2		7-Aug-06	34.189	-77.857
NOC-Masonboro	1		9-Aug-06	34.152	-77.849
Whiskey Creek	1, 2		9-Aug-06	34.161	-77.865
South Carolina					
Albergottie	1, 2, 3	17-Aug-05		32.448	-80.720
Bulls	1, 2, 3	29-Jun-05		32.825	-80.027
Guerin	1, 1, 2, 2, 3	5-Jul-05	20-Jun-06	32.944	-79.766
James Island	1, 1, 2, 2, 3	1-Aug-05	17-Aug-06	32.744	-79.974
Murrells Inlet	2, 3	22-Jun-05		33.564	-79.025
New Market	1	8-Aug-05	24-Jul-06	32.806	-79.940
NIWB-Town	1, 1, 2, 2, 3	20-Jun-05		33.339	-79.189
Okatee	1, 2, 3	18-Jul-05		32.287	-80.929
Orangegrove	1, 1, 2, 3	20-Jul-05		32.812	-79.978
Parrot	1, 1, 2, 3	7-Jul-05		32.733	-79.910
Shem	1, 2	29-Aug-05		32.801	-79.869
ACE-Village	1, 2, 3	3-Aug-05	5-Jul-06	32.419	-80.522
Georgia					
Burnett	1, 2		19-Jul-06	31.234	-81.538
SAP-Duplin	1, 2, 3		11-Jul-06	31.145	-81.285
SAP-Oakdale	1		11-Jul-06	31.481	-81.272
Postell	1		19-Jul-06	31.417	-81.375

National Land Cover Data (NLCD 2001, Homer et al. 2004) were downloaded for selected regions in SC, NC and GA using the MRLC web tool (http://gisdata.usgs.net/website/MRLC/viewer.php). The land cover and impervious cover data were matched to the watershed and subwatershed boundary data. Land cover data were determined from this layer and summed to obtain simplified categories of land cover. The impervious cover data were further modified by removing data that represented marsh and open water using National Wetlands Inventory (NWI). Specifically, three attribute classes from the NWI were selected and saved as a unique Shapefile® including Bay/Estuary, Non-Forested Wetland, and Open Water and then all attribute classes were merged to create one unique attribute class. This was used to separate marsh and open water (i.e., undevelopable areas) from the impervious cover data. All editing functions were performed in ArcEditor. Impervious cover levels were then calculated directly from the NLCD for all sub-watersheds and watersheds. These NLCD-derived impervious cover estimates were compared to values published in Holland et al. (2004). Both of these data sets used aerial photography from similar time frames (1999-

2001) to estimate impervious cover, but the NLCD impervious cover values underestimated the ground-truthed data reported in Holland et al. (2004). A similar underestimation has also been reported by Jarnagin et al. (2006). A quadratic relationship was developed ($y = 2.9301 + 2.16789x - 0.01611x^2$ where y is the adjusted impervious cover percent and x is NLCD-derived impervious cover, White et al. in prep), and used to adjust the NLCD-derived impervious cover percentage. Adjusted values are reported and used in this study.

Creek watersheds were classified at the largest order level into the following land use categories based on impervious cover as modified from Holland et al. (2004): (1) forested (< 10% impervious cover); (2) suburban (≥ 10% but < 35% impervious cover); (3) urban (≥ 35% impervious cover); and (4) salt marsh (emergent marsh instead of upland as the dominant land cover class). There were two exceptions to this classification. The Orangegrove watershed was estimated to have 37.3% impervious cover; however, since this was primarily light residential development and a small amount of upland (127 ha) relative to the total watershed size (322 ha), we categorized this as a suburban watershed. The Burnett watershed was estimated to have 11.8% impervious cover; however, since this was a superfund site designated by the United States Environmental Protection Agency (USEPA), it was categorized as an urban watershed.

The creek networks varied in regards to their size (i.e., number of orders and watershed size) and also with respect to the surrounding land use. It is important to note that several systems had upland creeks showing various levels of human development but also had creek segments that were dominated by salt marsh. Specifically, within the North Inlet, Guerin, Parrot, and Orangegrove networks, we sampled both upland and salt marsh creeks and they are treated separately in statistical analyses.

2.3 Stormwater Runoff Determinations

The flow curve number method developed by the U.S. Department of Agriculture (USDA) National Resources Conservation Service (NRCS) was used to estimate stormwater runoff volume for the 19 intertidal subwatersheds of upland creeks. The method is based upon the relationship between rainfall, runoff, and retention (rain not converted to runoff), and the hypothesis is that the ratio of actual retention to the potential maximum retention is similar to the ratio of actual direct runoff to potential maximum runoff (i.e., total rainfall. The flow curve number (CN) is a representation of the potential maximum retention and reflects the drainage characteristics of a watershed's soil and land cover. CN is determined by identifying the proportional composition of land cover categories and hydrologic soil groups within a watershed.

USDA-NRCS provides listings of numerous land cover categories, with each having 4 CN values based upon soil classification from 'A' (most pervious – sands) to 'D' (most impervious – clays). NLCD land cover categories were combined to match the CN method's applicable categories. For each land cover category, the USDA-provided CN number was modified by the proportion of hydrologic soil groups in the watershed, as determined by spatial soil data layers provided by USDA. The calculated CN was modified in the developed land cover categories by increasing the imperviousness by two grades to reflect soil compaction (Lim et al. 2006).

Once the CN was determined, watershed runoff volume was solved as follows (NRCS 2004):

$$S = \frac{1000}{CN} - 10$$

S = maximum potential retention after runoff begins

$$Q_d = \frac{(P - I_a)^2}{(P - I_a) + S}$$

Q_d = depth of runoff, inches
P = depth of rainfall (inches)
I_a = initial abstraction (rainfall lost to infiltration and surface depressions before runoff occurs (in.)

$$Q_{vol} = \frac{Q_d \times A_d}{12}$$

Q_{vol} = volume of runoff, acre-feet (af)
Q_d = depth of runoff, inches
A_d = drainage area, acres
12 = conversion factor for inches to feet

Two additional equations were used based on our modification of the I_a : S ratio from 0.2 to 0.05 (Woodward et al. 2003).

$$S_{0.05} = (1.33 \times S_{0.20})^{1.15} \qquad CN_{0.05} = \frac{100}{1.879 \times \left(\left(100 \div CN_{0.20}\right) - 1\right)^{1.15} + 1}$$

The solved value for runoff volume was converted from acre-feet to m^3 by applying a factor of 1233.48. Disparities in watershed areas were normalized by determining volume per unit area (km^2). For consistency in this report, all volumes were based on precipitation of 4.5 inches which is the rainfall designated by the National Weather Service as the two-year storm event for the general geographical location of the study watersheds.

The data calculated by applying the modified NRCS-CN method were used to examine the relationship between differences in the amount of runoff and in the amount of developed land cover among watersheds and to compare the mean and median differences among land use classes.

2.4 Sample Design

Each creek was sampled during the ebbing tide, approximately 2-3 hours prior to low tide over two consecutive days. The first order was sampled by foot, while the second and third orders were sampled by boat. Sampling was generally conducted in an upstream direction to minimize habitat disturbance. Within each creek order, samples were collected to quantify water quality, water column nutrients, pathogen indicators, macrobenthic infauna, resident nekton, sediment contaminants, oyster transcriptome profiles, and oyster pathogen and contaminant body burdens. Sampling stations were selected using a stratified random method. The number of samples collected in each order varied by sample type.

Water quality data (temperature, dissolved oxygen, salinity, pH, turbidity, chlorophyll-*a*) were collected in bottom waters (0.3 m above bottom) using a YSI 6600 data logger. A logger was deployed in the second reach of each creek order and collected data at 15 minute intervals for up to 2 full tidal cycles (25 hrs). Water samples for pathogen indicators were collected in the second reach in sterile 2 L polypropylene bottles. In addition, a water sample was collected in an acid-washed 500 mL polyethylene bottle within each reach of each creek order for nutrient determinations. All water grab samples were collected approximately 0.3 meters below surface, or mid-water column in water less than 0.3 m in depth. In addition, all water grab samples were collected by hand and in an upstream direction to prevent contamination. Bottle caps were removed immediately prior to sampling, and bottles were inverted until at sample depth.

Macrobenthic infauna were sampled using two different field methods. In intertidal creeks, the benthos was sampled approximately 1 m below mean high water (MHW) using a 0.0044 m^2 core sampler. A total of 9 cores (3 from each reach) were collected at randomly located stations to ensure sampling of the benthic fauna along the entire headwater habitat. A small scoop of mud was collected next to each core sample for sediment analysis [% sand, % silt, % clay, total organic carbon (TOC)). Additionally, 2 small cores (0.0009 m^2) were collected at each site and composited across all sites within each reach to quantify porewater ammonium (NH_4^+). In subtidal creeks, the infauna were sampled using a 0.04 m^2 modified Van Veen grab sampler. One grab sample was collected for benthos in each reach. Sediment samples for grain size analysis and porewater ammonia determination were taken from the top 2 centimeters of a second intact grab from each site.

Sediments were sampled for chemical contaminants only once in each creek order. In the second reach of intertidal creeks, the top 2 cm of sediment were carefully scraped off the surface of mud exposed at low tide and homogenized in a stainless steel bowl. In the second reach of subtidal creeks, the top 2 cm of a successful Van Veen grab were homogenized for chemical analysis. The homogenate was apportioned to appropriate pre-cleaned sample jars (i.e., metals in plastic and organics in glass) and placed on ice as soon as possible.

Nekton, predominantly fish and epibenthic crustaceans, were sampled using different field methods for different creek orders. In intertidal creeks, the nekton was sampled using a ¼ inch mesh seine net. One seine was pulled in each reach in an upstream direction for up to 25 meters. Every effort was made to stretch the net from bank to bank. In cases where this was not possible the seined width was estimated to the nearest meter. Water width and depth were measured at both the starting and end points in the seine to calculate the area and volume of the creek swept. In subtidal creeks, nekton were sampled using a 4-seam trawl (5.5 m foot rope, 4.6 m head rope, and 1.9 cm bar mesh throughout) pulled at a constant speed in the downstream direction for 250 m.

In 2006 only, oysters (*Crassostrea virginica*) were hand-collected in every creek order when present. If possible oysters were collected near the site where the data logger was deployed and sediment samples collected in the second reach. After collection, oysters were divided up for genomic transcriptome analyses (25 oysters), pathogen determination (~ 20 oysters), and chemical contaminant body burdens (~12 oysters).

2.5 Laboratory Processing Methods

2.5.1 Basic Water Quality

Basic water quality data (i.e., temperature, pH, DO, salinity, turbidity, depth, and chlorophyll-*a*) were downloaded from the data loggers and examined to remove data resulting from exposure at low tide (common in first order creeks). Data loggers were calibrated prior to deployment and a post-calibration check was conducted after retrieval to ensure the logger was functioning properly. Summary data (e.g., mean, maximum, minimum, range) for the measured parameters were calculated.

2.5.2 Nutrients and Phytoplankton

Both whole and filtered water samples were used for nutrient analyses. Whole water samples were analyzed for total nitrogen (TN) and total phosphorus (TP) using the persulfate digestion method (D'Elia et al. 1977). Additional samples were filtered through a 47 mm GF/F (Whatman) to quantify dissolved constituents (i.e., ammonium (NH_4^+), nitrite+nitrate ($NO_{2/3}$), total dissolved nitrogen (TDN), ortho-phosphate (PO_4^{3-}), total dissolved phosphorus (TDP), and silicate (DSi)). Ammonium was analyzed via the Berthelot Reaction using a Technicon AutoAnalyzer (Technicon Industrial Systems, 1986), and silicate was measured using the "molybdenum blue" method on the same AutoAnalyzer (Technicon Industrial Systems, 1986). Both ortho-phosphate and nitrate+nitrite were analyzed using standard methods (EPA methods 365.1 and 365.2, respectively, in USEPA, 1979). The material remaining on the filter paper was extracted in acetone and analyzed for chlorophyll-*a* (Chl-*a*) and phaeophytin (Phaeo) using fluorometric techniques (Welschmeyer 1994).

2.5.3 Pathogen Indicators

Water collected for pathogen indicators was analyzed for both bacterial and viral indicators within 24 hours. Fecal coliforms (FC) and enterococci (Ent) were enumerated by membrane filtration according to standard methods (APHA 1998). Coliphages were enumerated and characterized as described in Stewart et al. (2006). Both male-specific (F+) and somatic (F-) coliphages were enumerated by the single agar layer method, adapted from USEPA Method 1602 (USEPA 2001).

Oysters to be tested for pathogen body burdens were first homogenized and composited for each collection site to obtain at least 100 g (wet weight) tissue. The tissue liquor was tested for the microbial indicators FC, Ent, and also the viral indicators F+, F- coliphages using similar methods to water samples.

2.5.4 Chemical Contaminants

Sediments and oyster tissues were analyzed for a suite of 22 trace metals, 22 pesticides, 25 polycyclic aromatic hydrocarbons (PAHs), 79 polychlorinated biphenyls (PCBs), and 13 polybrominated diphenyl ethers (PBDEs) (Appendix A). PBDEs are used as flame retardants and considered to be an emerging contaminant of concern. Data quality was assured using a series of spikes, blanks, and standard reference materials (NIST 1944 for sediments, and NIST 1566b for tissues). Sediment samples were kept frozen at approximately - 40 ºC until analyzed. To thaw, samples were left in closed containers in a + 4 ºC cooler for approximately 24 hours. Samples were thoroughly homogenized using a ProScientific handheld homogenizer prior to any sample extraction. Tissues from multiple oysters (maximum of 12 oysters) were composited to obtain 15 g of wet weight and then frozen and stored at - 40 ºC until analysis. Oyster tissues were removed from the freezer and stored overnight at 4 ºC and allowed to partially thaw. The oyster tissue was well homogenized using a ProScientific homogenizer in 500 mL Teflon containers. The homogenized tissue sample was split into an organic (pre-cleaned glass container) and inorganic (pre-cleaned polypropylene container) sample and stored at - 40 ºC until extraction or digestion. A percent dry-weight determination was made gravimetrically on an aliquot of each wet sediment and tissue sample.

Inorganic sample digestion and analysis consisted of the following steps. Dried sediment was ground with a mortar and pestle and transferred to a 20 mL plastic screw-top container. A 0.25 g sub-sample of the ground material was transferred to a Teflon-lined digestion vessel and digested in 5 mL of concentrated nitric acid using microwave digestion. The sample was brought to a fixed volume of 50 mL in a volumetric flask with deionized water and stored in a 50 mL polypropylene centrifuge tube until instrumental analysis of Li, Be, Al, Fe, Mg, Ni, Cu, Zn, Cd, and Ag. A second 0.25 g sub-sample was transferred to a Teflon-lined digestion vessel and digested in 5 mL of concentrated nitric acid and 1 mL of concentrated hydrofluoric acid in a microwave digestion unit. The sample was then evaporated on a hotplate at 225 °C to near dryness, and 1 mL of nitric acid was added. The sample was brought to a fixed volume of 50 mL in a volumetric flask with deionized water and stored in a 50 mL polypropylene centrifuge tube until instrumental analysis for V, Cr, Co, As, Sn, Sb, Ba, Tl, Pb, and U. Selenium was analyzed by hotplate digestion using a 0.25 g sub-sample and 5 mL of concentrated nitric acid. Each sample was brought to a fixed volume of 50 mL in a volumetric flask with deionized water and stored in a 50 mL polypropylene centrifuge tube until instrumental analysis. Additionally, two to three grams wet tissue were microwave digested in Teflon-lined digestion vessels using 10 mL of concentrated nitric acid along with 2 mL of hydrogen peroxide. Digested samples were brought to a fixed volume with deionized water in graduated polypropylene centrifuge tubes and stored until analysis.

A separate inorganic aliquot was used for mercury analysis. Approximately 0.5 g of wet sediment or tissue was analyzed on a Milestone DMA-80 Direct Mercury Analyzer. All remaining elemental analysis was performed using an Inductively Coupled Plasma Mass Spectrometry (ICP-MS) except for silver, which was determined using Graphite Furnace Atomic Absorption (GFAA) spectroscopy. Data quality was controlled by using a series of blanks, spiked solutions, and standard reference materials including NRC MESS-3 (Marine Sediments) and NIST 1566b (freeze dried mussel tissue).

Organic extraction and analysis consisted of the following steps. An aliquot (10 g sediment or 5 g tissue wet weight) was extracted with anhydrous sodium sulfate using Accelerated Solvent Extraction (ASE) in either 1:1 methylene chloride:acetone for sediments or 100% Dichloromethane for tissues (Schantz 1997). Following extraction, samples were dried and cleaned using Gel Permeation Chromatography and Solid Phase Extraction to remove lipids and then solvent-exchanged into hexane for analysis. Samples were analyzed for PAHs, PBDEs, PCBs, and a suite of chlorinated pesticides using appropriate Gas Chromatograph and Mass Spectrometer (GC-MS) technology. Data quality was ensured by assessing a spiked blank, a reagent blank, and appropriate standard reference materials with each set of samples to ensure the integrity of the analytical method.

2.5.5 Macrobenthic Community

Benthic samples collected in the field were sieved through a 0.5 mm standard sieve, and the material retained on the screen was transferred to a polyethylene bottle and preserved in 10% formalin containing Rose Bengal. Samples were capped and stored until benthic macroinvertebrates could be sorted out from the detritus, identified down to the lowest practical taxonomic unit, and counted. Quality control / quality assurance (QA/QC) procedures were followed. One out of every 10 samples was re-sorted to ensure 90% sorting efficiency. If 10% of the organisms remained in the sample after sorting, then all 10 samples were resorted. The samples were then identified to the lowest taxonomic level via dissecting and compound microscopes. One out of every ten samples was re-identified by a taxonomist for QA/QC purposes. If 10% of the dominant organisms or 25% of the rare organisms were misidentified, then all 10 samples were re-identified.

2.5.6 Nekton Community

Nekton collected from seine (0.635 cm bar mesh) sampling were rinsed carefully, and retained fish and crustaceans were preserved in 10% formalin in seawater. Preserved organisms were sorted in the laboratory and identified to the lowest practical taxonomic level (usually species). Animals collected in trawls (1.9 cm bar mesh) were identified and counted in the field; one sample each day was returned to the laboratory for QA/QC. Procedures followed were 90% sorting accuracy and a 90% or 25% identification accuracy for abundant and rare taxa, respectively.

2.5.7 Oyster Tissue Genomics

For genomic analysis, 25 oysters were shucked, weighed, and dissected; samples of the gill and hepatopancreas tissues were excised, preserved in buffer (RNA *later*®, Ambion), and kept on ice. Oyster tissue samples were hard-frozen in liquid nitrogen as soon as possible and stored until genomic analysis. Total RNA was isolated from RNA*later*® (Ambion Inc, Austin, TX) stabilized oyster tissues using the RNeasy® Mini kits (Qiagen, Valencia, CA) with an on-column DNase (Qiagen, Valencia, CA) treatment. Extracted RNA was quantified by absorbance at 260nm using a NanoDrop® ND-1000 Spectrophotometer (NanoDrop Technologies, Wilmington, DE). One microgram of total RNA was used to produce Cy3-labeled aminoallyl RNA (Cy3-aRNA) probe using the Amino Allyl MessageAmp™ II aRNA Amplification Kit (Ambion Inc, Austin, TX), according to the manufacturer's instructions. Ten micrograms of the subsequently produced Cy3-aRNA was diluted (1:3) in hybridization buffer (50% formamide, 2.4% SDS, 4X SSPE, 2.5X Denhardt's solution, and 2 µl of blocking solution [1 µg Cot-1 DNA and 1 µg poly dA]). The probe was then boiled for 1 min and incubated in the dark for 1 h at 50 °C prior to hybridization.

The oyster cDNA microarray (Jenny et al. 2007) slides were prewashed with 0.2% SDS for 2 min, just-boiled Milli-Q water for 2 min, rinsed in 70% ethanol for 2 min, and then dried. Slides were pre-hybridized in a hybridization oven with a pre-hybridization buffer (33.3% formamide, 1.6% SDS, 2.X SSPE, 1.6X Denhardt's solution, and 0.1 µM salmon sperm DNA) in the dark for 1 h at 50 °C. Slides were hybridized with Cy-3-aRNA in the dark for 16 h at 50 °C. After the hybridization, slides were rinsed in 2X SSC, 0.1% SDS and soaked in 0.2X SSC, 0.1% SDS for 15 min in the dark at room temperature, followed by a rinse in 0.2X SSC, soaking in 0.2X SSC for 15 min, 0.1X SSC for 15 min and finally Milli-Q water for 5 min in the dark in order to remove carryover SDS. The microarrays were then dried and scanned with ScanArray™ Express and SpotArray software at 70 V photomultiplier tube (PMT) gain and analyzed with QuantArray software (Perkin Elmer, Boston, MA).

2.6 Data Summary and Statistical Analyses

Resulting data (e.g., land cover, nutrient concentrations, infaunal abundances, pathogen abundances) were entered into a relational database for storage. This database, a PostgreSQL database, was accessed by individual users via Microsoft Access frontends (White et al. 2008). Through the frontends, individual users could query the database for relevant summaries and datasets for statistical analysis. Data are available upon request.

Tidal creek data from both 2005 and 2006 summer sampling periods have been summarized into one data set; no attempt was made to examine year-to-year variability. The main unit of statistical inference is the creek order, and the resulting data set comprises 43 observations (24 from intertidal systems, 19 from subtidal systems). In cases involving multiple measures per order, data were first averaged within each order to obtain one value for each indicator. Creek data were further summarized by averaging across the second and third orders to get one value representing the larger subtidally-dominated habitats. Lastly, for creeks that were sampled across both years (i.e., Guerin, James Island School, New Market, and Village; Table 1), data were averaged across years resulting in a single value for each intertidal system and each subtidal system for any particular parameter.

Statistical analyses were designed to address two research questions: 1) Do measured parameters vary across the sampled land use classes? and 2) Do measured parameters vary along the creek longitudinal gradient? To address these research questions, we employed Analysis of Variance (ANOVA), Analysis of Covariance (ANCOVA), and regression. The basic ANOVA model was a two-way, fixed factor model, with Land Use Class Type (salt marsh, forested, suburban, urban) and Creek Order (intertidal, subtidal) as the main effects. The interaction term was included in all models tested and excluded if nonsignificant ($p \geq 0.05$). Pairwise differences were examined by comparing least square means (using PDIFF in SAS). For the macrobenthic community, ANCOVAs were used to account for the effects of sediment type and salinity levels prior to testing for differences among land use classes and between orders. Lastly, individual response variables were regressed against impervious cover by creek order to document significant predictive relationships. Regressions were considered significant at $p < 0.05$. The regressions were performed with the forested, suburban, and urban creeks. The salt marsh creeks were excluded from this analysis because they had no developable upland. If data were found to be non-normal or heteroscedastic, basic transformations (log, square root, arcsine) were attempted. If those transformations did not improve the distribution of the data, data were rank transformed. Analyses were performed using SAS 9.1 (SAS Institute Inc., Cary, NC) or Systat 11 (Systat Software, Inc., San Jose, CA).

To summarize sediment contaminant data, the values less than the method detection limits (MDL) were set to 0 before analysis. Total PAH, total PCB, and total PBDE concentration were determined by summing the 25, 79, and 13 individual analytes, respectively, measured for each group of contaminants (Appendix A). The concentrations of trace metals, PAHs, PCBs, and pesticides from the study creeks were compared to sediment quality guidelines. Long et al. (1995) developed sediment quality guidelines by summarizing the published literature on the effects of a suite of sediment contaminants on a wide range of marine biota and derived two threshold values, an effects range-low (ERL) and an effects range-median (ERM) for individual analytes. An ERL was defined as the sediment concentration of a given contaminant where 10% of all published studies have reported an adverse effect, and an ERM was defined as the sediment concentration where 50% of all published studies have reported an adverse effect. Values below the ERL would rarely be expected to be associated with measurable biological effects; values between the ERL and ERM represent a range in which there are possible biological effects for a wide range of organisms. Values above the ERM represent a range above which there are probable biological effects.

The mean ERM quotient (mERMQ) was calculated for all contaminants (Total mERMQ) and for each major class of contaminant (i.e., trace metals, PAHs, PCBs, and pesticides) using the 24 analytes outlined by Long et al. (1995, Appendix B). Calculations were made by (1) dividing the concentration of each analyte or analyte group by the published ERM value, (2) summing the ratios of analytes within each contaminant class (e.g., trace metals), and (3) dividing by the number of contaminants in that class. In addition, total mERMQ values, which encompassed all four contaminant classes, were calculated for each sample. These values were calculated in the same fashion except that analytes were not combined within contaminant classes, instead the ratios of all 24 analytes were summed, and the total was divided by 24 (Long et al. 1998). The use of these quotients provides a way to compare potential cumulative effects of contaminants after weighting them on a toxicological basis. In analyses, mERMQs were used.

3. Results

3.1 Stressors

The 19 tidal creek systems surveyed for this study consisted of one to five watersheds depending upon the number of intertidal and subtidal creek segments sampled (Table 2). In addition, each system had one or two land use designations depending upon the land surrounding the sampled creeks. Watersheds for creeks draining salt marsh were classified as marsh, and watersheds for creeks draining upland areas were classified as either forested, suburban, or urban.

Intertidal watersheds ranged in size from 28 ha (Parrot, marsh) to greater than 2400 ha (Burnett, urban and Okatee, suburban; Figure 3) and impervious cover ranged from 0% (salt marsh watersheds) up to about 70% (New Market, urban). Subtidal watersheds included the intertidal area and ranged in size from 59 ha (Orangegrove, marsh) to 5501 ha (Okatee, suburban) and impervious cover in these watersheds ranged from 0 to 47.7% (Shem, urban).

Land cover in each watershed was determined using NLCD categories of developed-high, developed-low, agricultural, bare land, forested, forested wetland, marsh, or water. The percent composition of land cover shows a progressive change along the salt marsh-forested-suburban-urban gradient (Figure 4). Creek watersheds classified as salt marsh were primarily marsh land cover, while the forested systems were primarily forested land cover. For suburban and urban watersheds, there was a general increase in urban land cover (both developed-low and developed-high) and a concomitant decrease in forested land cover.

Table 2. Creek system, land use class, watershed area, and watershed impervious cover for each creek segment. Subtidal watershed area includes the related intertidal area.

Creek System	Land Use Class	Watersheds	Order	Creek Segment	Area (ha)	Impervious Cover (%)
North Carolina						
Hewlitts	Suburban	Hewlitts-S	1	Intertidal	614	40.9
		Hewlitts-N	1	Intertidal	459	34.5
		Hewlitts	2	Subtidal	2782	33.4
Masonboro	Marsh	NOC-Masonboro	1	Intertidal	29	2.9
Whiskey Creek	Suburban	Whiskey	1	Intertidal	482	34.7
		Whiskey	2	Subtidal	712	32.4
South Carolina						
Albergottie	Suburban	Albergottie	1	Intertidal	558	8.1
		Albergottie	2 & 3	Subtidal	2096	23.9
Bulls	Urban	Bulls	1	Intertidal	369	40.5
		Bulls	2 & 3	Subtidal	510	38.1
Guerin	Marsh	Guerin	1	Intertidal	25	0.0
		Guerin	2	Subtidal	342	0.0
	Forested	Guerin	1	Intertidal	219	3.0
		Guerin	2 & 3	Subtidal	3427	3.0
James Island	Suburban	James Island-N	1	Intertidal	296	30.0
		James Island-N	2	Subtidal	773	29.1
	Suburban	James Island-S	1	Intertidal	144	41.3
		James Island-S	2 & 3	Subtidal	1820	29.5
Murrells Inlet	Urban	Murrells	2 & 3	Subtidal	1297	40.3
New Market	Urban	New Market	1	Intertidal	199	70.4
North Inlet	Marsh	NIWB-Clambank	1	Intertidal	55	0.0
		NIWB-Clambank	2	Subtidal	102	0.0
	Forested	NIWB-Crabhaul	1	Intertidal	184	2.9
		NIWB-Town	2 & 3	Subtidal	1860	2.9
Orangegrove	Marsh	Orangegrove	1	Intertidal	18	0.0
		Orangegrove	2	Subtidal	59	0.0
	Suburban	Orangegrove	1	Intertidal	61	39.2
		Orangegrove	2 & 3	Subtidal	322	37.3
Parrot	Marsh	Parrot	1	Intertidal	28	0.0
	Suburban	Parrot	1	Intertidal	62	21.2
		Parrot	2 & 3	Subtidal	501	17.7
Okatee	Suburban	Okatee	1	Intertidal	2415	17.9
		Okatee	2 & 3	Subtidal	5501	13.3
Shem	Urban	Shem	1	Intertidal	456	49.4
		Shem	2	Subtidal	1269	47.7
Village	Forested	Village	1	Intertidal	630	3.6
		Village	2 & 3	Subtidal	2016	4.0
Georgia						
Burnett	Urban	Burnett	1	Intertidal	2425	11.2
		Burnett	2	Subtidal	2589	11.8
Duplin	Forested	SAP-Duplin	1	Intertidal	385	3.0
		SAP-Duplin	2 & 3	Subtidal	1480	3.0
Oakdale	Forested	SAP-Oakdale	1	Intertidal	286	3.1
Postell	Urban	Postell	1	Intertidal	218	39.8

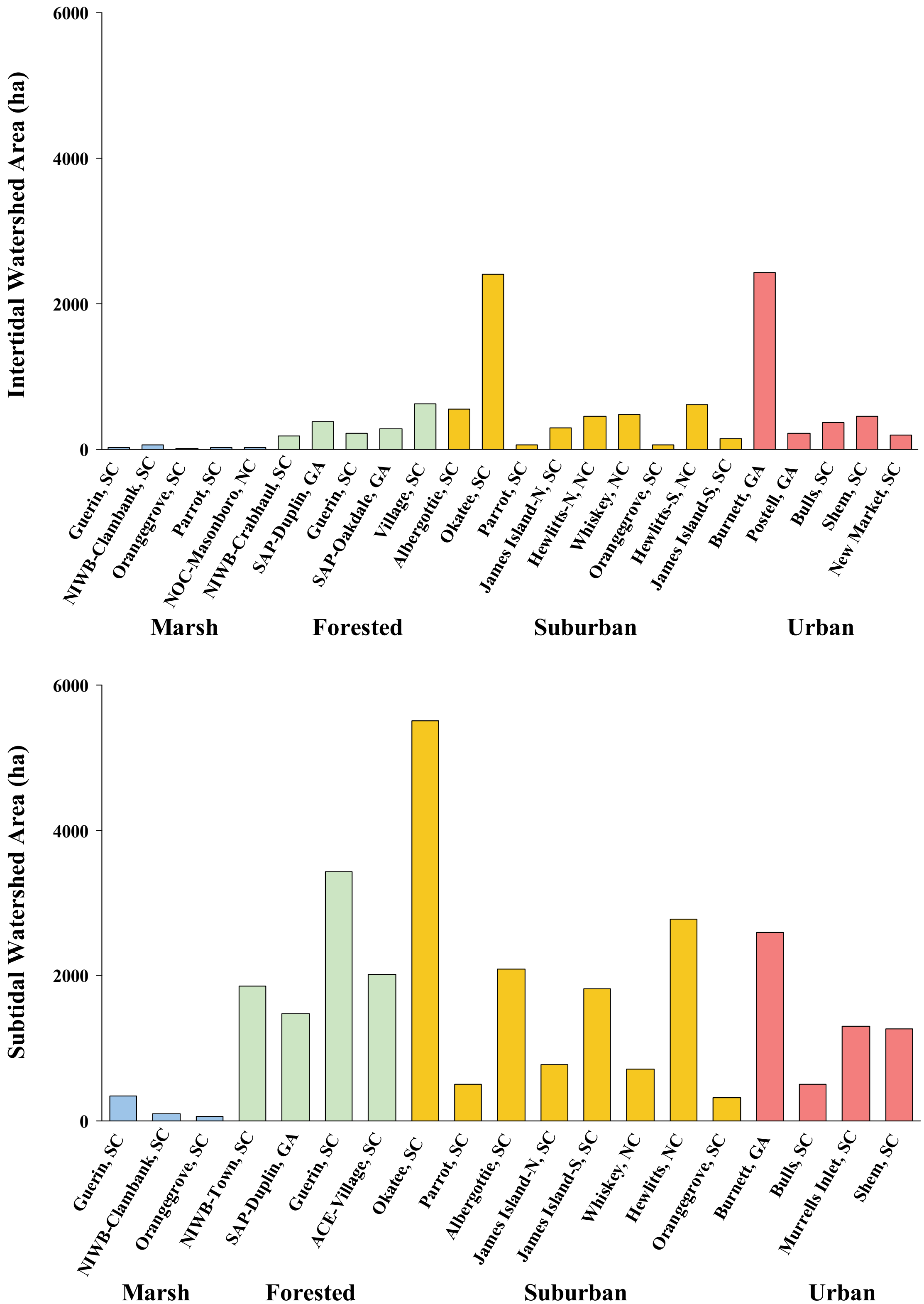

Figure 3. Area of intertidal (upper) and subtidal (lower) watersheds examined within this study. The subtidal watersheds include the associated intertidal land areas. Land use class is marked by color.

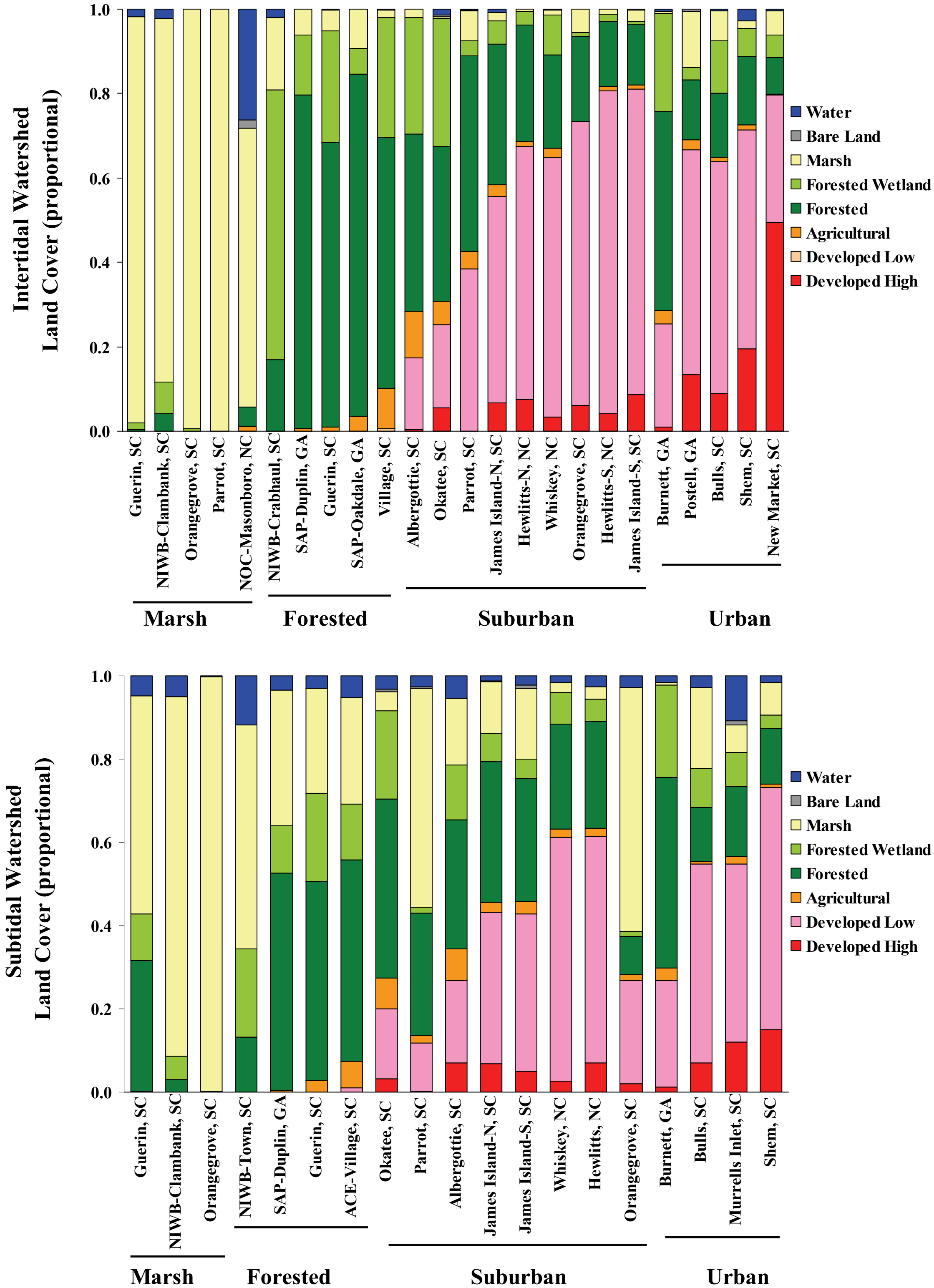

Figure 4. Proportional land cover categories (from NLCD 2001) within intertidal (upper) and subtidal (lower) watersheds examined within this study.

Land use classes were primarily determined using watershed impervious cover, and as expected, the amounts of impervious cover within each watershed class were significantly different from each other (ANOVA, $p<0.0001$). Across land use classes, there was no significant difference between the intertidal and subtidal impervious cover amounts.

Classifying watersheds based on impervious cover provides a useful framework for analyzing and interpreting study results. Similarly, impervious cover is a valuable direct indicator variable to describe the physical conditions and attributes of these study watersheds. For intertidal and subtidal creek watersheds, human population density (individuals ha^{-1}) is linearly related to the impervious cover (%) and explains 88% and 75%, respectively, of the total variability (Figure 5). Impervious cover is also strongly related to aspects of urbanization (i.e., the decrease in non-developed land cover classes and the increase in classes of developed land). For intertidal and subtidal creek watersheds, forested land cover was negatively related to impervious cover (Figure 6). Impervious cover appears to be a strong predictor of the overall watershed human population density and land cover attributes.

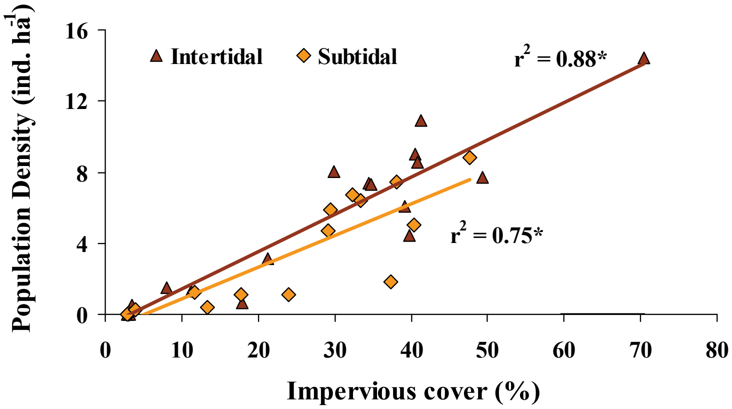

Figure 5. Relationship between population density and impervious cover within watersheds. Model r^2 is shown for each regression with asterisk (*) indicating significance ($p < 0.05$). Marsh watersheds are excluded owing to lack of impervious cover.

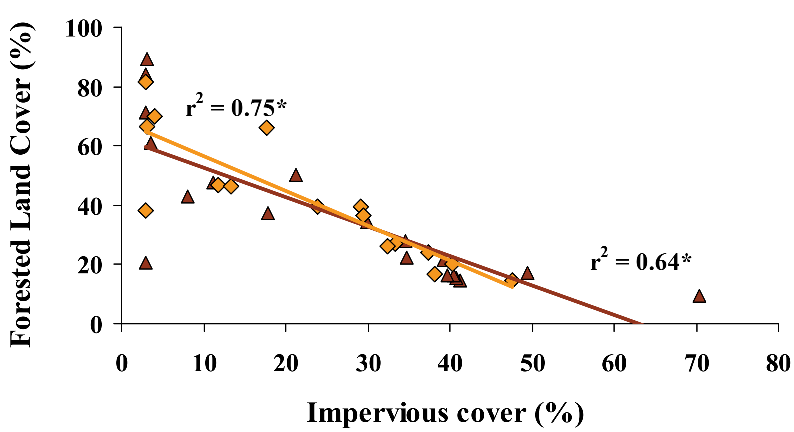

Figure 6. Relationship between forested land cover and impervious cover within watersheds. Impervious cover was regressed against forested land cover. Model r^2 is shown for each regression with asterisk (*) indicating significance ($p < 0.05$). Marsh watersheds are excluded owing to lack of impervious cover.

3.2 Exposures

3.2.1 Stormwater Runoff

Urbanization alters the hydrologic cycle or water budgets of watersheds. As land becomes covered with surfaces impervious to rain, water is redirected from groundwater recharge and evapotranspiration to stormwater runoff. This is a critical issue considering that nonpoint source (NPS) pollution is the leading cause of water quality degradation, and stormwater runoff accounts for most NPS pollution (USEPA 2002).

Stormwater runoff amounts for 4.5 in. rainfall were calculated for the 19 watersheds draining intertidal creek segments. In general, the runoff volume increased along the gradient of forested-suburban-urban land use classes (Figure 7). Average amounts from suburban and urban watersheds were 2.5 and 4 times greater, respectively, than from forested watersheds. Comparisons of medians among land use classes provide even larger differences, with runoff volume medians for suburban and urban watershed medians exceeding forested by factors of 6 and 8.4, respectively (Figure 8). More than 50% of runoff volumes from suburban and urban watersheds fell within a range of 28,000 to 45,000 m^3 km^{-2} while 50% of the forested watershed volumes were between 5,000 and 16,000 m^3 km^{-2}. Another way to consider stormwater runoff is in terms of the percent of the total rainfall that is converted to runoff for each watershed. Runoff percent generally increased with increasing impervious cover and ranged from 4.6% (SAP-Duplin and SAP-Oakdale, forested) to 63.3% (New Market, urban; Figure 9). A number of creeks did not follow this pattern due to naturally occurring variability, differences

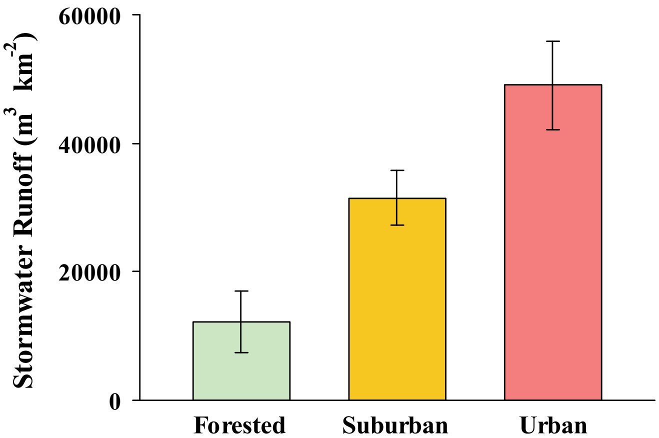

Figure 7. Stormwater runoff volume for intertidal watersheds grouped by land use class. Bars represent average runoff volumes. Error bars are ± 1 standard error. Volumes are standardized by watershed area and based on rainfall of 4.5 in.

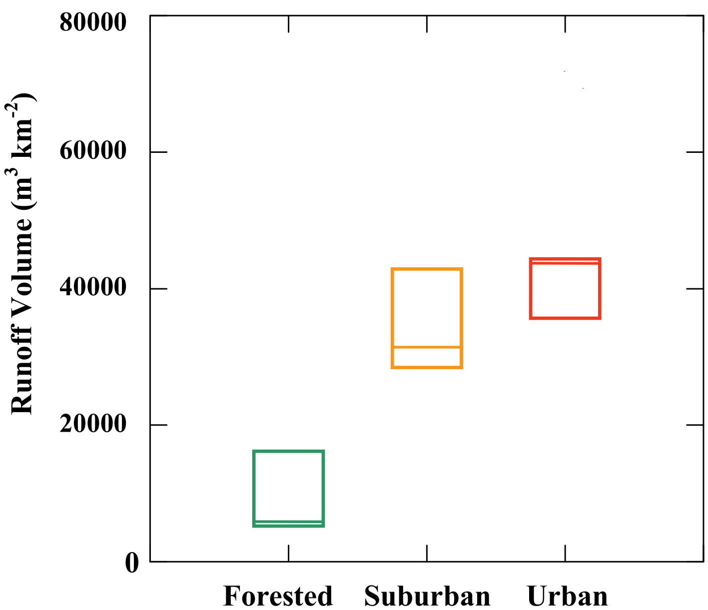

Figure 8. Stormwater runoff volume for intertidal watersheds grouped by land use class. The center horizontal line marks the median, and the length of each box shows the range within which the central 50% of the values fall. Calculated volumes are standardized by watershed area and based on precipitation of 4.5 inches.

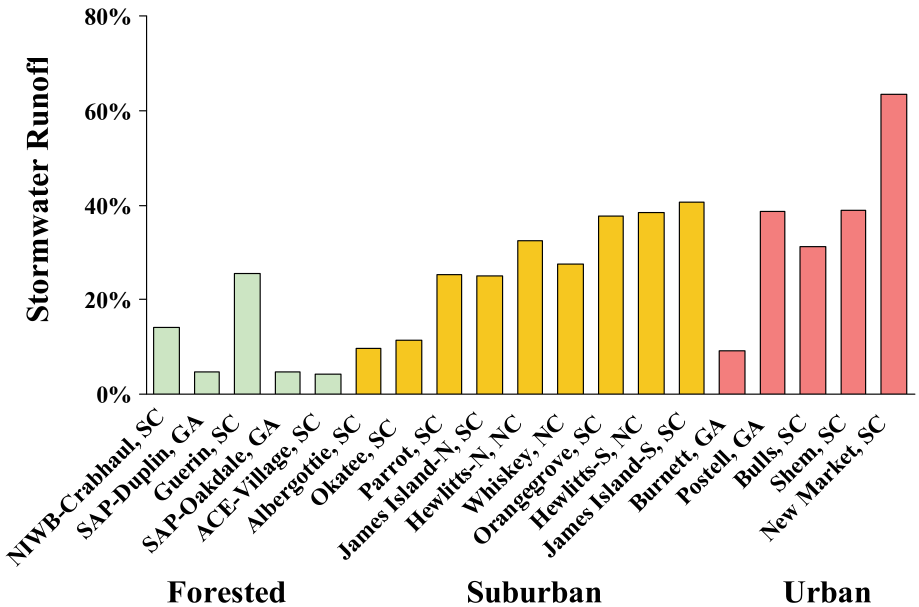

Figure 9. Rainfall percent that converts to stormwater runoff in intertidal watersheds. Land use class is marked by color. Percentages are standardized by watershed area and based on rainfall of 4.5 in.

in hydrologic soil groups, and land use considerations. Guerin and NIWB-Crabhaul (both forested systems) had runoff percentages greater than several watersheds classified as developed because both had a greater proportion of more impervious soils. Burnett (urban) had a runoff percentage (9.1%) lower than all but 3 forested watersheds because the watershed was classified as urban although impervious cover is only 11.2% based on its industrial history (creosote wood preserving).

Runoff volume calculated by the modified NRCS-CN method was significantly associated with percent of impervious cover in the respective watersheds (Figure 10). This close relationship suggests that relative differences in runoff volumes for watersheds can be predicted based upon percent of impervious cover. Greatest variation occurs in the forested watersheds with low impervious cover levels.

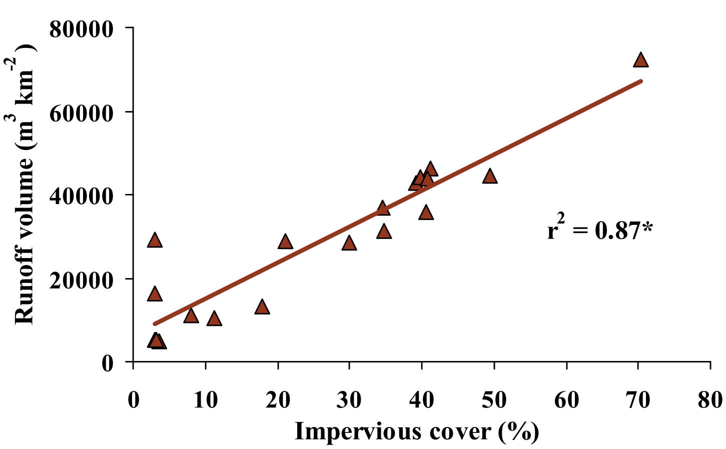

Figure 10. Relationship between stormwater runoff volume and impervious cover for intertidal watersheds. Calculated volumes are standardized by watershed area and based on precipitation of 4.5 inches. Model r^2 is shown with asterisk (*) indicating significance ($p < 0.05$).

3.2.2 Basic Water Quality

Basic water quality metrics were sampled including temperature, pH, salinity, and dissolved oxygen (DO). Temperature affects the rate of chemical reactions, and organisms have differing physiological tolerances to temperature. Extreme values of pH can occur when acids or caustic materials enter creek waters indicating the presence of pollutants. Salinity levels influence the distribution and diversity of many invertebrates and fish species and can be stressful to many organisms when large variations occur over short time periods. Low DO levels can limit distribution or survival of most biota, especially if conditions persist for extended periods (Van Dolah et al. 2004).

Averages for creek water temperature values were observed from 25.0° C (NIWB-Crabhaul, subtidal, forested) to 32.6° C (Albergottie, intertidal, suburban). Average pH values were observed from 6.52 (Okatee, intertidal, suburban) to 7.97 (NOC-Masonboro, intertidal, marsh, Figure 11). Average salinity values were observed from 0.51 ppt (Bulls, intertidal, urban) to 35.1 ppt (NIWB-Clambank, intertidal, marsh). Average DO values were observed to from 2.68 mg L^{-1} (James Island-S, intertidal, suburban) to 6.89 mg L^{-1} (Hewlitts-N, subtidal, suburban). Temperature ranges (maximum minus minimum) were observed from 1.24° C (Orangegrove, subtidal, suburban) to 13.18° C (Orangegrove, intertidal, suburban). pH ranges were observed from 0.18 (Bulls, intertidal, urban) to 1.53 (Hewlitts-N, intertidal, suburban). Salinity ranges were observed from 0.8 ppt (SAP-Duplin, subtidal, forested) to 31.3 ppt (Hewlitts-S, intertidal, suburban). DO ranges were observed from 2.68 mg L^{-1} (James Island-S, intertidal, suburban) to 15.02 mg L^{-1} (Hewlitts-N, intertidal, suburban).

In general, basic water quality averages and ranges for NOC-Masonboro, SAP-Duplin, and SAP-Oakdale were similar to the other creeks in marsh and forested watersheds. Salinity levels were high in NOC-Masonboro, SAP-Duplin, and SAP-Oakdale, similar to the other creeks with a high oceanic influence (Figure 11). Salinity ranges were also high in the Georgia and North Carolina intertidal creeks draining developed watersheds.

Average water quality levels were not significantly affected by land use class or longitudinal gradient. Land use had a significant effect on salinity range with the urban and suburban creeks having significantly larger ranges than the marsh and forested creeks (Table 3). In addition, salinity range as well as temperature range and DO range responded to the longitudinal spatial gradient sampled with the intertidal creeks having significantly larger ranges compared to the subtidal creeks.

Intertidal salinity ranges showed a significant relationship with the amount of impervious cover in the watersheds (Figure 12). This pattern is similar to previous research in SC intertidal creeks and has been attributed to flashier runoff from developed watersheds due to increased impervious cover (Lerberg et al. 2000, Holland et al. 2004). None of the other basic water quality metrics had statistically significant regressions.

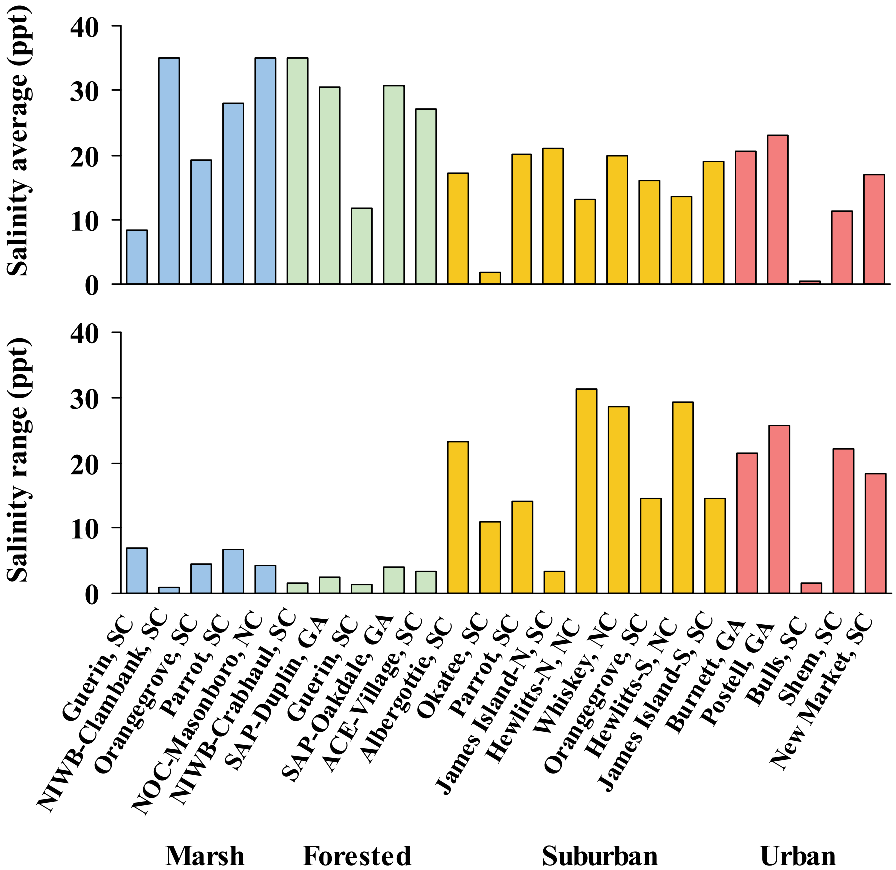

Figure 11. Salinity averages and ranges (maximum minus minimum) for intertidal creeks. Land use class is marked by color. Ranges were calculated from semi-continuous (every 15 minutes) sampling over a 24-hour period.

3.2.3 Nutrients and Phytoplankton
Sampled nutrients and phytoplankton metrics comprised ammonium (NH_4^+), nitrate plus nitrite ($NO_{2/3}$), total dissolved nitrogen (TDN), total nitrogen (TN), orthophosphate (PO_4^{3-}), total dissolved phosphate (TDP), total phosphate (TP), silicate (DSi), chlorophyll-*a* (Chl-*a*), and phaeophytin (Phaeo). Nutrient concentrations can be related to anthropogenic influences and adverse impacts on creek biota. In particular, stormwater runoff from developed land carries $NO_{2/3}$ and PO_4^{3-} from fertilizer applications into creek waters. High $NO_{2/3}$ concentrations indicate possible creek eutrophication which can lead to large algal blooms resulting in low and fluctuating dissolved oxygen levels that can adversely impact creek biota. Chl-*a* is a measure of phytoplankton biomass, and high concentrations can indicate the presence of excessive algal blooms from eutrophication.

Concentration levels for nutrients ranged from one to three orders of magnitude among the creeks sampled. TN ranged from 0.33 mg L^{-1} (Hewlitts, subtidal, suburban) to 4.65 mg L^{-1} (Orangegrove, intertidal, marsh). $NO_{2/3}$ ranged from 0.003 mg L^{-1} (SAP-Duplin, intertidal, forested) to 0.397 (Parrot, intertidal, suburban). Chl-*a* ranged from 0.57 µg L^{-1} (Orangegrove, intertidal, suburban) to 174.15 µg L^{-1} (Orangegrove, intertidal, marsh). From an individual creek standpoint, Orangegrove (intertidal, marsh) had far higher levels of phosphorous (PO_4^{3-}, TDP, TP) than any other creek. This could reflect the historic land use in the area of phosphate mining. Phaeophytin levels also were far higher in the Orangegrove marsh watershed creek than in other creeks (Figure 13).

Nutrient and phytoplankton concentrations for NOC-Masonboro (intertidal, marsh) were consistently low, similar to the other marsh watershed creeks that had no forests adjacent to the watershed (i.e., NIWB-Clambank, Parrot). Concentrations for SAP-Duplin and SAP-Oakdale creeks were similar to or higher than the other forested watershed creeks. SAP-Duplin (intertidal, forested) had the highest DSi concentrations (7.87 mg L^{-1}) of all creeks, developed and undeveloped. NOC-Masonboro (intertidal, marsh) had the lowest DSI concentrations (0.41 mg L^{-1}). Concentrations of the three phosphorous species were consistently low in all North Carolina creeks (marsh and suburban).

Table 3. Results of 2-way ANOVA on averages and selected ranges of water quality indicator variables sampled in summer, 2005 and 2006. Land use class factors are Marsh (M), Forested (F), Suburban (S), Urban (U). Order factors are Intertidal (I) and Subtidal (S). Post hoc multiple comparisons were performed using least squared means; model factors (arranged from low to high) with different superscripts are statistically different.

Parameter	Model p-value	r^2	Land Use p-value	Order p-value	Inter-action	Land Use LS means	Order LS means
Basic water quality – range (sonde data)							
Temperature	<0.001	0.609	0.111	<0.001	ns		I^a S^b
Salinity	0.001	0.366	<0.05	<0.05	ns	F^a M^a S^b U^b	S^a I^b
Dissolved oxygen (DO)	<0.001	0.436	0.303	<0.001	ns		S^a I^b
pH	0.059	0.208	0.057	0.183	ns		
Basic water quality – average (sonde data)							
Temperature	0.144	0.161	0.085	0.599	ns		
Salinity	0.061	0.206	0.071	0.120	ns		
Dissolved oxygen (DO)	0.500	0.082	0.774	0.121	ns		
pH	0.450	0.090	0.855	0.092	ns		
Nutrients / Phytoplankton (grab samples)							
Ammonium (NH_4^+)	0.003	0.332	0.169	0.001	ns		S^a I^b
Nitrate+nitrite ($NO_{2/3}$)	0.001	0.399	0.003	0.329	ns	F^a M^a S^b U^b	
Total dissolved nitrogen (TDN)	0.057	0.210	0.663	0.006	ns		S^a I^b
Total nitrogen (TN)	<0.001	0.417	0.687	<0.001	ns		S^a I^b
Ortho-phosphate (PO_4^{3-})	0.062	0.205	0.084	0.092	ns		
Total dissolved phosphorous (TDP)	0.059	0.208	0.111	0.056	ns		
Total phosphorous (TP)	0.003	0.338	0.120	0.001	ns		S^a I^b
Silicate (DSi)	0.149	0.159	0.871	0.014	ns		S^a I^b
Chlorophyll-*a* (Chl-*a*)	0.341	0.234	0.858	0.002	ns		S^a I^b
Phaeophytin (Phaeo)	0.004	0.330	0.677	<0.001	ns		S^a I^b
Pathogen Indicators (grab samples)							
Enterococcus (ENT)	0.001	0.367	0.015	0.002	ns	M^a $F^{a,b}$ S^b U^b	S^a I^b
Fecal coliform (FC)	<0.001	0.616	<0.001	<0.001	ns	M^a F^a S^b U^b	S^a I^b
F- coliphage (F-)	<0.001	0.493	<0.001	0.001	ns	M^a F^a S^b U^b	S^a I^b
F+ coliphage (F+)	0.007	0.301	0.004	0.300	ns	M^a F^a S^a U^b	

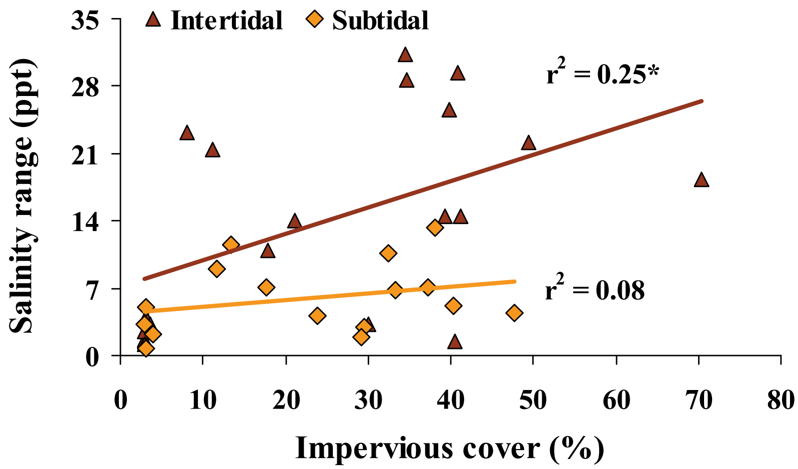

Figure 12. Relationship between salinity range and impervious cover for the study watersheds. Model r^2 is shown for each regression with asterisk (*) indicating significance ($p < 0.05$).

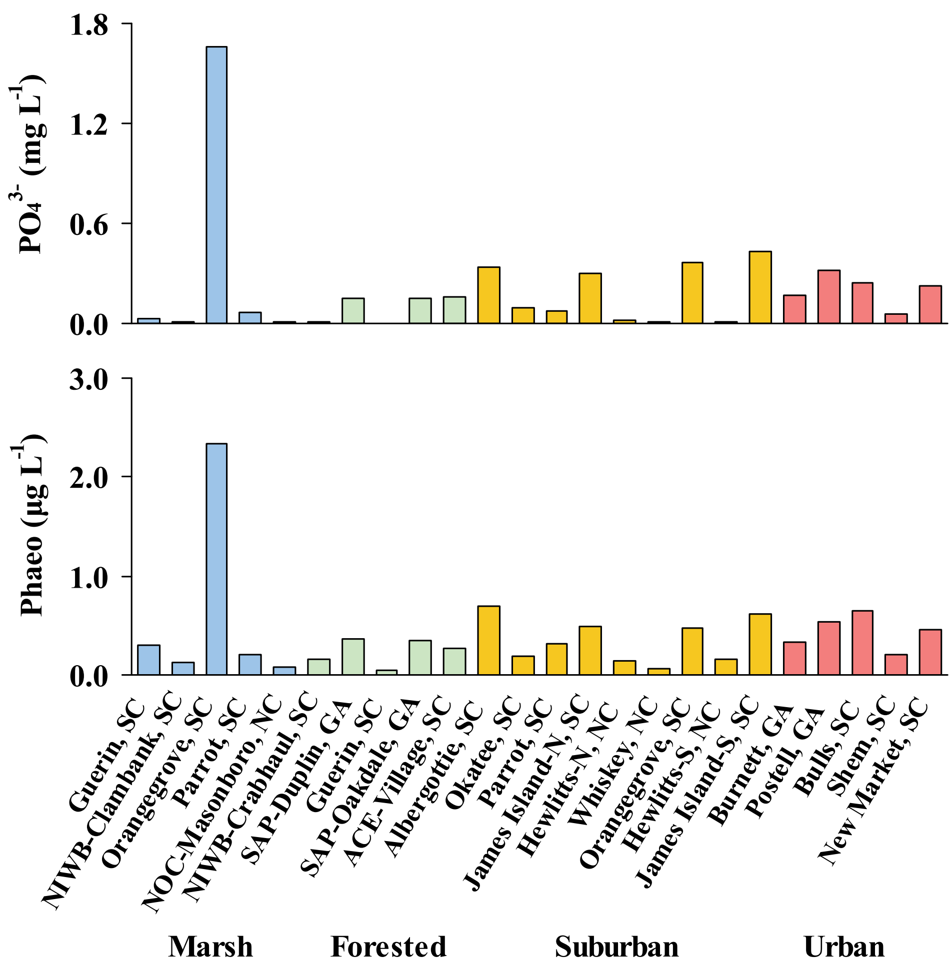

Figure 13. Intertidal nutrient and phytoplankton levels for individual creeks. Land use class marked by color. Bars represent average concentrations. PO_4^{3-}=orthophosphate, Phaeo=phaeophytin.

The type of land use surrounding the tidal creeks had little effect on most nutrient and phytoplankton concentrations, whereas the longitudinal spatial gradient sampled showed a more consistent significant effect (Table 3). Land use class did have a significant effect on $NO_{2/3}$ levels, and concentrations for creeks in marsh and forested watershed classes were significantly lower than in developed watershed classes. The results for $NO_{2/3}$ was probably related to the combined increase of fertilizer use and stormwater runoff in developed watersheds. All nutrient concentrations, with the exception of $NO_{2/3}$, exhibited similar spatial gradients, with intertidal creeks having significantly (or trending toward significance, $p < 0.10$) higher levels than subtidal creeks (Figure 14).

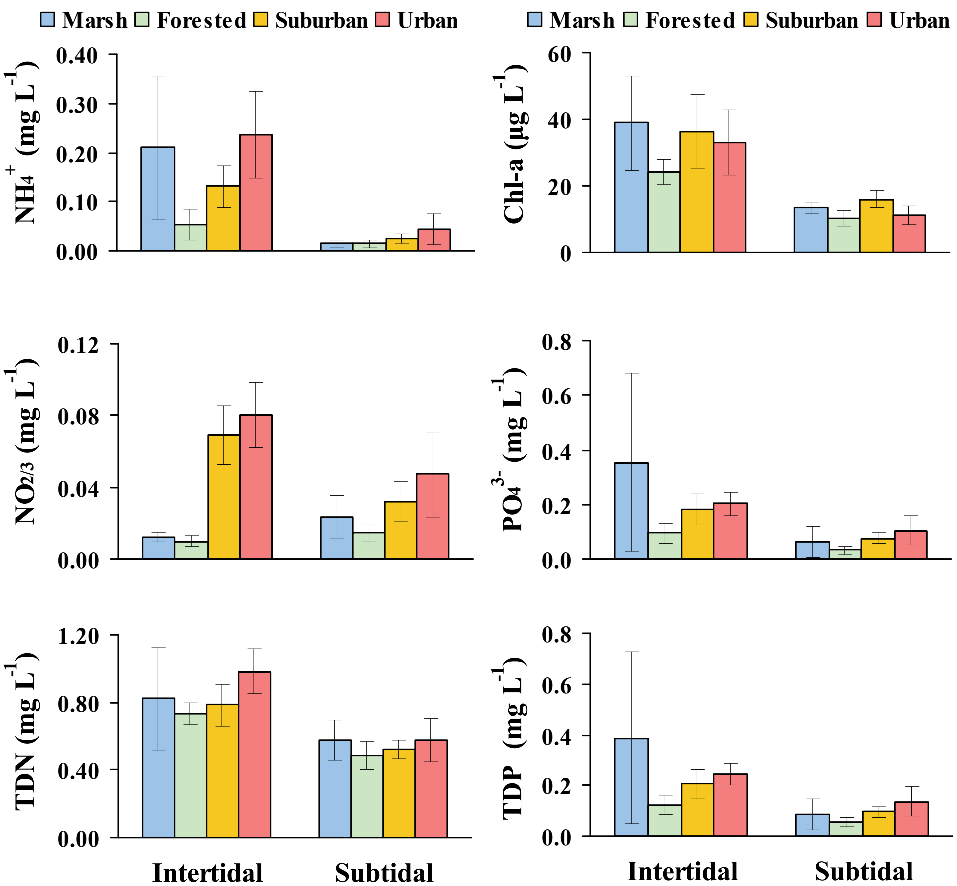

Figure 14. Nutrients and phytoplankton levels by land use class and longitudinal gradient. Bars represent average concentrations. Error bars are ± 1 standard error.

Intertidal concentrations of NH_4^+ and $NO_{2/3}$ and subtidal levels of $NO_{2/3}$ increased significantly with increasing levels of impervious cover in the watersheds (Figure 15). None of the other nutrients or phytoplankton measures were significantly related to impervious cover.

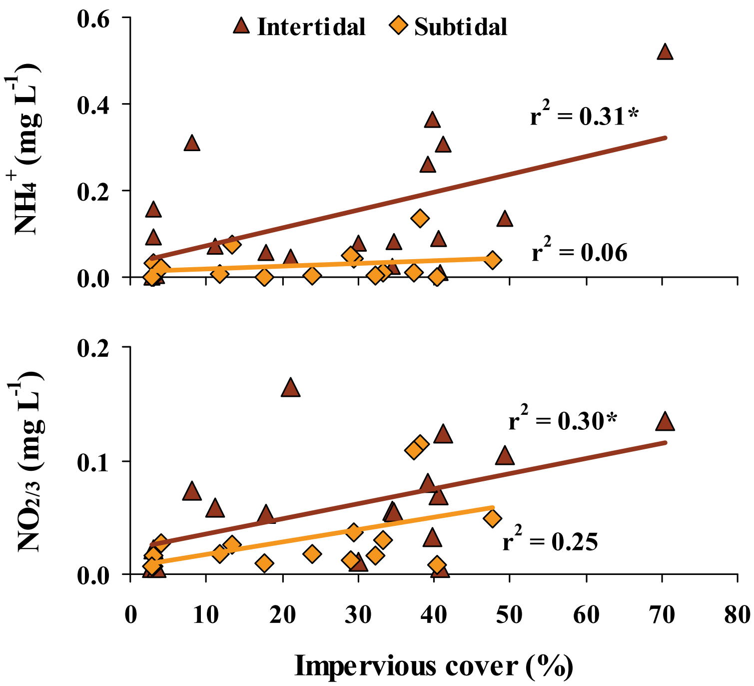

Figure 15. Relationship between nutrient concentrations and impervious cover for the study watersheds. Impervious cover was regressed against ammonium (NH_4^+; upper) and against nitrate plus nitrite ($NO_{2/3}$; lower). Model r^2 is shown for each regression with asterisk (*) indicating significance ($p < 0.05$).

Based on categorical guidelines developed for coastal waters by NOAA (Bricker et al. 1999), concentrations found in this study for TDN and TDP ranged from medium to high, and Chl-*a* from low to hypereutrophic. In general, intertidal creek concentrations were classified into the higher categories compared to the subtidal creeks (Figure 16). TDN concentrations for intertidal creeks draining suburban and urban watersheds were classed as medium for North Carolina study sites and either medium or high for South Carolina and Georgia study sites. TDN concentrations were classified as medium for all subtidal creeks and for intertidal creeks draining forested and marsh watersheds with one exception (Orangegrove, marsh, intertidal: high).

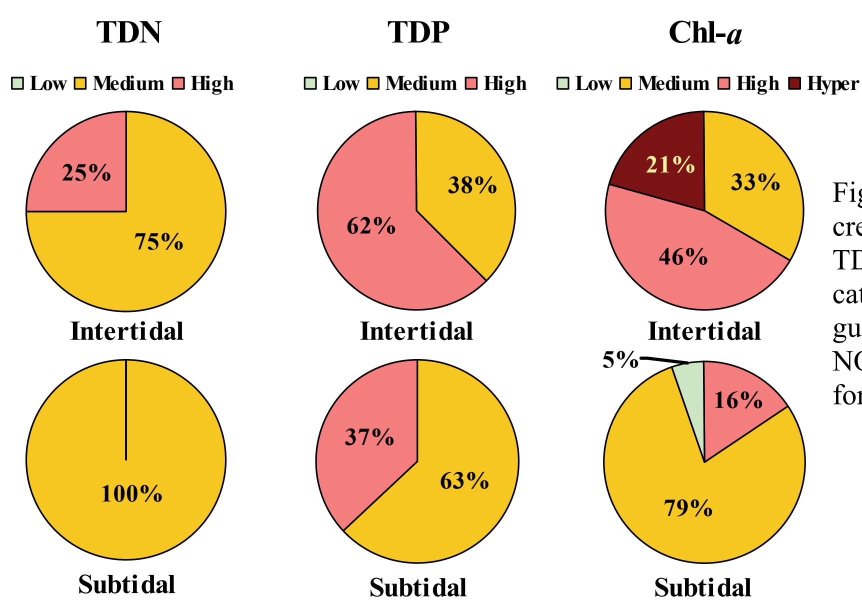

Figure 16. Percentage of creeks classified into TDN, TDP, and Chl-*a* qualitative categories based upon guidelines developed by NOAA (Bricker et al. 1999) for coastal waters.

TDP concentrations in South Carolina and Georgia generally were classified as high for intertidal creeks and as high and medium for subtidal creeks. All of the North Carolina sites were classified as medium. Intertidally the Chl-*a* concentrations ranged from medium to hypereutrophic across all of the land use classes. Subtidal creek concentrations were generally classified as medium.

3.2.4 Pathogens

Fecal coliform (FC) and Enterococcus (ENT) are bacteria that have been used extensively as indicators of fecal pollution and enteric pathogens. However, they may be inadequate indicators for all pathogens that are associated with fecal pollution, particularly enteric viruses. Coliphages, viruses that infect *Escherichia coli*, are being investigated to determine if they are a more appropriate indicator for water borne pathogens. F+ and F- are the two main types of coliphages.

FC concentrations ranged from <1 to 91,000 colony forming units (CFU) 100 ml^{-1} while ENT concentrations ranged from 3 to 21,000 CFU 100 ml^{-1}. Levels of measured viruses tended to be lower than those of the bacteria, ranging from 0 to 450 plaque forming units (PFU) 100 ml^{-1} and <1 to 1,200 PFU 100 ml^{-1} for F+ and F- coliphages, respectively.

In general, results for the National Estuarine Research Reserves' (NERR) creeks were similar to the other forested and marsh watershed creeks, with concentrations lower than developed creeks. However, concentrations of F+ and F- coliphages in the intertidal section of the SAP-Oakdale, (forested) were far higher than all other forested sites, and were similar to results from sampled creeks draining suburban watersheds (Figure 17). Of the creeks in the forested watersheds, intertidal SAP-Oakdale also had the highest levels of FC. ENT and FC concentrations for NOC-Masonboro (marsh) were low, similar to the other marsh watershed creeks that did not have forests adjacent to the watershed.

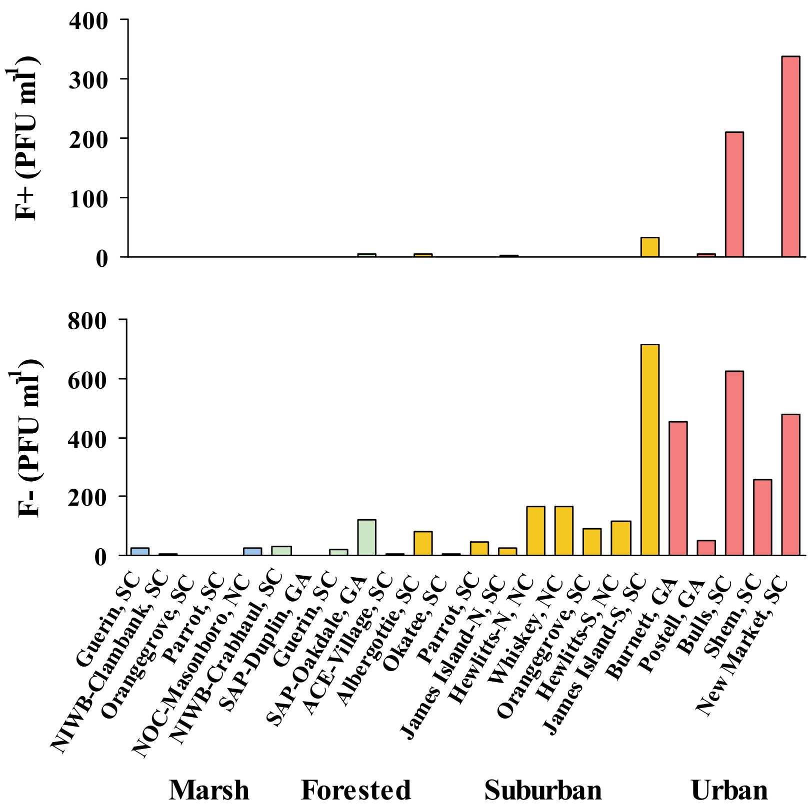

Figure 17. Intertidal F+ and F- coliphage levels for individual creeks. Land use class is marked by color. Bars represent average concentrations in plaque forming units (PFU) 100ml^{-1}.

The type of land use surrounding the tidal creeks and the spatial gradient sampled were found to affect both bacterial and viral pathogen indicator densities measured in this study.

Concentrations generally were lowest in salt marsh and forested watershed classes and highest in the urban watershed classes, a pattern that is most apparent in the intertidal creek sections. FC and F- coliphage levels differed significantly for the salt marsh and forested creeks compared to the developed (suburban and urban) watershed classes (Table 3). ENT concentrations were significantly higher in the suburban and urban classes compared to the salt marsh class, with the forested class similar to all of the other classes. F+ coliphage levels were <1 PFU 100 mL^{-1} in all salt marsh creeks, and the F+ concentrations in the marsh, forested and suburban classes were significantly lower than concentrations in the urban class. ENT, FC, and F- coliphage concentrations exhibited similar spatial gradients, with intertidal creeks having significantly higher densities of pathogen indicators than subtidal creeks. The F+ coliphage concentrations showed a similar trend but was not statistically significant (Figure 18, Table 3).

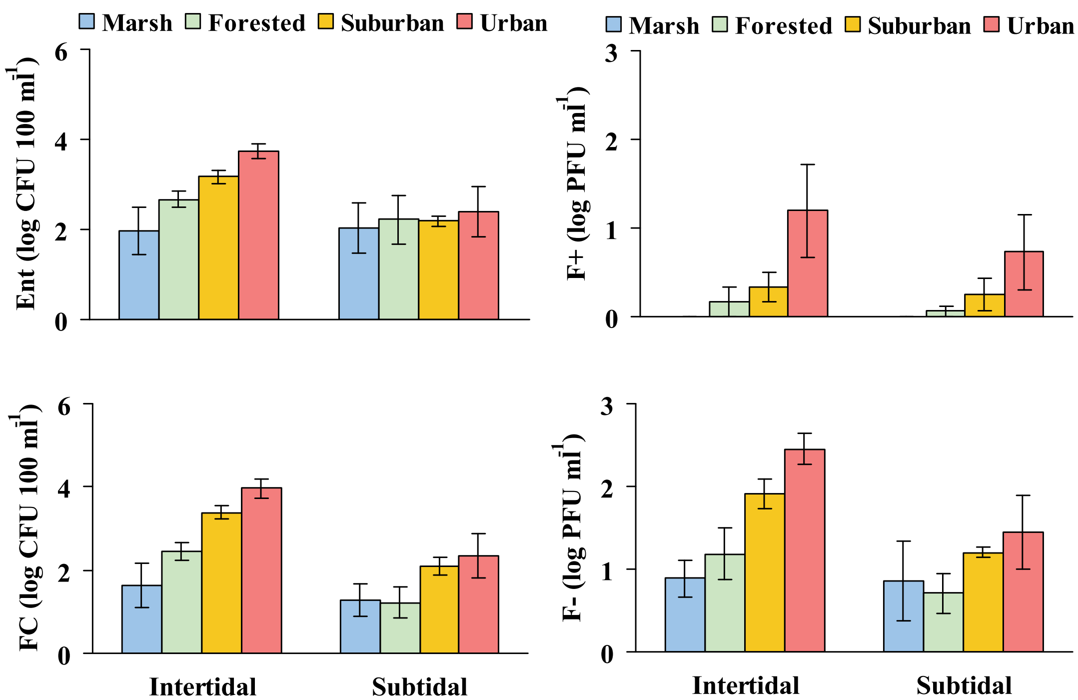

Figure 18. Bacterial and viral indicator levels by land use class and longitudinal gradient. Bars represent average concentrations and error bars are ± 1 standard error.
Ent = Enterococcus, FC = Fecal Coliform, F+ and F- = coliphages. Log transformation is x +1.

Concentrations of pathogen indicators increased with increasing levels of impervious cover in the watersheds (except ENT in subtidal areas), especially in the intertidal systems (Figure 19). In intertidal creeks, significant relationships were found between all of the pathogen indicators and the amount of impervious cover in the watershed. In the subtidal creeks, only the F+ coliphage showed a significant relationship with impervious cover in the watershed.

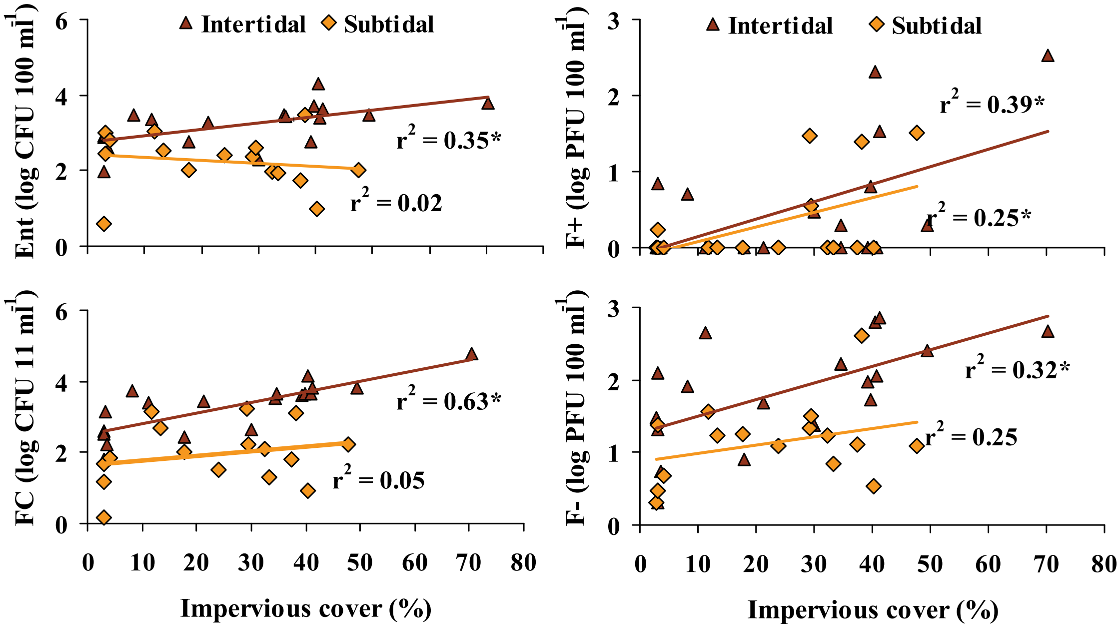

Figure 19. Relationship between pathogen indicators and impervious cover for the study watersheds. Model r^2 is shown for each regression with asterisk (*) indicating significance ($p < 0.05$). Marsh watersheds are excluded owing to lack of impervious cover. Ent = Enterococcus, FC = Fecal Coliform, F+ and F- = coliphages. Log transformation is x + 1.

3.2.5 Sediment Quality

Tidal creek sediments are often characterized as pluff mud which is a soft and mucky sediment rich in organic matter and high in clay content. These types of sediments are often repositories for chemical contaminants entering the tidal creeks from runoff and recreational use of the waters. Characterizing the sediments is an important aspect of interpreting sediment contaminant data. For example, trace metal contaminants bind to clay particles while organic contaminants bind to organic carbon (Beeftink et al. 1982, Boehm and Farrington 1984, Barrick and Prahl 1987). Therefore, higher clay and total organic carbon (TOC) levels will often be associated with higher contaminant loadings. Ammonious nitrogen (TAN) is also an important indicator of sediment quality and can cause toxicity to benthic organisms.

3.2.5.1 Sediment Composition

Sediment characteristics (e.g., % sand, % clay, % silt, and TOC) were evaluated using a 2-way ANOVA. The coarse grain composition (% sand) was not significantly related to either the surrounding land use or creek order. With respect to the finer particulates (% clay), sediment clay concentration ranged from 1.6% (NIWB-Clambank, subtidal, marsh) to 74.0% (Orangegrove, subtidal, marsh); these variable data did not show a significant relationship with either land use class or creek order (Tables 4, 5, 6). In comparison, the TOC ranged from 0.09% (Hewlitts, subtidal, suburban) to 10.7% (Hewlitts-S, intertidal, suburban) with significantly higher TOC concentrations in the intertidal creeks compared to the subtidal creeks (Tables 4, 5, 6). Sediment TOC was significantly related to creek order, with higher organic carbon concentrations in the intertidal creeks. There was no measurable effect of land use class.

Sediment TAN was significantly lower in forested creeks compared to suburban creeks (Table 6). Urban and marsh creeks were in between forested and suburban ones, but not significantly different from each other. Most creek types showed higher concentrations of sediment TAN in the intertidal habitats, although on average, there was no difference between intertidal and subtidal creeks with respect to sediment TAN.

Table 4. Characteristics and contaminant concentrations in tidal creek sediments for selected parameters. Total PAH is the sum of 23 analytes and total PCB is the sum of 79 analytes. Italicized numbers indicate concentrations that exceeded the ERL (as defined by Long et al. 1995) for that parameter.

		Sediment					Trace Metals					
Land Use	Creek	Clay (%)	TOC (%)	Total PAHs	Total PCBs	Total DDTs	As	Cr	Cu	Hg	Pb	Zn
Intertidal												
Salt Marsh	Guerin	47.40	5.10	114.7	0.40	0.00	***9.83***	61.0	33.5	0.081	23.0	65.2
	NOC-Masonboro	9.99	0.39	8.3	0.18	0.00	4.14	17.1	0.0	0.006	7.9	0.0
	NIWB-Clambank	60.77	4.31	112.5	0.00	0.00	***20.45***	59.6	16.1	0.048	22.1	68.6
	Orangegrove	64.68	8.26	1276.5	1.20	***1.71***	***16.50***	***92.6***	***35.7***	0.144	37.9	125.0
	Parrot	67.43	4.40	181.3	0.00	0.00	***23.60***	69.9	19.5	0.067	24.1	79.8
Forested	SAP-Duplin	40.94	6.33	15.4	1.02	0.00	***12.10***	33.4	9.6	0.037	13.2	42.9
	Guerin	51.87	4.15	45.4	0.19	0.00	6.41	55.8	8.2	0.058	20.6	43.5
	NIWB-Crabhaul	12.59	1.00	33.1	0.00	0.00	7.62	16.0	0.0	0.020	7.8	16.8
	SAP-Oakdale	18.54	1.66	45.1	0.64	0.00	5.91	22.0	0.0	0.022	10.5	31.4
	ACE-Village	4.32	0.18	9.7	0.34	0.00	0.96	14.0	0.0	0.004	7.8	0.0
Suburban	Albergottie	15.21	1.14	105.2	0.00	***2.03***	2.65	26.7	0.0	0.029	11.1	26.0
	Hewlitts-N	12.35	3.56	1749.4	1.48	***7.05***	4.66	24.9	23.6	0.047	18.2	44.3
	Hewlitts-S	18.67	10.68	555.9	0.36	***2.35***	4.50	23.1	0.0	0.039	13.0	41.0
	James Island-N	28.42	3.31	384.5	1.34	0.95	7.45	46.4	14.0	0.046	23.3	58.6
	James Island-S	22.16	2.93	1646.8	0.40	0.94	7.51	36.0	12.2	0.036	18.1	46.5
	Okatee	24.52	1.48	82.3	5.93	0.00	3.76	31.6	0.0	0.025	14.4	30.4
	Orangegrove	29.52	2.65	697.2	3.78	***2.24***	6.58	43.0	12.8	0.056	17.0	45.6
	Parrot	67.25	4.87	286.0	4.45	***3.54***	***19.60***	67.1	25.7	0.102	30.0	97.4
	Whiskey	20.57	3.43	482.8	2.10	***2.27***	7.02	27.6	15.8	0.044	13.6	57.0
Urban	Bulls	36.52	3.67	2000.9	0.79	1.18	7.79	56.0	19.2	0.081	28.2	84.0
	Burnett	15.82	2.28	2290.1	***60.87***	0.00	3.12	29.0	***61.5***	0.124	13.9	36.9
	New Market	35.38	6.69	***10237.7***	***107.26***	***23.28***	***14.18***	***93.9***	***51.9***	***0.366***	***92.9***	***197.0***
	Postell	5.33	0.30	223.2	2.62	0.87	5.67	23.3	0.0	0.022	14.4	37.2
	Shem	58.01	7.42	***8081.4***	1.30	***3.34***	***20.57***	73.9	***41.5***	0.133	32.9	144.0
Subtidal												
Salt Marsh	Guerin	3.47	0.35	18.4	0.74	0.00	4.34	21.8	0.0	0.011	9.2	23.1
	NIWB-Clambank	1.58	0.10	13.2	0.00	0.00	2.41	15.5	0.0	0.003	5.7	0.0
	Orangegrove	74.02	5.04	1095.9	0.70	1.36	***18.50***	***88.0***	***35.4***	0.137	36.0	122.5
Forested	SAP-Duplin	51.85	2.68	49.3	0.35	0.00	***15.40***	49.1	8.3	0.022	16.3	54.0
	Guerin	14.79	0.30	32.6	0.15	0.00	5.15	25.4	1.7	0.017	12.2	17.3
	NIWB-Town	1.79	0.12	12.4	0.00	0.00	2.48	11.3	0.0	0.003	4.8	0.0
	ACE-Village	11.69	0.33	104.0	5.29	***2.63***	2.69	19.0	1.6	0.009	7.4	8.5
Suburban	Albergottie	10.13	0.47	59.9	0.33	0.27	3.02	21.4	0.0	0.009	7.7	0.0
	Hewlitts	2.44	0.09	0.0	0.16	0.00	2.24	15.8	0.0	0.002	6.9	0.0
	James Island-N	6.07	0.35	292.4	0.16	0.46	2.75	15.4	0.0	0.012	8.4	21.1
	James Island-S	29.84	2.10	526.0	0.36	0.60	***8.54***	50.0	12.0	0.040	17.6	49.9
	Okatee	39.47	0.56	41.3	0.36	0.00	8.08	42.0	6.1	0.027	15.1	40.4
	Orangegrove	71.29	2.67	1956.8	0.74	***1.72***	***16.05***	77.5	31.0	0.125	33.3	107.0
	Parrot	22.48	2.09	117.9	0.00	0.00	6.83	33.2	3.7	0.022	13.2	30.7
	Whiskey	69.64	5.30	552.9	0.83	0.88	***15.80***	35.4	26.1	0.067	19.6	76.3
Urban	Bulls	37.78	2.98	941.1	1.00	0.93	***8.62***	53.3	17.9	0.077	25.1	74.2
	Burnett	21.59	2.77	39.9	2.71	0.00	0.56	6.3	0.0	0.009	3.9	0.0
	Murrells Inlet	6.06	0.28	198.4	0.00	0.00	4.28	30.3	0.0	0.006	11.5	10.9
	Shem	1.59	0.15	531.8	0.68	0.00	1.28	20.3	0.0	0.004	8.0	0.0

3.2.5.2 Sediment Contamination

Polycyclic aromatic hydrocarbons (PAHs), polychlorinated biphenyls (PCBs), pesticides, and polybrominated diphenyl ethers (PBDEs) were measured in this study. PAHs are a major component of lubricating oils and fossil fuels which are released into the environment when

Table 5. The number of ERL exceedences (as defined by Long et al. 1995) for each contaminant class at each sampling site. There are 13, 1, 2, and 8 individual parameters for the PAHs, PCBs, pesticides, and metals, respectively.

Land Use	Creek	PAH # >ERL	PCB # >ERL	Pesticides # >ERL	Trace Metals # >ERL
Intertidal					
Salt Marsh	Guerin	0	0	0	1
	NOC-Masonboro	0	0	0	0
	NIWB-Clambank	0	0	0	1
	Orangegrove	1	0	1	3
	Parrot	0	0	0	1
Forested	SAP-Duplin	0	0	0	1
	Guerin	0	0	0	0
	NIWB-Crabhaul	0	0	0	0
	SAP-Oakdale	0	0	0	0
	ACE-Village	0	0	0	0
Suburban	Albergottie	0	0	1	0
	Hewlitts-N	1	0	2	0
	Hewlitts-S	0	0	1	0
	James Island-N	0	0	0	0
	James Island-S	0	0	0	0
	Okatee	0	0	0	0
	Orangegrove	0	0	1	0
	Parrot	0	0	2	1
	Whiskey	0	0	1	0
Urban	Bulls	0	0	0	0
	Burnett	4	1	0	1
	New Market	13	1	2	6
	Postell	0	0	0	0
	Shem	9	0	2	2
Subtidal					
Salt Marsh	Guerin	0	0	0	0
	NIWB-Clambank	0	0	0	0
	Orangegrove	1	0	0	3
Forested	SAP-Duplin	0	0	0	1
	Guerin	0	0	0	0
	NIWB-Town	0	0	0	0
	ACE-Village	0	0	1	0
Suburban	Albergottie	0	0	0	0
	Hewlitts	0	0	0	0
	James Island-N	0	0	0	0
	James Island-S	0	0	0	1
	Okatee	0	0	0	0
	Orangegrove	2	0	1	1
	Parrot	0	0	0	0
	Whiskey	0	0	0	1
Urban	Bulls	1	0	0	1
	Burnett	0	0	0	0
	Murrells Inlet	0	0	0	0
	Shem	0	0	0	0

these products are spilled or combusted. Potential sources are runoff from highways and parking lots, street dust, fuel spills, marinas and recreational boating activities, and atmospheric fallout (reviewed by Weinstein 1996). In addition, natural sources of PAHs include forest fires. PCB production was banned in the 1970s, but PCBs have been reported to accumulate in estuarine environments (reviewed by Weinstein 1996). Most of the pesticides measured for this study are historical pesticides such as DDT, mirex, and chlordane (this group was banned in the 1970s). Current-use pesticides were not measured because they are water soluble and do not accumulate in sediments. Trace metals are naturally occurring elements and are influenced by natural weathering of basement rock (Williams et al. 1994). Several are anthropogenically enhanced from industrial and urban associated uses (e.g., lead, chromium, copper, cadmium, zinc, and mercury). Due to the large number of analytes measured, only the totals and a few of the anthropogenically introduced metals will be discussed (Tables 4, 5). PBDEs are contaminants of emerging concern that are used as flame retardants in furniture, plastics, and clothing.

Table 6. Results of 2-way ANOVAs performed on sediment characteristics and calculated Effects Range Median Quotient (ERMQ) values. ERMQ was calculated as the Total ERMQ, including PAHs, PCBs, Pesticides, and Trace metals; in addition, an ERMQ value was calculated for some of the subcomponents (PAHs, Metals, PCBs). See text for details. Land use factors are Marsh (M), Forested (F), Suburban (S), Urban (U). Order factors are Intertidal (I) and Subtidal (S). Post hoc multiple comparisons were done by comparing least squared means; model factors (arranged from low to high) with same superscripts are not statistically different.

Parameter	Model p-value	r^2	Land Use p-value	Order p-value	Inter-action	Land Use LS Means	Order LS means
Sediment TAN	0.061	0.21	0.052	0.207	ns	F^a U^{ab} M^{ab} S^b	
Sediment % Clay	0.291	0.12	0.776	0.051	ns		
Sediment TOC	0.014	0.27	0.533	0.001	ns		S^a I^b
Total ERMQ	0.009	0.29	0.055	0.009	ns	F^a M^{ab} S^b U^b	S^a I^b
PCB ERMQ	0.043	0.22	0.046	0.149	ns	F^a M^a S^a U^b	
Metal ERMQ	0.061	0.21	0.361	0.016	ns		S^a I^b
PAH ERMQ	0.001	0.49	0.007	0.040	0.016	F^a M^a S^a U^b	

Sediments collected in intertidal systems generally showed increasing concentrations of PAHs, PCBs and pesticides going from forested and salt marsh to suburban and urban creeks (Table 4). In particular, the intertidal portions of the most urbanized creeks including New Market, Shem, and Burnett creeks had a number of exceedences of the ERL for PAHs, PCBs and pesticides (Table 5). Burnett Creek is a current Superfund site that has a legacy of previous contamination from Brunswick Wood Preserving. A major spill of diesel oil and pentachlorophenol (PCP) into the creek occurred in 1989, and extensive contamination of the groundwater has occurred. Cu levels in the intertidal component of Burnett Creek were the highest of any sampled for this study, but Cu was undetectable in the subtidal component of the creek. This indicates that the Cu source was in the headwater area of this creek. With respect to the trace metals, though, creeks that exceeded the ERL level were more interspersed throughout all four land use classes. In particular, levels of As, a naturally occurring metal in these systems, were found to be the primary metal that exceeded the ERL. None of the sediment contaminant levels exceeded the ERM level for any parameter.

In subtidal systems, sediment contaminant levels were often below the established ERL (Table 5). Three creeks (SAP-Duplin, forested; ACE-Village, forested; Orangegrove, marsh) had elevated levels of Total DDT, suggesting a historical use of these pesticides in these areas. SAP-Duplin and ACE-Village creeks have past agricultural activities in their watersheds, while Orangegrove creek is located on the Ashley River, a river with a legacy of industrial contamination. With respect to the metals, at least one creek from each class exceeded the As ERL, and Orangegrove (marsh) had elevated Cr and Cu levels as well.

Sediment contaminant data suggest that the Sapelo Island NERR and North Carolina NERR sites were some of the least impacted of all the creeks studied. NOC-Masonboro had very low concentrations of PAHs (total PAH = 8.3 ng/g dry weight) and other analytes with no exceedences of the ERL (Tables 4, 5). Similarly, both SAP-Oakdale and SAP-Duplin creeks demonstrated very low levels of sediment contamination. None of the 24 analytes exceeded the

ERL level in SAP-Oakdale Creek. Arsenic consistently exceeded the ERL in both the upper portion and lower reaches of SAP-Duplin Creek. Arsenic often occurs naturally in these systems, and the arsenic ERL level was often exceeded in forested or otherwise relatively unimpacted salt marsh creeks.

The Total mERMQ value ranged from 0.0043 to 0.342 and 0.0041 to 0.085 in intertidal and subtidal creeks, respectively. When analyzed in a 2-way ANOVA, forested creeks had significantly lower Total mERMQ values than both suburban and urban creeks, and marsh creeks were between the forested and the suburban/urban creeks (Table 6, Figure 20). The generally

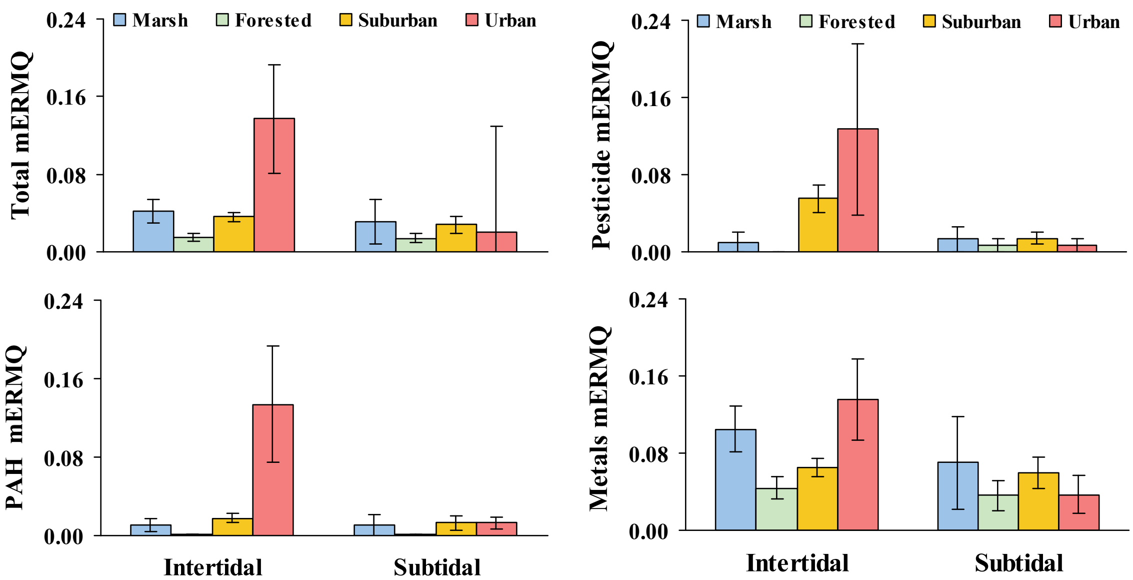

Figure 20. Characteristics and contaminant concentrations in tidal creek sediments. From top, the Total mean Effects Range Median Quotient (mERMQ), PAH mERMQ, Pesticide mERMQ, and Metals mERMQ for each land use and creek order class. Bars represent average concentrations. Error bars are ± 1 standard error.

low values in subtidal creeks in comparison to the intertidal creeks probably resulted in fewer statistical differences among land use classes. The intertidal creeks showed a clear trend of increasing values from forested to suburban to urban creeks. Salt marsh creeks were often similar to the suburban creeks which may be due to the high levels of TOC in these creeks. The Pesticide mERMQ and Metal mERMQ values were similar across land use classes, while the PAH mERMQ and PCB mERMQ values were significantly higher in the urban land use class compared to the other classes (Table 6, Figure 20). The intertidal creeks had significantly higher concentrations of overall contamination (Total mERMQ) as well as pesticide and metal contamination. The PAH mERMQ and PCB mERMQ values were found to be in similar concentrations down the length of the creek.

Hyland et al. (1999) provided evidence that mERMQ values greater than 0.020 represent a moderate risk of observing a degraded benthic community and values > 0.058 represent a high risk of observing a degraded benthic community. It should be noted that these estimates were derived from large, subtidal areas (orders 2 and 3 as well as open estuarine habitats) and may not accurately reflect the tolerances of the macrobenthic community in shallow, intertidal headwater creeks to sediment contaminants. In the intertidal creeks, 4 of the 5 salt marsh creeks exceeded the 0.020 threshold with one of those exceeding the 0.058 threshold; 2 of the 5 forested creeks exceeded 0.020; 8 of the 9 suburban creeks exceeded 0.020 with 2 of those exceeding 0.058; and 5 of the 5 urban creeks exceeded 0.020 with 4 of those exceeding 0.058. In the subtidal creeks, 1 of the 3 salt marsh creeks exceeded the 0.058 threshold; 1 (SAP-Duplin Creek) of the 4 forested creeks exceeded 0.020; 4 of the 8 suburban creeks exceeded 0.020 with 1 of those exceeding 0.058; and 1 of the 4 urban creeks exceeded 0.020.

Regression analysis demonstrated that mERMQ generally increased with increasing levels of impervious cover (Figure 21). Regressions of total mERMQ, metals mERMQ, and PAH mERMQ versus impervious cover were statistically significant in the intertidal creeks. In addition, PAH mERMQ was statistically significant in the subtidal creeks.

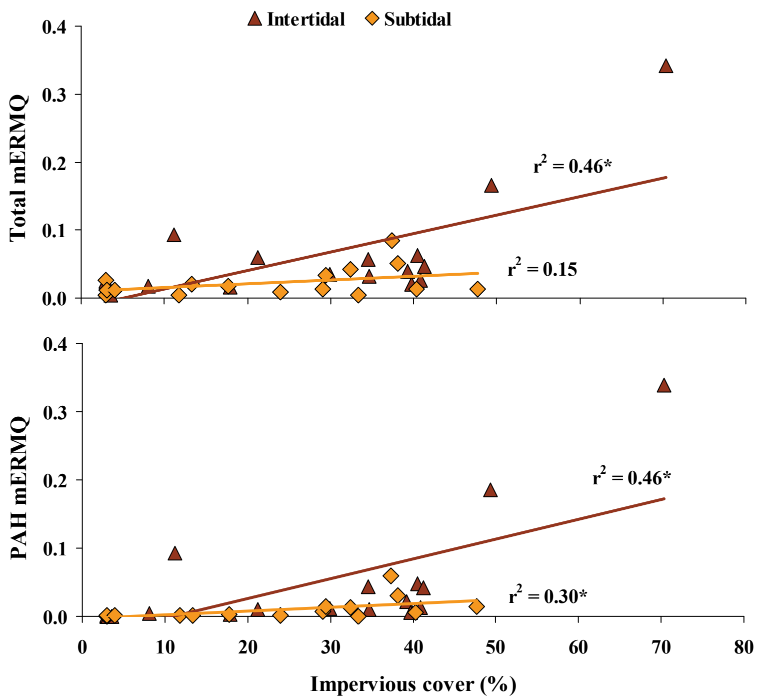

Figure 21. Relationship between contaminant quotients and impervious cover for the study watersheds. Impervious cover was regressed against Total mean Effects Range Median Quotient (mERMQ; upper) and against PAH mERMQ (lower). Model r^2 is shown for each regression with asterisk (*) indicating significance ($p < 0.05$).

PDBEs were only detected in the intertidal areas of the more developed creeks: James Island and Orangegrove (suburban) and Bulls, Shem, and New Market (urban, Figure 22). It appears, then, that these intertidal creeks are potentially valuable sentinels for detecting more of the emerging contaminants of concern as analytical methods are developed.

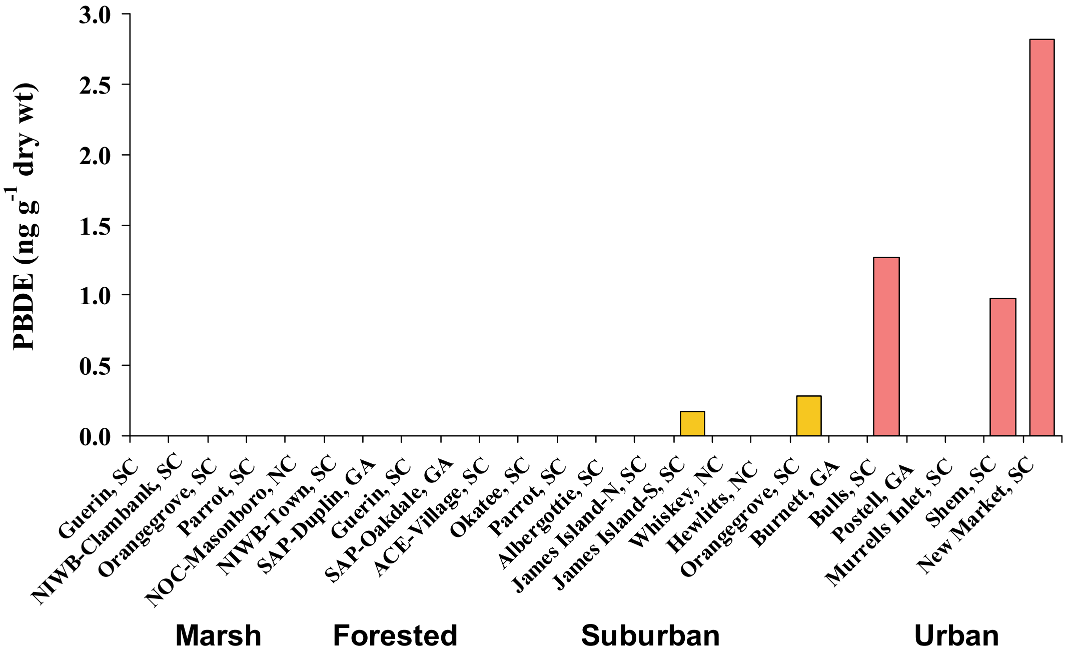

Figure 22. Concentration of polybrominated diphenyl ethers (PBDEs) in intertidal creek sediments. Land use class is marked by color. Bars represent measured concentrations.

3.3 Ecological Response

3.3.1 Macrobenthic Community

The macrobenthic community was sampled to examine how different levels of development affect abundances and distributions of these organisms. A total of 18,296 organisms representing 279 taxa were collected in 303 samples. Sampling methods varied between intertidally-dominated (intertidal) and subtidally-dominated (subtidal) components of the system, as did volume of sediment collected. Because samples collected in subtidal systems were ten times larger than those collected in intertidal systems, abundances were converted to density (expressed as ind. m^{-2}) prior to analysis.

Overall, oligochaetes composed 60% of the organisms sampled while polychaetes composed 35% of the organisms sampled. In the intertidal systems, oligochaetes composed 70% of the macrobenthic community while polychaetes comprised 28% of the community with the frequency of occurrence of each sample similar. Conversely, in the subtidal systems, polychaetes composed 65% of the community while oligochaetes composed only 10% with the frequency of occurrence higher for polychaetes than for oligochaetes (Table 7). Many other studies have found oligochaetes and polychaetes to be the numerically dominant organisms in southeastern tidal creeks (Sanger 1998, Lerberg et al. 2000, Holland et al. 2004, Gillett et al. 2007). The remaining 5% of the community consisted of crustaceans, mollusks, and nemerteans which will not be discussed further due to the relative minimum numbers of organisms obtained within the overall samples (Figure 23).

Table 7. Average number of ind. m^{-2} and the percent of samples in which the major taxonomic classes were collected in summer, 2005 and 2006. For creeks sampled in both years, average values were calculated.

Major Taxonomic Group	Intertidal Average (Ind. m^{-2})	Intertidal Frequency (%)	Subtidal Average (Ind. m^{-2})	Subtidal Frequency (%)
Polychaeta	1660.3	72.2	1535.0	95.4
Oligochaeta	4064.5	78.7	235.1	78.2
Amphipoda	15.8	3.7	264.1	58.6
Other	31.1	12.0	83.5	72.4
Molluska	39.3	9.3	127.2	64.4
Other Crustacea	15.3	4.6	82.5	51.7
Decapoda	1.0	0.5	15.4	36.8
Total	5827.4		2342.8	

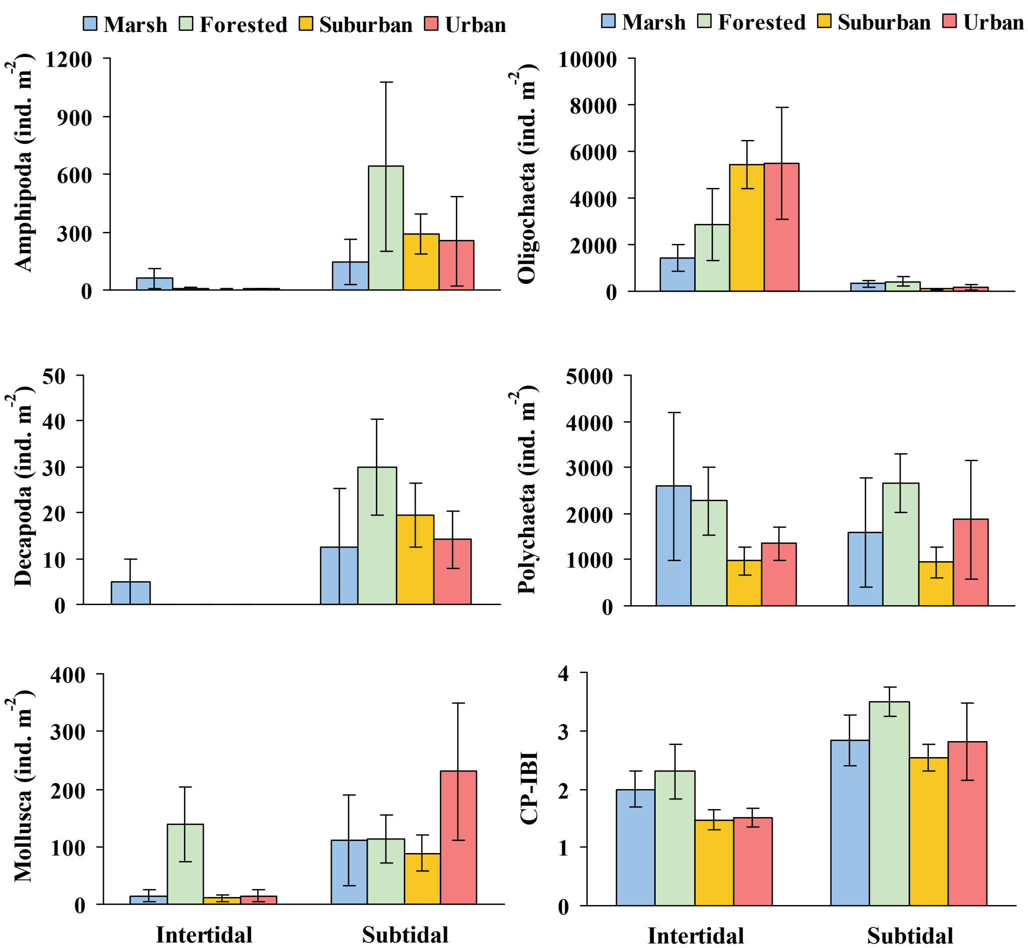

Figure 23. Average taxa abundances and the Carolinian Province Index of Biological Integrity (CP-IBI) by land use class and longitudinal gradient. Error bars are ± 1 standard error.

The ten most numerically abundant species and genera (5 oligochaetes and 5 polychaetes) composed over 80% of the individuals sampled (Table 8). The most abundant species was the oligochaete *Monopylephorus rubroniveus* which comprised 47% of the individuals collected. This species was found in 44% of the total samples collected; however, frequencies were much higher in intertidal systems relative to subtidal systems. The second most abundant species was the polychaete *Streblospio benedicti* which comprised 6.5% of the community and was found in 41% of the samples collected. Most of *M. rubroniveus* were found intertidally where they composed 65% of the community while most *S. benedicti* were found subtidally and composed 30% of the community (Table 8). These species have also been found to be the dominant benthic organisms in several other tidal creek studies (Sanger 1998, Lerberg et al. 2000, Holland et al. 2004, Gillett et al. 2007).

Table 8. Average number of ind. m^{-2} and percent of samples in which the twenty most abundant species were collected in the summer, 2005 and 2006. All species were in one of two classes of annelids: Oligochaeta (O), Polychaeta (P).

	Intertidal		Subtidal	
Scientific Name	Average (Ind. m^{-2})	Frequency (%)	Average (Ind. m^{-2})	Frequency (%)
Monopylephorus rubroniveus (O)	3220.01	59.72	7.71	4.60
Streblospio benedicti (P)	264.97	26.85	459.25	77.01
Tubificoides heterochaetus (O)	359.93	23.15	9.54	9.20
Laeonereis culveri (P)	349.72	30.56	1.04	4.60
Cirriformia sp. (P)	269.57	6.48	35.79	17.24
Tubificidae (O)	244.55	24.07	59.95	31.03
Capitella capitata (P)	206.77	28.24	16.20	10.34
Tubificoides brownae (O)	123.04	13.89	118.21	45.98
Heteromastus filiformis (P)	143.46	20.37	35.53	36.78
Tubificoides wasselli (O)	101.60	6.02	29.26	20.69
Nereis succinea (P)	77.60	14.35	70.40	44.83
Spionidae (P)	71.48	15.28	56.43	24.14
Mediomastus sp. (P)	2.04	0.93	208.99	45.98
Polycirrus sp. (P)	0.00	0.00	174.76	11.49
Fabricia sp. (P)	51.57	3.24	0.00	0.00
Leitoscoloplos fragilis (P)	42.89	6.94	5.09	16.09
Nemertea (P)	31.14	12.04	30.69	55.17
Scoletoma tenuis (P)	0.00	0.00	105.93	34.48
Spiophanes bombyx (P)	0.51	0.46	94.83	17.24
Capitomastus aciculatus (P)	35.74	7.41	0.26	1.15

Analyses were performed on five individual species as well as larger taxonomic groups. The four most abundant species were chosen for analyses and included the oligochaetes *Monopylephorus rubroniveus* and *Tubificoides heterochaetus* and the polychaetes *Laeonereis culveri* and *Streblospio benedicti*. The next two most abundant taxa were *Cirriformia sp*. and Tubificidae, but they were not analyzed because *Cirriformia sp*. was only found in large abundances in North Inlet, and Tubificidae is a grouping of possibly many species that could not be identified to a lower taxonomic class. Thus the fifth species analyzed was *Capitella capitata* which was the 7th most abundant organism and is common in tidal creek systems. It should be noted that these species were the most abundant when examining total abundances as well as abundances in the intertidal systems. The species compositions subtidally were very different than intertidal systems (Table 8).

The macrobenthic community in NOC-Masonboro (marsh) was similar to the other salt marsh creeks sampled. Abundances of *M. rubroniveus* were approximately 1.5 times higher (~1500 ind. m^{-2}) in NOC-Masonboro than the other four salt marsh creeks studied; however, the proportion of oligochaetes and polychaetes were similar in all salt marsh creeks. In comparison, SAP-Oakdale (forested) had the lowest abundances of all five species for all forested creeks. In particular, *M. rubroniveus* had an average abundance in forested creeks (2000 ind. m^{-2}) nearly two orders of magnitude greater than the abundance found in SAP-Oakdale (25 ind. m^{-2}). Finally, SAP-Duplin (forested) appeared similar to other forested creeks in the subtidal systems except for having higher abundances of *S. benedicti* than any other forested creek. Conversely, the intertidal SAP-Duplin had over twice as many organisms (~10,000 ind. m^{-2}) as all other intertidal forested creeks sampled except NIWB-Crabhaul (~7,000 ind. m^{-2}). However, of the 9 samples, two in each creek drove this abundance. One sample collected contained nearly 80% (60,000 ind. m^{-2}) of the oligochaetes collected in SAP-Duplin intertidally while another sample had over 70% (9,000 ind. m^{-2}) of the polychaetes collected illustrating the patchiness of these systems.

Two-way ANCOVAs were performed on the selected species, as discussed above. Factors were land use class and order, and covariates were salinity and sediment composition. Salinity and sediment composition have often been shown to greatly influence benthic communities (Seys et al. 1999, Lerberg et al. 2000, Gillett et al. 2007). Salinity and sediment composition were significant covariates for *C. capitata*, salinity was significant for *T. heterochaetus* and *S. benedicti*, and neither covariate was significant for *M. rubroniveus* and *L. culveri*. Order differences were significant for all species except *S. benedicti* with higher densities observed in the intertidal creeks than the subtidal creeks. *Streblospio benedicti* had an opposite trend with higher abundances of organisms subtidally (Table 9, Figure 23). Intertidal creeks are often considered more stressful than subtidal creeks because they are subjected to wider ranges of environmental factors as well as higher levels of chemical contaminants due to their proximity to the uplands (Lerberg et al. 2000, Mallin et al. 2000, Holland et al. 2004). *Monopylephorus rubroniveus* and *S. benedicti* were not significantly different among land use classes. However, more than twice the average number of *M. rubroniveus* was found in suburban and urban creeks (~4500 ind. m^{-2}) compared to forested creeks and this was more than five times the average of individuals found in salt marsh creeks. In contrast, abundances of *S. benedicti* were most abundant in forested creeks (625 ind. m^{-2}) which had 2 to 3 times higher abundances than salt marsh, suburban, or urban systems (Figure 24). *Capitella capitata* was found to be significantly higher in forested creeks relative to suburban and urban creeks. *Tubificoides heterochaetus* and *L. culveri* had higher abundances in the suburban and urban creeks relative to salt marsh creeks with differing significance among classes for the two species (Table 9). Lerberg et al. (2000) also found *M. rubroniveus* and *L. culveri* to be more abundant in degraded systems and *S. benedicti* to be more abundant in less degraded systems.

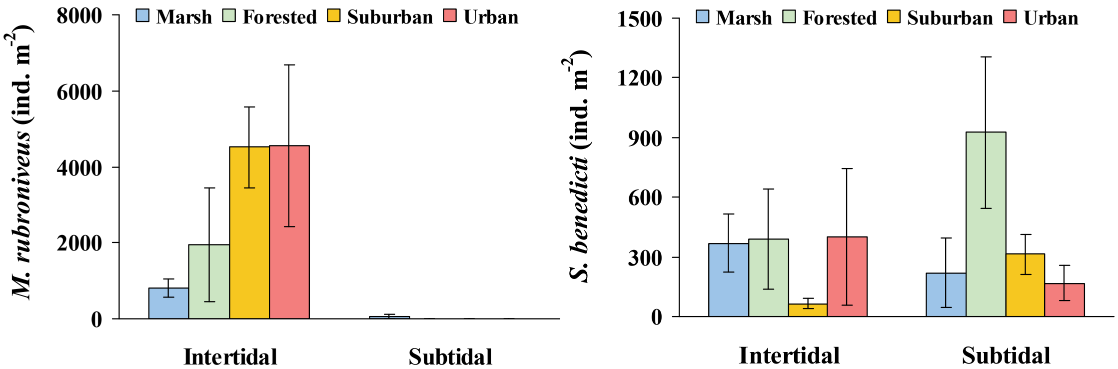

Figure 24. *Monopylephorus rubroniveus* and *Streblospio benedicti* (the two most abundant species) abundances by land use class and longitudinal gradient. Bars represent average concentrations. Error bars are ± 1 standard error.

Table 9. Results of 2-way ANCOVA on average densities (except percent composition for Oligochaeta and Polychaeta) of macrobenthic taxa and diversity metrics sampled in summer, 2005 and 2006. Land use (LU) factors are Marsh (M), Forested (F), Suburban (S), Urban (U). Order factors are Intertidal (I) and Subtidal (S). Covariates are percent mud and salinity. Post hoc multiple comparisons were performed using least squared means; model factors (arranged from low to high) with different superscripts are statistically different.

Parameter	Model p-value	r^2	Land Use p-value	Order p-value	% Mud p-value	Salinity p-value	Interactions	Land Use LS Means	Order LS means
Total Organisms	0.003	0.44	0.044	0.001	ns	ns	LU*order	$S^a\ U^{ab}\ M^{ab}\ F^b$	$S^a\ I^b$
M. rubroniveus	<0.001	0.84	0.502	<0.001	ns	ns	LU*order		$S^a\ I^b$
S. benedicti	0.013	0.32	0.700	0.180	ns	0.009	ns		
T. heterochaetus	<0.001	0.61	0.109	0.012	ns	<0.001	LU*sal	$M^a\ F^{ab}\ S^{ab}\ U^b$	$S^a\ I^b$
L. culveri	<0.001	0.61	0.007	<0.001	ns	ns	ns	$M^a\ F^{ab}\ S^{bc}\ U^c$	$S^a\ I^b$
C. capitata	<0.001	0.65	0.018	<0.001	0.067	0.074	ns	$M^a\ S^{ab}\ U^b\ F^b$	$S^a\ I^b$
Oligochaeta	<0.001	0.41	0.180	<0.001	ns	ns	ns		$S^a\ I^b$
Polychaeta	0.006	0.34	0.209	0.126	ns	0.011	ns	$S^a\ U^a\ M^a\ F^b$	
H'	<0.001	0.64	0.194	0.533	ns	0.025	order*sal		$I^a\ S^b$
J'	0.020	0.26	0.085	0.014	ns	ns	ns	$U^a\ S^{ab}\ F^b\ M^b$	$I^a\ S^b$
Mao-Tao	<0.001	0.58	0.039	<0.001	0.019	ns	order*mud	$S^a\ U^{ab}\ M^b\ F^b$	$I^a\ S^b$

Analyses were performed on taxonomic groups to examine higher-taxa changes in the macrobenthic communities as well. For these analyses, sums of oligochaete and polychaete species in each sample were used. Both taxa had higher abundances intertidally but only oligochaetes were significantly different ($p < 0.0001$). Neither group showed significant differences between land use classes; however, oligochaetes generally had higher abundances in the suburban and urban systems while polychaetes generally had higher abundances in the salt marsh and forested systems (Table 9, Figure 23).

The abundances of each dominant organism were regressed against the level of impervious cover for each creek order. The r^2 values were generally low (0.05-0.15), but most taxa showed an increase or decrease in abundance as impervious cover increased. Both oligochaete species examined, *M. rubroniveus* and *T. heterochaetus*, as well as the polychaete *L. culveri* increased in abundance as impervious cover increased, but only intertidally. On the other hand the other two polychaete species, *S. benedicti* and *C. capitata* had decreasing abundances with increasing impervious cover; however, the regression of subtidal *S. benedicti* was the only one that was significant.

Species diversity was also examined in the creek systems. The 9 intertidal cores were summed and the 3 subtidal cores were averaged to evaluate species diversity because of the differences in sample size (i.e., core is ~1/10 the grab). Three measures were used for most analyses which included Shannon-Wiener diversity (H'), evenness (J'), and Mao-Tau. Mao-Tau uses analytical formulas to compute expected species accumulation curves to calculate expected species richness (Colwell et al. 2004, Mao et al. 2005). All measures were significantly higher in the subtidal systems compared to intertidal systems. H' and J' detected no significant differences between land use classes, and the Mao-Tau measure of diversity found significant differences. All measures had higher numbers in forested creeks relative to urban and/or suburban systems. Both J' and Mao-Tau also indicated significantly higher diversities in the salt marsh systems relative to urban or suburban creeks, respectively (Table 9).

The Benthic Index of Biological Integrity developed for the Carolinian Province (Cape Henry, VA - St. Lucie Inlet, FL; CP-IBI) as described in Van Dolah et al. (1999) was also used to examine the ecological status of the benthos. The CP-IBI is a measure of the health of a community, and higher numbers represent less impaired communities. A significant difference between orders was found with higher CP-IBI scores in the subtidal systems (Figure 23). There was also a significant difference between land use classes with forested and marsh systems having higher CP-IBI scored than suburban and urban systems. This index was developed to measure benthic communities in open water systems, and species composition in intertidal creeks differs from that of open water. Therefore, significant differences among land use classes in intertidal systems may not signify differences in community health. According to the CP-IBI, all intertidal creek systems are degraded regardless of land use class. This result is probably driven by the fact that the metric was developed for subtidal waters and not for the more stressed headwater creeks.

Indices for intertidal communities were developed by Lerberg et al. (2000) for pollution indicator and pollution sensitive species. Pollution indicator species included *M. rubroniveus*, *L. culveri* and *Tubificoides brownae* which are species often associated with stressful environments while pollution sensitive species included *T. heterochaetus*, *S. benedicti*, Nemertinea, *Heteromastus filiformis* and *Tharyx acutus*. Two-way ANOVAs found significant differences among land use classes in pollution tolerant species with a significantly higher proportion of pollution tolerant organisms being found in suburban and urban systems. Pollution indicator species increased significantly in abundance as creek watersheds increased in impervious cover, and pollution sensitive species decreased significantly in abundance as impervious cover increased (Figure 25).

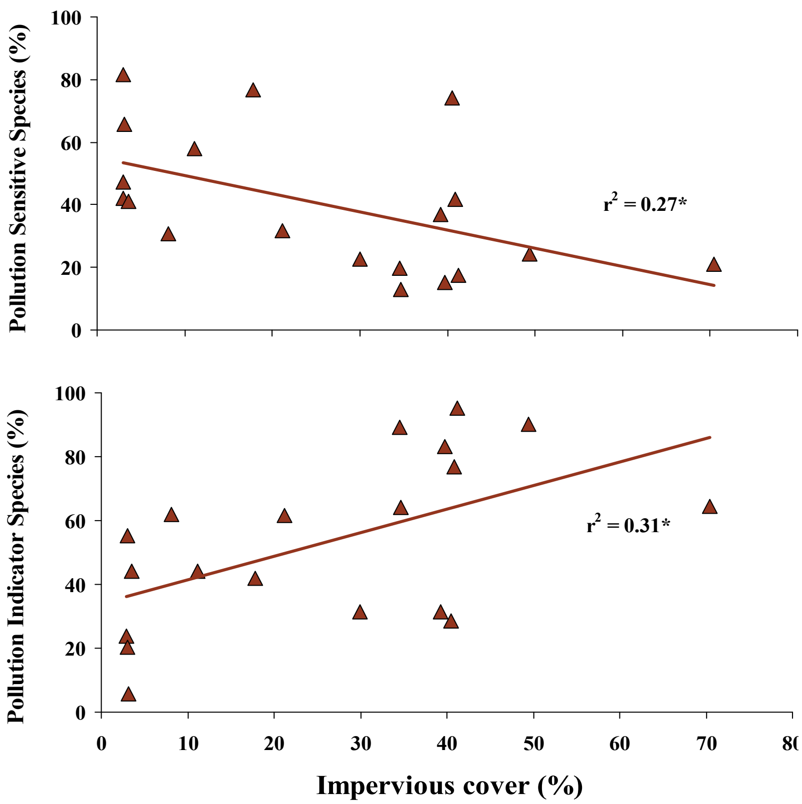

Figure 25. Relationship between pollution species and impervious cover for intertidal watersheds. Impervious cover was regressed against pollution sensitive species (upper) and against pollution indicator species (lower). Model r^2 is shown for each regression with asterisk (*) indicating significance ($p < 0.05$).

3.3.2 Nekton Community

The nekton assemblages in intertidally-dominated (intertidal) and subtidally-dominated (subtidal) portions of a system differed substantially (Table 10). The two different components of these systems differ in water quality and volume. This reflects in part the distinct habitat structure in each creek segment type. Sampling methods could also add to the differences; intertidal assemblages were sampled by seine and subtidal by trawl. Fifty-nine intertidal and fifty-nine subtidal species were identified; however, only species occurring most commonly across the creeks were analyzed statistically and discussed in this section. For the intertidal systems, abundances are reported as individuals per square meter (ind. m^{-2}), and the species occurring most commonly were *Callinectes sapidus* (blue crabs), *Fundulus heteroclitus* (mummichog), *Leiostomus xanthurus* (spot), *Palaemonetes* spp. (grass shrimp), *Litopenaeus setiferus* (white shrimp) and *Farfantepenaeus aztecus* (brown shrimp). For the subtidal systems, units are individuals per hectare (ind. ha^{-1}), seven species occurred most commonly: *Bairdiella chrysoura* (silver perch), *Callinectes sapidus*, *Lagodon rhomboids* (pinfish), *Leiostomus xanthurus*, *Lolliguncula brevis* (brief squid), *Farfantepenaeus aztecus* (brown shrimp), and *Litopenaeus setiferus* (white shrimp).

In the intertidal creeks, a resident creek species, *Palaemonetes* spp., was the most abundant, and densities ranged from 0.01 ind. m^{-2} (NIWB-Clambank, marsh) to 24.04 ind. m^{-2} (SAP-Duplin, forested). Transient Penaeidae (*Litopenaeus setiferus* and *Farfantepenaeus aztecus*) ranked second in abundance with densities ranging from 0 ind. m^{-2} (Okatee, suburban) to 6.58 ind. m^{-2} (SAP-Oakdale, forested). SAP-Duplin (forested) and SAP-Oakdale had higher abundances

Table 10. Average abundance per area of the top ten most abundant nekton species collected for each land use class (Forested, Marsh, Suburban, Urban) and creek order (intertidal, subtidal). Intertidal creek abundances are individuals per square meter, and subtidal abundances are individuals per hectare.

Species	Marsh	Forested	Suburban	Urban
Intertidal (ind. m^{-2})				
Bairdiella chrysoura	0.59	0.16	0.01	0.08
Callinectes sapidus	0.02	0.14	0.06	0.05
Fundulus heteroclitus	0.45	0.42	0.61	0.65
Gambusia holbrooki	0.00	0.03	0.07	0.18
Gerreidae	0.04	0.14	0.00	0.02
Lagodon rhomboides	0.10	0.00	0.01	0.01
Leiostomus xanthurus	0.07	0.03	0.23	0.04
Palaemonetes sp.	2.27	6.28	1.45	4.44
Penaeidae	0.62	2.09	1.20	2.40
Poecilia latipinna	0.00	0.08	0.14	0.93
Subtidal (ind. ha^{-1})				
Anchoa mitchilli	27.29	23.75	126.88	4.83
Bairdiella chrysoura	528.90	18.12	599.49	4.03
Callinectes sapidus	22.72	10.06	27.07	26.81
Lagodon rhomboides	278.31	13.69	47.92	143.99
Leiostomus xanthurus	1283.95	49.08	330.38	313.24
Lolliguncula brevis	35.43	101.05	59.19	104.67
Micropogonias undulatus	0.00	10.87	3.34	38.82
Farfantepenaeus aztecus	10.29	101.02	121.85	213.90
Litopenaeus setiferus	1741.01	48.71	1130.69	137.68
Stellifer lanceolatus	82.13	6.84	16.72	1.61

of the two Penaeid shrimp, *Palaemonetes* spp. and *F. heteroclitus* compared to other creeks draining forested watersheds, and *C. sapidus* densities were four times greater in SAP-Oakdale (0.94 ind. m^{-2}) than the next highest density (0.21 ind. m^{-2}, Parrot, suburban) observed in the study (Figure 26). NOC-Masonboro (marsh) densities generally were low compared to the other creeks draining marsh watersheds. These differences may have been driven by the geographical location of NOC-Masonboro at the northern range of the sampling area, especially since Cape Hatteras, North Carolina is a dividing line for many coastal fish assemblages.

In the subtidal creeks, *L. setiferus* was most abundant, ranging from 0 ind. ha^{-1} (NIWB-Clambank, marsh) to 3,368 ind. ha^{-1} (Okatee, suburban). Second in abundance, *L. xanthurus* density ranged from 0 ind. ha^{-1} (Hewlitts, suburban) to 2,576 ind. ha^{-1} (Orangegrove, marsh). SAP-Duplin was the only creek draining a marsh or forested watershed with no presence of *L. setiferus*. Four creeks within developed watersheds had no presence of *L. setiferus*, including three located in NC and GA. SAP-Duplin and three creeks in developed watersheds in NC and GA also had low or no densities for *L. xanthurus*.

In the intertidal creeks, *Palaemonetes* spp. trended toward a significant land use class effect with forested creeks having the highest abundance while suburban creeks were significantly lower than the other land use classes (Table 11). Several factors appear to be important in explaining the variation of the grass shrimp, including geographic location, although the patterns were not statistically significant (Figure 27).

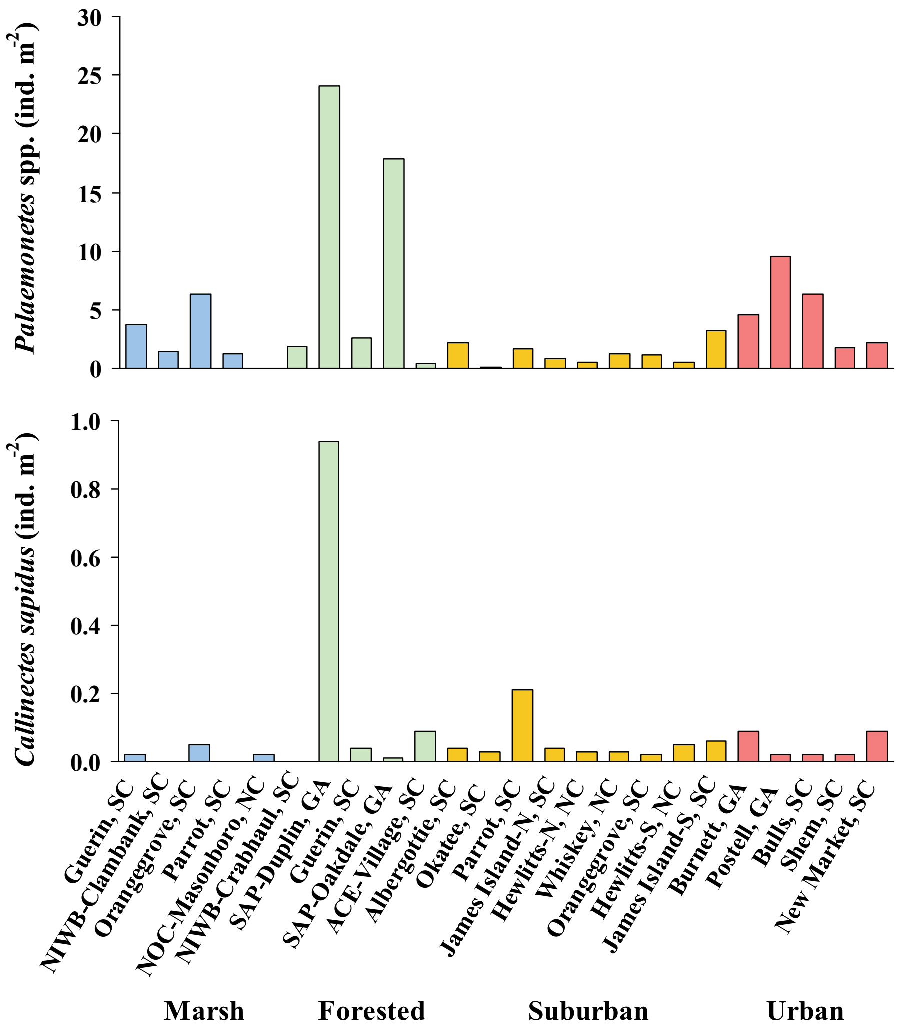

Figure 26. *Palaemontes* spp. and *Callinectes sapidus* abundances in the intertidal sections. Land use class is marked by color. Bars represent average abundances.

In the subtidal creeks, *L. rhomboides* was the only species with a significant land use class effect (Table 11). The forested and suburban classes had significantly lower abundances of this species compared to the marsh class, and the the urban class was similar to the other classes. The general trend regarding land use class was that the abundances of individual species in marsh creeks were different from the abundances in the other three land use classes.

Surveys previously conducted in intertidal creeks in South Carolina found reduced numbers of Penaeidae in watersheds with higher impervious cover levels (Holland et al. 2004). None of the regression analyses of the nekton species for intertidal or subtidal creeks was significant.

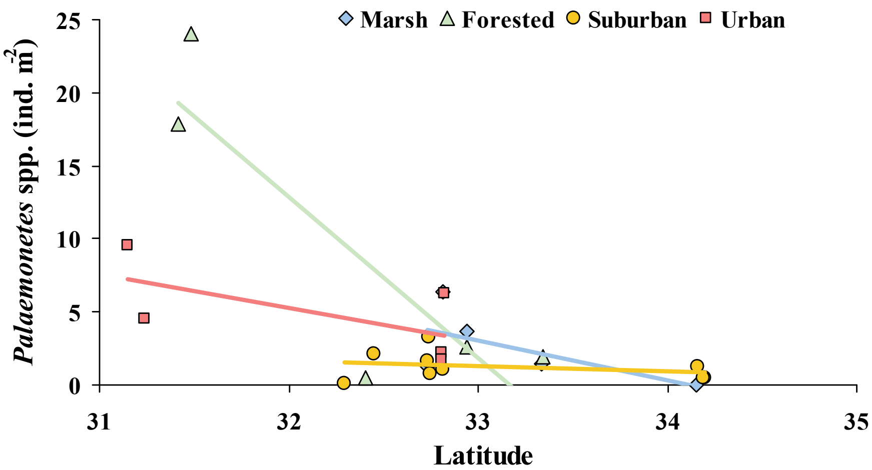

Figure 27. Relationship between *Palaemonetes* spp. abundance and creek latitude by land use class. Regression results were not significant.

Parameter	Model p-value	r^2	Land Use LS means
Intertidal			
F. heteroclitus	0.884	0.03	
C. sapidus	0.297	0.17	
L. xanthurus	0.539	0.10	
Palaemonetes spp.	0.064	0.30	S^a M^{ab} U^b F^b
Penaeidae	0.233	0.19	
Subtidal			
B. chrysoura	0.422	0.17	
C. sapidus	0.304	0.21	
L. rhomboides	0.044	0.41	F^a S^a U^{ab} M^b
L. xanthurus	0.179	0.27	F^a U^{ab} S^{ab} M^b
L. brevis	0.462	0.15	
P. aztecus	0.851	0.05	
P. setiferus	0.416	0.17	
Penaeidae	0.806	0.06	

Table 11. Results of 1-way ANOVA examining differences in average abundance in intertidal and subtidal creeks separately by land use class. Land use class factors are Marsh (M), Forested (F), Suburban (S), Urban (U). Post hoc multiple comparisons were performed using least squared means; model factors (arranged from low to high) with different superscripts are statistically different.

3.4 Human Responses

3.4.1 Oyster Tissue Pathogens

Oysters were only collected during the 2006 sampling period from 11 tidal creek systems: SAP-Duplin, SAP-Oakdale, Guerin, Burnett, Postell, New Market, James Island, ACE-Village, Hewlitts, Whiskey, and NOC-Masonboro. Tissues were examined for pathogen body burdens, and the same pathogen indicators measured in the water column were measured in the oyster tissues.

Pathogen concentrations for any particular parameter varied over 3-4 orders of magnitude. FC ranged from 23 CFU 100 g^{-1} tissue weight in ACE-Village (subtidal, forested) to 220,000 CFU 100 g^{-1} tissue weight in New Market (interidal, urban). ENT concentrations varied from 3,240 CFU 100 g^{-1} tissue weight to 320,000 CFU 100 g^{-1} tissue weight in James Island (subtidal, suburban) and SAP-Duplin (subtidal, forested), respectively. F- coliphages in a few cases were not detected, but reached 6,136 PFU 100 g^{-1} tissue weight in New Market Creek (urban). F+ coliphages were only detected in 4 samples; the highest concentrations were observed in SAP-Oakdale (intertidal, forested; 4,816 PFU 100 g^{-1} wet tissue weight).

Samples collected from NERR locations were difficult to classify with respect to the quality of the resource. For instance, FC, F+, and F- measured in NOC-Masonboro oysters were some of the lowest concentrations observed; yet, the ENT concentration (60,000 CFU 100 g^{-1} tissue weight) was in the middle of the observed values for all creeks. SAP-Oakdale showed concentrations higher than the median concentration for FC, F-, and F+ indicators; ENT concentrations (6,000 CFU 100 g^{-1} tissue weight) were fairly low compared to the other sites. The subtidal portion of SAP-Duplin (forested) had the highest ENT concentrations (320,000 CFU 100 g^{-1} tissue weight) observed in any of the samples.

The overall sample size was small, as oysters were only collected from a subset of the sampled systems. Nonetheless, ANOVA results indicated a significant land use class effect in F- coliphages, with concentrations in oysters collected from forested watersheds being lower than those collected from either suburban or urban watersheds (Table 12).

Table 12. Results of 2-way ANOVA examining differences in concentrations of selected pathogen indicators measured in tissue from oysters collected in study creeks. Land use factors are Forested (F), Suburban (S), Urban (U). The one sample from a Marsh creek (NOC_Masonboro) was excluded from these analyses. Order refers to creek order (intertidal, subtidal). Post hoc multiple comparisons were performed using least squared means; model factors (arranged from low to high) with different superscripts are statistically different.

Parameter	Model p-value	r^2	Land Use p-value	Order p-value	Inter-action	Land Use LS means	Order LS means
Enterococcus	0.563	0.15	0.375	0.774	ns		
Fecal Coliform	0.181	0.32	0.419	0.078	ns		
F- Coliphage	0.005	0.65	0.004	0.076	ns	F^a U^b S^b	
F+ Coliphage	0.352	0.23	0.677	0.153	ns		

Regression analysis showed that there was a significant ($p < 0.05$) positive relationship between watershed impervious cover and FC concentrations in oysters collected in intertidal creeks (Figure 28). For F- coliphages, there were significant relationships in both creek orders. There were no other significant regressions.

For exploratory purposes, the indicator concentrations from oyster tissue were plotted versus water concentrations for each system. Oysters are presumed to be bioconcentrators of water column materials, and the hypothesis that concentrations in oyster tissues would be higher than in the water was evaluated with biplots. Results indicated that oysters collected in these systems were consistently above the 1:1 line (assuming that conc g^{-1} and conc ml^{-1} are equivalent) for ENT (Figure 29). For FC and F-, the concentrations were more likely to fall on the 1:1 line, although oysters seem to be concentrating FC in water that had low concentrations. Data were scarce for F+ coliphages, but 4 of the 5 oyster samples where F+ coliphages were found had much higher concentrations than in the water.

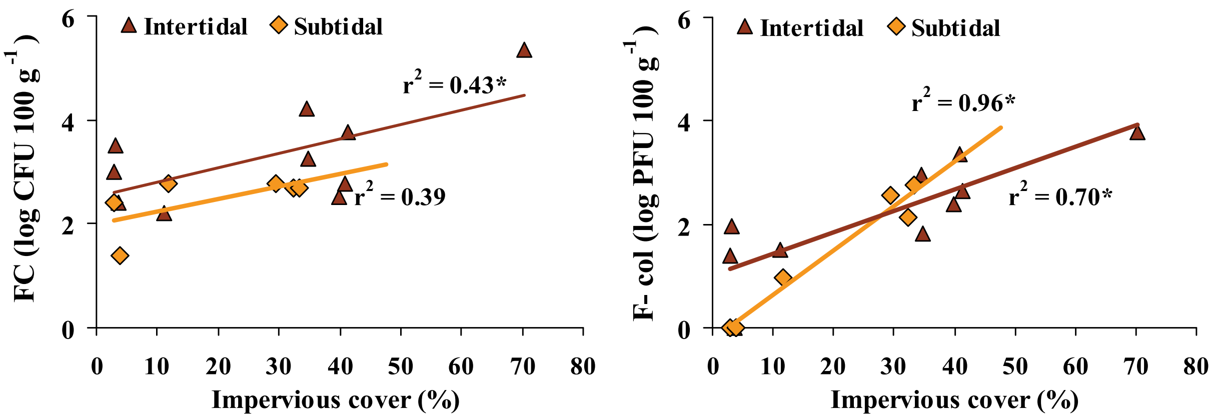

Figure 28. Relationship between pathogen indicator levels in oyster tissues and impervious cover for the study watersheds. Impervious cover was regressed against fecal coliform (FC; left) and against F-coliphage (right). Model r^2 is shown for each regression with asterisk (*) indicating significance ($p < 0.05$). Log transformation is x +1.

3.4.2 Oyster Tissue Contaminants

Oyster tissues were collected from the 11 tidal creek systems sampled in 2006. Samples represent all orders in the 12 intertidal and 7 subtidal creek segments. Tissues were analyzed for lipids and contaminants including PAHs, PCBs, PBDEs, pesticides, and metals. Statistical analyses were not performed on these data due to the high number of non detects and low sample size.

Oyster tissue lipid concentrations ranged from 5.7% (SAP-Duplin, subtidal, forested) to 38% (ACE-Village, subtidal, forested). In general, higher values were observed in the forested creeks compared to the marsh and developed creeks (Figure 30). Exceptions were SAP-Duplin (intertidal and subtidal, forested) with levels similar to the developed systems, and James Island (intertidal, suburban) with levels similar to the forested systems. Overall lipid concentrations were higher than those observed in the May River when it was listed as an Outstanding Resource Waterway by South Carolina Department of Health and Environmental Control (Van Dolah et al. 2004). In addition, the lipid concentrations tended to be a little lower in the subtidal creeks compared to the intertidal creeks.

Total PAH tissue concentrations ranged from 0 (Guerin, intertidal, forested; Hewlitts, subtidal, suburban) to 2,161 ng g^{-1} dry weight (Burnett, intertidal, urban). Naphthalene, a low molecular weight PAH, was most commonly detected and was found in marsh, forested and developed systems. The other PAHs detected included acenaphthylene, anthracene, benzo(a)anthracene, benzo(b)fluoranthene, benzo(g,h,i)perylene, fluoranthene, and pyrene. All except acenaphthylene are high molecular weight PAHs typical of pyrogenic sources, and were found in developed systems, except for one forested system (SAP-Duplin, subtidal). SAP-Duplin (subtidal) had high levels of benzo(g,h,i)perylene (488 ng g^{-1} dry weight).

Total PCB concentrations ranged from 0 to 244.3 ng g^{-1} dry weight (Burnett, intertidal, urban). Concentrations above the detection limit were found in the three urban intertidal creeks and in the one urban subtidal creek; low concentrations (<2 ng g^{-1} dry weight) were found in Guerin (subtidal, forested) and Whiskey (subtidal, suburban). PBDEs, flame retardants, were detected in oyster tissue at only one site, New Market (intertidal, urban). Pesticide concentrations in oyster tissues were dominated by DDT and its derivatives. Total DDT concentrations ranged from 2.53 (Guerin, intertidal, forested) to 20.54 ng g^{-1} dry weight (New Market, intertidal, urban) (Figure 31). In general, total DDT tissue concentrations were higher in the developed systems compared to the forested and marsh systems. The only other detectable pesticide contaminants were mirex (3.49 ng g^{-1} dry weight) in Guerin (intertidal, forested), endosulfan I (2.38 ng g^{-1} dry weight) in ACE-Village (intertidal, forested), and dieldrin (3.82 ng g^{-1} dry weight) in Postell, (intertidal, urban).

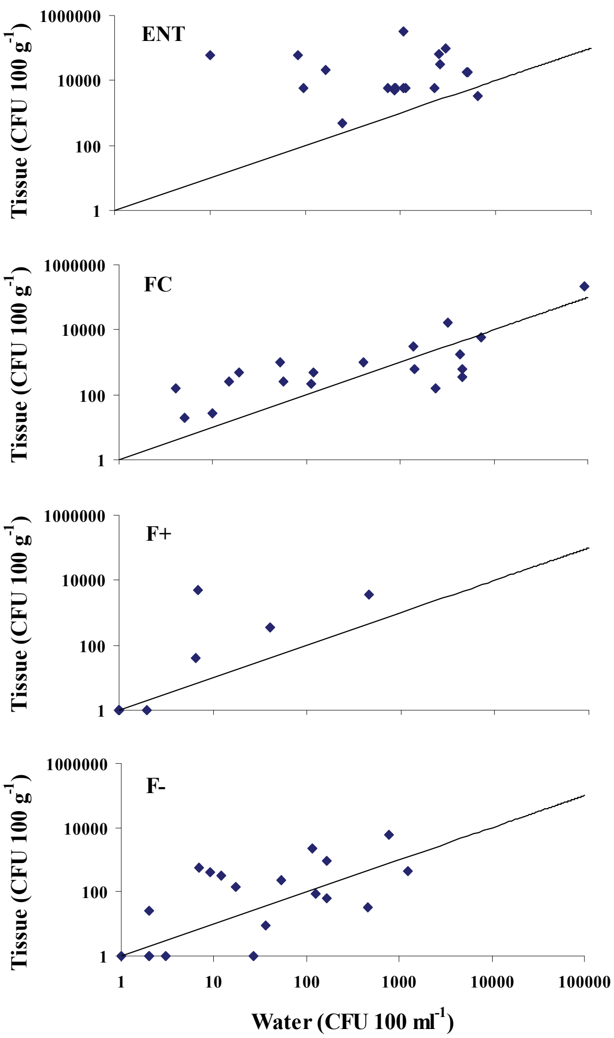

Figure 29. Biplots of pathogen indicators comparing water column concentrations with oyster tissue concentrations. Ent = Enterococcus, FC = Fecal Coliform, F+ and F- = coliphages.

To evaluate the potential for bioaccumulation of PCBs, PCB congeners were grouped by number of chlorines and then compared to sediment PCBs. PCBs with 7 or more chlorines have an increased potential for transfer up the food web (Oliver and Niimi 1988). The sediment total PCB concentration in New Market (intertidal, urban) was the highest of all creeks (107 ng g^{-1} dry weight) and consisted primarily of lower chlorinated compounds (hexa- and tetrachlorobiphenyls). New Market's oyster tissue concentrations were comparatively low (approximately 52 ng g^{-1} dry weight). This difference corresponds with conclusions that lower chlorinated compounds found in the sediments are not bioaccumulating in tissues. In comparison, Burnett (urban, intertidal) had the second highest total PCB concentration in sediments (61 ng g^{-1} dry weight) and consisted primarily of higher chlorinated compounds. Oyster tissue concentrations were high in Burnett (244 ng g^{-1} dry weight), reflecting high bioaccumulation in this system.

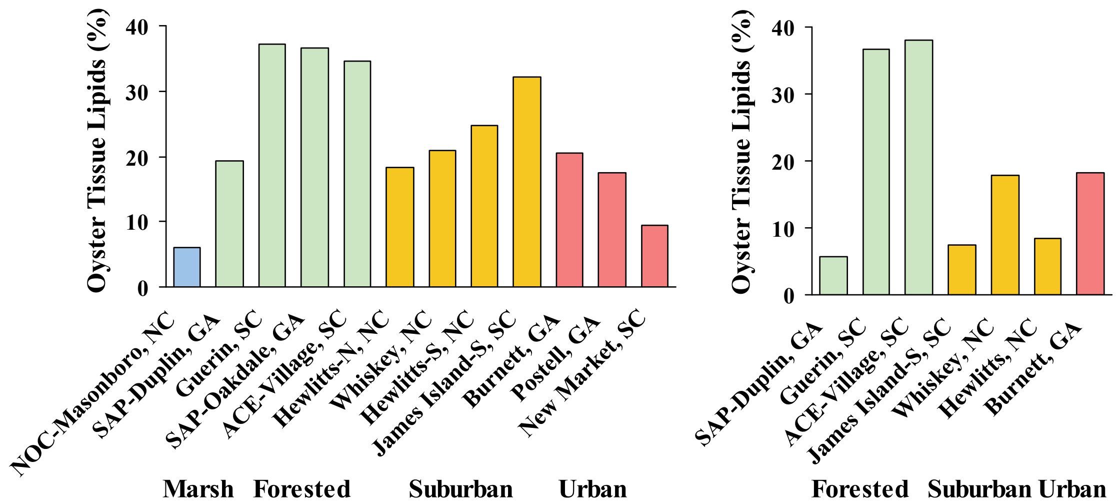

Figure 30. Lipid level percentage in oyster tissues for intertidal (left) and subtidal (right) creeks. Land use class is marked by color.

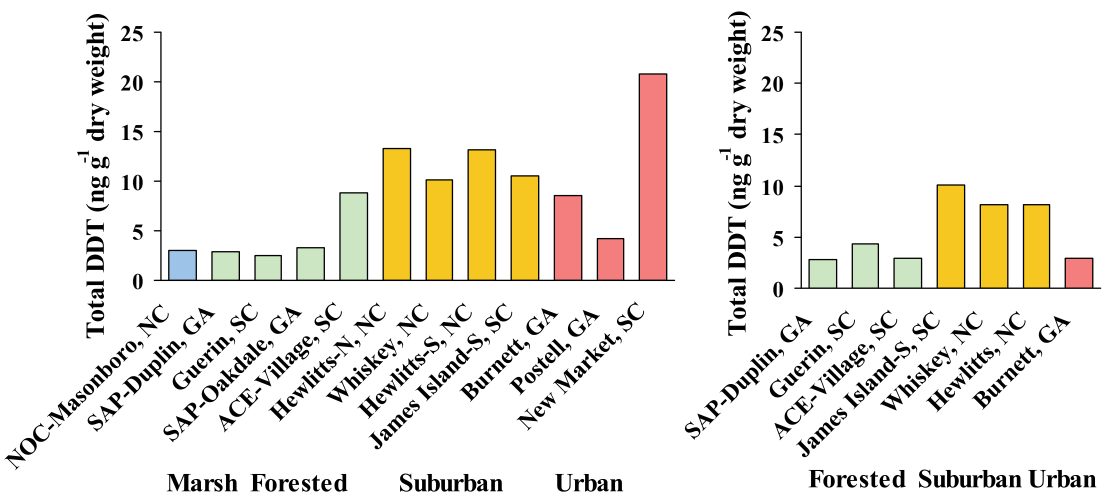

Figure 31. DDT levels in oyster tissues for intertidal (left) and subtidal (right) creeks. Land use class is marked by color.

Concentration data for only a few metals (As, Cd, Cr, Cu, Pb, Hg) are discussed here. Except for arsenic, these are the metals which are often elevated from anthropogenic sources. Lead concentrations were similar across the different land use classes and were generally low (< 0.7 µg g^{-1} dry weight) except for New Market (intertidal, urban: 1.58 µg g^{-1} dry weight). Mercury concentrations were similar across land use classes and were generally low (< 0.19 µg g^{-1} dry weight) except for Burnett (urban) in both the intertidal (0.35 µg g^{-1} dry weight) and subtidal (0.42 µg g^{-1} dry weight) creek segments. The highest concentrations of arsenic were found in the North Carolina creeks, similar to the fish tissue contamination findings of Cooksey et al. (2008). The cadmium, copper, and chromium concentrations were generally higher in the forested and suburban creeks compared to the marsh and urban creeks. In particular, Guerin, a forested creek in the Francis Marion National Forest, had some of the highest concentrations of these metals.

In addition, the oyster tissue concentrations on wet weight basis were compared to Food and Drug Administration (FDA, FDA 2001) environmental chemical contaminant action levels and the USEPA (2000) human-health consumption limits for cancer and non-cancer endpoints (Table 13). The FDA action levels are simply threshold values for comparison against tissue concentrations (non-consumption based). None of the concentrations observed in oyster tissue exceeded the molluscan bivalve or any fish actions levels for As, Cd, Cr, Pb, Ni, methyl mercury, PCBs, DDT, heptachlor epoxide, or mirex. The USEPA values are based on a consumption rate of four 8-ounce meals of fish per month for an adult population. It should be noted that we are comparing oyster tissue to fish tissue values; however, the comparison represents a level of potential risk. It should also be noted that for a number of these systems the shellfish are closed for harvest. Inorganic arsenic (estimated as 2% of total arsenic (USEPA 2004) and total DDT values exceeded the cancer endpoint for all sites sampled for oyster tissue. Dieldrin values exceeded the cancer endpoint at one site (Postell, intertidal, urban). Total PCB values exceeded the cancer endpoint at three sites (New Market, intertidal, urban; Whiskey, subtidal, suburban; Burnett, subtidal, urban) with one site exceeding the non-cancer endpoint (Burnett, intertidal, urban).

Table 13. Contaminant wet weight concentrations in tidal creek oysters. Concentrations given in ng g^{-1} for PCB and DDT, and in µg g^{-1} for metals (As, Cd, Hg, and Se). Inorganic Arsenic calculated as 2% of total arsenic. Methyl mercury calculated as 10% of total mercury. Italicized numbers indicate concentrations that exceeded the EPA cancer-endpoint consumption limits for fish, based on four 8-ounce meals per month. Bold and italicized numbers indicate concentrations that exceed non-cancer endpoints. No measurements were found to exceed FDA levels of concern for shellfish.

Land Use	Creek	Total PCBs	Total DDTs	Total As	Estimated Inorganic As	Total Cd	Total Hg	Total Methyl Hg	Total Se
Intertidal									
Marsh	Masonboro NERR	0.00	*0.34*	3.15	*0.06*	0.10	0.01	0.001	1.08
Forested	Duplin Creek	0.00	*0.30*	0.79	*0.02*	0.07	0.01	0.001	1.09
Forested	Guerin Creek	0.00	*0.28*	0.63	*0.01*	0.33	0.02	0.002	0.95
Forested	Oakdale Creek	0.00	*0.32*	1.07	*0.02*	0.08	0.01	0.001	1.42
Forested	Village Creek	0.00	*1.04*	1.01	*0.02*	0.10	0.01	0.001	1.21
Suburban	Hewlitts Creek S	0.00	*1.31*	1.43	*0.03*	0.08	0.01	0.001	0.74
Suburban	Hewlitts Creek N	0.00	*1.14*	1.23	*0.02*	0.08	0.01	0.001	0.56
Suburban	James Island Creek	0.00	*0.98*	0.85	*0.02*	0.09	0.01	0.001	1.22
Suburban	Whiskey Creek	0.00	*0.93*	1.57	*0.03*	0.06	0.01	0.001	1.07
Urban	Burnett Creek	***29.93***	*1.04*	0.57	*0.01*	0.12	0.04	0.004	0.76
Urban	New Market Creek	*3.81*	*1.52*	0.90	*0.02*	0.11	0.01	0.001	0.65
Urban	Postell Creek	0.00	*0.30*	0.67	*0.01*	0.08	0.01	0.001	0.64
Subtidal									
Forested	Duplin Creek	0.00	*0.29*	1.02	*0.02*	0.12	0.01	0.001	1.30
Forested	Guerin Creek	0.00	*0.36*	0.86	*0.02*	0.28	0.02	0.002	1.05
Forested	Village Creek	0.00	*0.30*	1.47	*0.03*	0.24	0.01	0.001	1.65
Suburban	Hewlitts Creek	0.00	*1.02*	4.15	*0.08*	0.10	0.01	0.001	1.10
Suburban	James Island Creek	0.00	*0.94*	1.16	*0.02*	0.15	0.01	0.001	1.27
Suburban	Whiskey Creek	*0.19*	*0.96*	2.55	*0.05*	0.05	0.01	0.001	1.53
Urban	Burnett Creek	*20.14*	*0.18*	0.70	*0.01*	0.13	0.05	0.005	1.03

3.4.3 Oyster Tissue Genomics

Modern biotechnological tools such as DNA micro-arrays provide highly sensitive means for monitoring human health. These techniques also appear to have great potential as tools for detecting responses of keystone organisms to environmental stresses associated with anthropogenic or climatic perturbations. Unfortunately, there is presently little basis for interpreting these genomic cues in terms of their ecological consequences at relevant scales of natural populations, communities and ecosystems. To integrate these innovative tools into future environmental monitoring, assessment and management programs, it is essential that we gain an understanding of the ecosystem context for these genomic measurements. This is particularly important under present federal initiatives that seek to use "ecosystem-based" approaches for managing coastal waters, including key habitats and exploited animal populations. As a proof of concept, we evaluated the utility of an oyster microarray developed as a part of the NOAA Oceans and Human Health Initiative to discriminate between first order creeks with differing land use classes and levels of impervious cover.

There are several motivating questions. Do genome-scale changes in gene expression exhibit clear responses to different types and levels of stress? Are patterns of DNA micro-array gene expressions related in qualitative and quantitative terms to ecological signals observed at higher levels of biological organization such as populations and ecosystems? What is the nature of these relationships across biological scales, and how do they change under different regimes of environmental stress?

Preliminary analysis (neural networks followed by receiver operating characteristic (ROC) curve) of gene expression profiles of the 12 creeks (2006 only) classified by the amount of impervious cover in the surrounding watershed indicated that unique expressed sequence tags (EST; 215 in gill, 202 in hepatopancreas) were important contributors to discrimination in more than 20% of the pairwise comparisons. The weakest discrimination (between <10% and 30-50% impervious cover in hepatopancreas tissues) was still significantly different from random, and this must be viewed as a significant deviation ($p < 0.01$, Figure 32). The lower level of discrimination in this comparison probably stems from combining creeks into the 30-50% category which have significant differences in contaminant loads. This combination should result in high variance in expression profiles within the group, leading to a lower discriminatory power (Table 14).

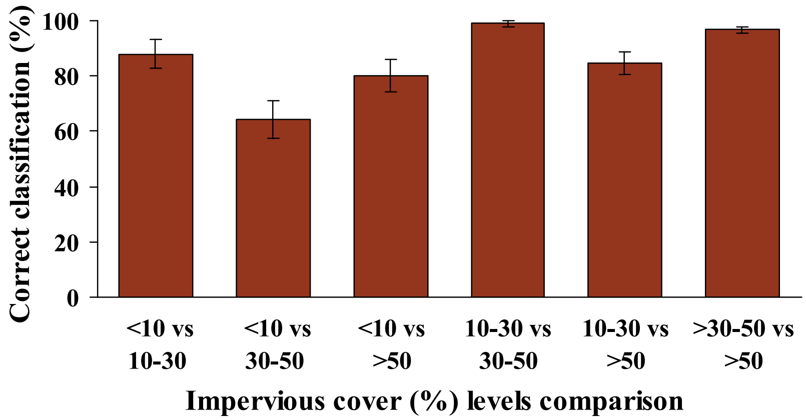

Figure 32. Ability of gene expression profiles (hepatopancreas tissues) to discriminate creek types based upon levels of impervious cover. Percent of correct classifications are shown for each comparison of levels. Error bars are ± 1 standard error.

Table 14. Oyster tissue analysis. Assignment of individual creeks to impervious cover and land use classes based upon watershed characteristics.

Creek	Impervious Surface	Land Use	Sample Size
Guerin	<10%	Forested	8
ACE-Village	<10%	Forested	9
SAP-Duplin	<10%	Forested	14
SAP-Oakdale	<10%	Forested	24
Postell	>30%-50%	Urban	24
Burnett	10%-20%	Industrial	12
New Market	50%	Urban	25
Hewletts	30%-50%	Suburban	17
NOC-Masonboro*	<10%	Forested	25
Whiskey	30%-50%	Suburban	13
James Island-S	30%-50%	Suburban	9

*NOC-Masonboro is a marsh creek classified as forested for the genomics analysis.

The discriminatory power gene expression profiles based upon the classification scheme of Holland et al. (2004) is illustrated in Figure 33. Both the gill and hepatopancreas comparisons were significantly greater than random expectations. In three of the six total comparisons, however, the gill data was significantly better than hepatopancreas in discriminating between land use classes. The weakest discrimination was between forested sites and the single industrial site and this may stem from the limited number of samples examined from Burnett Creek (n=12).

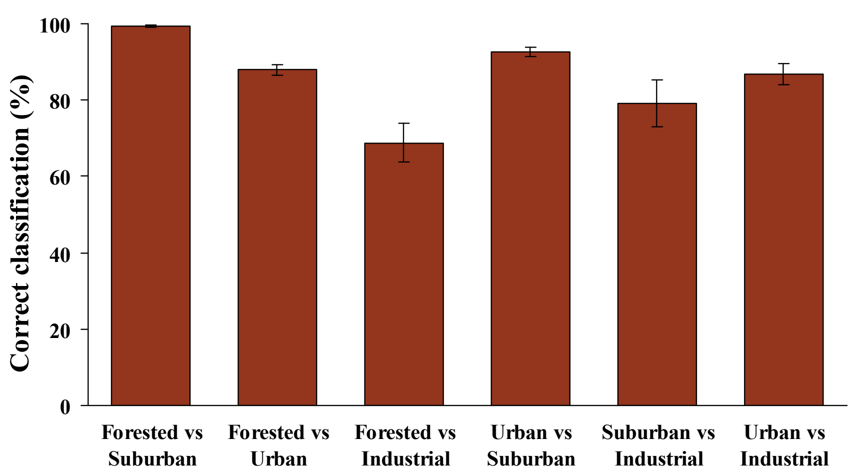

Figure 33. Ability of gene expression profiles (hepatopancreas tissues) to discriminate creek types based upon land use class. Percent of correct classifications are shown for each land use class comparison. Error bars are ± 1 standard error.

Research Highlight – Related Research in Sapelo Island NERR

Preliminary Results from Bottlenose Dolphin Studies in Sapelo Island National Estuarine Research Reserve and the Turtle/Brunswick River Estuary

An effort to examine chemical contaminant exposure for bottlenose dolphins which utilize the Sapelo Island, Georgia NERR was initiated in August 2007. This effort was coordinated with the Integrative Ecological Assessment efforts presented in this report in order to provide supplemental information on a top level predator in the NERRS. Dolphin exposure to persistent organochlorine contaminants (POCs) is of particular interest due to the magnification of these compounds through the food chain, and the dolphins' propensity to accumulate persistent contaminants in their lipid-rich blubber. Because of these exposures and the fact that many dolphin populations show a high degree of site-fidelity to a localized region, dolphins represent good integrative indicators of estuarine and coastal ecosystem health. Contaminant levels measured in dolphin tissues reflect the contamination of the habitat in which they reside.

This study is examining chemical contaminant exposure in bottlenose dolphins in the Turtle/Brunswick River estuary (TBRE), a highly polluted site approximately 20 miles southwest of the Sapelo Island NERR, and dolphins residing within and near the Sapelo Island NERR. Samples of dolphin skin and blubber are being obtained by remote biopsy sampling.

Initial results indicate extremely high concentrations of PCBs in dolphins sampled from both the TBRE and the Sapelo NERR (Figure A). Geometric mean PCB concentrations from dolphins sampled within and near the Sapelo NERR measured to date are comparable to concentrations measured from dolphins in northern Biscayne Bay, previously reported as some of the highest in US estuaries (Litz et al. 2007). Geometric mean concentrations measured from dolphins in the TBRE were over 2-fold higher than those measured in northern Biscayne Bay. The maximum concentration measured from the TBRE (2822 μg/g lipid) exceeded the prior worldwide maximum concentration (2100 μg/g lipid) for bottlenose dolphins that had been measured in a stranded dolphin from a highly polluted area of the Mediterranean Sea (Corsolini et al. 1995, Aguilar et al. 2002).

The geometric mean PCB concentration measured from dolphins near the Sapelo NERR (156.7 μg/g lipid) was approximately 3-4 times higher than concentrations reported from dolphins sampled in other southeast U.S. estuaries: Charleston, South Carolina (42.1 μg/g lipid for male & juvenile) and Beaufort, North Carolina (31.7 μg/g lipid for male & juvenile). Dolphins sampled in the TBRE (401 μg/g lipid) were nearly 10-fold higher than those reported for Charleston and Beaufort.

PCB congener profiles from both TBRE and Sapelo were indicative of an Aroclor 1268 signature, with a high prevalence of octa- and nonachlorobiphenyls. Signatures with these highly chlorinated homologs have been described in prior studies of sediments and biota and linked to the LCP Chemicals Superfund site near Brunswick, GA (Kannan et al. 1997, Kannan et al. 1998). Our preliminary results suggest a strong Aroclor 1268 signature for dolphins sampled in the TBRE, with 59% of the total PCBs being contributed by octa- and nonachlorobiphenyls. The proportion of these same homologs was slightly less in the Sapelo dolphins (41%), but still in stark contrast to other southeast sites where octa- and nonachlorobiphenyls comprise a small percentage (~9%) of the total PCBs.

Concentrations of PBDEs and the most prevalent pesticide compounds were several times lower than total PCBs and were comparable to values measured in other southeast U.S. sites (Figure B). Total PBDE concentrations were comparable to those measured from northern Biscayne Bay. Measured ΣDDT concentrations were high (20.1 and 17.7 μg/g lipid for males from Sapelo and TBRE, respectively), but were comparable to other nearby sites in the Carolinas.

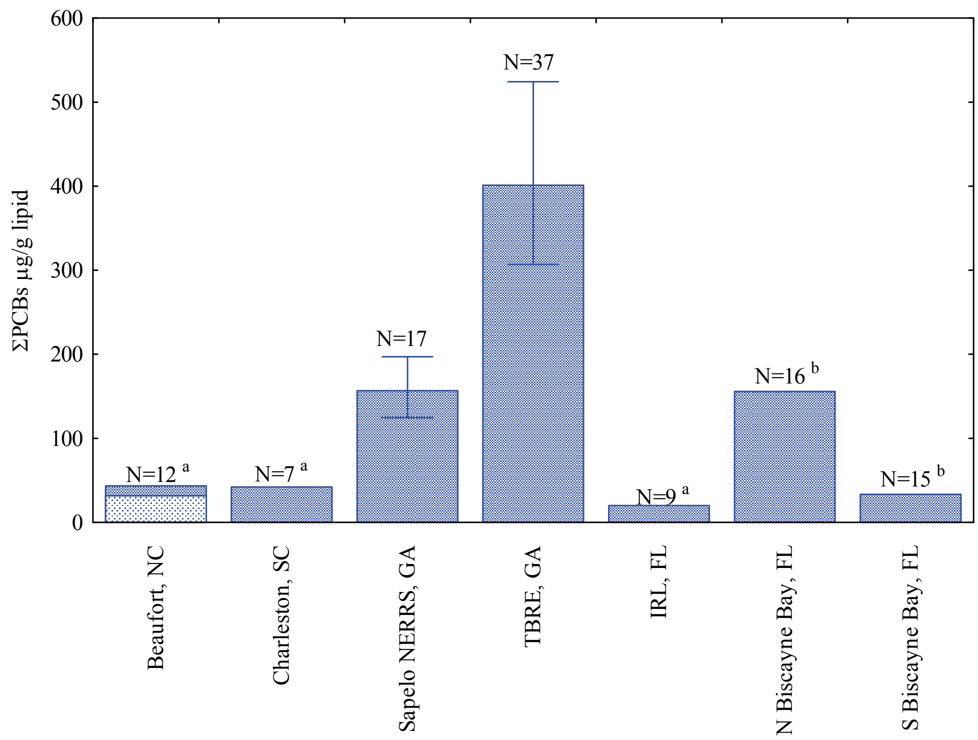

Figure A. Comparison of ΣPCB concentrations measured in male/juvenile bottlenose dolphins sampled from southeast U.S. estuaries. Bars represent geometric means calculated for this study or reported in published literature, whiskers represent 95% CIs around the geometric mean calculated for this study. Data sources: a=Hansen et al. 2004; b= Litz et al. 2007. Lightly shaded overlay for Beaufort, NC represents geometric mean for Σ27 PCB congeners reported by Hansen et al. 2004; darker shaded area represents an estimated increase in total if 66 congeners were to be included (unpublished data, Beaufort, NC).

Significance: *Threshold tissue concentrations to elicit physiological effects have been proposed for PCBs in marine mammal blubber: 14-17 μg/g lipid (Kannan et al. 2000, Schwacke et al. 2002). For both male and female dolphins in the TBRE, 100% of samples exceeded these thresholds (minimum concentration = 19.8 and 374.4 μg/g lipid for females and males, respectively). In fact, all of the male samples from TBRE exceeded the threshold by more than an order of magnitude and the highest male sample measured (2822 μg/g lipid total PCBs) was 100-fold greater than proposed toxic thresholds. Even more alarming is that dolphins that utilize the Sapelo NERRS, a protected area dedicated for the preservation of wildlife habitat, are also being exposed to high levels of the same PCBs. All of the males and all but one of the females sampled in Sapelo exceeded the toxic threshold for PCBs. The minimum concentration (76.5 μg/g lipid) measured in male samples from Sapelo was more than 4-fold higher and the maximum concentration (332 μg/g lipid) was nearly 20-fold higher than proposed toxic thresholds.*

At this point, it is unclear whether the dolphins sampled in Sapelo are exposed to high PCB levels because they range into the TBRE or whether their high PCB concentrations result from a movement of contaminated prey biota.

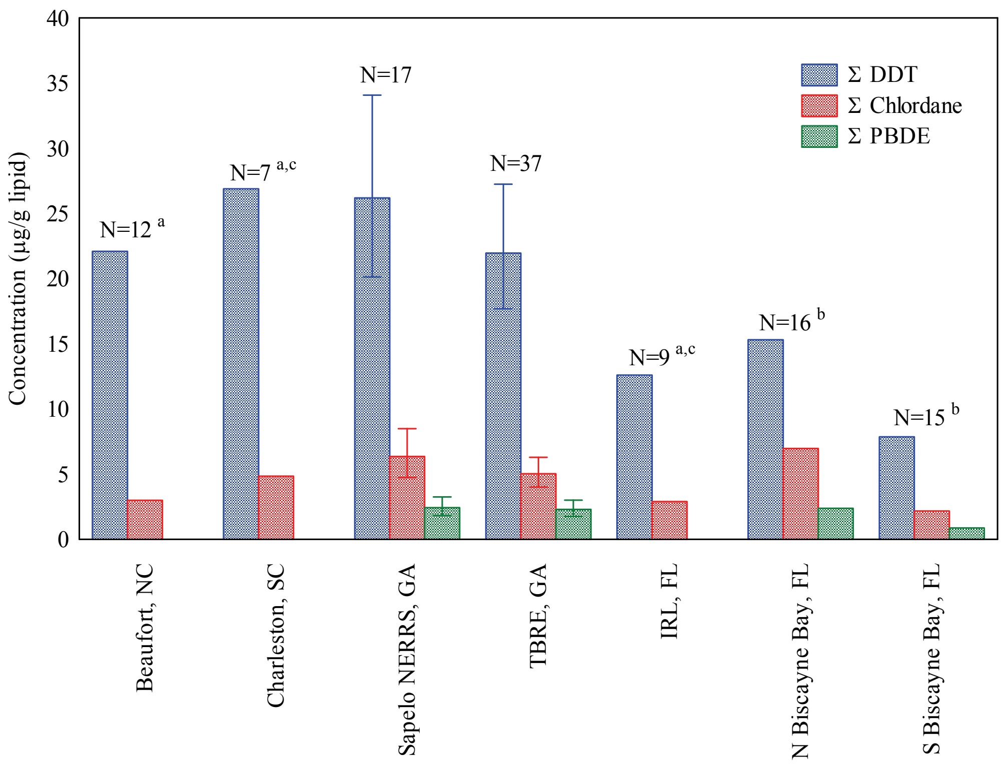

Figure B. Comparison of Σ DDT, Σ Chlordane and Σ PBDE concentrations measured in male/juvenile bottlenose dolphins sampled from southeast U.S. estuaries. Bars represent geometric means calculated for this study or reported in published literature, whiskers represent 95% CIs around the geometric mean. Data sources: a=Hansen et al. 2004; b= Litz et al. 2007.

Project Status & Plans: *Research on dolphins in these areas is continuing: (1) evaluation of mercury levels from skin samples is currently being performed, and (2) photo-identification studies have been initiated to elucidate dolphin distribution and movements. A final report summarizing biopsy and photo-identification efforts in the TBRE and Sapelo NERR is scheduled to be available in December 2009.*

Acknowledgments: *The bottlenose dolphin study within the Sapelo NERR and TBRE is a collaborative effort of the NOAA Cooperative Center for Marine Animal Health. Collaborators include NOAA/NOS/NCCOS, NOAA Fisheries OPR and SEFSC, NIST, NOAA/NERRS, and GA DNR. Student funding for the photo-identification effort is also being provided the Chicago Zoological Society (CZS). For further information, contact Lori Schwacke (Lori.Schwacke@noaa.gov).*

4. Discussion

4.1 Sentinel Habitats

Recent comprehensive reports of the condition of the Nation's coastal resources have noted measurably diminished ecosystem condition and potential human health risks (USEPA 2001, NMFS 2002, Pew 2003). Major sources of impairment across all regions and habitats included chemical and microbial contamination, increased "flashiness" in freshwater inflows, nutrient over-enrichment and hypoxia, increased frequency of harmful algal blooms, habitat modification and degradation, wetland loss, increased abundance of non-native species, over-harvesting of fisheries, and impaired biological communities. Most of these reports conclude that the major environmental threats to coastal resources are from diffuse or non-point sources of pollution. In addition, the cumulative effects of multiple stressors, including the interactions among them, have been identified as a major contributor to diminished resources. Most of these assessments were based on broad scale coastal sampling using standard ecosystem condition indicators that reliably document existing conditions. These monitoring networks are, however, insensitive to many stressors and do not provide early warning of ensuing harm to the ecosystems or the humans that rely on and use them. Further, these existing environmental monitoring networks also provide little reliable information about impacts on public health and other human resources or the specific source(s) of the stress.

The lack of early warning of degradation is related to choices made in monitoring open water habitats; however, sampling sentinel habitats in nearshore environments can provide an early warning of any ensuing harm to the larger coastal ecosystem. There are several principal attributes of sentinel habitats. They should be key structural components of ecosystems that have important roles in sustaining overall ecosystem function (e.g., nursery habitat, feeding grounds, mineral cycling, biological productivity). The amount and complexity of the sentinel habitat has a major influence on the condition of the ecosystems in which it exists. This complexity and the impacts on the habitat are at least partially understood by scientists and recognized by the public. They generally receive high exposure to stressors (e.g., shallow water in environments with limited capacity to dilute pollutants or modulate environmental change) and are sensitive to changes in environmental conditions, particularly multiple stressors. Sentinel habitats should also be easily observed to ensure changes can be measured. Close proximity to land and shallow depth are other useful attributes.

Tidal creeks and salt marshes are the interface between the landscape and estuaries, resulting in dynamic environments that are renowned for their ecological complexity and seafood production. In the Southeast US, watersheds containing tidal creeks are among the most rapidly developing in the Nation. The headwater regions of these creeks are the receiving waterbody for pollution released into the environment. The integrity and productivity of headwater portions of tidal creek environments are impaired by land use changes and the related non-point source pollution years to decades in advance of similar signals occurring in deeper open estuarine waters, suggesting these habitats are valuable sentinels of ensuing degradation, including a myriad of potential public health threats.

Previous research in tidal creek ecosystems has demonstrated the environmental quality of these systems, particularly the intertidally-dominated portions or headwaters, to be sensitive to land use change within the watershed (Sanger et al. 1999a, 1999b, Lerberg et al. 2000, Mallin et al. 2000, Holland et al. 2004). Because they are sensitive to local changes, these systems provide an early warning of the degradation from upland land use well before changes would be detected in larger coastal waters (e.g., tidal rivers, estuaries). Tidal creeks are therefore useful sentinels for monitoring the impacts of human activities on coastal habitats. The current study demonstrates that the sensitivity of tidal creeks to changes in these small coastal watersheds diminishes down their length (i.e., from small headwater creeks to larger tidal creeks). This spatial variability must be recognized before assessing the environmental quality of these habitats.

For many of the measured parameters, the intertidally-dominated or headwater portions of tidal creeks were found to respond differently than the subtidally-dominated or larger portions of tidal creeks. For example, water quality parameters (including basic water quality, nutrients, and pathogen indicators) showed many differences between the intertidal and subtidal segments of these creek networks. The smaller intertidal creeks generally had higher concentrations of nonpoint source pollutants, which is likely an indication of an upland runoff component as well as an estuarine dilution influence (i.e., tidal flushing) in the larger creeks. Biological parameters measured (e.g., nekton and benthos) also demonstrate significant variability along the headwaters to tidal river gradient. There is a marked shift in the benthic fauna, from one dominated by oligochaetes in the headwaters to one dominated by polychaetes in the larger water bodies. The nekton also appears to shift along this gradient, from more resident and nursery species in the headwaters to larger transient organisms in deeper creeks. Recognizing this spatial variability not only allows valid comparisons to be made across similar creek classes (with respect to surrounding land use for example) but also may provide information about which tools or indices may be useful for specific faunas. The environmental quality in intertidally-dominated portions of tidal creeks was also found to have stronger relationships with land cover changes than the subtidally-dominated habitats. These relationships may serve as the basis for future predictive models, and the significant relationships common in these habitats (both subtidal and intertidal, although mostly intertidal) will serve a valuable function in developing predictive models of land use impacts on these valuable coastal habitats.

In addition to accounting for the spatial variability down the length of a creek, the type of land cover in the watershed is also an important factor to consider when comparing creeks. The creeks draining watersheds with only salt marsh land cover were found to respond differently than creeks draining watersheds with forested upland land cover. The input of pollution is very different between these two systems. Creeks draining only salt marsh primarily receive contaminants from downstream sources. In comparison, creeks draining terrestrial areas receive significant freshwater input and contaminants from the upstream and upland areas. The input of freshwater is a critical factor to consider when assessing the impacts of land use change. Freshwater input in the form of stormwater runoff increases with increasing levels of impervious cover, carrying increased pollutants from the surrounding watershed into tidal creeks (Figure 34).

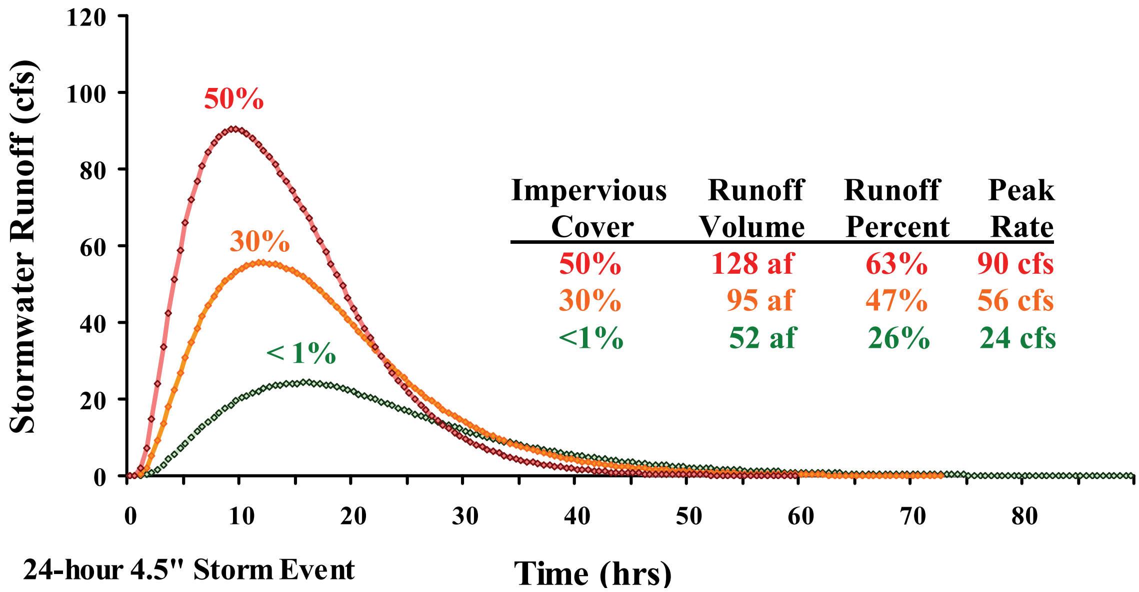

Figure 34. Hydrographs of modeled stormwater runoff from Guerin Creek, a forested study watershed. Runoff is shown at the current level of impervious cover (<1%) and also at levels of 30% and 50%. cfs = cubic feet per second, af.= acre-foot.

4.2 Conceptual Model

As noted above, tidal creek networks are the primary hydrologic link between estuaries and land based activities. As the first zone of impact for non-point source pollution runoff, the potential for microbial and chemical contamination in tidal creek habitats is great. Developing a conceptual model is a critical step for identifying and evaluating monitoring and management strategies including what parameters to measure and when and where measurements should be taken (Saila 1979, NRC 1990, Barnthouse and Brown 1994). Holland et al. (2004) developed a conceptual model to describe the source-receptor links between the origin of an environmental problem (e.g., human activity, extreme natural event, linkages between ocean processes) and anticipated impacts on ecosystems. The model was based on the EPA Ecological Risk Assessment paradigm with stressors leading to changes in the physical-chemical environment (i.e., exposures) which in turn leads to a biological response.

The Holland et al. (2004) conceptual model for intertidally-dominated tidal creeks in South Carolina did not include a number of new indicators (e.g., nutrients, emerging contaminants of concern) or show the relationship the environment has on the health and welfare of humans. Historically, scientists have only looked at human impacts on the environment but are now beginning to recognize how environmental changes impact humans. Based on the data collected by this study, the conceptual source-receptor model has been expanded (Figure 35). This model provides an overview of the linkages between coastal development and associated human activities, "key" changes in the physical-chemical environment, and anticipated responses of tidal creek ecosystems and human systems. This model continues to be updated and revised as new data and information become available. This model was developed using an integrated weight of evidence approach based on the information collected. The stressor or coastal development activities in the surrounding watersheds found in this study include changes in the land cover and increases in the population density and impervious cover as development occurs.

The exposure or changes in the physical chemical environment associated with increasing development include increases in the salinity range, levels of $NO_{2/3}$ and NH_{4+}, the amount of stormwater runoff, bacterial and viral pathogen indicators, and chemical contamination of the sediment including some emerging chemicals of concern with increasing development. The biological response or impacts on the living resources identified in this study include impacts on the macrobenthic communities such as changes in the species composition, number of organisms and diversity with increasing levels of development and impaired oyster health as evidenced by the changes in gene expression. Finally, the societal response of impacts to the health and well-being observed in this study include increased flooding from increases in development and increased public health risk from bacteria in the water and shellfish.

The tidal creek conceptual model identified adverse changes generally occurred in the physical and chemical environment (e.g., water quality indicators such as indicator bacteria for sewage pollution or sediment chemical contamination) when impervious cover levels in the watershed reach 10-20%. Ecological processes responded to and were generally impaired when impervious cover levels exceeded 20-30% in suburban and urban watersheds (Figure 35). There is an emerging consensus that patterns of coastal development are associated with evidence of increasing fecal pollution in tidal creeks, estuaries, and bathing beaches (this study, Mallin et al. 2000, Karn and Harada 2001, Holland et al. 2004, Mallin 2006). From a human health perspective, the accumulation of pathogens in the water, sediments, and organisms may render

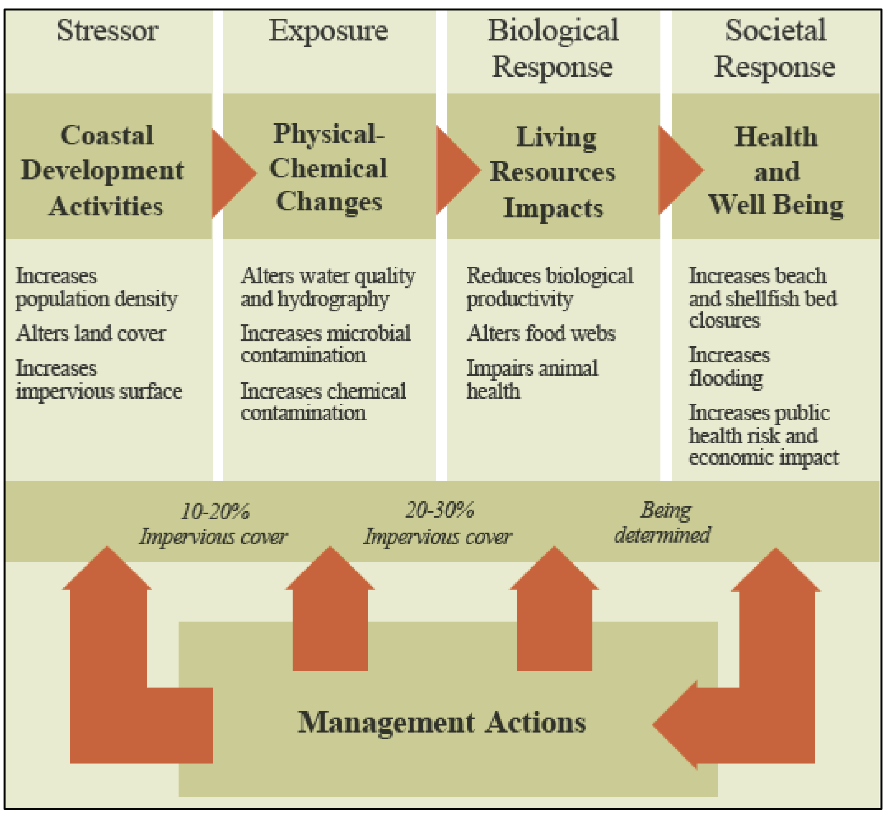

Figure 35. Conceptual model identifying linkages between development of the upland and ecological and human well-being of southeastern U.S. tidal creeks. Ranges of impervious cover percent denoting transition from one model phase to the next are shown toward the bottom of the model.

seafood products unsafe to eat and water unsafe for contact recreation. Flooding vulnerability, public health risk, and economic impacts are also being considered. Estimates of impervious cover levels defining where human uses are impaired are currently being determined, but it generally appears that shellfish bed closures and the flooding vulnerability of headwater regions become a concern when impervious cover values exceed 10-30%. This research project has validated the model for the southeastern U.S.

The tidal creek conceptual model also provides a framework for defining system feedback loops and identifying which level of government is responsible for ensuring appropriate actions are taken to remediate and restore impaired systems (Figure 35). County and municipal governments are responsible for regulating land use activities and making most zoning decisions and are the agencies controlling impervious cover levels. State and federal governments mainly influence physical-chemical exposures (water and sediment quality) but also play a large role in enforcement and permitting activities related to near-marsh development.

A pamphlet was created to summarize this tidal creek research for decision-makers including resource managers and land use planners. The NERRS has strong education and stewardship programs through its Coastal Training Program (CTP) and Stewardship Coordinators. Therefore, an opportunity exists to work with the North Carolina and Sapelo Island NERRs to provide this information to the public and decision makers through presentations and the pamphlet. The information has been presented to numerous audiences to encourage wise decision making as well as encourage the general public, elected officials, and county/city administrators to become stewards of the environment.

4.3 NERRs as Regional References

Much of our understanding of the impacts of anthropogenic activities on natural environments relies heavily on the concept of reference sites or the reference condition (Stoddard et al. 2006). For stream ecosystems, for example, biological criteria used to characterize the quality of a stream segment are developed by comparison to a population of reference sites (e.g., Hughes 1995, Barbour et al. 1995). No such effort has been made to document appropriate coastal reference sites to evaluate the impacts of human activity in the coastal zone and adjacent marine waters. This study, and the partnership with the NERRS, was undertaken in part to explore tidal creeks as sentinel habitats and to evaluate whether this network of protected coastal habitats could serve as reference sites for future research.

The two NERRs, North Carolina and Sapelo Island, that were sampled as part of this study were found to be reasonable regional references; for many of the sampled parameters, data showed that creeks within (or adjacent to) these NERRs differed from the typical condition of other suburban or urban creeks. However, in some cases, the land use (both current and historical) within and around these areas appears to impact the reserves and may diminish their capacity as regional references. For example, Oakdale Creek within the Sapelo Island NERR (accessible only by boat) had high levels of a number of the pathogen indicators in both water and oyster tissues. The sources of these pathogens are unidentified; however, there are many possibilities. Free ranging cattle and high densities of whitetail deer roam the island, a large stork and egret rookery is located in the immediate sampling vicinity, a small Gullah-Geechee (descendants of West African slaves) community is located upland of the sampling site, and the island is also occupied by research and management communities and visitors. Determining the sources of these pathogens is important to develop future management decisions (e.g., excluding cattle from salt marshes/tidal creeks, septic maintenance programs) regarding these habitats and their watersheds. In addition, land use decisions outside of the NERR boundaries can also influence the habitats within the Reserves. For example, the suburban development adjacent to the North Carolina Masonboro site has the potential to impact the reserve. Educating decision-makers on land use change is an important activity to ensure that the potential impacts of the land use decisions are considered.

The dolphin contaminant study highlighted in this report also identifies other considerations in using the NERRs as regional references. Dolphin tissue levels of PCBs around the Sapelo Island NERR were found to be high compared to most other areas previously studied (Hansen et al. 2004, Litz et al. 2007); however, it is unknown if these levels are due to dolphin or prey movements between contaminated sites (e.g., Turtle River/Brunswick) and the Sapelo Island NERR. The research on dolphin tissue levels and migration patterns in these areas is continuing.

4.4 Forecasting

The relationship between watershed development and the ecological condition of the headwater areas of tidal creeks in SC is fairly well-understood (Sanger et al. 1999a, 1999b, Lerberg et al. 2000, Holland et al. 2004), but spatial and temporal variability and patterns in ecological condition along tidal creek networks are poorly characterized. Effective monitoring, assessment, and prediction of the effects of coastal urbanization on tidal creeks and estuaries require that this variability be understood. Stratification of tidal creek networks into units that represent relatively homogenous environments or creek classes is one tool for characterizing and understanding the variability within tidal creek networks. This stratification is crucial for understanding at what scale land use impacts can be observed. Classifying watersheds that drain into specific creek networks based on the degree and type of development that has occurred is a tool and requirement for understanding variability among creek networks and forecasting the impacts of development.

In addition to using a classification system, other metrics must also be added to provide forecasting ability. One particularly important component is modeling the volume and rate of runoff leaving the upland. Smith (2005) found that runoff into creeks with developed watersheds occurred over hours immediately following the rain event compared to a reference creek with an undeveloped watershed where runoff occurred more slowly over a period of a day. However, measuring the volume and rate of runoff entering into tidal creeks is an expensive and time consuming activity. Therefore, other opportunities such as modeling the volume and rate of runoff need to be explored to determine the potential impact of the volume and rate of water entering into tidal creek ecosystems. Existing models such as the Natural Resources Conservation Service's flow curve number and dimensionless unit hydrograph methods have been modified for the southeastern US region to estimate the runoff volume and rate for a variety of rainfall events and under different development scenarios (i.e., changes in the impervious cover) (Figure 34). This modeling of stormwater runoff entering headwater tidal creeks will allow for future predictions of the runoff and pollutant loadings as watershed impervious cover levels are increased. The runoff modeling used in this study is being validated and future work includes the development of a tool that resource managers can use to evaluate the impact of development on a tidal creek.

Most importantly, the tidal creek study and conceptual model provide a framework for forecasting the changes in ecological and public health indicators based on watershed attributes and impervious cover associated with coastal development. Ecological forecasting has begun to emerge as an organizing principle in many research efforts (Clark et al. 2001). For example, with freshwater resources stretched thin in many areas of the world, the effects of urbanization on stream ecosystems is a critical area for research on forecasting. The opportunities are great, but integrating traditionally disparate disciplines to develop appropriate forecasting models is a challenge (Nilsson et al. 2003). The tidal creek research described here provides a roadmap for forecasting. Information on watersheds and the impervious surface associated with coastal development (Figure 36a) relates strongly to hydrographic characteristics of creeks (Figure 36b) and ultimately on indicators of water quality and ecosystem condition such as bacterial indicators (Figure 36c). Based on these relationships, it is possible to use simple linear relationships to forecast the likely effects that increasing impervious cover may have on critical indicators of the ecosystem and public health and well-being (Figure 36d). The forecast models can be used by planners, developers, and managers to understand the potential impacts of a development before building instead of years after (Clark et al. 2001).

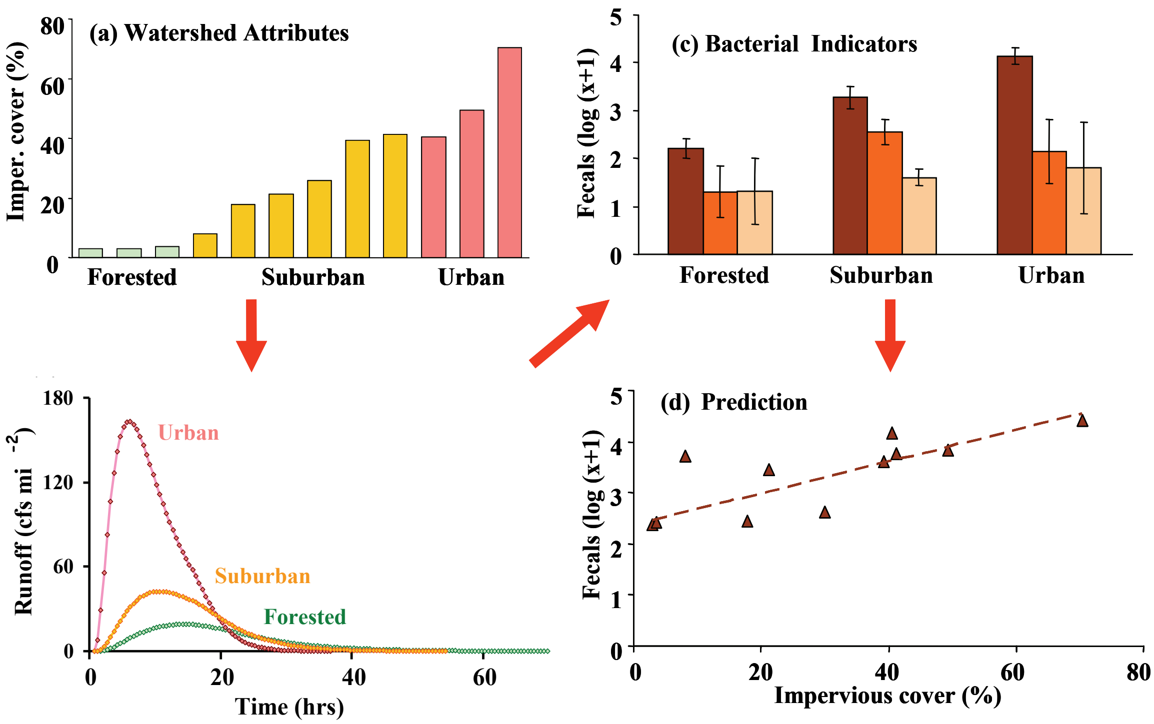

Figure 36. Graphical presentation forecasting the impacts of land use change on tidal creek environmental quality.

4.5 Summary

In the Southeastern US, coastal uplands adjacent to tidal creeks and salt marshes are increasingly popular locations for building new homes, resorts, retirement destinations, and recreational facilities. These tidal creek networks are also critical feeding grounds, spawning areas, and nursery habitats for many species of fish, shellfish, birds, and mammals. Tidal creeks also form the primary hydrologic link between estuaries and land-based activities and, as such, reflect the impacts of coastal development earlier than larger coastal waterbodies. Nonpoint source pollution (e.g., stormwater runoff) carries sediments, chemicals, bacteria, viruses, and other pollutants into tidal creeks and salt marshes and degrades water quality. The relationships between development levels and the environmental quality and public health and well-being indicators evaluated were strongest in the shallow, intertidally-dominated headwater creeks.

The scale of our tidal creek study watersheds (100s to 1000s ha) is also the spatial scale at which coastal land use decisions and remediation actions typically occur. If creeks draining watersheds this size, especially the headwater portions of those watersheds, are valuable indicators of impacts from land use activities and urbanization, managers or land use planners are afforded a valuable tool to predict the impacts of developments on pathogen indicator concentrations in nearby tidal creeks and thereby inform the decision-making process.

5. Overall Project Summary and Conclusions

The present report is part of a two-volume set summarizing results of a collaborative NCCOS-NERRS effort to assess the status of ecosystem conditions and potential stressor impacts at NERRS sites in the southeastern U.S., and to provide this information as a basis for monitoring future conditions in these same areas or in other NERRS locations. There are two complementary components of this overall initial effort: (1) a sentinel habitats study designed to evaluate the impacts of development on tidal creek ecosystems, including potential impacts to human health and well-being; and (2) a probabilistic monitoring component to assess the spatial extent of ecological condition throughout sub-tidal estuarine waters, based on the status of various measured ecological indicators relative to specific management thresholds. The tidal creek component, discussed in the present Volume I, was conducted on NERRS sites at Sapelo Island, Georgia and Masonboro Island, North Carolina and was coordinated with results of related tidal creek work in South Carolina. The sub-tidal probabilistic component, discussed in Volume II (Cooksey et al. 2008), was conducted throughout all four North Carolina NERR locations (Currituck Sound, Rachel Carson, Masonboro Island, and Zeke's Island). Together, the two project components are intended to provide a demonstration of the utility of the complementary assessment tools, one serving as a sentinel of environmental signals in areas of estuaries where signals are likely to occur (i.e., tidal creeks close to pollutant sources), and the other providing a quantitative basis for assessing the relative proportions of degraded vs. non-degraded conditions throughout a targeted resource category (i.e., sub-tidal estuarine waters of a reserve) relative to the various measured indicators and associated management thresholds. While providing new information on the status of ecological condition and human health risks in several southeastern U.S. NERRS locations, the results also are intended to serve as a useful framework of assessment strategies that could be applied systematically across other reserves to support broader regional and national comparisons.

5.1 Summary Points from Volume I: The Impacts of Coastal Development on the Ecology and Human Well-being of Tidal Creek Ecosystems of the US Southeast (Present Volume)

Southeastern tidal creeks are sensitive to coastal development and provide an early warning of potential degradation from upland land uses well before adverse conditions would be detected in larger coastal waters (e.g., tidal rivers, bays). Accounting for the spatial variability in hydrological and other watershed attributes among individual tidal creek systems is an important factor to consider in assessing the environmental quality of these habitats. Thus, the application of an appropriate tidal creek classification scheme in the sampling design process to accommodate such variability was a critical aspect of the research described in Volume I. One important finding of this research was that the sensitivity of tidal creeks to changes in the environmental quality of the surrounding watersheds diminishes downstream toward the mouths of the creeks. Smaller intertidal creeks generally had higher concentrations for indicators of non-point source pollutants (e.g., water quality, nutrients, and pathogen indicators), which likely reflect both the greater upland runoff component and estuarine dilution influence (i.e., tidal flushing) associated with larger creeks. Additionally, indicators of deteriorating environmental quality were found to vary directly with increasing levels of impervious cover, the latter allowing greater inputs of runoff and associated pollutants from the surrounding watershed into tidal creeks, particularly in headwater regions. The integrity and productivity of headwater portions of tidal creek environments are often impaired by land use changes and associated non-point source pollution, suggesting that these habitats serve as valuable early-warning sentinels of ensuing stress including ecological and potential public health threats (e.g., seafood consumption advisories, swimming advisories). A conceptual model of these linkages was validated and expanded for the southeastern US. Lastly, the tidal creek study and its associated conceptual model provide a useful framework for forecasting potential changes in ecological and human health indicators within these systems in relation to varying watershed attributes and land use patterns.

5.2 Summary Points from Volume II: Assessing Ecological Condition and Stressor Impacts in Subtidal Waters of the North Carolina NERRS (from Cooksey et al. 2008.)

This component of the project was aimed at assessing the status of ecological condition and stressor impacts in subtidal estuarine waters throughout the four North Carolina NERR locations (Currituck Sound, Rachel Carson, Masonboro Island, and Zeke's Island). Sampling incorporated multiple indicators of ecosystem condition including measures of water quality, sediment quality, biological condition, and potential threats to human health and well-being (e.g., fish-tissue contaminant levels relative to human health consumption limits, various aesthetic properties). A probabilistic sampling design permitted statistical estimation of the spatial extent of degraded versus non-degraded condition across these estuaries relative to specified threshold levels of the various indicators (where possible). With some exceptions, the status of this reserve appeared to be in relatively good to fair condition overall, with the majority of the area (about 54%) having various water quality, sediment quality, and biological (benthic) condition indicators rated in the healthy to intermediate range of corresponding guideline thresholds. Only three of the 30 stations sampled, representing 10.5% of the area, had one or more of these indicators rated as poor/degraded in all three categories. However, although co-occurrences of adverse biological and abiotic environmental conditions were limited spatially, at least one indicator of ecological condition rated in the poor/degraded range was observed over a broader area (35.5% represented by 11 stations). In addition, fish-tissue contaminant data were not included in these overall spatial estimates; however, the majority of samples (77% of fish that were analyzed, from 79%, of stations where fish were caught) contained inorganic arsenic above the consumption limits for human cancer risks, though most likely derived from natural sources. Such symptoms reflect a growing realization that North Carolina estuaries are under multiple pressures from a variety of natural and human influences. These data also suggest that, while the current status of overall ecological condition appears to be good to fair, long-term monitoring is warranted to track potential changes in the future. This study establishes an important baseline of overall ecological condition within the NC NERR that can be used to evaluate any such future changes and to trigger appropriate management actions in this rapidly evolving coastal environment.

5.3 Coastal Management Applications and Opportunities

NCCOS's mission, as defined in its FY05-09 Strategic Plan (NCCOS 2004), is to provide coastal managers with scientific information and tools needed to balance society's environmental, social, and economic goals. The NCCOS Strategic Plan also calls for baseline assessments of ecological resources and potential stressor impacts in NERRS and other NOAA protected areas. The NERRS' mission is to practice and promote coastal and estuarine stewardship through innovative research and education activities focused on the NERRS (NERRS 2005). The present collaborative studies sought to address common research and management goals supportive of both program missions. The two studies provide new information on the current status of the ecological condition and human health risks at NERRS sites and neighboring waters in Georgia, South Carolina, and North Carolina as well as a set of complementary assessment tools for monitoring future conditions in these same areas or in other NERRS locations. The tidal creek study and its associated conceptual model (Volume I) provide a framework for forecasting potential changes from coastal development on the ecological and human health and well-being within these systems. The tidal creek study also exemplifies the utility of these habitats as a sentinel of environmental signals in areas of estuaries (e.g., in upper reaches close to pollutant sources) where signals are most likely to occur. The probabilistic sampling approach used in the subtidal assessment (Volume II) provides an additional unbiased statistical basis for quantifying the spatial extent of condition relative to the various measured indicators and desired management thresholds, throughout a targeted resource category, and thus a quantitative baseline for monitoring how the relative proportions of healthy vs. degraded areas may be changing with time. Thus, in addition to providing new information on the status of ecosystem conditions in specific southeastern U.S. NERRS locations, the results also are intended to serve as a framework of assessment strategies that could be applied systematically across other reserves to support national comparisons. Such ecological assessment tools would also complement system-wide, water-quality monitoring program (SWMP) and other site-specific research activities currently underway in the NERRS program.

Such baseline assessments of the status of ecosystem conditions and stressor impacts also provide the first steps in the implementation of Integrated Ecosystem Assessments (IEAs) of NERRS sites. An IEA is a synthesis and quantitative analysis of information on relevant natural and socio-economic factors in relation to specified ecosystem management goals (NOAA 2007, Murawski and Menashes 2007). The NERRS, as a system of protected areas, offers an ideal series of place-based sites for an IEA, which begins with the assessment of baseline conditions defining current ecosystem status, as well as the assessment of stressor impacts and their links to source drivers and pressures (as was the focus here). NOAA has placed an emphasis on conducting IEAs to support improved ecosystem approaches to management (EAM) within its protected coastal resources in order to offer coastal managers a more comprehensive framework to their coastal decision making. Such an approach requires increased understanding of these complex systems and improved integration and collaboration in their management.

Related to the above point, as a NOAA protected resource, the NERRS offers an ideal opportunity to become a suite of place-based reference sites across the nation for documenting status and trends in coastal ecosystem conditions among reserves and in comparison to other non-protected areas. Results of the present two studies have provided new information on the status of conditions at NERRS sites in the southeastern U.S. These areas were found to be reasonable regional references, with the caveat that some symptoms of stress were detectable and thus the realization that such areas and surrounding watersheds are under multiple pressures from a variety of natural and human influences. Long-term monitoring is warranted to track potential changes in the future. The data and assessment strategies provided here can be applied in any such efforts for these same areas, as well as in any future surveys in other NERRS sites to support broader-scale regional and national comparisons.

Another underlying project goal is to make the present information and assessment tools readily available for meeting NERRS research and management needs. There is a tremendous opportunity to achieve this goal through the educational and outreach resources of NERRS, including their strong education programs, Coastal Training Programs (CTP), and stewardship coordinators. Any related efforts to inform the public and coastal management community through targeted presentations and products derived from these studies should help to expand their utility toward addressing important coastal management and human health concerns.

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Appendix A. The expected range of method detection limits (MDLs) based on extracted sample mass for the sediment and oyster tissue contaminant analyses.

The units for sediment metals and organics are µg g^{-1} dry weight and ng g^{-1} dry weight, respectively. The units for oyster tissue metals and organics are µg g^{-1} dry weight and ng g^{-1} dry weight, respectively.

Analyte	Sediment MDL Range	Tissue MDL Range
Aluminum	0.008 - 0.090	5.35 - 10
Antimony	0.786 - 0.804	0.196 - 0.368
Arsenic	0.091 - 0.933	0.023 - 0.124
Barium	0.108 - 1.1	0.026 - 0.050
Beryllium	0.159 - 0.167	0.019 - 0.037
Cadmium	0.0036 - 0.0038	0.002 - 0.004
Chromium	0.161 - 1.64	0.201 - 0.376
Cobalt	0.079 - 0.082	0.049 - 0.093
Copper	6.26 - 6.56	0.779 - 6.49
Iron	0.051 - 0.532	31.6 - 59.1
Lead	0.051 - 0.518	0.012 - 0.024
Lithium	0.652 - 0.683	0.081 - 0.152
Manganese	4.96 - 5.2	0.617 - 1.16
Mercury	0.0006 - 0.008	0.006 - 0.014
Nickel	1.1 - 1.16	0.027 - 0.051
Selenium	0.161 - 0.165	0.103 - 0.194
Silver	0.269 - 0.282	0.18 - 1.12
Thallium	0.030 - 0.032	0.019 - 0.036
Tin	0.141 - 0.144	0.035 - 0.066
Uranium	0.047 - 0.048	0.029 - 0.055
Vanadium	0.06 - 0.061	0.015 - 0.028
Zinc	16 - 16.8	99.5 - 186
2,4'-DDD	0.315 - 1.32	5.11 - 8.98
2,4'-DDE	0.0787 - 0.33	1.28 - 2.25
2,4'-DDT	0.355 - 1.49	5.76 - 10.1
4,4'-DDD	0.265 - 1.11	4.29 - 7.55
4,4'-DDE	0.263 - 1.1	4.26 - 7.49
4,4'-DDT	0.789 - 3.31	12.8 - 22.5
Aldrin	0.527 - 2.21	8.56 - 15
Hexachlorobenzene	0.214 - 0.897	3.47 - 6.1
Chlorpyrifos	0.0447 - 0.187	0.724 - 1.27
cis-Chlordane	1.39 - 5.85	22.6 - 39.8
Dieldrin	0.116 - 0.486	1.88 - 3.31
Endosulfan I	0.127 - 0.531	2.05 - 3.61
Endosulfan II	0.26 - 1.09	4.23 - 7.43
Endosulfan sulfate	0.38 - 1.59	6.16 - 10.8
Heptachlor	0.205 - 0.861	3.33 - 5.86
Heptachlor epoxide	0.585 - 2.45	9.49 - 16.7
Gamma-HCH (g-BHC, lindane)	0.087 - 0.366	1.41 - 2.49
Mirex	0.178 - 0.745	2.88 - 5.07
Trans-nonachlor	1.43 - 6	23.2 - 40.8
PBDE 100	0.124 - 0.522	2.02 - 3.55
PBDE 138	0.372 - 1.56	6.04 - 10.6

Appendix A (cont.)

Analyte	Sediment MDL Range	Tissue MDL Range
PBDE 153	0.119 - 0.5	1.93 - 3.4
PBDE 154	0.685 - 2.87	11.1 - 19.5
PBDE 17	0.106 - 0.446	1.72 - 3.03
PBDE 183	0.176 - 0.741	2.86 - 5.04
PBDE 190	0.735 - 3.08	11.9 - 21
PBDE 28	0.158 - 0.665	2.57 - 4.52
PBDE 47	0.142 - 0.598	2.31 - 4.07
PBDE 66	0.134 - 0.562	2.17 - 3.82
PBDE 71	0.14 - 0.589	2.28 - 4
PBDE 85	0.732 - 3.07	11.9 - 20.9
PBDE 99	0.134 - 0.562	2.17 - 3.82
PCB 101	0.157 - 0.66	2.55 - 4.49
PCB 103	0.148 - 0.62	2.4 - 4.22
PCB 104	0.128 - 0.535	2.07 - 3.64
PCB 105	0.14 - 0.589	2.28 - 4
PCB 107/108	0.272 - 1.14	4.42 - 7.77
PCB 110	0.197 - 0.825	3.19 - 5.61
PCB 114	0.231 - 0.968	3.74 - 6.58
PCB 118	0.485 - 2.03	7.87 - 13.8
PCB 119	0.234 - 0.981	3.79 - 6.67
PCB 12	0.189 - 0.794	3.07 - 5.4
PCB 123	0.926 - 3.89	15 - 26.4
PCB 126	0.164 - 0.687	2.66 - 4.67
PCB 128	0.095 - 0.401	1.55 - 2.73
PCB 130	0.174 - 0.732	2.83 - 4.98
PCB 132/168	0.161 - 0.674	2.6 - 4.58
PCB 138	0.089 - 0.375	1.45 - 2.55
PCB 141	0.125 - 0.526	2.04 - 3.58
PCB 146	0.473 - 1.99	7.68 - 13.5
PCB 149	0.319 - 1.34	5.17 - 9.1
PCB 15	0.165 - 0.691	2.67 - 4.7
PCB 151	0.151 - 0.633	2.45 - 4.31
PCB 153	0.394 - 1.66	6.4 - 11.3
PCB 154	0.14 - 0.589	2.28 - 4
PCB 156	0.12 - 0.504	1.95 - 3.43
PCB 157	0.11 - 0.459	1.78 - 3.13
PCB 158	0.183 - 0.767	2.97 - 5.22
PCB 159	0.090 - 0.379	1.47 - 2.58
PCB 169	0.119 - 0.5	1.93 - 3.4
PCB 170	0.14 - 0.589	2.28 - 4
PCB 172	0.13 - 0.544	2.1 - 3.7
PCB 174	0.154 - 0.647	2.5 - 4.4
PCB 177	0.169 - 0.709	2.74 - 4.82
PCB 18	0.422 - 1.77	6.85 - 12
PCB 180	0.115 - 0.482	1.86 - 3.28
PCB 183	0.149 - 0.625	2.41 - 4.25
PCB 184	0.102 - 0.428	1.66 - 2.91

Appendix A (cont.)

Analyte	Sediment MDL Range	Tissue MDL Range
PCB 187	0.086 - 0.361	1.4 - 2.46
PCB 188	0.092 - 0.388	1.5 - 2.64
PCB 189	0.129 - 0.54	2.09 - 3.67
PCB 193	0.191 - 0.803	3.1 - 5.46
PCB 194	0.087 - 0.366	1.41 - 2.49
PCB 195	0.254 - 1.07	4.12 - 7.25
PCB 198	0.173 - 0.727	2.81 - 4.95
PCB 2	0.146 - 0.611	2.36 - 4.16
PCB 20	0.134 - 0.562	2.17 - 3.82
PCB 201	0.117 - 0.491	1.9 - 3.34
PCB 202	0.115 - 0.482	1.86 - 3.28
PCB 206	0.103 - 0.433	1.67 - 2.94
PCB 207	0.296 - 1.24	4.79 - 8.43
PCB 209	0.116 - 0.486	1.88 - 3.31
PCB 26	0.125 - 0.526	2.04 - 3.58
PCB 28	0.383 - 1.61	6.21 - 10.9
PCB 29	0.224 - 0.941	3.64 - 6.4
PCB 3	0.098 - 0.415	1.6 - 2.82
PCB 31	0.325 - 1.37	5.28 - 9.28
PCB 37	0.184 - 0.772	2.98 - 5.25
PCB 44	0.116 - 0.486	1.88 - 3.31
PCB 45	0.231 - 0.968	3.74 - 6.58
PCB 48	0.107 - 0.451	1.74 - 3.06
PCB 50	0.21 - 0.883	3.42 - 6.01
PCB 52	0.146 - 0.611	2.36 - 4.16
PCB 56/60	0.216 - 0.906	3.5 - 6.16
PCB 61/74	0.285 - 1.2	4.62 - 8.13
PCB 63	0.218 - 0.915	3.54 - 6.22
PCB 66	0.215 - 0.901	3.48 - 6.13
PCB 69	0.372 - 1.56	6.04 - 10.6
PCB 70	0.44 - 1.85	7.14 - 12.6
PCB 76	0.292 - 1.23	4.74 - 8.34
PCB 77	0.156 - 0.656	2.54 - 4.46
PCB 8	0.367 - 1.54	5.95 - 10.5
PCB 81	0.188 - 0.79	3.05 - 5.37
PCB 82	0.215 - 0.901	3.48 - 6.13
PCB 84	0.342 - 1.44	5.55 - 9.77
PCB 87	0.203 - 0.852	3.29 - 5.8
PCB 88	0.219 - 0.919	3.55 - 6.25
PCB 9	0.403 - 1.69	6.54 - 11.5
PCB 92	0.134 - 0.562	2.17 - 3.82
PCB 95	0.102 - 0.428	1.66 - 2.91
PCB 99	0.182 - 0.763	2.95 - 5.19
Acenaphthene	0.048 - 21.97	117.76 - 207.16
Acenaphthylene	0.064 - 7.63	40.88 - 71.91
Anthracene	0.063 - 8.63	46.25 - 81.36
Benzo(b)fluoranthene	0.063 - 4.6	24.68 - 43.41
Benzo(a)anthracene	0.065 - 5.42	29.05 - 51.1

Appendix A (cont.)

Analyte	Sediment MDL Range	Tissue MDL Range
Benzo(a)pyrene	0.054 - 13.61	72.97 - 128.36
Benzo(e)pyrene	0.054 - 16.72	89.64 - 157.68
Benzo(g,h,i)perylene	0.051 - 9.16	49.07 - 86.32
Benzo(k+j)fluoranthene	0.133 - 11.87	63.62 - 111.91
Biphenyl	3.49 - 8.61	46.15 - 81.18
Chrysene+Triphenylene	0.047 - 14.07	75.39 - 132.63
Dibenz(a,h)anthracene	0.158 - 17.94	96.17 - 169.17
Dibenzothiophene	0.043 - 8.27	44.34 - 77.99
Fluoranthene	24.62 - 60.71	325.39 - 572.4
Fluorene	0.052 - 22.97	123.11 - 216.56
Naphthalene	0.037 - 4.64	24.88 - 43.77
1,6,7 Trimethylnaphthalene	0.123 - 32.6	174.73 - 307.36
1-Methylnaphthalene	0.036 - 17.35	93 - 163.6
2,6 Dimethylnaphthalene	0.03 - 18.7	100.23 - 176.31
2-Methylnaphthalene	0.059 - 27.06	145.02 - 255.1
Indeno(1,2,3-cd)pyrene	0.167 - 22.72	121.78 - 214.22
Perylene	2.33 - 5.75	30.84 - 54.25
Phenanthrene	4.06 - 10.02	53.7 - 94.46
1-Methylphenanthrene	0.037 - 11.12	59.59 - 104.83
Pyrene	4.62 - 11.38	61.01 - 107.32

Appendix B. The 24 analytes used to calculate the mean ERM quotient. For total PCB and DDT, refer to Appendix A for the individual analytes used.

Analyte
Arsenic
Cadmium
Chromium
Copper
Lead
Mercury
Silver
Zinc
4,4'-DDE
Total DDT
Acenaphthene
Acenaphthylene
Anthracene
Benzo(a)anthracene
Benzo(a)pyrene
Chrysene+Triphenylene
Dibenz(a,h)anthracene
Fluoranthene
Fluorene
Naphthalene
2-Methylnaphthalene
Phenanthrene
Pyrene
Total PCB

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