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EPISODIC NUTRIENT LOADING IMPACTS ON EUTROPHICATION OF THE SOUTHERN PAMLICO SOUND: THE EFFECTS OF THE 1999 HURRICANES

by
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ABSTRACT

Extreme hydrologic events, such as the sequential hurricanes Dennis, Floyd and Irene in the fall of 1999, disrupt the filtering capacity of the sub-estuaries of Albemarle and Pamlico Sounds and may allow considerable nutrient loading to the sounds and adjacent coastal waters. We began an extensive monitoring program of water quality in southern Pamlico Sound following the storms and flooding in 1999 and continued sampling through May 2002. We tracked hydrographic variables, concentrations of dissolved and particulate forms of carbon, nitrogen and phosphorus, and several indicators of the phytoplankton community to evaluate mechanisms by which the sound could be episodically eutrophied by storms and flooding. The sound was diluted with freshwater and required nearly two years to return to salinities greater than 20 psu. Dissolved inorganic nutrients were elevated shortly after the storms and quickly decreased to low concentrations. Particulate matter and chlorophyll *a* concentrations increased after the storms and remained elevated for months. Blooms of phytoplankton may have developed in subsequent seasons and promoted nutrient cycling. Temporal changes were found in phytoplankton taxonomic composition, chemically defined by pigments, with freshwater forms being more numerous shortly after the storms. Overall, significant amounts of nutrients were inferred to enter the sound in dissolved inorganic, particulate and planktonic forms. Loading of sediments and subsequent remineralization of associated nutrients may have occurred but could not be readily demonstrated. Our monitoring program has provided a significantly improved understanding of the water quality of Pamlico Sound compared to previous studies. We used our findings to recommend a series of steps to enhance this understanding in the future.

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SUMMARY AND CONCLUSIONS

We conducted a thirty-one month study on the water quality of southern Pamlico Sound. This study was prompted by the series of hurricanes and subsequent flooding that occurred in the fall of 1999 and addresses several hypotheses concerning how nutrients move into the sound from its sub-estuaries and how these nutrients are processed. The extended period of monitoring has also allowed us to (1) establish the current environmental and water quality conditions of the southern sound and relate these conditions to the ability of the sub-estuaries to protect the sounds from nutrient loading, (2) infer mechanisms by which the sound may be nutrified during extreme hydrologic events, and (3) establish a baseline against which to gauge ecological change.

We postulated that the mechanism by which nutrients entered the sound could be inferred by the response of standing stocks of materials in the sound subsequent to the storms of 1999. The sound may receive this episodic loading by at least 3 mechanisms: (1) Flow-through of unprocessed nutrients, (2) Resuspension and transport of sediments with subsequent biological and chemical release of nutrients, and (3) Movement of phytoplankton blooms down the sub-estuary to near the mouth allowing algal biomass to enter the sound for processing. Nutrients were inferred to enter the sound in dissolved inorganic, particulate and planktonic forms. Dissolved inorganic nutrients quickly decreased to low concentrations. Blooms of phytoplankton appeared to continue through subsequent seasons and promoted nutrient cycling. Loading of sediments and subsequent remineralization of associated nutrients may have occurred but could not be readily demonstrated.

The study reported on here is one of the first to reveal the ecological response of a segment of Pamlico Sound to the combined effects of anthropogenic nutrient inputs and climatic perturbations, in the form of 3 sequential hurricanes in fall 1999. The southern portion of Pamlico Sound appears to have better water quality than its sub-estuaries, as defined by the concentrations of nutrients and chlorophyll *a*. It has the capacity for hypoxia during summer months, although the extent could not be determined by our sampling. The system can be perturbed during extreme hydrologic events as experienced in fall 1999 and these events are predicted to increase over the next few decades. Nutrient concentrations recovered quickly, but algal blooms continued for much of the following year. We cannot predict how frequently the sound could respond to repetition of such events with such resilience. Only continued study and better understanding will provide such predictive ability. While the comprehensiveness of the spatial and temporal coverage of Pamlico Sound was restricted due to limited funding and logistic constraints, information was gained as a guideline for filling future water quality monitoring needs.

RECOMMENDATIONS

Pamlico Sound and its sub-estuaries form the second largest estuarine system in the United States. Arguably, this is one of the country's most important fisheries, nursery and recreational assets. It is under increasing pressure from growing human development in its watersheds and is at the forefront of climatic change, including a predicted increase in both the frequency and intensity of tropical storms and hurricanes (Goldenberg et al. 2001), and sea level rise due to global warming. As such, it is imperative and timely that we obtain scientifically based information on water quality and habitat condition in order to develop a baseline against which we can gauge anticipated changes. Despite the sound's enormous ecological and economic importance and the lack of knowledge of how this system functions and responds to external perturbations, neither the State of North Carolina nor the federal government has routine water quality monitoring and assessment programs in place to fill these important needs. It is unfortunate that at this important juncture of man-induced and climatic change, Pamlico Sound is the United States' largest coastal body of water of which we know the least.

This study is, therefore, one of the first to reveal the ecological response of a segment of this large system to the combined effects of anthropogenic nutrient inputs and climatic perturbations, in the form of 3 sequential hurricanes that struck in fall 1999. While the comprehensiveness of the spatial and temporal coverage of Pamlico Sound was restricted due to limited funding and logistic constraints, the following information was gained as a guideline for filling essential water quality monitoring needs:

- Given the observed spatial gradients in nutrient loads and concentrations, as well as phytoplankton productivity, biomass and community compositional responses among the stations employed in this study, it is essential to develop sampling transects spanning the major tributaries (Neuse and Pamlico River estuaries, Chowan-Roanoke Rivers and Albemarle Sound) to the inlets at the Outer Banks. The prescribed number of sampling stations remains undetermined, but should include at least 4 to 6 stations in each of the sounds' sub-basins (southern, western and northern), positioned in such a manner that both large-scale circulation and effects of outflows from the sub-estuaries can be captured.

- Temporally, it is essential that seasonal patterns and shifts as well as episodic events in nutrient loading, primary productivity, floral and faunal compositional responses and benthic biogeochemical conditions (i.e., hypoxia, anoxia) be accounted for. Based on this study, we recommend at least bi-monthly sampling, possibly concentrated during the spring through fall months (March-November). These sampling frequencies will ensure adequate assessments of nutrient loads (determining nutrient fluxes and budgets), biological responses to nutrient loads, and community succession patterns.

- The sound-wide monitoring program should be coordinated in time and space to take advantage of other efforts that gauge the ecological condition of Pamlico Sound. These include the University of North Carolina (UNC)/Duke University/North Carolina

Department of Environment and Natural Resources (NCDENR)/Department of Transportation (DOT) ferry-based water quality monitoring program (FerryMon; www.ferrymon.org), which employs ferry crossings at 3 locations: Cherry Branch-Minnesott Beach (Neuse River estuary), Cedar Island-Ocracoke (southern Pamlico Sound basin) and Swan Quarter-Ocracoke (western Pamlico Sound basin). This program is a temporally intensive, spatially restricted surface water monitoring effort. Critical parameters included are similar to those used in this current study and include temperature, salinity, dissolved oxygen, pH, turbidity, chlorophyll *a*, soluble and particulate nutrients and diagnostic algal photopigments. In addition, the U.S. Geological Survey, Raleigh, has deployed at least one moored monitoring module that collects similar water quality parameters, and there are plans for the placement of additional modules. The U.S. Environmental Protection Agency (EPA) and the National Oceanographic and Atmospheric Administration (NOAA) Charleston Laboratory conducts a one-time-a-year (midsummer) water column and benthic sampling in Pamlico Sound for water quality/habitat assessment as part of the Coastal Environmental Monitoring and Assessment Program (EMAP, www.usepa.emap.gov). Lastly, the Neuse River estuary has an ongoing water quality monitoring program in place (www.marine.unc.edu/neuse/modmon). Pamlico Sound monitoring could be coordinated in time and space to extend synchronized data sets from the upper reaches of each estuary to the inlets at the outer banks, facilitating tracking and quantification of nutrient loads, productivity responses, algal blooms and other water quality responses (e.g. hypoxia). Spatial and temporal coordination will also be a tremendous asset for ground-truthing aircraft and satellite remote sensing efforts currently underway (NOAA-NASA-EPA).

- In addition to standard hydrographic and nutrient-productivity water quality parameters, the Pamlico Sound should be routinely monitored for common toxic substances known to be emitted from agricultural, urban and industrial sources (i.e., EPA's priority toxics "hit list"), potentially toxic heavy metals (e.g., Cd, Hg, Pb), and higher fauna (zooplankton, benthic invertebrates and infauna, larger crustaceans, commercial and recreational fishes). Surveys for microbial pathogens (bacterial and viral), submersed aquatic plants, benthic microalgae and macroalgae should also be considered. Most likely, these surveys can be conducted at lower frequencies (seasonally or semi-annually) than nutrient-productivity studies. Pathogen surveys can be performed along with routine water quality surveys. At present, however, *no* routine program is in place to monitor these substances and biotic entities.

- Monitoring and survey programs for Pamlico Sound should take advantage of the evolving, chemotaxonomic (e.g., ChemTax), biochemical and molecular indicators, as well as other rapid diagnostic tools being developed for large-scale systems. These indicators are currently being applied across estuarine and coastal waters exhibiting varying hydrologic residence times (flushing rates) and nutrient loads, both regionally (www.aceinc.org) and nationally (http://glei.nrri.umn.edu/eagle). Some of these indicators can be coupled (as ground-truthing and calibration sources) to remote sensing, enabling researchers and managers to "scale up" for large systems like the Pamlico Sound.

- Due to limited resources and logistic constraints, water quality monitoring efforts on large coastal systems like Pamlico Sound should, by nature, be interdisciplinary and multi-institutional. This would include State-Private Sector-University partnerships, successful examples of which are already in place (e.g., the UNC-NCDENR-Weyerhaeuser ModMon Program on the Neuse River estuary and FerryMon for Pamlico Sound). North Carolina has a vehicle in place for fostering development and funding for such a collaborative effort, namely the EPA's National Estuarine Program (NEP), administered through NCDENR. The Pamlico Sound is one of 22 designated estuaries deemed important enough to be in this program.

In summary, information obtained from this project can be used to help develop a much needed water quality monitoring and assessment program for the Pamlico Sound. Given parallel efforts in automated monitoring, ecological indicator development, application of remote sensing and the potential for a cooperative program via the NCDENR and NEP; the stage is set for developing a long overdue, yet timely water quality monitoring program for this State's most important aquatic resource, Pamlico Sound.

INTRODUCTION

Extreme hydrologic events, such as the sequential hurricanes Dennis, Floyd and Irene in the fall of 1999, disrupt the filtering capacity of the sub-estuaries of Albemarle and Pamlico Sounds and may allow considerable nutrient loading to the sounds and adjacent coastal waters. These sounds combine to form the second largest estuary in the United States of America and by far the largest body of water in North Carolina. Their importance to the economy and well being of the State is substantial (Copeland and Gray 1991). Pamlico Sound (PS) is the larger of the two systems. It is a major nursery and refuge for the State's and Mid- and Southeast Atlantic commercial and recreational fisheries, and a tourism and recreational center that is highly revered for its beauty and environmental quality. Despite funding in the 1980's and early 90's for studies on the Albemarle-Pamlico Sound system (USEPA-NCDENR) Albermarle Pamlico Estuarine Study (APES), little attention has been given to the sounds themselves. This has been due to the logistical difficulties of working in such large systems and to the belief that the sounds possess better environmental health than the sub-estuaries (i.e., the Chowan, Pamlico and Neuse River estuaries). Until recently, there has been limited evidence supporting this belief (Bales and Nelson 1988, Paerl et al. 2000, 2001). There is a need to (1) establish the current environmental and water quality conditions of the sounds, (2) relate these conditions to the ability of the sub-estuaries to protect the sounds against environmental perturbations (including extreme hydrologic conditions), (3) establish a baseline against which to gauge change, and (4) develop a management plan that includes both an understanding of the interrelationships between water quality in the sounds and sub-estuaries as well as an awareness of the combined effects of anthropogenic and natural disturbances.

Cultural eutrophication of the sub-estuaries of PS has occurred gradually and continuously over recent decades (Hobbie et al. 1972, Hobbie and Smith 1975, Kuenzler et al. 1979, Witherspoon et al. 1979, Tedder et al. 1980, Matson et al. 1983, Paerl 1983, 1987, Harned and Davenport 1990, Copeland and Gray 1991, Christian et al. 1986, Stanley 1988). These sub-estuaries act as a filter for nutrient loads and thus reduce the amount of nutrients delivered to PS (Paerl et al. 1990, Rudek et al. 1991, Christian et al. 1991, Boyer et al. 1994, Christian and Thomas 2000). However, this capacity to remove nitrogen may decrease with increased loading associated with storm-induced flood events. The nutrient buffering afforded by the sub-estuaries is dependent on the slow movement of water through them, as long water residence time maximizes retention and removal of nitrogen (e.g., through denitrification)(Stanley 1992, Christian and Thomas 2000, Seitzinger et al. 1993). However, Christian and Thomas (2000) inferred from network analysis of nitrogen cycling in the Neuse River estuary that the fraction of loaded nitrogen escaping into PS would increase as river discharge increases. By increasing the hydrologic energies through the sub-estuaries and decreasing residence times and thus the biological processing of nutrients, extreme storms may episodically and radically accelerate eutrophication of PS.

During the fall of 1999, three hurricanes (Dennis, Floyd and Irene) inundated eastern North Carolina with up to 1 m of rainfall within a 6 week period, causing a 50 to 500-

year flood event in the watershed of PS (Bales et al. 2000). This acute but high magnitude disturbance represented a unique opportunity to evaluate the effects of changes in hydrology and associated nutrient loading on the water quality of PS. Although nutrient loading in this case resulted from sequential storm events, inferences may be made about the effects of anthropogenic nutrient loading. The results of accelerated nutrient loading via the sub-estuaries into PS may include deterioration of its water quality with consequences similar to what has been witnessed in the sub-estuaries (Hobbie and Smith 1975, Kuenzler et al. 1982; Paerl 1987; Christian et al. 1988, Stanley 1988; Dodd et al. 1993; Paerl et al. 1995, 1998). However, because of its size, limited exchange with the ocean, and long (~1 year) freshwater residence time, recovery from the loss of environmental quality, would be more difficult to overcome than in the sub-estuaries.

The research described in this report relates environmental conditions in the southern portion of PS to those observed in a major nutrient input source, the Neuse River estuary. It also addresses the need for long-term monitoring of the sound. Furthermore, considerable nutrient loading appears to have entered the sound in association with the hurricanes of the fall of 1999 (Paerl et al. 2001). The sound's ecosystem response to this loading was expected to extend well beyond the time of the storm and elevated stormwater runoff. Our study evaluated the extent to which PS was affected. We also examined how storm-related discharge of different nutrients impacted phytoplankton community composition and potentially the trophodynamics of the sound.

BACKGROUND

Water quality studies

This work builds on the considerable research conducted on the Neuse River estuary by Paerl et al. (e.g., Paerl and Fogel 1994, Paerl et al. 1995, 1998, Pinckney et al. 1997, 1998, 1999), Christian et al. (e.g., Christian et al. 1991, Boyer et al. 1993, 1994, Christian and Thomas 2000) and others (e.g., Lebo 1999, Reckhow 1999, Bales et al. 2000, Luettich et al. 2000, Glasgow and Burkholder 2000). Almost all of this research has been in support of environmental management of the estuary proper but not the PS. The estuary contains a highly productive fishery and serves the State as a nursery habitat and recreational area. Despite its obvious ecological and economic importance, it has been the site of numerous hypoxic events (Paerl et al. 1995, 1998), fish kills (Paerl et al. 1998, NCDENR-DWQ data base on fish kills), and nuisance algal blooms (Paerl 1983, 1987, Burkholder and Glasgow 1997). These environmental problems have initiated actions by the State to protect and improve water quality. A major part of the State's management plan that has emerged from these research results is to reduce Total Maximum Daily Loading (TMDL) of nitrogen to the Neuse River estuary. The impacts of such a reduction on PS are not known. Research proposed in this study helps establish a critically important baseline against which we can start to gauge potential responses of the sound to future nutrient reductions in its sub-estuaries.

Pamlico Sound proper has little water quality information with which to develop models or make rational management decisions regarding its eutrophication. Relatively little hydrographic and water quality data have been collected for the open waters of the PS. Before 1963, temperature and salinity were the only hydrographic variables that had been monitored in PS (Winslow 1889, Grave 1904, Coker 1907, Roelofs & Bumpus 1953. Then, Woods (1967) collected monthly temperature, salinity, dissolved oxygen (DO), chlorophyll *a*, and total phosphorus data until 1966. Apparently, these are the earliest DO and nutrient data collected in the sound. Data for the open waters of Albemarle Sound are also sparse (see Bales and Nelson 1988), except for a two-year period of intensive sampling during the early 1970's (Bowden and Hobbie 1977). Currently, there is no state or federal water quality monitoring program in place, and prior to this study all research activities have been *ad hoc* teaching and exploratory research cruises (L. Crowder and J. Ramus, Duke University Marine Lab; H. Paerl and R. Luettich, UNC-Chapel Hill Institute of Marine Science (UNC-CH IMS). The only other routine monitoring that has taken place is an annual visit from the USEPA-NOAA EMAP program, which collects standard hydrographic data (temperature, salinity, dissolved oxygen, dissolved inorganic nitrogen and phosphorus, and chlorophyll *a*), as well as benthic and water column samples for biological inventories during a one-week period at a location in the western PS (Balthis et al. 1998). NCDENR-DWQ monitoring includes only a few monthly sampling locations near the mouths of the Neuse and Pamlico Rivers, and university researchers have also shied away from the sound as a site for their studies. The lack of research in PS stems from its vast size and the perception that most serious water quality problems are confined to its tributary estuaries.

Mechanisms of nutrient import and effects

To our limited knowledge, PS has not demonstrated the same degree of eutrophication as its sub-estuaries. The sub-estuaries typically remove nutrients before they can enter the sound. However, flooding associated with major storms such as hurricanes Dennis, Floyd and Irene, can short-circuit this filtration capacity. These acute events can lead to long-term nutrient loading to PS that may last from months to years. As a result, the ecosystem response to this type of loading may extend well beyond the duration of the storm and elevated stormwater runoff. The sound may receive this episodic loading by at least 3 mechanisms:

1. Flow-through of unprocessed nutrients
2. Resuspension and transport of sediments with subsequent biological and chemical release of nutrients
3. Movement of phytoplankton blooms down the sub-estuaries to near the mouth allowing algal biomass to enter the sound for processing

These mechanisms affect the timing, quantity and quality of the nutrients discharged to the sound, and hence the type of ecosystem response. Although a weather event may occur as a pulse with a characteristic short time scale, the resultant loading may be extended and occur with its own temporal distribution depending upon the nature of the mechanisms.

- □*Mechanism 1.* The temporal distribution of flow-through, unprocessed inorganic nutrients will be similar to that of freshwater discharge lagged by the hydrogeomorphology of the sub-estuary. Unprocessed inorganic nutrients are readily available to autotrophic processes in the sound, but terrestrially-derived organic matter may be more slowly made available or not available at all to phytoplankton in the sound. Both inorganic and organic forms will enter the sub-estuaries in quantities that represent a balance between exposure of new sources of nutrients to the rivers (e.g., overland flooding or overflow of waste lagoons) and dilution by floodwater. The timing of the loading of nutrients would be dependent on the ability of the estuarine biota to metabolize them, which in turn is dependent on seasonality, biodegradability and residence time.

- □*Mechanism 2.* Large hydrologic energies from high flows and potentially from winds can resuspend sediments upstream and within the sub-estuaries and sound. The temporal distribution of dissolved nutrients from sediment-related loading will be bi-modal. The first release will be associated with physicochemical release from pore waters and desorption from the disturbance itself. The second release will result from biological mineralization of the disturbed and transported sediments. This latter release may be delayed and related to seasonal temperature patterns. The rates of ammonium and phosphate release and denitrification from sediments will, in turn, be dependent on stratification, low oxygen conditions and nitrate supply in bottom water.

- □*Mechanism 3.* Nutrients assimilated in the sub-estuaries may enter the sound as biota. In particular, phytoplankton entering into the sound can be significant nutrient sources. Phytoplankton contributions are seasonal and dependent on community composition (Pinckney et al. 1998). The location of the phytoplankton bloom determines how much primary producer organic matter enters the sound and is not released through trophic dynamics. The extensive freshwater volume associated with the event may also change the taxonomic composition of the phytoplankton. For example, bloom-forming blue-green algae (cyanobacteria) may become a more predominant fraction under these reduced salinity conditions (Paerl 1987, Christian et al. 1988). This may have profound effects on the food web (Fulton and Paerl 1987) and hence the ability to process the organic nutrients.

We have attempted to assess the response within the sound. Some of the consequences and expected results relating to each mechanism are summarized in Table 1.

Table 1. Summary of expected results of Hurricanes Dennis, Floyd and Irene on the eutrophication of southern Pamlico Sound given different loading mechanisms.

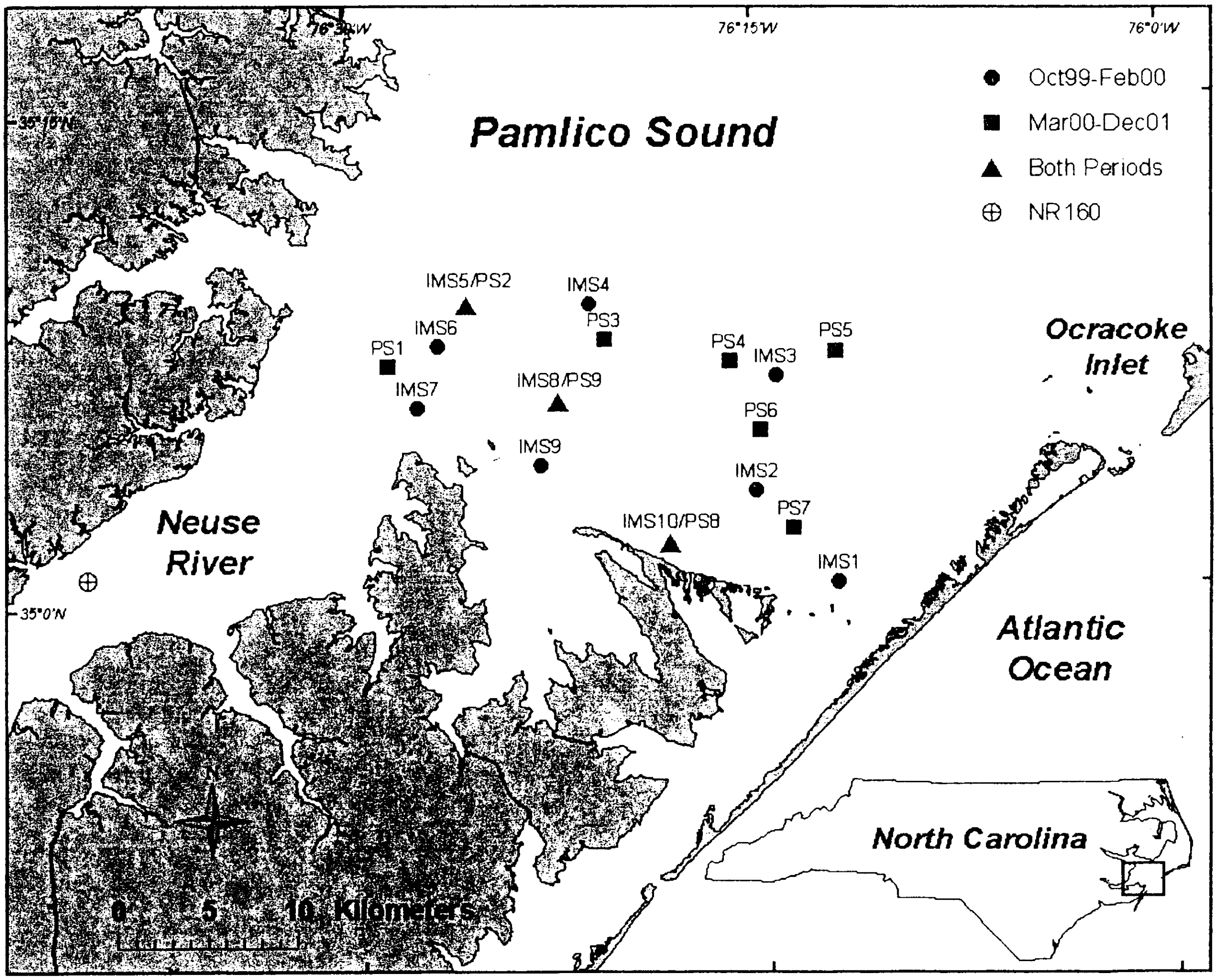
	Nutrient Loading Mechanism		
Result Type	1. Flow-through of unprocessed dissolved nutrients	2. Sediment resuspension and transport	3. Transport as phytoplankton biomass
Temporal distribution nutrients or algal	Immediate peak with tail traceable by freshwater plume dissipation	Bimodal: immediate peak with short tail and seasonally dependent extended rise	Seasonally dependent blooms
Ecosystem response	Low productivity of sub-estuary; transfer of productivity, bloom and hypoxia potential to the sound	Displacement of sediment nutrients and hypoxia potential from sub-estuary to the sound	Shifts along nutrient and salinity gradients with brackish bloom forms displaced to the sound

METHODS

Study Site

Pamlico Sound has a watershed of 80,000 km^2 in North Carolina and Virginia. It has a surface area of (4,500 km^2) and is a shallow (mean depth of 4.6 m) lagoonal system (Fig. 1). It has limited exchange with the Atlantic Ocean through a few small inlets along the Outer Banks of North Carolina. The dominant source of fresh water for its southern portion is the Neuse River estuary (Bales et al. 2000), but other major sources farther north are the Pamlico River estuary and Albemarle Sound. Our study focuses on the relationship between the Neuse River estuary and the southern portion below Brant Island Shoal. This shoal has a depth of 2-3 m and helps to separate water masses above and below it. The Neuse River estuary is 400 km^2 with a basin size of 16,000 km^2. It is also shallow with a mean depth of 3.4 m. Astronomical tides are dampened by its lagoonal nature, and residence times of water are long in both systems as a result of the limited water exchange with the Atlantic Ocean.

Figure 1. Map of Pamlico Sound and study area. Time period for each station is indicated. Station NR160 is part of the long term ModMon study.



Field Studies

The large size of PS precluded detailed broad scale sampling. Thus, we concentrated on the southern portion of the sound linked to the farthest downstream monitoring station of the ModMon program on the Neuse River estuary and to the crossing of the Cedar Island to Ocracoke ferry (Fig. 1). This region of PS is largely under the influence of the Neuse River estuary and hydrologically separated from the northern water masses by Brant Island Shoal. We acknowledge that there are other sources of water and nutrients to PS, including the Pamlico River and Albemarle Sound. It was not our intention to create a budget for the sound or assess all sources of loading.

Monthly samplings were performed within a grid extending from the mouth of the Neuse River estuary towards Ocracoke Inlet using the R/V Capricorn (UNC-IMS) (Fig. 1). Ten stations were visited at least monthly immediately following the hurricanes from early October 1999 until February 2000. Beginning in March of 2000, the stations were relocated and reduced to nine (Fig. 1). The new locations were chosen to overlap with other research group stations and the track of the NCDOT Cedar Island to Ocracoke ferry (Buzzelli et al. 2003; www.ferrymon.org). Trips continued at roughly monthly intervals through April 2002. We also used ModMon station 160 (http://www.marine.unc.edu/neuse/modmon) as the closest reference site for water quality information prior to September 1999.

The sampling stations distributed from the mouth of the Neuse River estuary to Ocracoke Inlet were divided into 4 groups including West, Middle, East, and Southeast. The West group is the first to receive flow from the Neuse River and from there, water flows through the Middle, East, Southeast stations before exiting the sound through Ocracoke Inlet.

Sampling at each station involved vertical profiles of physical, chemical and biological variables. Physical variables included temperature, salinity, and photosynthetically active radiation (PAR). Chemical variables profiled were nitrate + nitrite ($NO_3^{-} + NO_2^{-}$), ammonium (NH_4^{+}), phosphate (PO_4^{-3}), silicic acid (SiO_3^{-2}), dissolved oxygen (DO), particulate matter [particulate carbon (POC), particulate nitrogen (PN) and particulate phosphorus (PP). The biological variable profiled was phytoplankton biomass. Also at each station, primary productivity rates were measured by ^{14}C incorporation. Measurements of diagnostic photosynthetic pigments by high performance liquid chromatography (HPLC) equipped with an in-line photodiode array spectrophotometer (PDAS) were used to assess phytoplankton community structure and function. We have used HPLC-PDAS/ChemTax in conjunction with primary productivity measurements to characterize phytoplankton community compositional responses to hydrologic and nutrient perturbations and trends in seasonal bloom dynamics in the Neuse River estuary since 1994 (Pinckney et al. 1999). During this period, the Neuse River estuary has experienced the combined stresses of anthropogenic nutrient enrichment, droughts (reduced flushing combined with minimal nutrient inputs), and since 1996, elevated tropical storm and hurricane activity (high flushing accompanied by elevated nutrient inputs). These distinct perturbations as well as the hurricanes of 1999 reported on here have provided excellent research opportunities for examining the impacts of both anthropogenic and natural stressors on phytoplankton community structure at the functional group level, a level pertinent to changes in carbon and nutrient (N, P) flux as well as food web changes.

The different forms of phosphorus were determined monthly, at the surface and bottom of the water column, from July 2000 to March 2002 with the exception of March, July, and December of 2001. Measurements of phosphate or soluble reactive phosphorus (PO_4^{-3}), PP, and total dissolved phosphorus (TDP) were taken for each sample. Dissolved organic phosphorus (DOP) was calculated by subtracting PO_4^{-3} concentrations from the TDP. PO_4^{-3}plus DOP plus PP constitute the total amount of phosphorus in a water sample (TP).

Table 2. Physical, chemical, and biological parameters that were measured in this study.

Parameter	Method/Instrument
Salinity, pH, temperature, depth, conductivity, dissolved O_2, turbidity, chlorophyll fluorescence	YSI 6600 Multiparameter Water Quality Monitor
Irradiance (PAR)	LiCor LI-1000 with 4 π sensor
Dissolved inorganic nutrients	Lachat QuickChem 8000 AutoAnalyzer
Particulate organic C and N	Perkin-Elmer 2400 Elemental Analyzer
Dissolved inorganic and organic C	Shimadzu TOC 5000
Primary productivity	Simulated *in situ* ^{14}C method
Diagnostic photosynthetic pigments	Shimadzu HPLC-PDAS (M10AV)
Particulate and dissolved total P	Digestion to phosphate

Analytical Methods

Protocols for routine physical, chemical, and biological measurements are summarized in Table 2. Nutrient analyses (NO_3^-+NO_2^-, NH_4^+, and PO_4^{-3}) were done by high sensitivity flow injection analysis using a Lachat Quickchem 8000 (QuikChem Methods 31-107-04-1-C, 31-107-06-1-A/B, 31-115-01-1-F/G, respectively). The detection limits were approximately 0.08 µM, 0.3 µM, and 0.01 µM for [NO_3+NO_2], [NH_4], and [PO_4], respectively. Nutrient concentrations below these values were reported as one half of the method detection limits. Particulate carbon and nitrogen were determined on GF/F filtered samples using a Perkin Elmer Elemental Analyzer. Inorganic carbon was removed by vapor phase acidification of the filters. Particulate and total dissolved phosphorus were digested and measured as phosphate. Particulate phosphorus was Kjeldahl digested, and TDP was digested in persulfate (Franson 1995). Dissolved organic phosphorus was estimated from the difference between TDP and PO_4^{-3}. Measurements for comparison of forms of P were conducted at East Carolina University, using similar chemistry for PO_4^{-3} but with an Orion Scientific Instrument segmented flow analyzer. Dissolved organic carbon (DOC) concentrations were measured using a Shimadzu TOC-5000A Analyzer. This instrument uses high temperature catalytic oxidation followed by non-dispersive infrared analysis of the CO_2 produced. Samples were acidified to pH<2 and sparged with air before being analyzed for non-volatile organic carbon.

Phytoplankton chlorophyll *a* concentrations were measured using the modified in vitro fluorescent technique in EPA Method 445.0 (Arar et al. 1997) that uses narrow bandpass excitation/emission filters and no acidification. Samples (50-75 mL) were collected on 25 mm GF/F filters (vacuum filtration, <25 kPa), blotted dry, and frozen immediately. Chlorophyll *a* was extracted from the filter using a tissue grinder and 90% aqueous acetone. The samples remained in the acetone overnight at -20 °C. The extracts were filter-clarified and analyzed on a TD700 fluorometer (Turner Designs, Sunnyvale, CA). The fluorometer was calibrated with chlorophyll *a* (Sigma Chemical Co.) after determining the concentration using a Shimadzu UV160U Spectrophotometer and the extinction coefficients of Jeffrey and Humphrey (1975). The calibration was checked daily against a solid secondary standard (Turner Designs, proprietary formula).

For phytoplankton community composition, we used HPLC to quantify the relative biomass of taxonomic groups in the phytoplankton community. High performance liquid chromatography can rapidly and accurately separate and quantify photopigments from mixed algal samples (Wright et al. 1991, Millie et al. 1993, Jeffrey et al. 1997). An in-line photodiode array spectrophotometer (PDAS) allowed for rapid identification of individual photopigments based on characteristic absorption spectra. Protocols for this method are detailed in Van Heukelem et al. (1994) and Pinckney et al. (1996). Chemotaxonomic photopigments (chlorophylls and carotenoids) were an effective means for determining the relative abundance of major algal groups in phytoplankton communities (Rowan 1989, Jeffrey et al. 1997). Recently developed statistical procedures (ChemTax) were applied to give estimates of the relative contribution of each algal group in units of chlorophyll *a* (Pinckney et al. 1998). ChemTax is a matrix factorization program used to calculate the absolute and relative abundances of major algal groups from concentrations of chemosystematic photopigment biomarkers (Mackey et al. 1996). This program uses a steepest descent algorithm to determine the best fit based on an initial estimate of pigment ratios for algal classes (Table 1 in Mackey et al. 1996). Using the concentrations of alloxanthin, antheraxanthin, chlorophyll b, total chlorophyll *a* [chlorophyll *a* + chlorophyllide *a*], fucoxanthin, lutein, peridinin, violaxanthin, and zeaxanthin, ChemTax partitioned the total community biomass (total chlorophyll *a*) into group specific biomass of five major algal divisions (Cryptophyta, Cyanophyta, Bacillariophyta, Dinophyta, and Chlorophyta). The analyses were grouped by depth level and season (Pinckney et al. 1998) and were conducted on data through November 2001.

Photosynthetic rates were measured using an adaptation of Steemann Nielsen's (1952) ^{14}C method. The adaptation is detailed in Paerl et al. (1998) with the following additional changes. Surface water samples were incubated in 125 mL polyethylene terephthalate copolyester (PETG) media bottles (total volume 168 mL). Final concentrations of $NaH^{14}CO_3$ (50-60 mCi $mmol^{-1}$, ICN Pharmaceuticals, Costa Mesa, CA) ranged from 0.01 to 0.03 μCi (370-1110 Bq) mL^{-1}. Each bottle was filtered entirely, unless particle concentrations were high, in which case measured aliquots were removed for filtering. Dissolved inorganic carbon (DIC) concentrations were determined with a Shimadzu TOC-5000A in inorganic carbon mode.

Data analysis

Space-time plots were produced using Surfer version 7.0 (Golden Software, Inc.). Stations were given a radial distance from station PS1 at the mouth of the Neuse River (Fig. 1). Interpolated grid files were created from the distance and date data using inverse distance to the fourth power and an anisotropy ratio of five. Contour plots of the grid files were then created using high smoothing.

Stations within PS were sampled regularly beginning in the fall of 1999 and results were analyzed by comparing monthly conditions for one year after the hurricanes to those months beyond one year. Water quality variables examined for hurricane effects were POC, PN, DOC, NO_3^- + NO_2^-, NH_4^+, PO_4^{-3}, chlorophyll *a* (Chl *a*), and primary productivity. Data were pooled by variable for all stations by month from October 1999 to August 2000, and for September 2000 to May 2002. We then compared rankings for each month from the first group with corresponding months in the second group. For example we compared ammonium concentrations during November 1999 with those pooled for November 2000 and 2001. Data from the two periods were compared using Kruskal-Wallace non-parametric analysis, and differences in medians were estimated by inspection. We realize the limitations of non-parametric analyses but used the Kruskal-Wallace test because of concerns of violations of assumptions of parametric testing, given the sampling design and distributions of data in both time and space.

Comparisons between concentrations from surface and bottom waters were made by paired Student's t-tests. Pairing was for each unique station and sampling date combination.

Station 160 within the Neuse River estuary has been sampled regularly since summer 1997 and represented an opportunity to compare both before and after hurricane conditions. Data were pooled by month for years before mid-September 1999, mid-September 1999-2000, and after mid-September 2000. Data from the three periods were compared using Kruskal-Wallace non-parametric analysis, and differences in medians were estimated by inspection.

RESULTS

A considerable amount of data was collected throughout this study. Because of this, it has been a challenge to present the data in a concise but informative way. We have chosen two modes of presentation. First, we developed "Surfer" plots where concentrations of each variable are represented by shading within a plane delineated as distance from the mouth of the Neuse River estuary (from PS1 in Fig. 1) and time. These representations are provided for description of time vs. distance trends in the variables. Second, we compared potential nutrifying effects of the hurricanes. Each variable by month for the year following the event is statistically compared against like months from later times. This is done for the composite of samples of all stations within the Sound and for Station 160 from the Neuse River estuary, from which pre-hurricane water quality data was available.

The distribution of phosphorus among chemical species was not determined immediately after the hurricanes, because funding for this component was not available. We have included a section comparing the concentrations of phosphorus in soluble reactive, particulate and dissolved organic forms for the period of measure. This was done to provide a baseline of the phosphorus distribution within southern PS.

Space-Time Plots

Hydrography

Salinity was reduced across most of the southern sound shortly after the hurricanes (Fig. 2). Both surface and bottom waters had salinities below 10 psu during November 1999, and surface waters remained at these salinities into January 2000. Recovery occurred more rapidly near Ocracoke Inlet and in bottom waters. One year after the hurricanes, salinity across the southern sound was greater than 15 psu, and by year 2 it was generally greater than 20 psu. This level of salinity was maintained through the end of the sampling period. Differences in patterns between surface and bottom waters were such that stratification was frequently observed. This was most evident near Ocracoke Inlet.

One effect of stratification can be low bottom water DO concentrations. Concentrations less than 4 mg L^{-1} were common through 2001 (Fig. 3). Low concentrations were most common during warmer months, although occasional hypoxic events (<4 mg L^{-1}) occurred in colder months. Even though stratification may have been more evident near Ocracoke Inlet, hypoxia generally occurred further upstream.

Light attenuation integrates the effects of color and turbidity. The diffuse light attenuation coefficient, K_d, was at its highest (least light penetration) immediately after the hurricanes and during late winter and spring 2000 (Fig. 4). The coefficient was above 2 m^{-1} at these peak times. After these times the coefficient was generally less than 1 m^{-1} with occasional increases to between 1 and 2 m^{-1}. No pattern was discernible with distance from the Neuse River estuary mouth.

Figure 2. Space-time plot for salinity in preferred salinity units (psu). Y-axis denotes distance in km from station PS1 at the Neuse River mouth. Date is on the x-axis. Diamonds indicate sample location and time; these will be left out for clarity on subsequent space-time plots. Upper panel is near surface data and lower panel is data closest to the bottom.

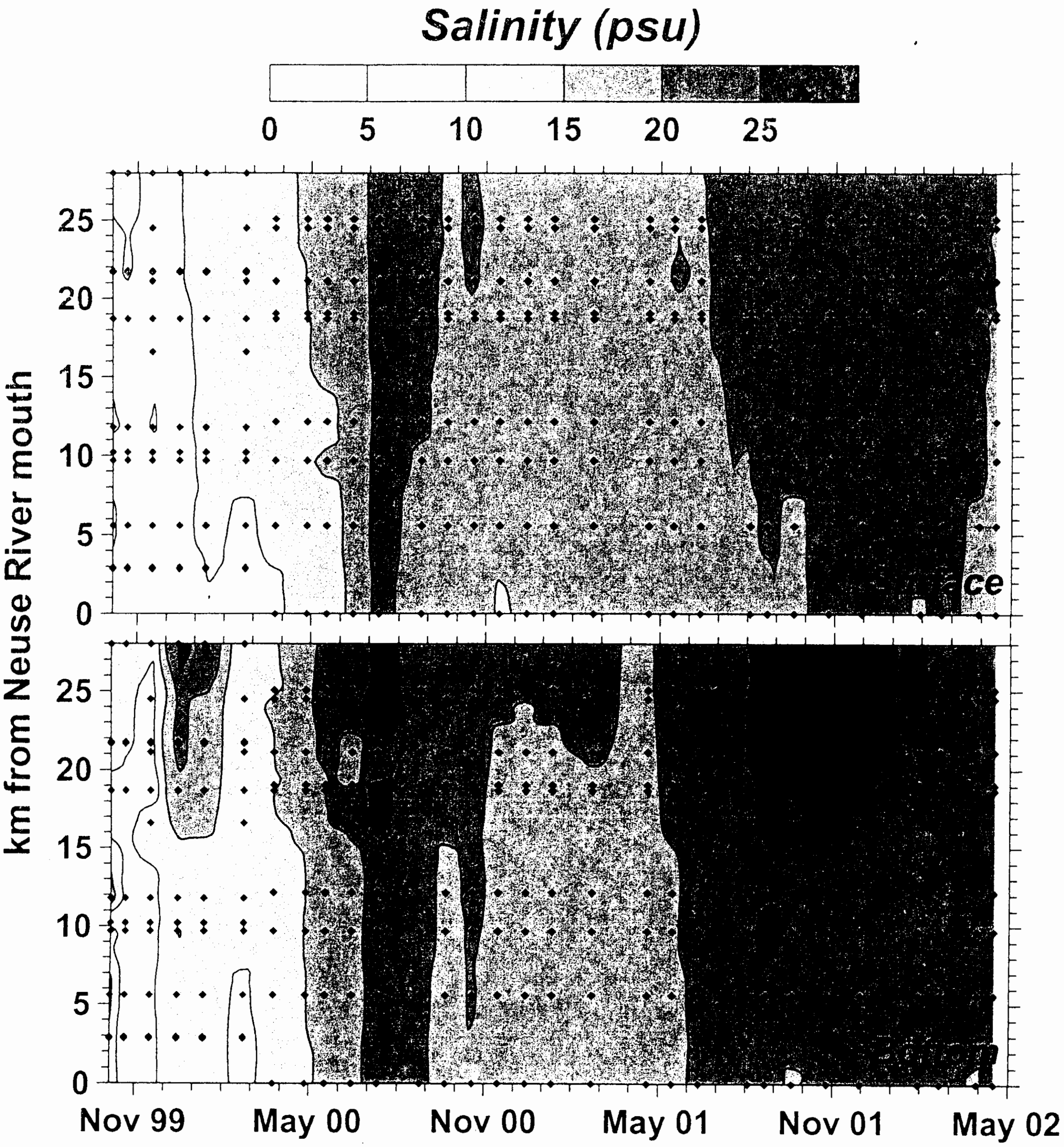


Figure 3. Space-time plot for dissolved oxygen nearest the bottom in mg L^{-1}. Axes as in Fig. 2. Dark contour lines indicate the 4 mg L^{-1} level below which is defined as hypoxic.

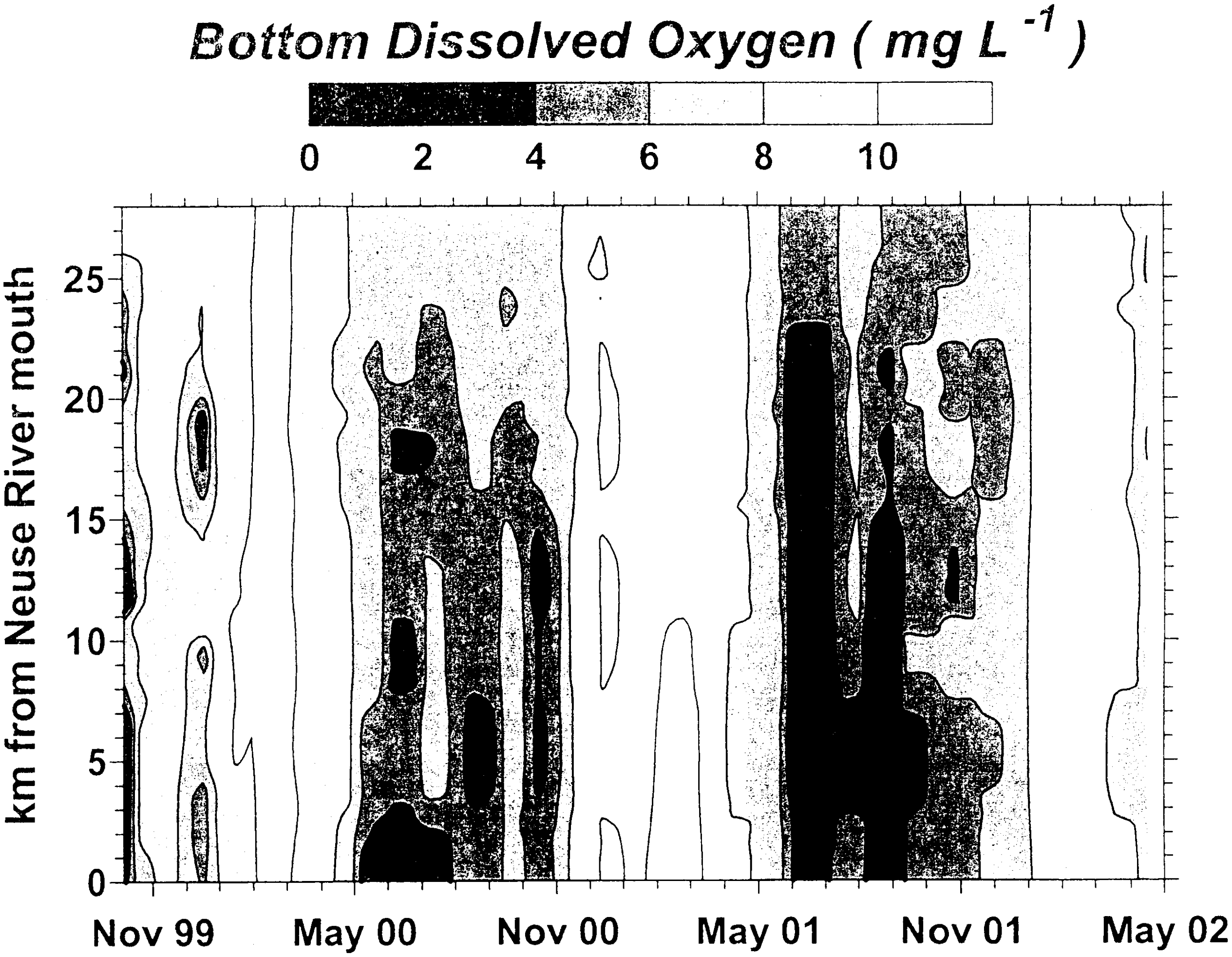
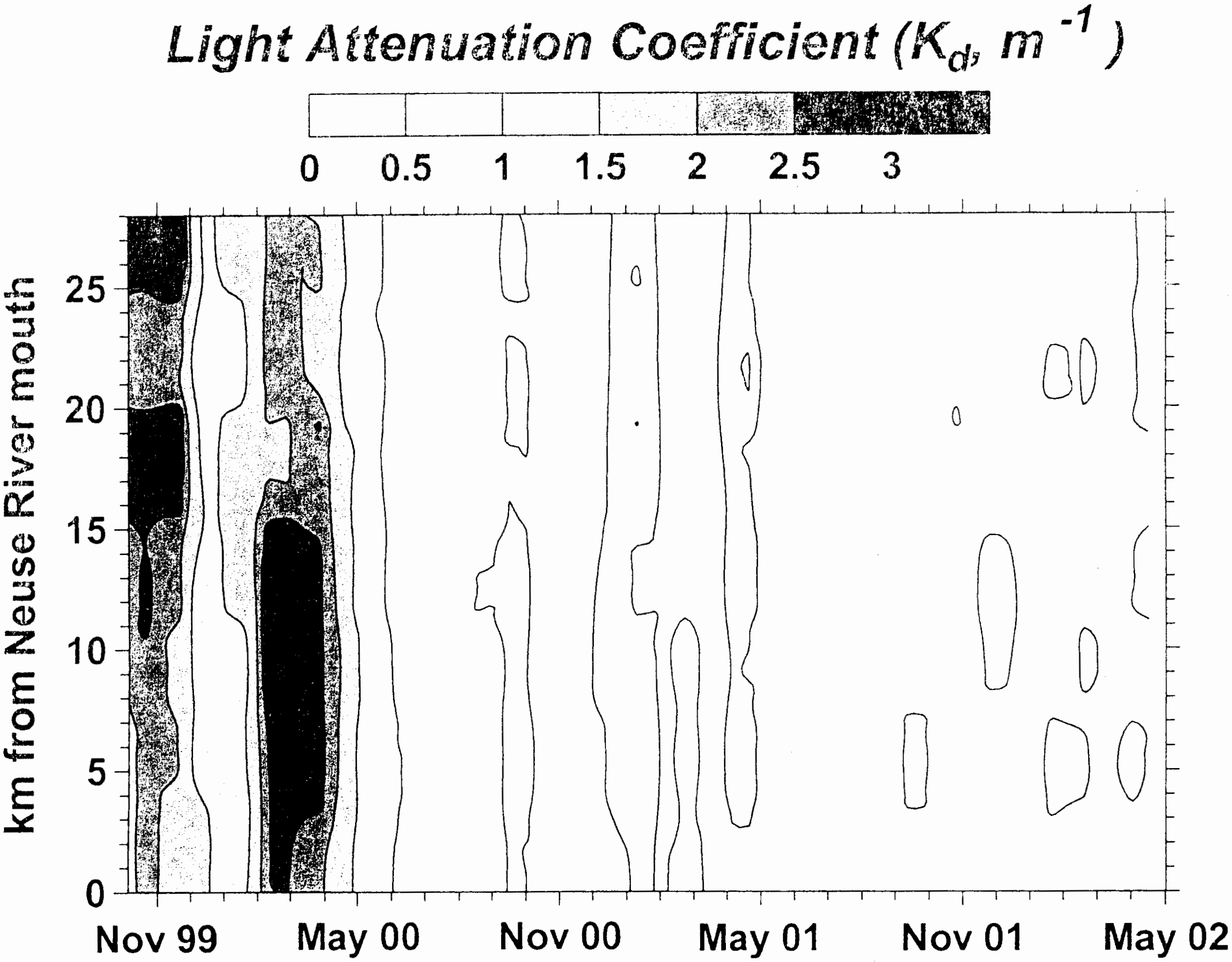


Figure 4. Space-time plot for diffuse light attenuation coefficient, K_d (m^{-1}). Higher numbers correspond to lower light transmission. Axes as in Fig. 2.



Dissolved nutrients

Concentrations of each dissolved nutrient (NO_3^- + NO_2^-), NH_4^+, PO_4^{-3}, and DOC were similar across the southern sound during the study and were relatively elevated after the hurricanes (Figs. 5-10). Nitrate concentrations in surface waters were generally less than 1.5 μM in the southern sound (Fig. 5). During much of 2000 and fall 2001 through spring 2002, concentrations were reduced to less than 0.1 μM. Highest concentrations were found during the two months after the hurricanes and near the mouth of the Neuse River estuary.

Concentrations of ammonium were generally higher than those of nitrate. As with nitrate, the highest concentrations in the surface were during the first samplings immediately after the hurricanes (Fig. 6). Concentrations during this time exceeded 5 μM in both surface and bottom waters (Figs. 6 and 7). Ammonium concentrations at both depths were then generally less than 2 μM, except during summer 2001. During this time, ammonium concentrations in the bottom water peaked again. Bottom water ammonium concentrations often exceeded concentrations in surface water, but aside from this summer period of high bottom ammonium concentrations, differences of more than 1 μM were rare.

Figure 5. Space-time plot for surface water nitrate concentrations (μM). Axes as in Fig. 2.

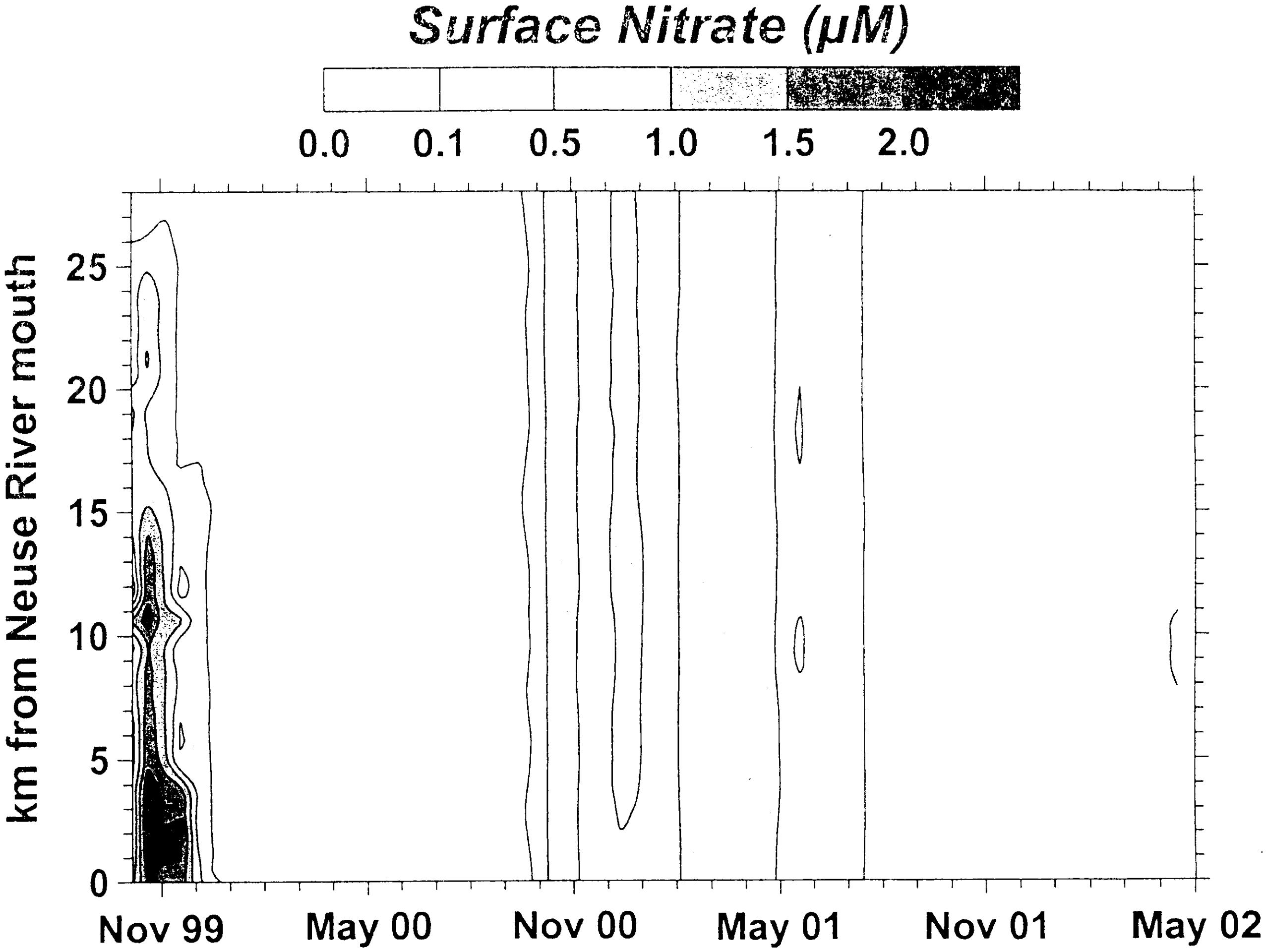


Figure 6. Space-time plot for surface water ammonium concentrations (μM). Axes as in Fig. 2.

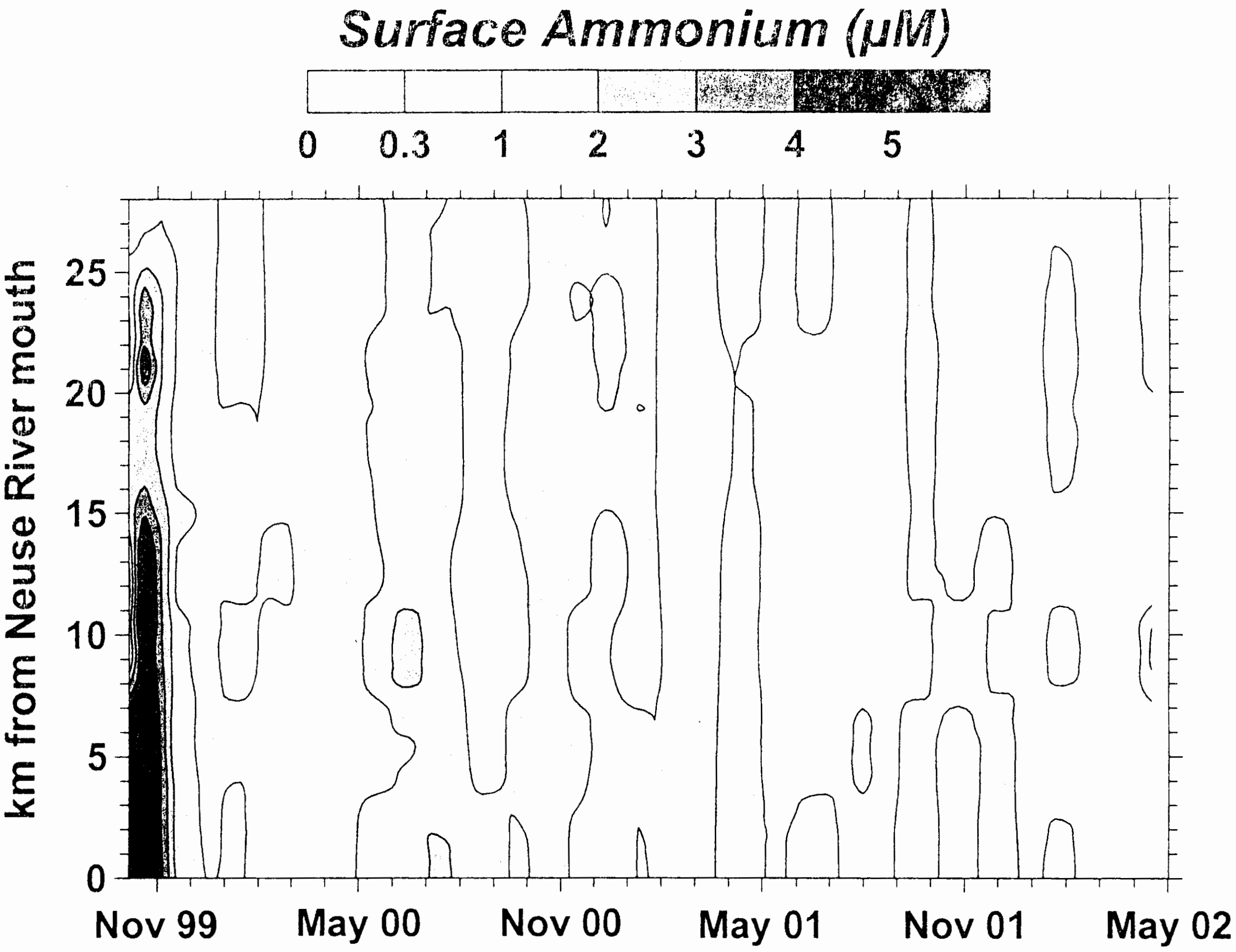
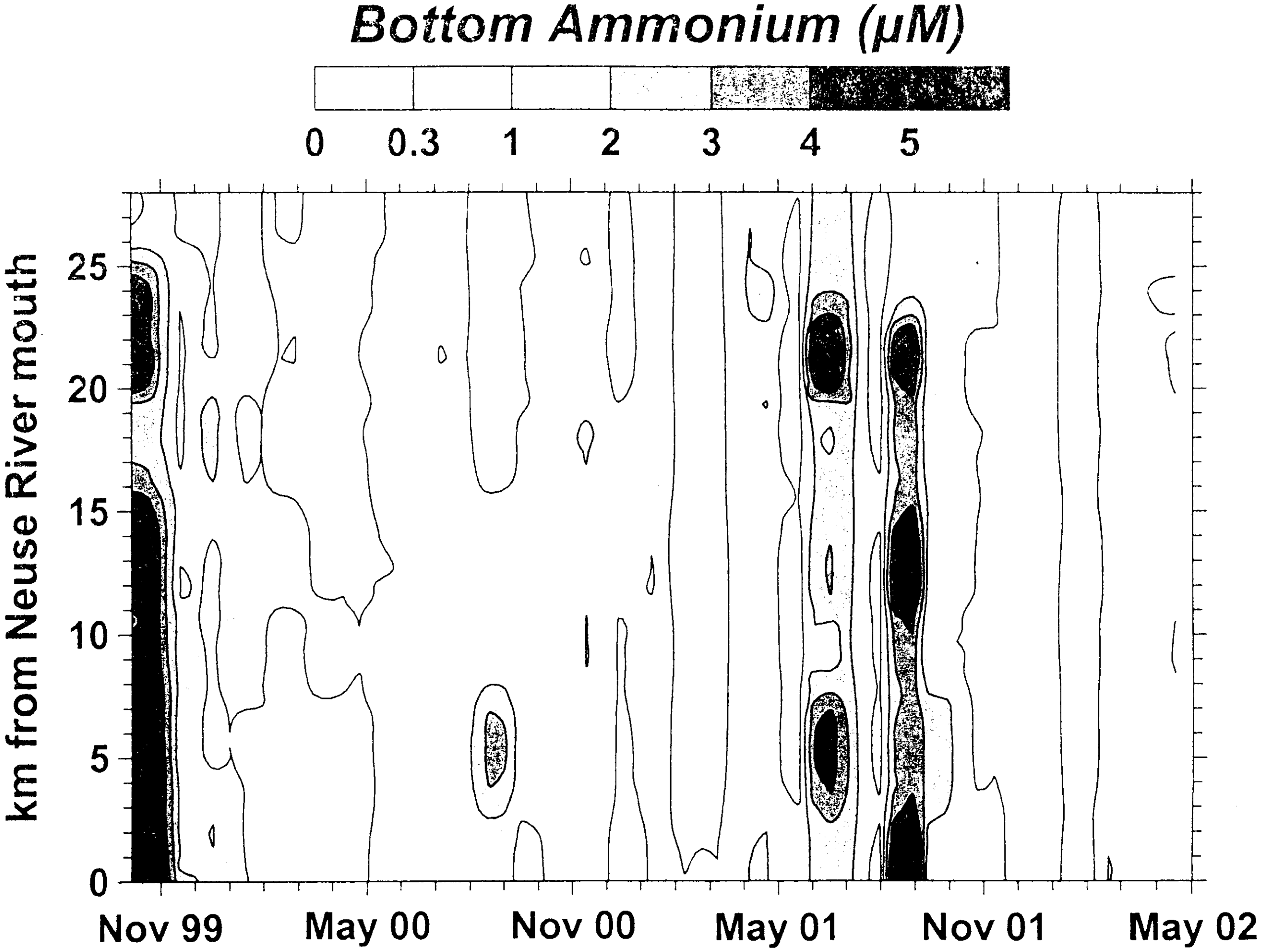


Figure 7. Space-time plot for bottom water ammonium concentrations (μM). Axes as in Fig. 2.



Soluble reactive phosphorus showed few differences with distance from the mouth of the Neuse River estuary, but showed differences between sampling dates (Figs. 8 and 9). Trends were similar in surface and bottom waters. The highest concentrations of PO_4^{-3} were not immediately after the hurricanes, but during late summers 2000 and 2001. At these times concentrations exceeded 1 μM, whereas generally they were less than 0.5 μM. Concentrations during the winters and springs of 2000-2001 and 2001-2002, after the peak, were generally higher than the winter/spring 1999-2000. Concentrations for the fall months immediately after the hurricanes were elevated compared to that winter.

Dissolved organic carbon concentrations generally ranged from 400 to 800 uM until late fall 2001 when concentrations generally decreased to below 400 uM (Fig. 10). Dissolved organic carbon was relatively constant with occasional patches of higher concentrations. No spatial or temporal trends were evident at this level of resolution.

Figure 8. Space-time plot for surface water phosphate (PO_4^{-3}) concentrations (μM). Axes as in Fig. 2.

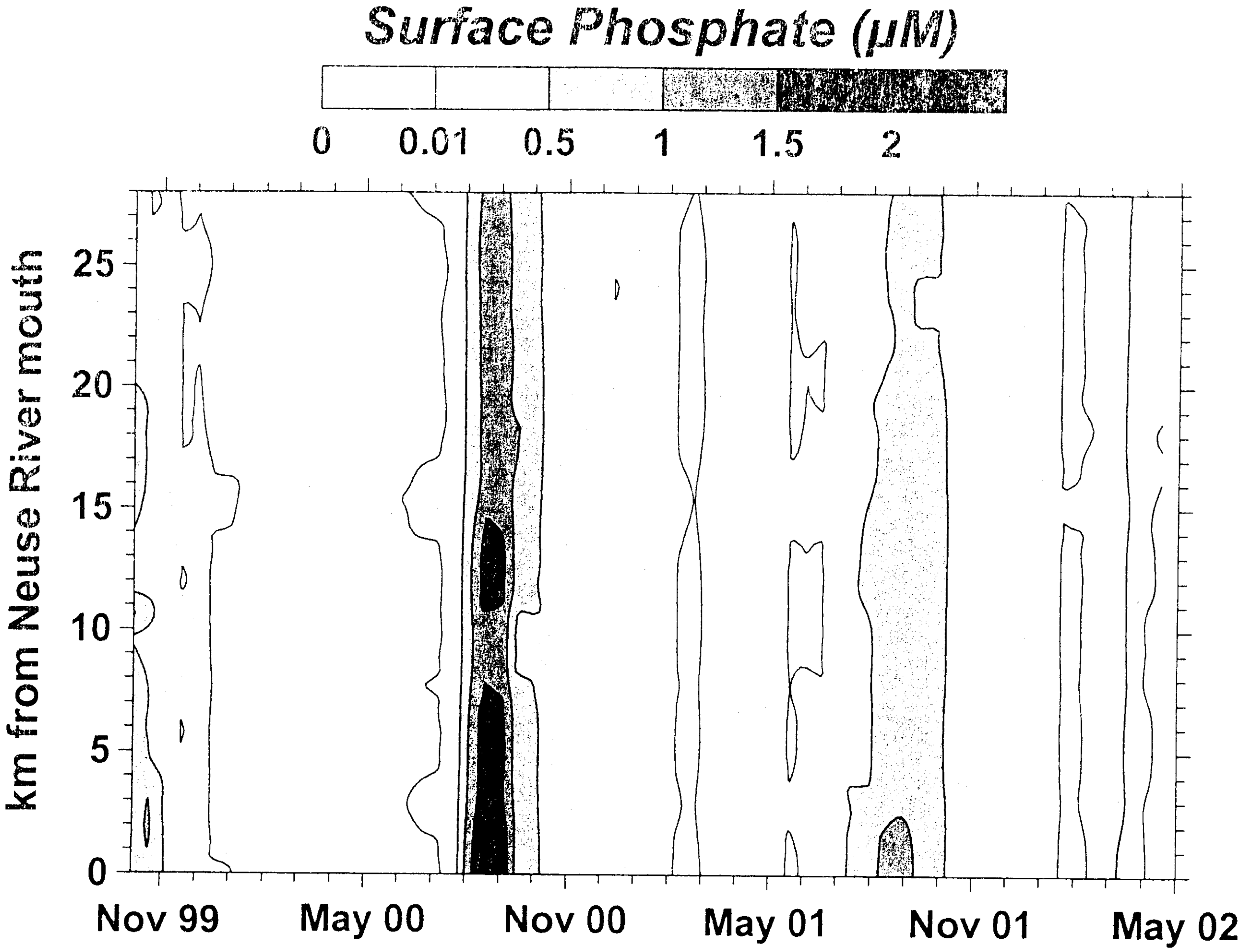


Figure 9. Space-time plot for bottom water phosphate (PO_4^{-3}) concentrations (µM). Axes as in Fig. 2.

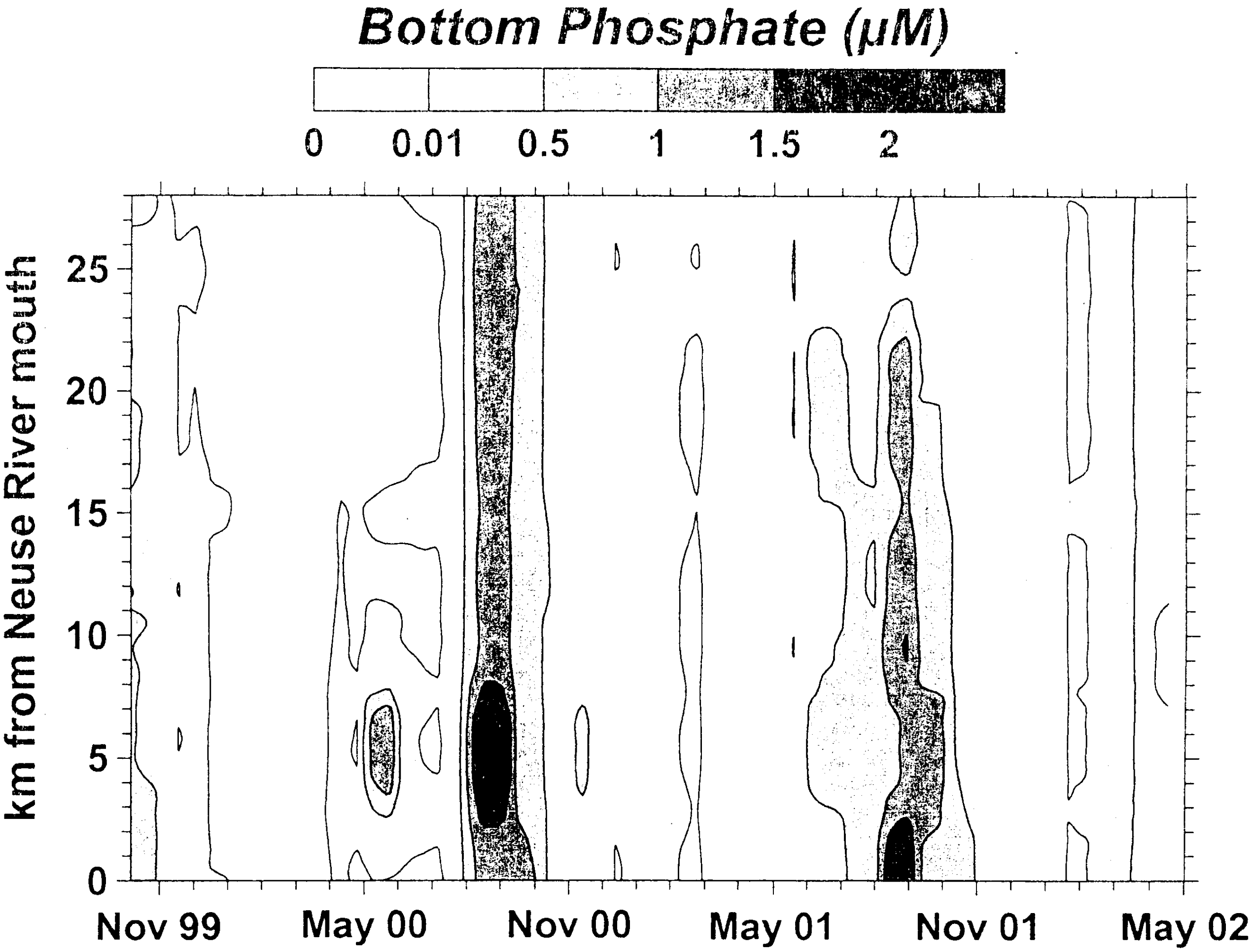
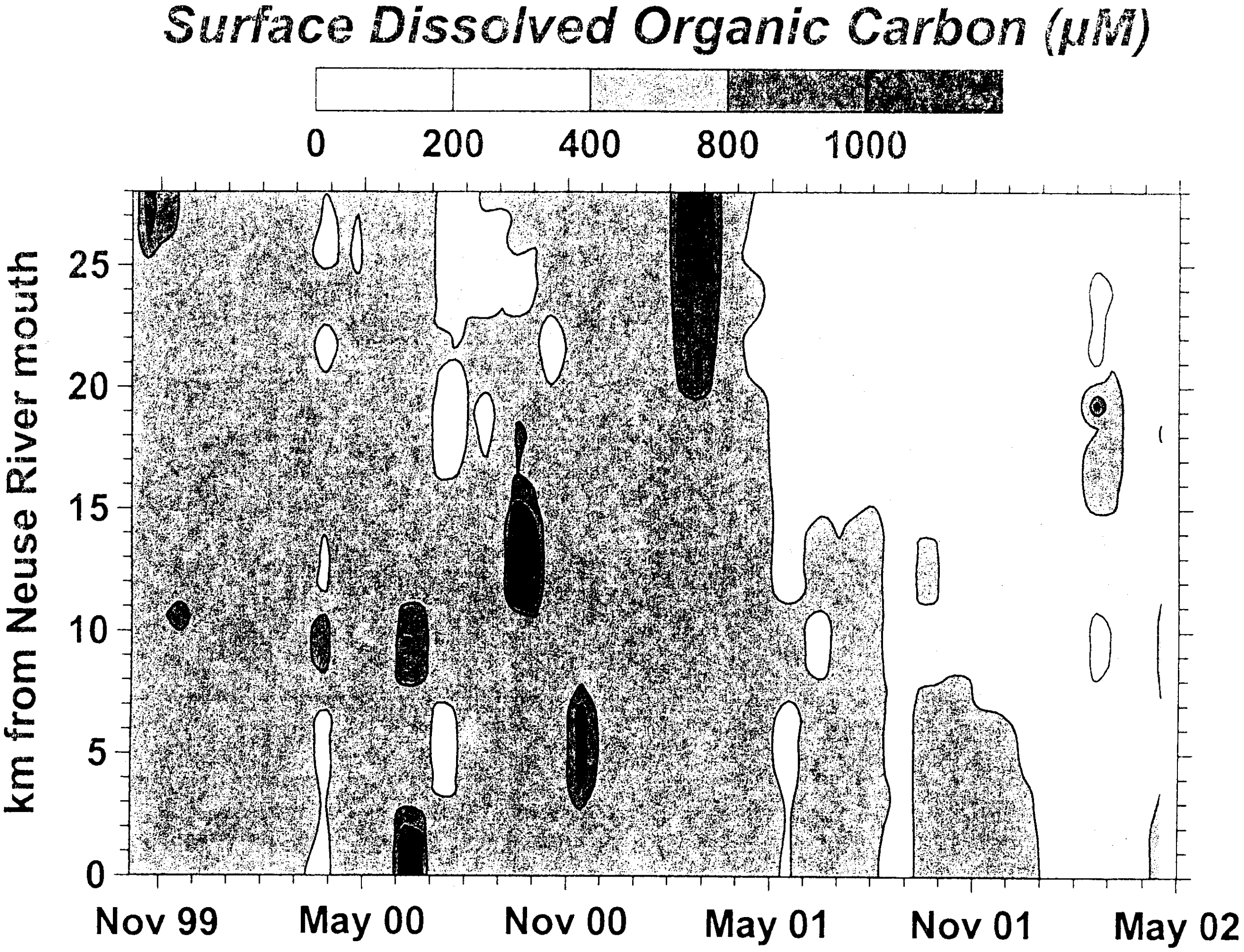


Figure 10. Space-time plot for surface water dissolved organic carbon (DOC) concentrations (µM). Axes as in Fig. 2.



Particulate nutrients

POC, PN, and PP were used to assess the seston. Samples for POC and PN were collected from the time of the hurricane, whereas the latter was only collected upon funding by WRRI.

POC concentrations in surface water showed peaks in late winter and spring in 2000 and in late summer and fall 2000 (Fig. 11). These peaks were more dramatic than concentrations immediately after the hurricanes. Peaks occurred at different locations in the sound and showed no obvious trends with distance from the Neuse River estuary. In spring 2000 the peak was near the mouth of the Neuse River estuary, whereas later in 2000 peaks were found farther downstream. These peaks exceeded 2,000 µg L^{-1}, but most samples had concentrations less than 1,500 µg L^{-1}. Concentrations were below 1,000 µg L^{-1} after fall 2001.

The pattern of PN was similar to that of POC (Fig. 12). PN showed the same peak in late winter and spring 2000 as POC. Lesser peaks were in late summer and fall of that year, and the peaks were in the same locations for both POC and PN. These peaks exceeded 300 µg L^{-1}. Most all other samples were less than 300 µg L^{-1}, although some patchiness is evident. Concentrations became low for 2002 with values less than 100 µg L^{-1} during that summer.

Figure 11. Space-time plot for surface water particulate organic carbon (POC) concentrations (µg L^{-1}). Axes as in Fig. 2.

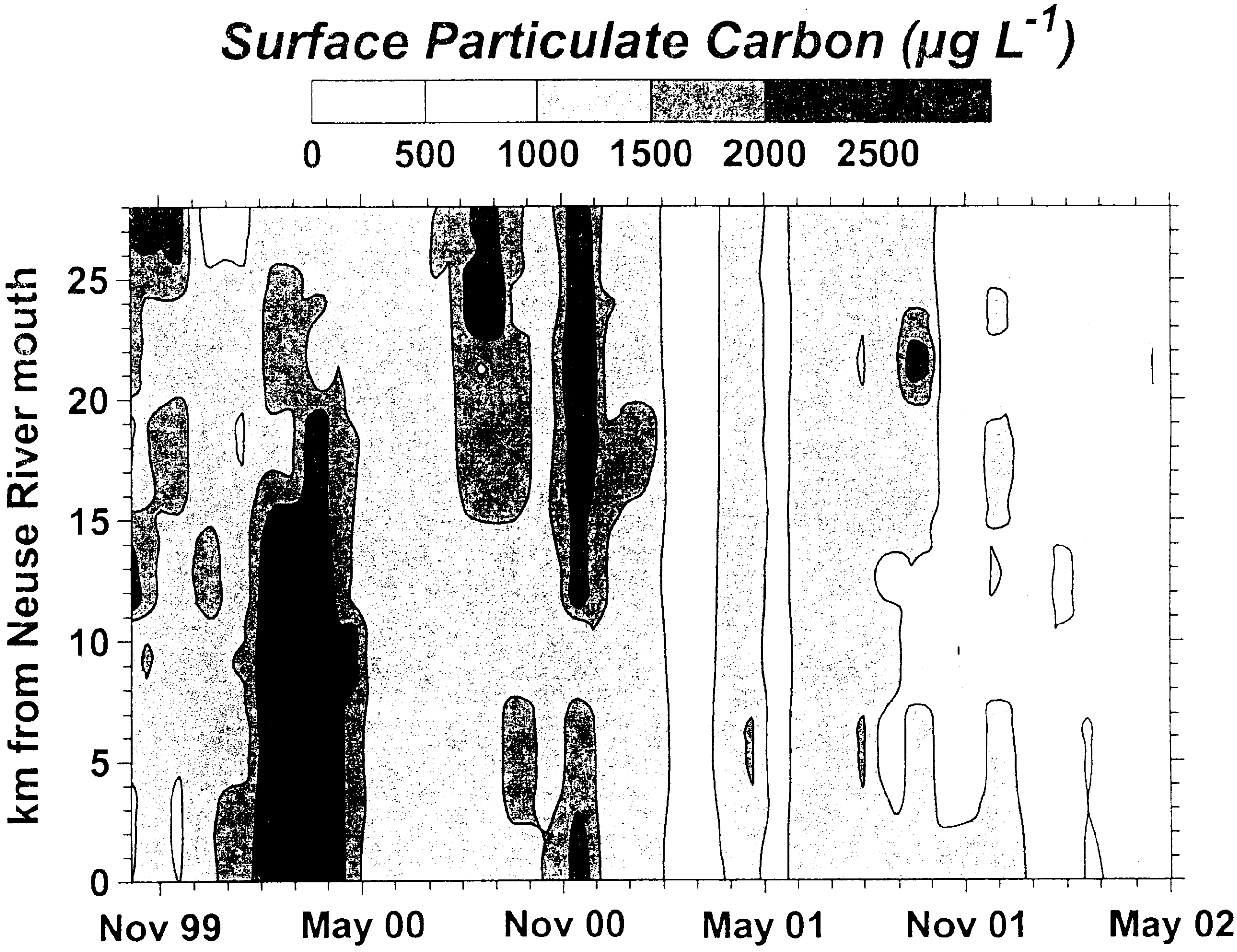
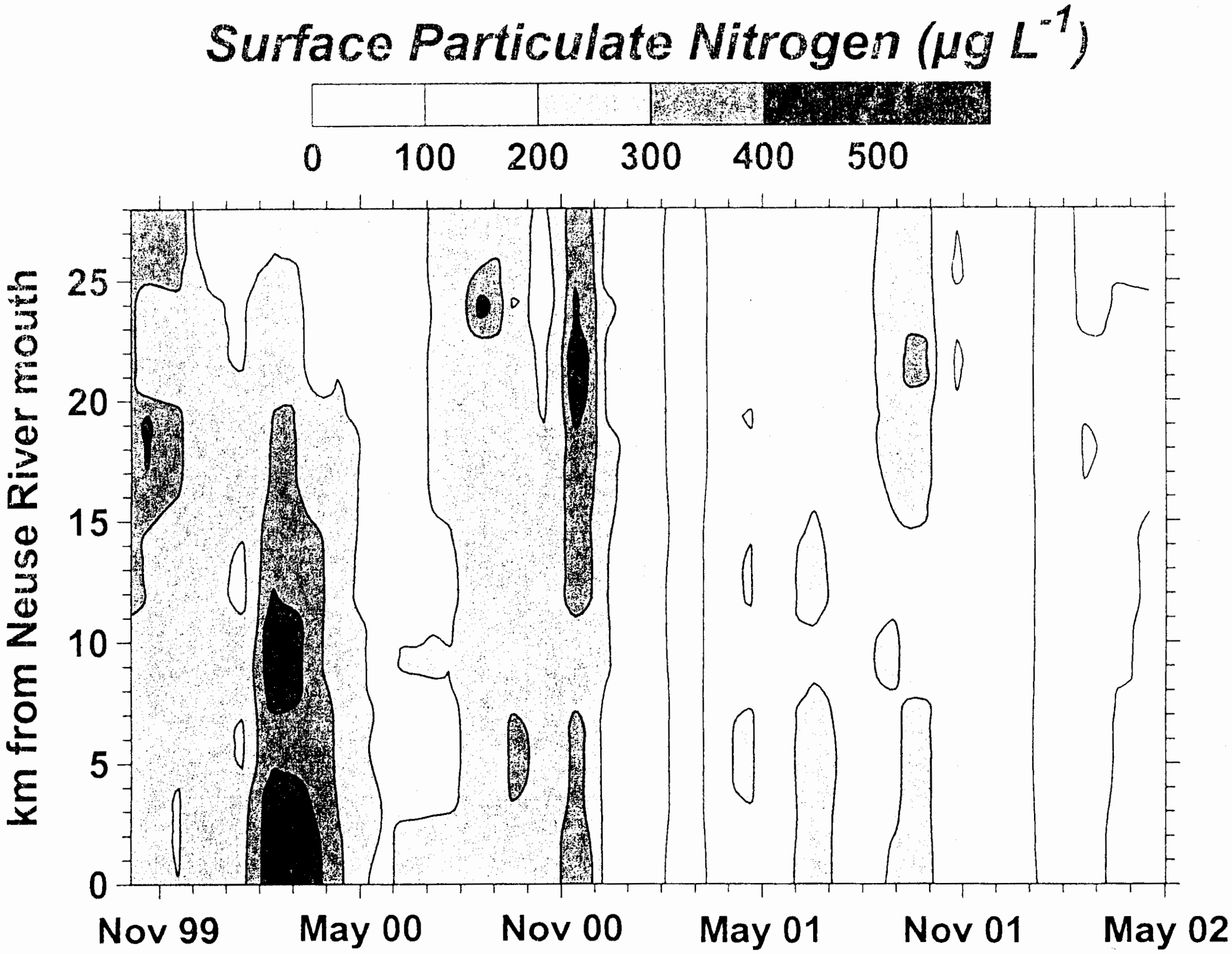


Figure 12. Space-time plot for surface water particulate nitrogen (PN) concentrations (μg L^{-1}). Axes as in Fig. 2.



Phytoplankon

Total phytoplankton standing crop was estimated from pigment concentrations (Chl *a*) using two methods, *in vitro* fluorometry and HPLC. Surface water Chl *a* concentrations, as measured by *in vitro* fluorometry, were elevated immediately following the storms with a substantial bloom that persisted from late winter through early spring 2000 (Fig. 13). Another bloom also occurred in fall 2000. During non-bloom periods, chlorophyll *a* concentrations were almost always less than 10 μg L^{-1}. The highest concentrations exceeded 30 μg L^{-1} near the mouth of the Neuse River estuary during spring 2000. Concentrations during this winter/spring bloom decreased from the mouth of the NRE to Ocracoke Inlet. Concentrations during 2002 dropped to <5 μg L^{-1}. Vertical differences in Chl *a* were minimal except for isolated locations in July 2000 and August 2001.

As would be expected, total Chl *a* concentrations, as measured by HPLC, showed similar patterns to those just described, only the absolute concentrations were less (Fig.

14). Chl *a* concentrations estimated from *in vitro* fluorometry are generally greater than when determined by HPLC because of non-specific fluorescence. These results are included since the total chlorophyll *a* concentration equals the sum of the separate group-specific biomass values.

Figure 13. Space-time plot for chlorophyll *a* (μg L^{-1}) measured by in vitro fluorometry. Axes as in Fig. 2.

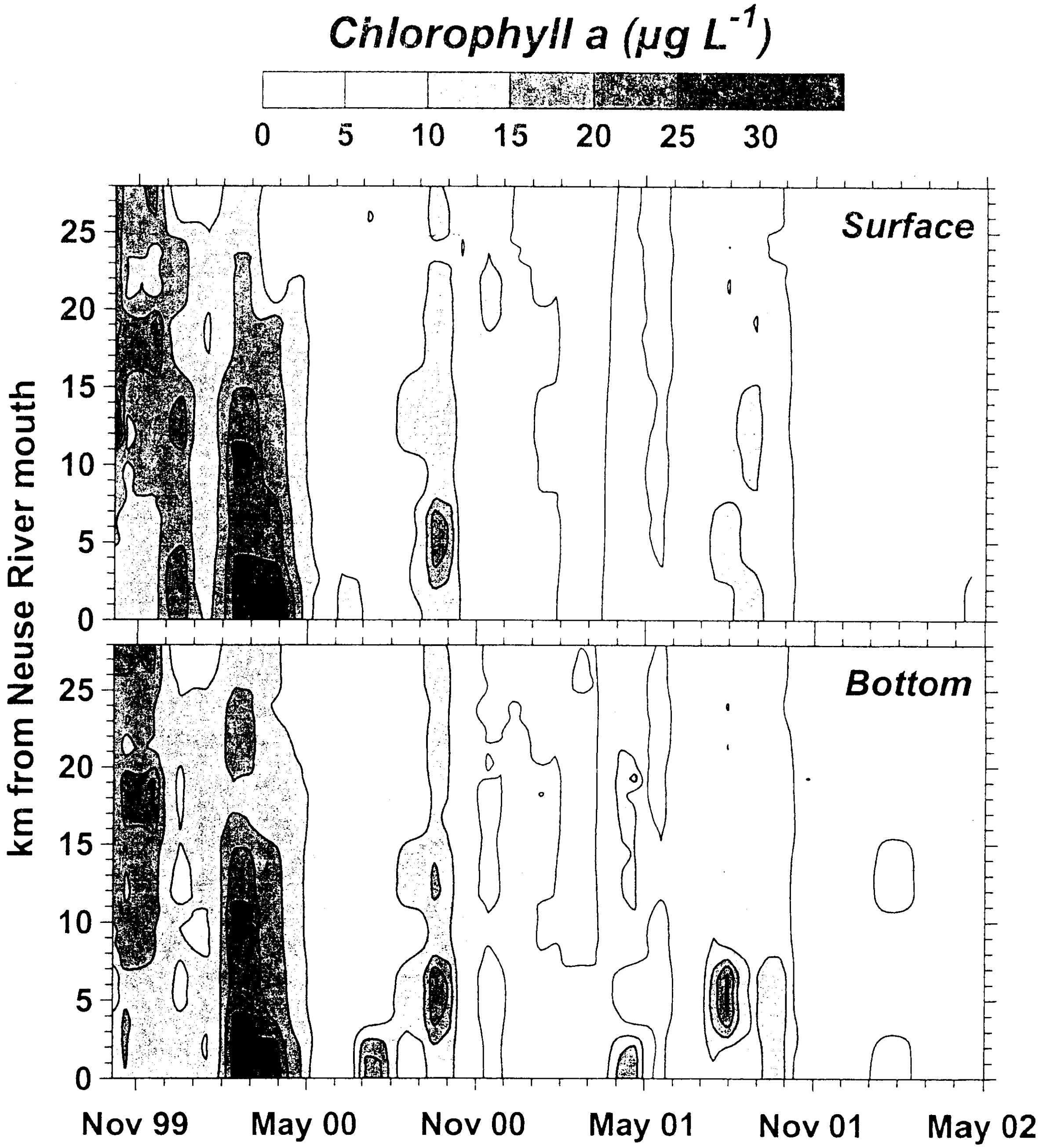
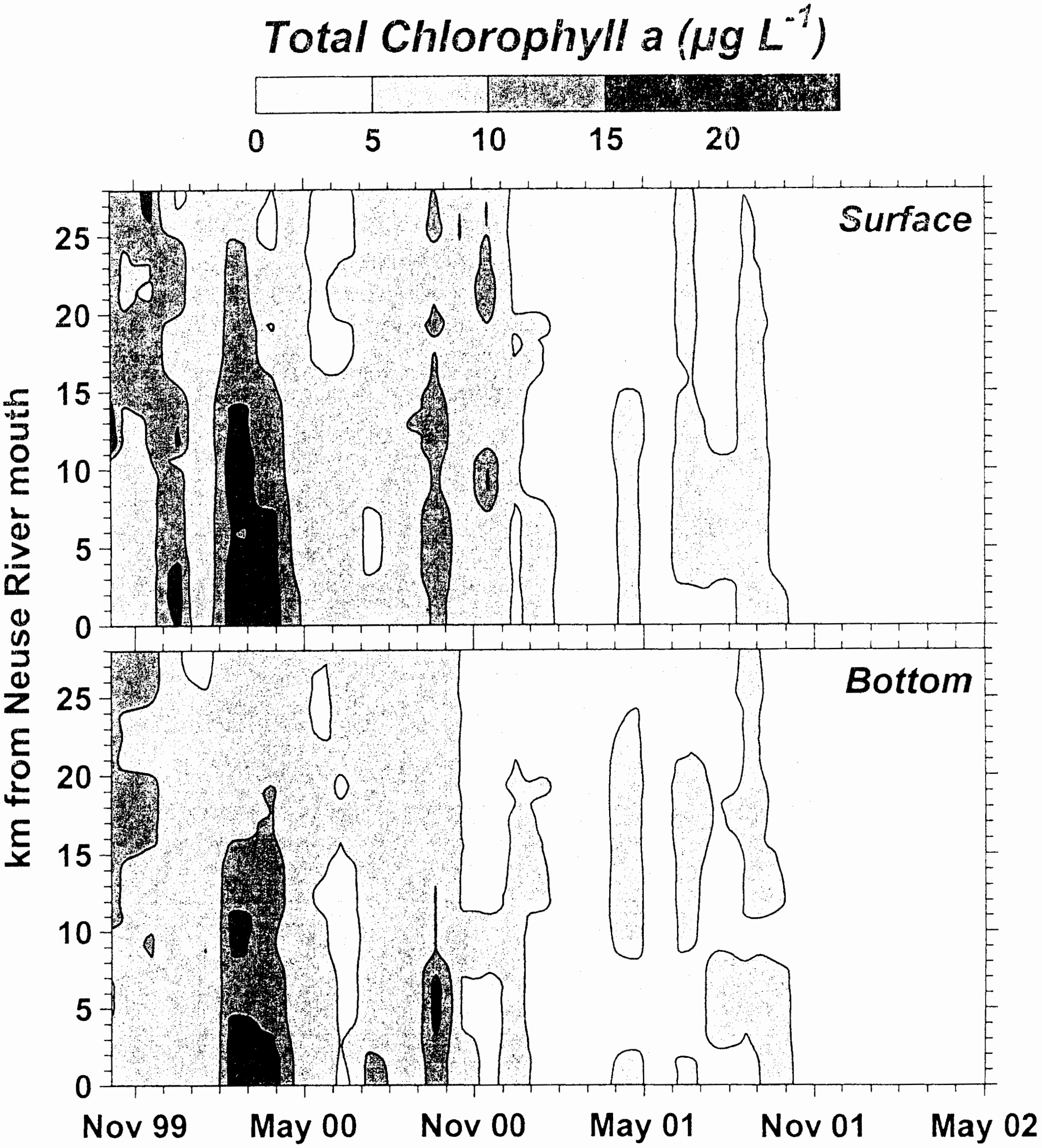


Figure 14. Space-time plot for total chlorophyll *a* (µg L^{-1}, sum of chlorophyll and chlorophyllide *a*) measured by HPLC. Axes as in Fig. 2.



Diagnostic pigment concentrations (chlorophylls and carotenoids) that were measured by HPLC were converted to Chl *a* equivalents of the major phytoplankton taxonomic groups by ChemTax analysis. Immediately following the storms, the phytoplankton community was an approximately equal mixture of cryptomonads (Cryptophytes), cyanobacteria (Cyanophytes), diatoms (Bacillariophytes), and green algae (Chlorophytes) (Figs. 15 to19). Cryptomonads gradually decreased through the study period, except for a large peak in surface samples in November 2000 (Fig. 15). In spring and summer 2000, cryptomonad biomass tended to be highest near the Neuse

River mouth. Cyanobacteria showed a cyclical pattern ranging from highest biomass in the warm periods to almost non-existent concentrations in the winter months (Fig. 16). Vertical differences in cyanobacteria were notable in fall 2000 (greater bottom concentrations) and summer/fall 2001 (greater surface concentrations). With concentrations of > 4 µg L^{-1}, cyanobacteria dominated the community in summer and fall 2001. The pattern of diatom biomass was similar to total chlorophyll *a*, with little difference between surface and bottom waters and fairly even distribution across the sound (Fig. 17). The bloom of diatoms in spring 2000 dominated the community and was also greatest near the river mouth. Green algae also bloomed after the hurricanes with surface peaks that exceeded 5 µg L^{-1} in January and March 2000 (Fig. 18). Bottom biomass was lower during this period. After spring 2000, green algal biomass rarely exceeded 2 µg L^{-1}. Dinoflagellate biomass was low in the sound except for some localized blooms (Fig. 19). Concentrations of dinoflagellate Chl *a* >2 µg L^{-1} were noted in late February/March 2000 and again at one station in August 2001. These blooms were closest to the mouth of the Neuse River estuary.

Figure 15. Space-time plot for group-specific biomass of cryptomonads (Cryptophyta) calculated with ChemTax. Units are in µg chlorophyll *a* L^{-1}. Axes as in Fig. 2.

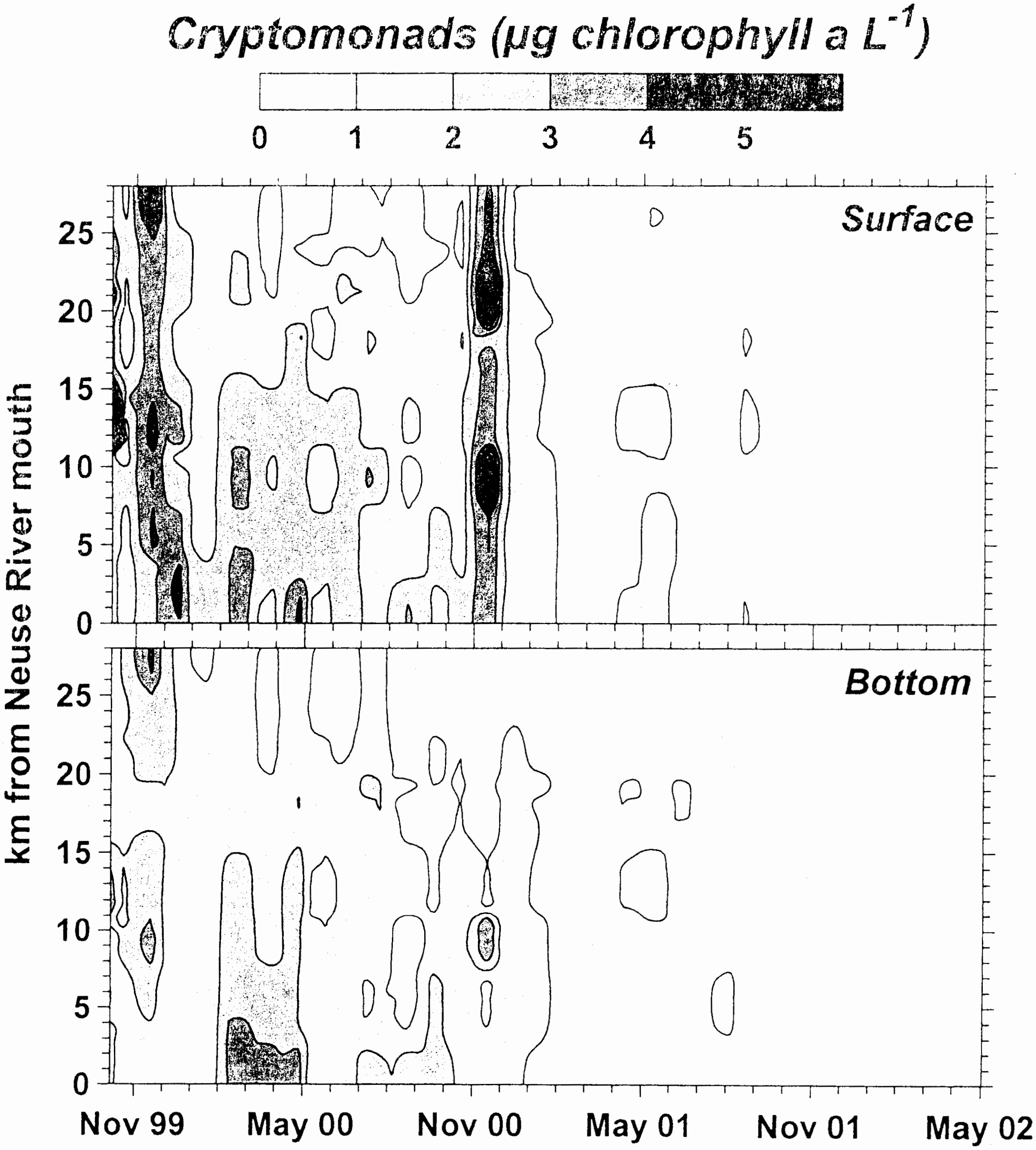


Figure 16. Space-time plot for group-specific biomass of cyanobacteria (Cyanophyta) calculated with ChemTax. Units are in μg chlorophyll *a* L^{-1}. Axes as in Fig. 2.

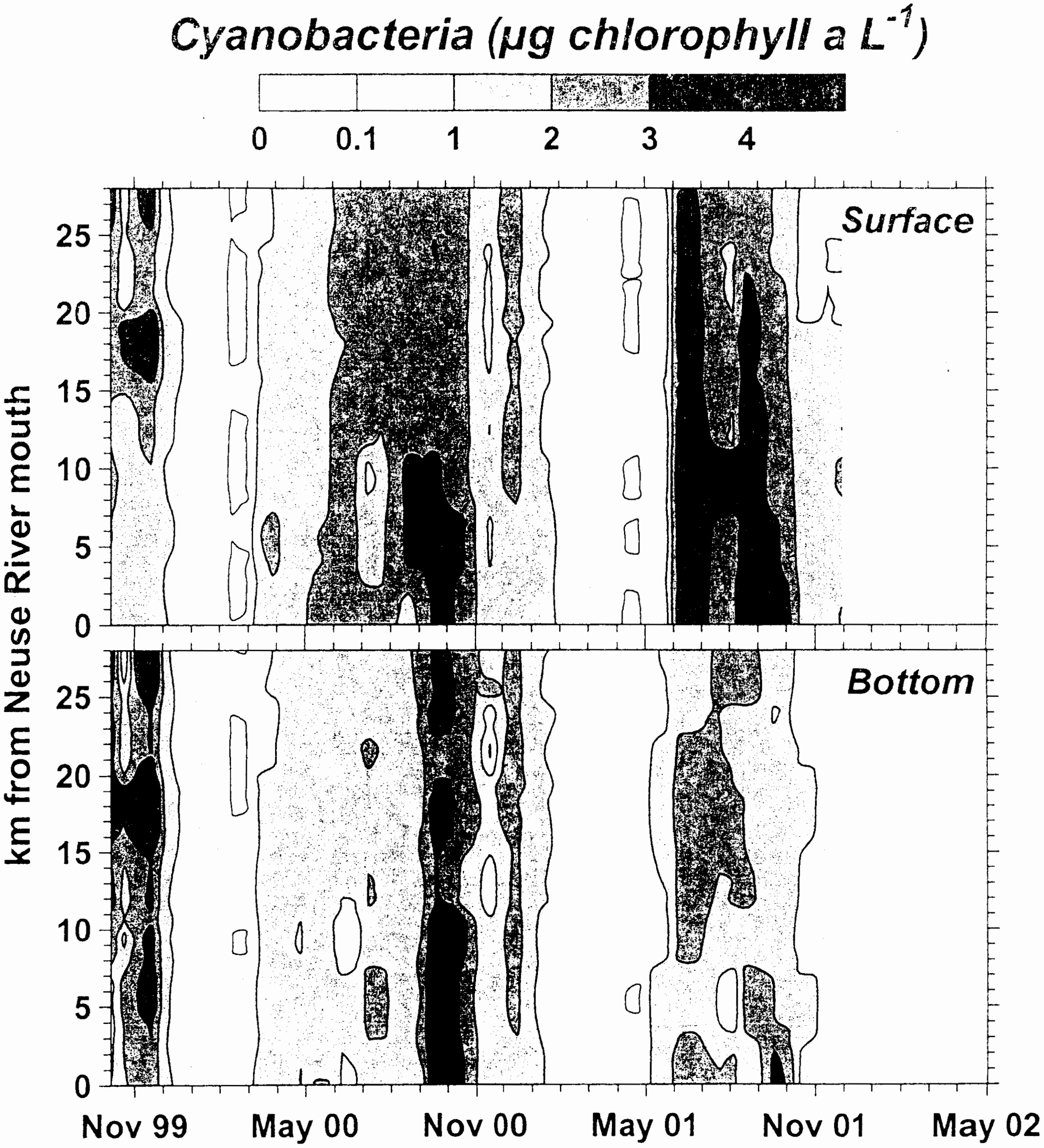


Figure 17. Space-time plot for group-specific biomass of diatoms (Bacillariophyta) calculated with ChemTax. Units are in µg chlorophyll *a* L^{-1}. Axes as in Fig. 2.

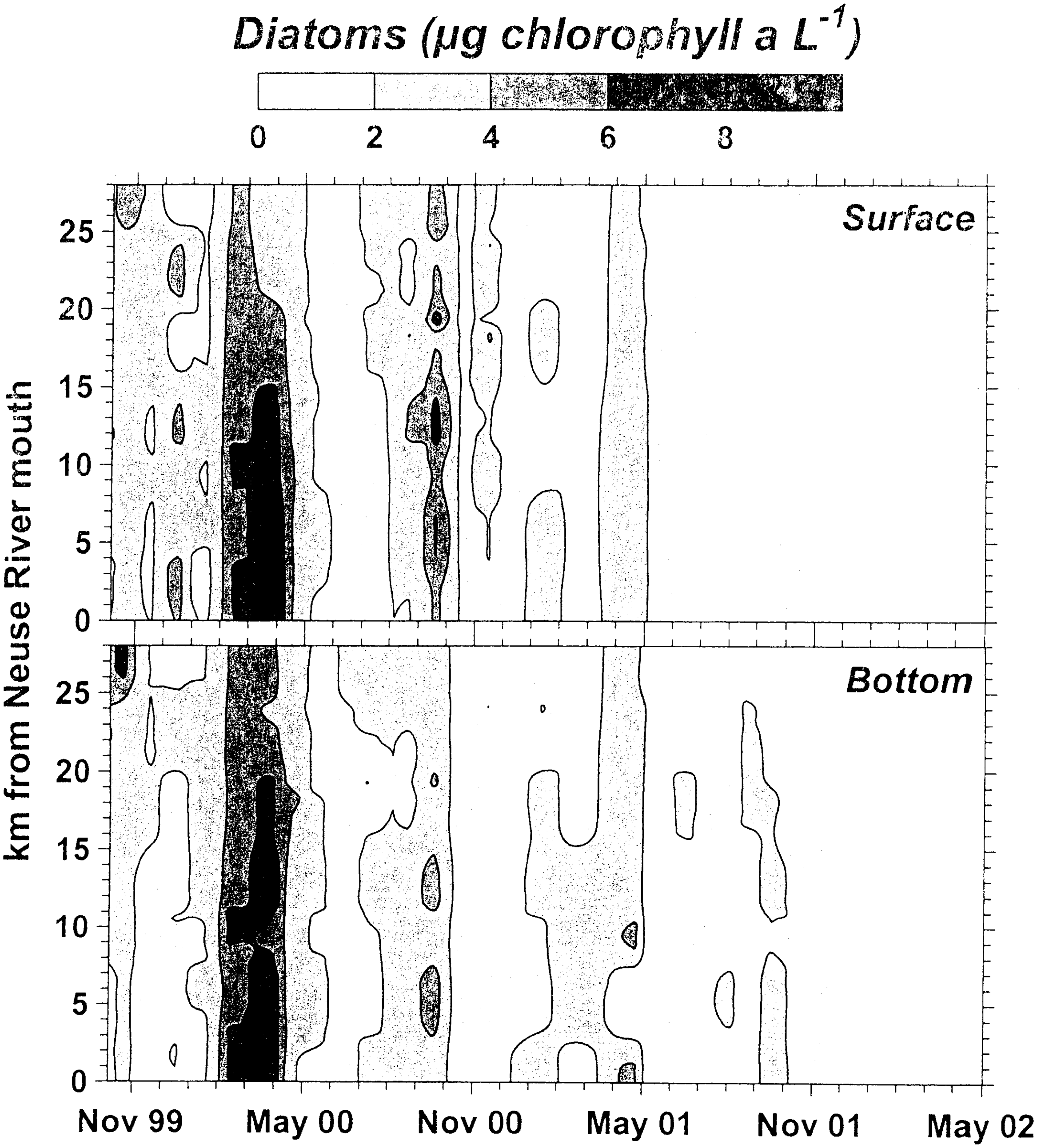


Figure 18. Space-time plot for group-specific biomass of green algae (Chlorophyta) calculated with ChemTax. Units are in µg chlorophyll *a* L^{-1}. Axes as in Fig. 2.

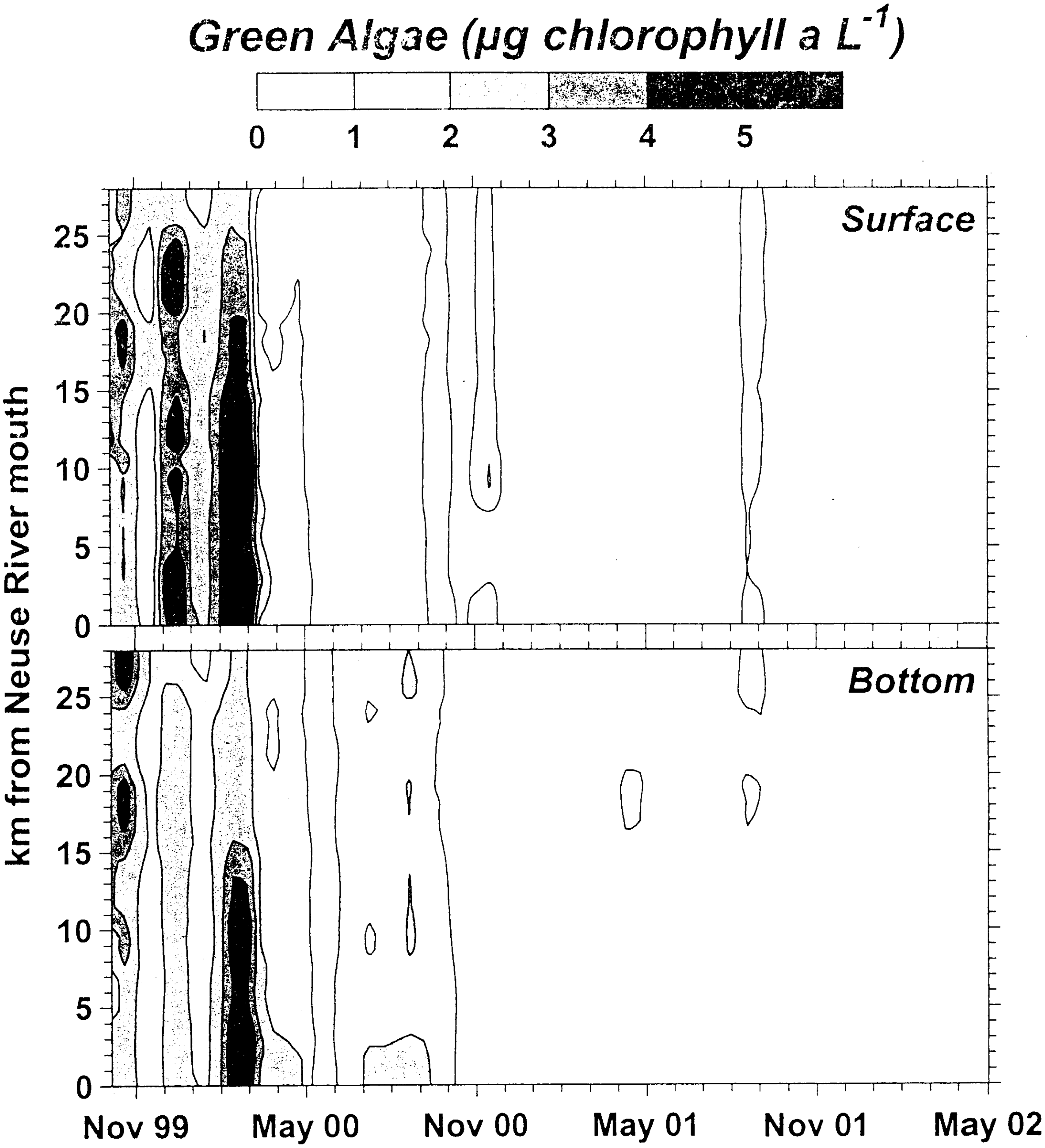
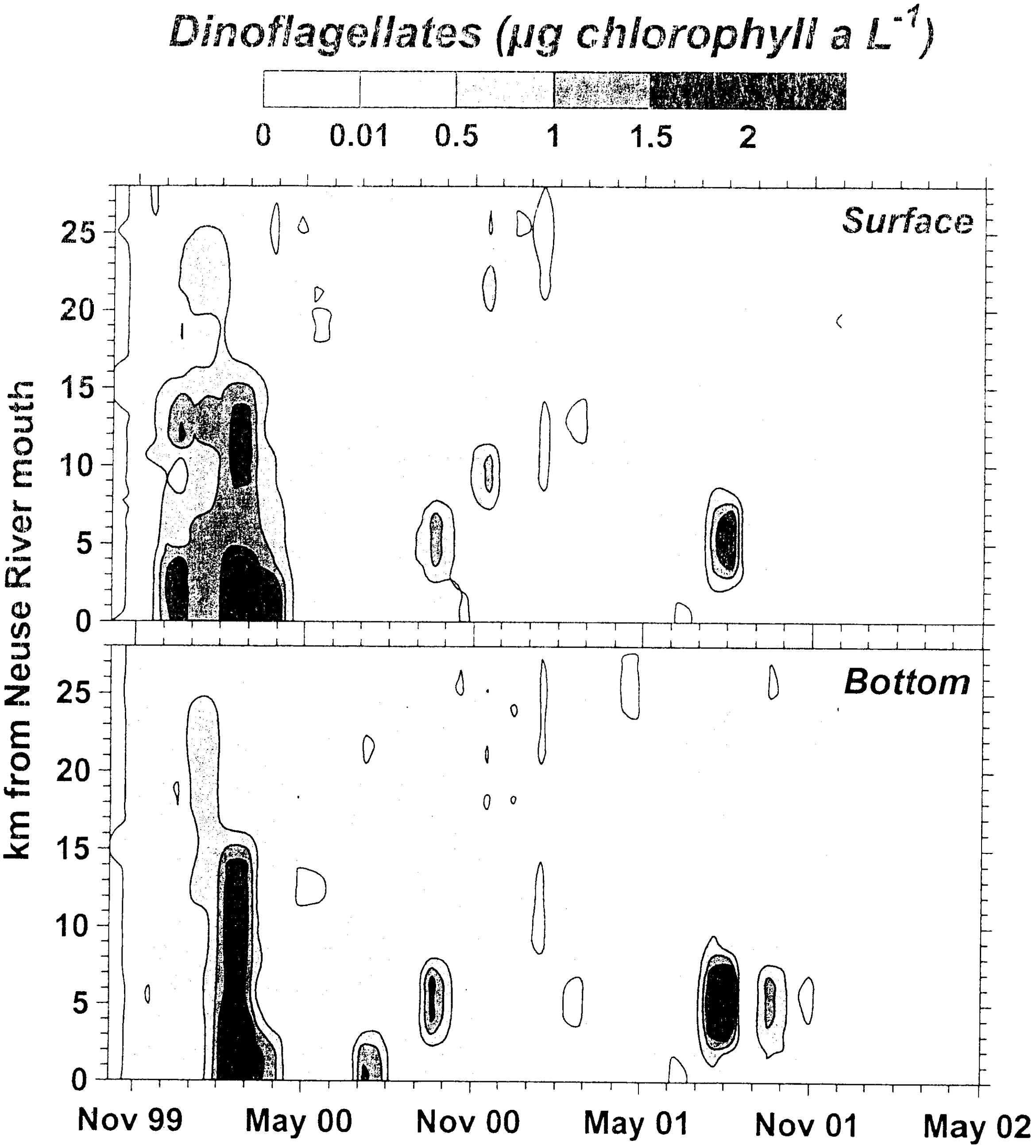


Figure 19. Space-time plot for group-specific biomass of dinoflagellates (Dinophyta) calculated with ChemTax. Units are in µg chlorophyll *a* L^{-1}. Axes as in Fig. 2.



Phosphorus distributions

The average, standard deviation, and range of values for the forms of phosphorus appear in Table 3. The period included in this analysis was from July 2000 to March 2002. The lower limit of detection for all phosphorus analysis at the Central Environmental Laboratory at East Carolina University (ECU) was determined to be 0.2 μM. Soluble reactive phosphorus was the least abundant of the chemical forms, while PP and DOP were comparable in concentration and individually were about twice the concentration of PO_4^{-3}. Temporal and spatial trends of PO_4^{-3} are addressed elsewhere in the report, so the following results will focus only on PP and DOP.

Table 3. Mean, standard deviation, and range of values measured for surface and bottom samples for each form of phosphorus. Units are in μM P.

	Surface			Bottom		
	Mean	St. Dev.	Range	Mean	St. Dev.	Range
Soluble reactive phosphorus	0.304	0.366	<0.2 - 2.23	0.325	0.330	<0.2 - 1.84
Particulate phosphorus	0.595	0.420	<0.2 - 4.58	0.745	1.45	<0.2 - 16.7
Dissolved organic phosphorus	0.607	0.371	<0.2 - 2.36	0.611	0.456	<0.2 - 3.68

Means for each of the 3 forms of phosphorus were similar between surface and bottom samples. Although the average concentrations from bottom waters were slightly higher than from surface waters for all forms of phosphorus, PO_4^{-3} was the only form found to be statistically significant ($p<0.05$) using a paired t-test (Table 3). Standard deviations from the means were all close to 0.4 except for the bottom PP values. This may be due to the occurrence of 3 very high values in the data set.

Dissolved organic phosphorus and PP concentrations showed few trends through space and time. In general, DOP concentrations are below 1.2 μM with occasional peaks throughout. For bottom DOP, the majority of peaks occur in the stations close to the river mouth (Fig. 20). The concentrations of DOP were consistently highest between July and Oct (Fig. 20). The average DOP concentrations showed an increasing trend with distance from the mouth of the river in July of 2000, January, May, June, September, and October of 2001, and February of 2002. Particulate phosphorus concentrations were generally below 0.8 μM with occasional peaks throughout and no spatial trends (Fig. 21). Particulate phosphorus concentrations showed no trends through time except that the lowest concentrations consistently occur in January and February.

Figure 20. Space-time plot for dissolved organic phosphorus (μM). Upper panel is surface data and lower panel is bottom data. Axes as in Fig. 2.

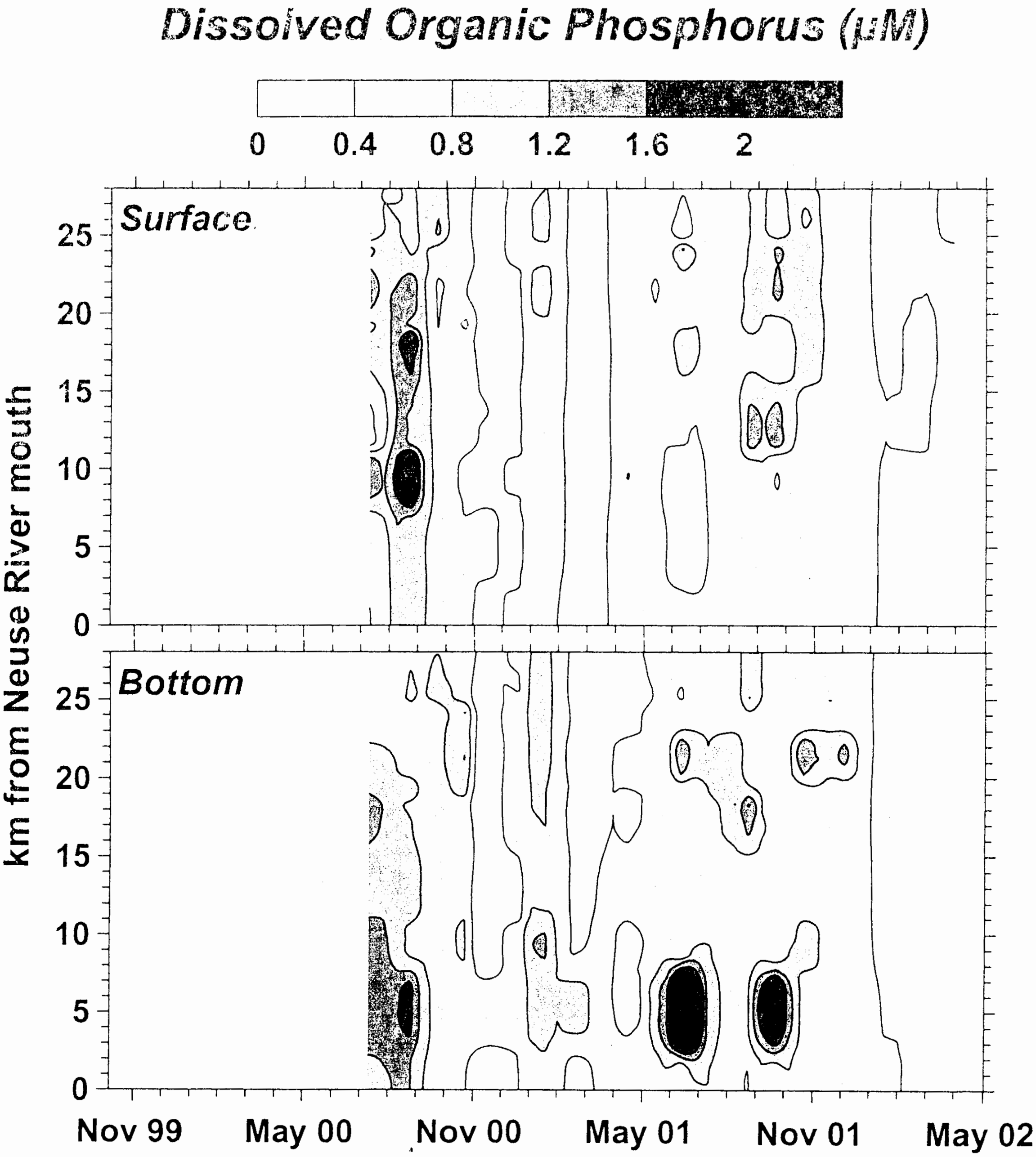
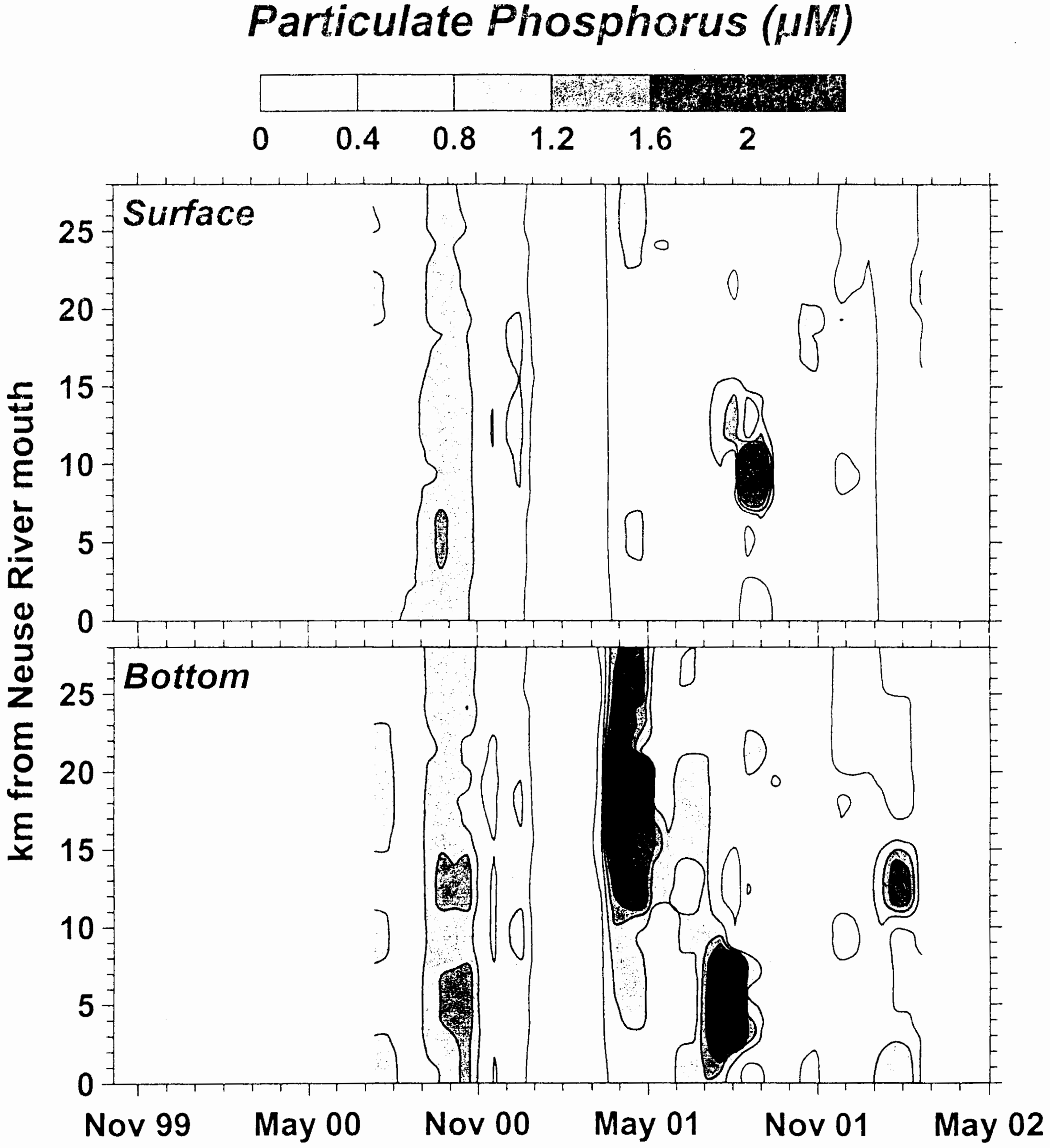


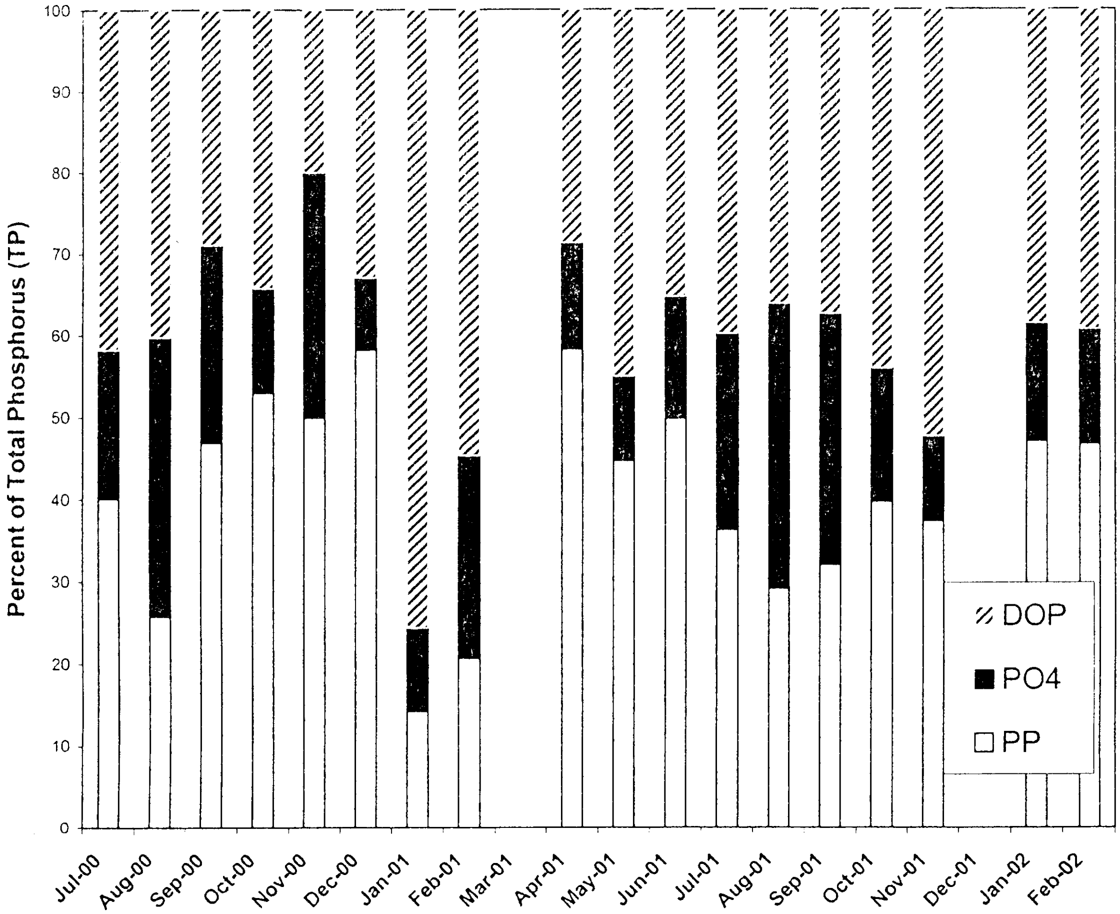
Figure 21. Space-time plot for particulate phosphorus (µM). Upper panel is surface data and lower panel is bottom data. Axes as in Fig. 2.



For each date, the percentages that each form of phosphorus contributed to the total phosphorus were averaged across all stations (Fig. 22). The grand mean percentage that each form of phosphorus contributed to TP in the southern sound was 40.5% PP, 19.2% PO_4^{-3} and 40.3% DOP. However, considerable variability occurred from month to month. PO_4^{-3}which is the form often used as the biologically-available form of phosphorus in water quality analyses, never constituted the majority and was rarely above 30% of the TP in the sound (Figure 22). Dissolved organic phosphorus for each month was greater than 50% of TP in the sound in January, February, and November 2001 (Fig. 22). Some of the lowest levels of TP were measured for these dates.

Particulate phosphorus for each month was greater than 50% of TP in October, November, and December 2000, and April and June of 2001 (Figure 22).

Figure 22. Percent distribution of total phosphorus as dissolved organic, phosphate, and particulate phosphorus. Each bar represents all stations and depths for each time point.



Nitrogen distributions

Much of the distributions of nitrogen species have been discussed elsewhere in this report, as the data for nitrogen spans the entire sampling period. Here we summarize the relative abundance of the three measured forms (i.e. ammonium, nitrate and PN) (Table 4). Particulate nitrogen dominated the concentrations of nitrogen, comprising on average 90% of the nitrogen pool considered. Ammonium was the next most abundant nitrogen species, averaging 3 to 5 times the concentration of nitrate. All forms had concentrations that were significantly greater in bottom waters than in surface waters (paired t-test, $p<0.05$, df =275). This included nitrate concentrations where grand mean for each depth was 0.3 µM. Dissolved organic nitrogen was not measured in this study, and therefore, its contributions are unknown.

Table 4. Mean, standard deviation, and range of values measured for surface and bottom samples for each form of nitrogen. Units are μM N.

	Surface			Bottom		
	Mean	St. Dev.	Range	Mean	St. Dev.	Range
Ammonium	1.0	2.4	<0.2 - 33.6	1.6	2.3	<0.2 - 23.4
Nitrate	0.3	0.5	<0.1 - 3.8	0.3	0.5	<0.1 - 3.3
Particulate N	14.3	6.9	4.8 - 53.1	16.8	10.1	4.8 - 82.3

Hurricane related effects

Conditions near the mouth of the Neuse River estuary (Station 160)

Station 160, located near the mouth of the Neuse River estuary, has been sampled regularly since summer 1997 and represented an opportunity to compare both before and after hurricane conditions. Data were pooled by month for years before mid-September 1999, mid-September 1999 through mid-September 2000, and after mid-September 2000. Data from the three periods were compared using Kruskal-Wallace non-parametric analysis, and differences in medians were estimated by inspection (Table 5). Eight water quality variables were evaluated. Although 26 of 96 possible comparisons were significant at $p<0.05$, only 16 were considered to represent effects of the hurricanes of 1999. Effects attributed to the hurricanes were determined by comparing the medians of the variables between the three time intervals (A, B, and C in Table 5). When the median of a variable for a month during mid-September 1999 through mid-September 2000 (Interval B) was much larger or smaller than the other time intervals (before mid-September 1999 (Interval A) and after mid-September 2000 (Interval C), the effect was viewed as a response to the fall 1999 hurricanes. For example, the median salinity during October 1999 was 2.1 psu, whereas it was 14.5 psu for the October months before the hurricanes and 16.1 psu for the October months after the hurricanes. The Kruskal-Wallace test was significant ($p<0.05$), and the median post-hurricane was different than for the other intervals. This was considered to be a hurricane effect. Significant differences not unique to this middle time interval were not considered as being representative of hurricane effects.

Table 5. Monthly median values of variables for Neuse River Station 160 for years before mid-September 1999 (A), mid-September 1999-2000 (B), and after mid-September 2000 (C). **BOLD** number groupings indicate significance at $p<0.05$. ***Bold, Italicized*** and ***Underlined*** number groupings represent variables and months that were considered to demonstrate hurricane effects (i.e., By inspection, interval B appeared to be different from the other intervals).

Month	Interval	Salinity (psu)	POC (µg C L^{-1})	PN (µg N L^{-1})	NO_3^- (µM)	NH_4^+ (µM)	PO_4^{-3} (µM)	Chl. *a* (µg L^{-1})	PPR (mg C m^{-3} h^{-1})
9	A	15.3	1774	288	***0.86***	***4.03***	1.22	**6.5**	35.2
	B	6.8	819	121	***2.04***	***13.08***	2.14	**6.2**	23.0
	C	15.3	1706	304	***0.04***	***0.87***	1.30	**13.7**	79.4
10	A	***14.5***	1394	236	***0.71***	**5.57**	***0.71***	**3.8**	25.4
	B	***2.1***	1480	205	***8.56***	**5.84**	***1.53***	**10.1**	47.2
	C	***16.1***	1380	245	***0.04***	**0.85**	***0.51***	**10.1**	19.9
11	A	***12.2***	1273	211	***1.01***	1.64	**0.64**	***2.9***	12.8
	B	***4.4***	974	169	***11.49***	1.54	**0.76**	***19.9***	50.2
	C	***17.8***	1167	204	***0.01***	0.79	**0.18**	***6.2***	18.7
12	A	***15.0***	1139	178	***0.64***	0.82	0.94	***3.1***	18.3
	B	***8.3***	1418	242	***2.96***	1.61	0.30	***25.0***	85.0
	C	***17.3***	1226	187	***0.08***	0.74	0.36	***11.2***	14.3
1	A	13.0	1727	239	0.46	0.81	0.70	***7.9***	38.7
	B	8.5	2370	435	0.04	1.06	0.01	***42.2***	92.4
	C	13.7	2067	207	0.49	0.54	0.54	***15.9***	12.6
2	A	**7.3**	**2330**	***305***	2.55	**1.05**	0.52	***8.0***	84.8
	B	**7.2**	**2210**	***380***	3.30	**0.74**	0.37	***31.1***	35.4
	C	**13.3**	**1067**	***215***	0.04	**0.36**	0.65	***6.1***	16.1
3	A	10.6	1800	***240***	0.04	0.79	0.01	***9.2***	27.6
	B	8.7	3586	***439***	0.04	0.51	0.01	***45.5***	60.6
	C	11.8	1610	***211***	0.04	0.89	0.01	***10.0***	21.3
4	A	10.8	1603	248	0.12	0.55	0.27	8.4	25.5
	B	9.4	1926	298	0.04	1.24	0.01	17.8	33.3
	C	13.3	957	183	0.62	1.02	0.12	6.7	31.2
5	A	11.2	1577	236	0.13	0.90	0.08	5.9	16.5
	B	11.6	1806	278	0.04	1.06	0.01	15.5	49.8
	C	13.1	1256	226	0.09	0.61	0.05	11.0	12.8
6	A	**9.4**	1710	238	0.27	1.21	0.47	6.7	26.8
	B	**14.0**	1657	290	0.04	1.98	0.90	10.2	89.4
	C	**16.5**	2132	358	0.39	1.03	0.25	8.6	16.9
7	A	14.6	1480	274	**0.41**	1.16	1.16	9.2	28.6
	B	17.2	1556	266	**0.04**	0.99	1.49	11.9	34.4
	C	16.3	1229	215	**0.04**	1.24	1.08	25.3	15.7
8	A	14.7	1708	336	**0.31**	1.80	**0.86**	9.1	44.6
	B	16.7	1252	232	**0.04**	2.01	**2.99**	8.9	50.6
	C	18.1	1412	288	**0.04**	0.88	**1.81**	14.9	24.2

Hurricane effects were considered for the following variables: salinity, concentrations of $NO_3^- + NO_2^-$, NH_4^+, PO_4^{-3}, POC, PN, Chl *a*, and primary productivity (PPR). Salinity did not demonstrate a significant hurricane effect until October 1999, but this effect continued through December. However, during the latter part of September 1999, median salinity at Station 160 was less than one half that for September of other years, which is in broad agreement with other post-hurricane studies carried out in this region (Paerl et al. 2000, 2001). Nitrate and ammonium concentrations were significantly higher in the latter part of September 1999 than in September of years before and after 1999. Nitrate concentrations stayed significantly higher through December 1999, whereas ammonium concentrations did not differ from previous years in October. No evidence of a significant hurricane effect was found for ammonium during subsequent months. Soluble reactive phosphorus was significantly higher only in October 1999 but demonstrated trends of high PO_4^{-3} during the months before and after that in 1999.

Particulate carbon and PN did not demonstrate strong responses to the hurricanes. No significant hurricane effects were found for POC. Potential hurricane effects were noted for February and March 2000. Statistically significant differences were found, and the highest concentrations were during 2000 for both months.

The longest significant effect of the 1999 hurricanes occurred for Chl *a*. Chlorophyll *a* concentrations remained relatively high from November 1999 through March 2000. Median concentrations during this period ranged from 19 to 46 $\mu g\ L^{-1}$, whereas the range for other years during these months was 3 to 16 $\mu g\ L^{-1}$. Higher medians were also found for April through June 2000, but these were not significantly different from those of other years. In contrast PPR showed no significant differences for any month. Our inability to obtain statistically significant effects for PPR may rest in the inter-annual variability of this measure. The highest median value for each month was found for the post-hurricane year for all months except September and February. Thus, there was a trend for enhanced primary productivity from the hurricanes, but this could not be substantiated statistically by our analysis.

Conditions of southern Pamlico Sound

We wanted to directly and quantitatively evaluate the effects of the hurricanes on the water quality of PS in an effort to test our hypotheses relating timing of concentration change to mechanism of impact. Temporal changes appeared more dramatic than spatial changes, based on the surfer plots shown earlier. Therefore, we pooled data from all stations for any particular time interval (i.e., month). We tested whether water quality variables for each month for one year after the hurricanes (October 1999-August 2000) were significantly different from subsequent months (October 2000-April 2002), using Kruskal-Wallace non-parametric analysis. Initially, data for any calendar month (October 2000-April 2002) were pooled for all years (e.g. data from February 2001 and 2002 were pooled). We reasoned that by combining years, we would incorporate normal inter-annual variation with which to compare the hurricane year. Seventy-two of 99 possible comparisons were statistically significant at $p<0.05$ as tested (data not shown). However, given the number of significant results, we were concerned about our pooling scheme and its consequences for interpretation. We therefore compared the three year intervals separately: October 1999-August 2000, September 2000-August 2001, and September 2001-April 2002.

Data from the three periods were compared using Kruskal-Wallace non-parametric analysis, and differences in medians were estimated by inspection (Table 6). Nine water quality variables were evaluated. Eighty-seven of 108 possible comparisons were significant at $p<0.05$, but it was difficult to isolate effects of the hurricanes of 1999. Months between May and August were sampled only during two years after the hurricane, and September was sampled only after one year from the hurricanes (e.g., 2000 and 2001). Potential hurricane effects were based on the criteria that: (1) the Kruskal-Wallace test was significant <0.05 and (2) the median of a variable during a month within 1 year after the hurricanes was higher or lower than that month in the subsequent years or year.

Some trends were noted in the patterns of significance (Table 6). Salinity was significantly lower during most of the year after the hurricanes. This was particularly apparent from October 1999 though April 2000, although some differences were noted later in the year.

Particulate carbon and PN were higher in October 1999 than for that month in other years, but this was not seen for November. However, significantly higher concentrations of PN were found in December and of both were found again from January 2000 through March 2000. Higher concentrations of one or both also occurred during subsequent months in the year following the hurricanes.

Dissolved organic carbon demonstrated significant differences among years for all months except July. However, the pattern for a particular month was not necessarily as one would predict for a hurricane effect. Concentrations of DOC were similar for October and November 1999 and 2000 but these four measures were higher than for October or November 2001. DOC concentration was significantly higher during December, January, March, May, June, and August after the hurricanes than during those months in subsequent years. Thus, the effects of the hurricanes on DOC concentrations may have been long term.

Concentrations of dissolved inorganic nutrients ($NO_3^- + NO_2^-$, NH_4^+, PO_4^{-3}) were all higher in October 1999 than in October 2000 and 2001, but patterns after that month were nutrient dependent. Nitrate and ammonium concentrations decreased to near those of other years during November 1999, and no consistent pattern of higher concentration was seen in the following year. In fact, nitrate during December 1999, May 2000 and June 2000 was significantly lower than during those months in the subsequent year. However, nitrate concentrations in PS from November 2000 to June 2001 were generally higher than during the year before or after. We do not know if this is a delayed effect of the hurricanes or part of the natural variation of the system. PO_4^{-3} decreased to concentrations significantly lower than the subsequent years during November and December 1999. Low PO_4^{-3} concentrations were also found during other post-hurricane months, but these were not necessarily lower than for months of both subsequent years. Thus, dissolved inorganic nutrients were removed rapidly after being pulsed into the sound.

Chlorophyll *a* concentrations and primary productivity were elevated for an extended period after the hurricanes. Both were significantly higher from October 1999 until May 2000, compared to later corresponding months. Differences were seen less frequently later in the year.

Table 6. Monthly median values of variables measured in Pamlico Sound for mid-Sep. 1999 to Aug.2000 (B), September 2000 to Aug. 2001 (C), and after Aug. 2001 (D). **Bold** numbers indicate significance at $p<0.05$. ***Bold, italicized*** and ***underlined*** number groupings indicate hurricane values that appear uniquely different by inspection.

Month	Interval	Salinity (psu)	POC (μg C L^{-1})	PN (μg N L^{-1})	DOC (μM)	NO_3^- (μM)	NH_4^+ (μM)	PO_4^{-3} (μM)	Chl. *a* (μg L^{-1})	PPR (mg C m^{-3} h^{-1})
10	B	***9.6***	***1622***	***303***	**679**	***1.01***	***5.74***	***0.48***	***16***	***38.6***
	C	***20.5***	***1172***	***183***	**665**	***0.89***	***1.37***	***0.24***	***6***	***18.1***
	D	***23.2***	***744***	***117***	**449**	***0.04***	***0.94***	***0.27***	***3***	***9.4***
11	B	***8.8***	**1510**	**280**	**688**	**0.04**	**1.06**	***0.01***	***15***	***28.7***
	C	***17.2***	**2309**	**343**	**692**	**0.33**	1.19	***0.25***	***7***	***23.6***
	D	***23.7***	**799**	**111**	**365**	**0.04**	**0.89**	***0.13***	***3***	***5.4***
12	B	***9.9***	1325	***232***	***672***	***0.04***	0.78	***0.01***	***13***	***28***
	C	***17.4***	1394	***199***	***560***	***0.61***	0.88	***0.03***	***6***	***14.7***
1	B	***11.7***	***1514***	***259***	***698***	**0.04**	**1.19**	**0.01**	***11***	***34.3***
	C	***17.0***	***1319***	***120***	***588***	**0.42**	**1.15**	**0.04**	***5***	***7.8***
	D	***22.6***	***558***	***86***	***319***	**0.04**	**0.15**	**0.01**	***3***	***11.7***
2	B	***10.8***	***3251***	***438***	**677**	**0.04**	0.59	**0.01**	***22***	***51.3***
	C	***17.6***	***752***	***91***	**647**	**0.58**	0.59	**0.01**	***3***	***5.6***
	D	***22.4***	***746***	***103***	**242**	**0.04**	0.55	**0.04**	***4***	***11.8***
3	B	***12.2***	**2208**	***287***	***634***	0.04	0.53	0.01	***16***	***35.5***
	D	***22.0***	**756**	***95***	***281***	0.04	0.51	0.01	***3***	***11.8***
4	B	***14.9***	**1608**	**211**	**462**	**0.04**	**0.82**	**0.01**	***11***	***30.3***
	C	***17.4***	**1490**	**212**	**471**	**0.76**	**1.53**	**0.27**	***8***	***19.2***
	D	***22.5***	**589**	**100**	**302**	**0.04**	**0.44**	**0.01**	***3***	***14.2***
5	B	19.0	***1171***	***173***	***510***	***0.04***	***1.10***	0.01	***7***	***16.2***
	C	19.3	***820***	***129***	***418***	***0.11***	***0.62***	0.01	***4***	***10.8***
6	B	***17.7***	1369	**180**	***668***	***0.04***	1.28	***0.01***	7	14.8
	C	***20.3***	1351	**201**	***457***	***0.33***	1.31	***0.10***	7	10.2
7	B	22.3	***1305***	***212***	483	0.04	***1.14***	***0.01***	***9***	21.8
	C	22.2	***1091***	***147***	470	0.04	***0.61***	***0.44***	***7***	16.8
8	B	***22.1***	***1392***	214	***536***	0.04	0.84	***1.34***	9	28.5
	C	***23.5***	***1089***	212	***409***	0.04	0.98	***0.86***	9	18.5
9	C	**18.7**	**1545**	235	**586**	0.04	**0.99**	0.82	**13**	**41.9**
	D	**23.2**	**1101**	220	**433**	0.04	**1.19**	0.72	**7**	**27.4**

DISCUSSION

We conducted a thirty-one month study of the water quality of southern Pamlico Sound. This study was prompted by the series of hurricanes and subsequent flooding that occurred in the fall of 1999 and addresses several hypotheses concerning how nutrients move into the sound from its sub-estuaries and how these nutrients are processed. However, the extended period of monitoring has also allowed us to:

(1) establish the current environmental and water quality conditions of the southern sound and relate these conditions to the ability of the sub-estuaries to protect the sounds from nutrient loading
(2) infer mechanisms by which the sound may be nutrified during extreme hydrologic events, and
(3) establish a baseline against which to gauge ecological change.

Our experiences provide insight into future environmental monitoring and management of this important resource to the State of North Carolina.

Environmental and water quality conditions of the southern Pamlico Sound and its relation to the Sound's sub-estuaries

Under normal flow conditions, the nutrient filtering capacity of the sub-estuaries of PS has been well established (Christian et al. 1991, Stanley 1992, Paerl et al. 1998, Christian and Thomas 2000, Glasgow and Burkholder 2000). The concentrations of dissolved inorganic nutrients are often observed to decrease along a gradient from the head of the sub-estuaries to their mouth. These observations have been used to infer that PS is nutrient poor and unlikely to demonstrate the problems of eutrophication (e.g., harmful algal blooms, hypoxia, and fish kills) found in its sub-estuaries. These inferences have been largely unsupported by measurements of water quality in the sound (Paerl and Ramus 1998). Prior to this study, research activities have been primarily *ad hoc* teaching and exploratory research cruises (L. Crowder and J. Ramus, Duke University Marine Lab; H. Paerl and R. Luettich, UNC-CH IMS)(excepting EMAP, Balthis et al. 1998). Only hydrographic variables had been monitored in the sound prior to 1963 (Winslow, 1889, Grave 1904, Coker 1907, Roelofs and Bumpus 1953). Woods (1967) collected limited water quality data from 1963 to 1966. These limited studies have supported the contention that the sound is less nutrient rich than the sub-estuaries. However, this study is the first intensive and extensive effort using methods that are compatible with those used in the sub-estuaries.

We have grouped the measured variables into 4 classes (i.e., hydrographic, dissolved nutrients, particulate nutrients, and phytoplankton) and assessed temporal and spatial conditions of variables within each class. The hydrographic variables reflect the wide, shallow morphology of the sound, where vertical stratification is controlled by wind mixing. When conditions permit, vertical differences in salinity at one station can often be similar to or larger than the salinity gradient across the sound from the mouth of the Neuse River estuary to Ocracoke Inlet. Hypoxia (< 4 mg DO L^{-1}) often ensues when vertical stratification occurs during warmer months. Hypoxia can extend for tens to perhaps hundreds of km^2 and last for weeks to perhaps months. Thus, the sound ecosystem has the metabolic capability to deplete dissolved oxygen from water under common physical conditions of temperature and stratification. The frequency of sampling, however, has not allowed us to estimate how quickly hypoxia can develop once proper physical conditions form. Therefore, we have demonstrated the occurrence and potential for wide-spread hypoxia; however, more detailed studies would be required to better assess its significance.

Dissolved inorganic nitrogen and phosphorus appear to have a short turnover time within the sound. Concentrations are maintained at sub µM to a few µM, with the exception of the period shortly after the storms of fall 1999. The pulse of relatively high concentrations after the storms disappeared rapidly, and subsequent months often had lesser concentrations than during the same months in later years. These reduced concentrations may have been caused by the algal blooms and associated nutrient assimilation that occurred in late winter and early spring 2000. Warm weather was associated with increases in concentrations of nitrate, ammonium and PO_4^{-3}, found at least during some years. These were not necessarily associated with storms and loading through the sub-estuaries. Bottom waters had higher concentrations during these times than in surface waters, indicative of in situ remineralization (Rizzo and Christian 1996, Luettich et al. 2000). Thus, high concentrations may have two causes: loading from the sub-estuaries associated with storm events and internal loading from remineralization.

Distributions of the various inorganic nitrogen species in the sound were similar to those at the mouth of the Neuse River or Pamlico River estuaries (Christian et al. 1991, Stanley 1992). Concentrations of DIN in the sound were similar or less than in the sub-estuaries. The water quality in the sound and at the mouth (e.g., station 160) of the sub-estuaries is much different than for the heads of the sub-estuaries. In the sound and at the mouths, ammonium concentrations were higher than for nitrate, even immediately after the hurricanes. Although nitrate tends to dominate dissolved inorganic nitrogen loaded into the sub-estuaries, hurricanes and large storms have been known to shift the loading towards dominance by ammonium (Paerl et al. 1998). Internal loading from remineralization would also be dominated by ammonium. Recycling of nitrogen in the sub-estuaries has been shown to be extremely important (Boyer et al. 1994, Christian and Thomas 2000) and would result in loading ammonium preferentially to nitrate, as water leaves the sub-estuaries and enters the sound. Thus, only the most intense storms and flooding are likely to cause the southern sound to have greater standing stocks of nitrate than ammonium.

Dissolved phosphorus includes inorganic and organic forms. The concentrations of the most abundant inorganic form, PO_4^{-3}, are generally low with the highest concentrations during warm months and strong freshwater inputs from storms. Concentrations of PO_4^{-3} in the sound were generally less than previously reported for the mouth of the Neuse River estuary (Christian et al. 1991) and reported here for station 160. Highest concentrations were found in bottom waters during warm months, another indication of the importance of remineralization to the nutrient budgets of the sound. Soluble reactive phosphorus is the most commonly measured dissolved form of phosphorus and likely the most biologically active. However, dissolved organic phosphorus (TDP minus PO_4^{-3}) often was more abundant than PO_4^{-3}. On average DOP composed 40% of total phosphorus (including PP) and PO_4^{-3} composed 19%. Soluble reactive phosphorus never exceeded 30% of the TP concentrations. Thus, a potentially important fraction of dissolved phosphorus has been unmeasured in studies of the estuarine systems of North Carolina (Christian et al. 1991, Paerl et al. 1998). We do not know how DOP responded to the storms of 1999, as funding for this portion of the project did not begin until later in 2000.

The pattern of DOC concentrations is in contrast to the rapid changes seen for the other dissolved nutrients. Dissolved organic carbon concentrations ranged by only a factor of about 3 (monthly medians of 242-698 µM) and no peak was seen in the few months following the hurricanes. Elevated concentrations were found later in winter and in summer 2000, however. It is difficult to discern the causes for the pattern. This is due to the myriad of molecules and their turnover times that comprise DOC (Amon and Benner 1996). The flooding freshwater did not appear to have high concentrations of DOC and subsequent increases in concentration may have been associated with biological activity in the sound. Algal blooms were noted, and these may have produced measurable increases in DOC concentration.

Particulate carbon and PN displayed an immediate response to the hurricanes with high concentrations in October 1999. This reflected the freshwater flooding of the sound and was similar to the Chl *a* response. However, whereas Chl *a* concentrations remained high, concentrations of POC and PN decreased in November 1999. Thus, phytoplankton were unlikely to be the sole contributor to the particulate nutrients initially. Allochthonous particulate matter coming in during the fall of 1999 may have included considerable, but unquantified, detritus. However, C:N for the months following the hurricanes were not high. The median molar ratios of C and N remained below 7 for the remainder of 1999 after the hurricanes. During the rest of the study, monthly medians rarely exceeded 10. High C:N would be indicative of terrestrially or macrophyte derived material (Paerl et al. 1998). Because C:N was low consistently, terrestrially derived detritus did not appear to dominate the particulate matter, even after the hurricanes. Particulate matter and phytoplankton may have been closely linked, during the remainder of the study. Spearman's rank correlations between chorophyll *a* and either POC or PN were significant at $p< 0.001$ for each month except August (neither significant at alpha = 0.05), September (PN not significant at alpha = 0.05) and December (POC not significant at alpha = 0.05). Thus, particulate concentrations in PS would appear to be related to the primary producer standing crops and their activity.

Phytoplankton community biomass was enhanced after the hurricanes beginning with a rapid increase from pre-storm Chl *a* levels of 5 µg L-1 or less (Paerl et al. 2001). The blooms were likely initiated through the input of floodwater N (Pinckney et al. 1998), although many factors are known to regulate estuarine phytoplankton (Day et al. 1989). Light may have been limiting in the fall and late winter after the storms (euphotic zone calculated from K_d ranged from 1 to 2.8 m deep), but considering that this is a wind-driven system, phytoplankton would probably be mixed enough to satisfy their light requirements. The early fall bloom was shifted away from the river mouth, probably through advection with the floodwaters (Tester et al. 2003). After a brief decline, Chl *a* peaked again in spring 2000, this time centered near the river mouth. This bloom was probably related to nutrient loading as the peak coincided with another pulse of freshwater, but nutrient uptake was so rapid that concentrations remained low or below detection during this period. Chlorophyll *a* decreased after spring 2000, with some small seasonal increases. The drop in biomass may reflect a return to more nutrient limited conditions, but it is also possible that the herbivore community re-established itself and began to graze on the phytoplankton community. This is supported by observations of large populations of gelatinous and crustacean zooplankton that appeared in net hauls from spring 2000 onward (G. Kleppel, personal communication).

Total phytoplankton biomass returned to pre-hurricane levels within about 8 months (Paerl et al. 2001), but the phytoplankton community composition continued to change over the study period. The initial post-hurricane community was a mixture of all the taxonomic groups except dinoflagellates. Previous bioassay work using Neuse River phytoplankton assemblages revealed a similar community composition under N enriched conditions (Pinckney et al. 2001). In the sound, green algae and diatoms responded to the floodwaters with dramatic biomass increases. Green algae became unimportant after February 2000, perhaps resulting from increased salinity, decreased nutrients, or increased selective grazing. Diatoms maintained biomass dominance throughout the following year with peak biomass linked to river discharge peaks. This may result from rapid utilization of external N (Collos 1989, Pinckney et al. 1999). Dinoflagellates, which are a seasonally dominant component of local estuarine communities (Pinckney et al. 1998, Litaker et al. 2002), had only a modest bloom in the first winter/spring period and were otherwise rare. Either this group could not meet its resource requirements after the flood, or an efficient grazer community kept dinoflagellate biomass low. As a group, cyanobacteria have relatively slow growth rates (i.e., long doubling times) and show a strong preference for relatively warm conditions (>15 °C) (Paerl 1999). This is reflected in both their lack of immediate response to the floodwaters and relatively large growth responses throughout the sound during the following two summers (2000 & 2001). It is unclear why cryptomonads maintained a significant presence in the first year and fall 2000, yet were much lower in 2001.

Mechanisms by which the Sound may be nutrified during extreme hydrologic events

We postulated that the mechanism by which nutrients entered the sound could be inferred by the response of standing stocks of materials in the sound subsequent to the storms of 1999. The sound may receive this episodic loading by at least 3 mechanisms:

(1) Flow-through of unprocessed nutrients
(2) Resuspension and transport of sediments with subsequent biological and

chemical release of nutrients
(3) Movement of phytoplankton blooms down the sub-estuary to near the mouth allowing algal biomass to enter the sound for processing

Evidence for all three mechanisms were observed.

The highest concentrations of dissolved inorganic nutrients were found in the sound during the first samplings after the storm sequence. These appear to be the direct result of loading and the flow-through of unprocessed nutrients. Within a month or two later, however, dissolved inorganic nutrients had returned to base concentrations. Christian and Thomas (2000) studied nitrogen cycling within the Neuse River estuary using ecological network analysis. They found that considerable recycling of nitrogen normally occurs within the Neuse River estuary, but this recycling decreases as loading increases and residence time decreases. Greater amounts of nitrogen escape to the Sound as more water flows through the sub-estuary. This finding agrees with other studies that have related discharge or residence time with the export of nutrients from estuaries (Nixon et al. 1996, Dettmann 2001). The work of Christian and Thomas (2000) was extended by Peierls et al. (2003) to the period of the storms. They inferred that little recycling occurred during that period and that all or almost all of the nitrogen entering the Neuse River estuary was exported to PS. Thus, the predicted patterns of dissolved inorganic nutrient concentrations in the sound were found to support the hypothesis that the sound was nutrified by this mechanism.

Nutrients were also postulated to enter the sound by resuspension and transport of sediments with subsequent biological and chemical release of nutrients. The proposed pattern of support would be a pulse of particulate matter in the Sound followed by an increase in dissolved nutrients during warmer months. The patterns of particulate matter and dissolved inorganic nutrients do not clearly support this hypothesis. Concentrations of PN and POC were elevated significantly during October 1999 relative to subsequent Octobers, while concentrations in November 1999 were not higher than subsequent Novembers. The molar C:N values of particulate matter after the storms were less than 10, whereas the surface sediments of the Neuse River estuary range from 10 to 17 (Luettich et al. 2000). Particulate carbon and nitrogen concentrations rose again in winter 1999-2000 beyond that for winter months in subsequent years. However, this rise was associated with algal blooms. Thus, evidence of significant inputs of sediment is lacking. Given the high density of sediment, however, it is possible that large amounts were moved into the sound and deposited before the first sampling in October 1999.

Evidence for the release of nutrients from remineralized sediment during warmer months is also not clear. Both ammonium and PO_4^{-3} concentrations were found to increase during summer months. Ammonium concentrations were significantly higher during May and July 2000 than during those months in 2001. PO_4^{-3} concentrations were higher in August 2000 than August 2001, but significantly less in June and July 2000 compared to 2001. Thus, we did not see a consistent pattern of highest concentrations during the warmer months during the year following the storms. This may have been the result of not having much sediment moved into the sound and remineralized within the first year. But other factors may be at work. Remineralized nutrients may have been rapidly removed by phytoplankton or microphytobenthos. Blooms were common during the year after the storms. Also, sediments may have entered the sound, but remineralization may act with a longer time scale than months. Associated nutrients may take years to be released.

Lastly, nutrients may enter the sound fixed into the organic matter of living organisms (primarily phytoplankton and other plankton). Given that the river flow was so high following the storms, it is highly likely that plankton would be transported from the sub-estuary to the sound. This was apparent in the seaward shifted peaks in algal biomass early in the flood period and importance of algae associated with freshwaters. The spring bloom near the Neuse mouth could also be in part due to advection of biomass produced in the lower reaches of the river. We do not know how much of the phytoplankton biomass arose from *in situ* growth and how much from import from the sub-estuary. The appearance of phytoplankton rare to the sound (dinoflagellates, green algae) may indicate transport of nutrients in living organic matter, but there is no way to eliminate the possibility that these taxa were produced within the sound. *In situ* growth would involve nutrient import from other sources.

In summary, nutrients were inferred to enter the sound in dissolved inorganic, particulate and planktonic forms. Dissolved inorganic nutrients quickly decreased to low concentrations. Blooms of phytoplankton continued through subsequent seasons and promoted nutrient cycling. Loading of sediments and subsequent remineralization of associated nutrients could not be readily demonstrated.

Establishing a baseline of information on the water quality of southern Pamlico Sound.

The study reported on here is one of the first to reveal the ecological response of only a segment of Pamlico Sound to the combined effects of anthropogenic nutrient inputs and climatic perturbations, in the form of 3 sequential hurricanes in fall 1999. The southern portion of PS appears to have better water quality than its sub-estuaries, as defined by the concentrations of nutrients and chlorophyll *a*. It does have the capacity for hypoxia during summer months, although the extent could not be determined by our sampling. The system can be perturbed during extreme hydrologic events as we had in fall 1999 and these events are predicted to increase over the next few decades (Gray et al. 1996, Landsea et al. 1999, Goldenberg et al 2001). Nutrient concentrations recovered quickly, but algal blooms continued for much of the following year. We cannot predict how frequently the sound could respond to repetition of such events with such resilience. Only continued study and better understanding will provide such predictive ability.

While the comprehensiveness of the spatial and temporal coverage of the PS was restricted due to limited funding and logistic constraints, the following information was gained as a guideline for filling essential water quality monitoring needs:

(1) Given observed spatial gradients in both nutrient loads and concentrations as well as phytoplankton productivity, biomass and community compositional responses among the stations employed in this study, it is essential to develop sampling transects spanning the major tributaries (Neuse and Pamlico River estuaries, Chowan-Roanoke Rivers and Albemarle Sound) to the inlets at the Outer Banks.

(2) Temporally, it is essential that seasonal patterns and shifts as well as episodic events in nutrient loading, productivity, floral and faunal compositional responses and benthic biogeochemical conditions (i.e., hypoxia, anoxia) be accounted for.

(3) The sound-wide monitoring program should be coordinated in time and space to take advantage of other efforts to gauge the ecological condition of the sound and its sub-estuaries.

(4) In addition to standard hydrographic and nutrient-productivity water quality parameters, Pamlico Sound should be routinely (but perhaps at low frequency) monitored for common toxic substances known to be emitted from agricultural, urban and industrial sources (i.e., EPA's priority toxics "hit list"), potentially toxic heavy metals (e.g. Cd, Hg, Pb), higher fauna, microbial pathogens, submersed aquatic plants, benthic microalgae and macroalgae.

(5) Monitoring and survey programs for Pamlico Sound should take advantage of evolving, chemotaxonomic (e.g., ChemTax), biochemical, molecular and other rapid diagnostic indicators being developed for large-scale systems.

(6) Due to limited resources and logistic constraints, water quality monitoring efforts on large coastal systems like Pamlico Sound should, by nature, be interdisciplinary and multi-institutional.

In summary, information obtained from this project can be used to help develop a much needed water quality monitoring and assessment program for the Pamlico Sound. Given parallel efforts in automated monitoring, ecological indicator development, application of remote sensing and the potential for a cooperative program via the NCDENR and NEP; the stage is set for developing a long overdue yet timely water quality monitoring program for this State's most important aquatic resource, Pamlico Sound.

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