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Algal and Invertebrate Community Composition along Agricultural Gradients: A Comparative Study from Two Regions of the Eastern United States

Scientific Investigations Report 2008–5046

U.S. Department of the Interior
U.S. Geological Survey

Front cover: Ogeechee River, Burke County, Georgia
Photograph by Alan M. Cressler, USGS

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By Daniel L. Calhoun, M. Brian Gregory, and Holly S. Weyers

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Foreword

The U.S. Geological Survey (USGS) is committed to providing the Nation with credible scientific information that helps to enhance and protect the overall quality of life and that facilitates effective management of water, biological, energy, and mineral resources (*http://www.usgs.gov/*). Information on the Nation's water resources is critical to ensuring long-term availability of water that is safe for drinking and recreation and is suitable for industry, irrigation, and fish and wildlife. Population growth and increasing demands for water make the availability of that water, now measured in terms of quantity and quality, even more essential to the long-term sustainability of communities and ecosystems.

The USGS implemented the National Water-Quality Assessment (NAWQA) Program during 1991 to support national, regional, State, and local information needs and decisions related to water-quality management and policy (*http://water.usgs.gov/nawqa*). The NAWQA Program is designed to answer: What is the condition of our Nation's streams and ground water? How are conditions changing over time? How do natural features and human activities affect the quality of streams and ground water, and where are those effects most pronounced? By combining information on water chemistry, physical characteristics, stream habitat, and aquatic life, the NAWQA Program aims to provide science-based insights for current and emerging water issues and priorities. During 1991–2001, the NAWQA Program completed interdisciplinary assessments and established a baseline understanding of water-quality conditions in 51 of the Nation's river basins and aquifers, referred to as Study Units (*http://water.usgs.gov/nawqa/studyu.html*).

Multiple national and regional assessments are ongoing in the second decade (2001–2012) of the NAWQA Program as 42 of the 51 Study Units are reassessed. These assessments extend the findings in the Study Units by determining status and trends at sites that have been consistently monitored for more than a decade, and filling critical gaps in characterizing the quality of surface water and ground water. For example, increased emphasis has been placed on assessing the quality of source water and finished water associated with many of the Nation's largest community water systems. During the second decade, NAWQA is addressing five national priority topics that build an understanding of how natural features and human activities affect water quality, and establish links between sources of contaminants, the transport of those contaminants through the hydrologic system, and the potential effects of contaminants on humans and aquatic ecosystems. Included are topics on the fate of agricultural chemicals, effects of urbanization on stream ecosystems, bioaccumulation of mercury in stream ecosystems, effects of nutrient enrichment on aquatic ecosystems, and transport of contaminants to public-supply wells. These topical studies are conducted in those Study Units most affected by these issues; they comprise a set of multi-Study-Unit designs for systematic national assessment. In addition, national syntheses of information on pesticides, volatile organic compounds (VOCs), nutrients, selected trace elements, and aquatic ecology are continuing.

The USGS aims to disseminate credible, timely, and relevant science information to address practical and effective water-resource management and strategies that protect and restore water quality. The USGS hopes this NAWQA publication will provide insights and information to meet your needs, and will foster increased citizen awareness and involvement in the protection and restoration of our Nation's waters.

The USGS recognizes that a national assessment by a single program cannot address all water-resource issues of interest. External coordination at all levels is critical for cost-effective management, regulation, and conservation of our Nation's water resources. The NAWQA Program, therefore, depends on advice and information from other agencies—Federal, State, regional, interstate, Tribal, and local—as well as nongovernmental organizations, industry, academia, and other stakeholder groups. Your assistance and suggestions are greatly appreciated.

Robert M. Hirsch
Associate Director for Water

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Conversion Factors

Multiply	By	To obtain
	Length	
centimeter (cm)	0.3937	inch
meter (m)	3.281	foot (ft)
kilometer (km)	0.6214	mile (mi)
meter (m)	1.094	yard (yd)
	Area	
square kilometer (km^2)	247.1	acre
square kilometer (km^2)	0.3861	square mile (mi^2)
	Volume	
liter (L)	0.2642	gallon (gal)
cubic meter (m^3)	264.2	gallon (gal)
cubic meter (m^3)	0.0002642	million gallons (Mgal)
cubic meter (m^3)	35.31	cubic foot (ft^3)
cubic meter (m^3)	0.0008107	acre-foot (acre-ft)
	Flow rate	
cubic meter per second (m^3/s)	70.07	acre-foot per day (acre-ft/d)
cubic meter per second (m^3/s)	35.31	cubic foot per second (ft^3/s)
meter per second (m/s)	3.281	foot per second (ft/s)
liter per second (L/s)	15.85	gallon per minute (gal/min)
cubic meter per day (m^3/d)	264.2	gallon per day (gal/d)
cubic meter per second (m^3/s)	22.83	million gallons per day (Mgal/d)
	Mass	
gram (g)	0.03527	ounce, avoirdupois (oz)
kilogram (kg)	2.205	pound avoirdupois (lb)

Vertical coordinate information is referenced to the National Geodetic Vertical Datum of 1929 (NGVD 29) and the North American Vertical Datum of 1988 (NAVD 88).

Horizontal coordinate information is referenced to the North American Datums of 1927 and 1983 (NAD 27, NAD 83).

Altitude, as used in this report, refers to distance above the vertical datum.

Specific conductance is given in microsiemens per centimeter at 25 degrees Celsius (µS/cm at 25°C).

Concentrations of chemical constituents in water are given either in milligrams per liter (mg/L) or micrograms per liter (µg/L).

Acronymns and Abbreviations

ACFB	Apalachicola–Chattahoochee–Flint River Basin NAWQA Study Unit
ADAS	Algal Data Analysis System
BMPs	best management practices
DIN	dissolved inorganic nitrogen
DOC	dissolved organic carbon
DP	Delmarva Peninsula
EPT	Ephemeroptera, Plecoptera, and Trichoptera
EWI	equal-width increment
GAFL	Georgia–Florida NAWQA Study Unit
GCP	Georgia Upper Coastal Plain
GIS	Geographic Information System
IDAS	Invertebrate Data Analysis System
MDS	non-metric multidimensional scaling
NAWQA	National Water-Quality Assessment
NEET	Effects of Nutrient Enrichment on Stream Ecosystems Topical study
NH_4	ammonium
NHD	National Hydrography Dataset
NLCD	National Land Cover Dataset
NO_2	nitrite
NOx	nitrate-nitrite nitrogen
NWQL	National Water-Quality Laboratory
OP	orthophosphate
PCA	Principal Components Analysis
PODL	Potomac–Delmarva NAWQA Study Unit
SC	specific conductance
TKN	total Kjeldahl nitrogen
TN	total nitrogen
TN/TP	total nitrogen to total phosphorus ratio
TP	total phosphorus
USEPA	U.S. Environmental Protection Agency
USGS	U.S. Geological Survey
WSA	Wadeable Streams Assessment

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Abstract

Benthic algal and invertebrate communities in two Coastal Plain regions of the Eastern United States—the Delmarva Peninsula (27 sites) and Georgia Upper Coastal Plain (29 sites)—were assessed to determine if aspects of agricultural land use and nutrient conditions (dissolved and whole-water nitrogen and phosphorus) could be linked to biological community compositions. Extensive effort was made to compile land-use data describing the basin and riparian conditions at multiple scales to determine if scale played a role in these relations. Large differences in nutrient condition were found between the two study areas, wherein on average, the Delmarva sites had three times the total phosphorus and total nitrogen as did the sites in the Georgia Upper Coastal Plain. A statistical approach was undertaken that included multivariate correlations between Bray-Curtis similarity matrices of the biological communities and Euclidean similarity matrices of instream nutrients and land-use categories. Invertebrate assemblage composition was most associated with land use near the sampled reach, and algal diatom assemblage composition was most associated with land use farther from the streams and into the watersheds. Link tree analyses were conducted to isolate portions of nonmetric multidimensional scaling ordinations of community compositions that could be explained by break points in abiotic datasets. Invertebrate communities were better defined by factors such as agricultural land use near streams and geographic position. Algal communities were better defined by agricultural land use at the basin scale and instream nutrient chemistry. Algal autecological indices were more correlated with gradients of nutrient condition than were typically employed invertebrate metrics and may hold more promise in indicating nutrient impairment in these regions. Nutrient conditions in the respective study areas are compared to draft nutrient criteria established by the U.S. Environmental Protection Agency. Substantial reductions in some nutrients would be required to meet proposed reference conditions on the Delmarva Peninsula.

Introduction

Even after being linked to a disproportionate share of water-quality impairments in the United States during the early 1990s (U.S. Environmental Protection Agency, 1992), agriculture continues to be a major source of nutrients affecting streams and rivers. More recently, it was estimated that agriculture was responsible for impairments in approximately 18 percent (78,000 kilometers [km]) of stream lengths assessed and for up to 48 percent of all reported water-quality problems during 2000 (U.S. Environmental Protection Agency, 2000c). The U.S. Geological Survey's (USGS) National Water-Quality Assessment (NAWQA) Program reported that streams draining agricultural areas were transporting some of the highest concentrations of both phosphorus and nitrogen seen in rivers in the United States. Streams in agricultural areas typically were transporting up to 20 percent of the phosphorus and up to 50 percent of the nitrogen that had been applied annually to the land (Mueller and Spahr, 2006). The U.S. Environmental Protection Agency (USEPA) Wadeable Streams Assessment (WSA) Program found that phosphorus and nitrogen concentrations were intermediately to highly elevated in 47 and 53 percent of the stream lengths studied, respectively, nationwide (U.S. Environmental Protection Agency, 2006). The WSA indicated that, within the Southeastern Coastal Plain ecoregion, total phosphorus was a "leading indicator of stress" to aquatic systems and was found to be at intermediate levels in 13 percent of stream lengths and at high levels in 29 percent. Although not as widespread a problem as phosphorus, nitrogen levels were found to be at intermediate to high levels in the Southeastern Coastal Plain ecoregion in 28 percent of stream lengths studied.

Paralleling this accumulation of scientific data, which have implicated agriculture as a major source of water-quality impairments, has been an increased understanding of riparian ecology and the key functional role that riparian ecosystems play at the interface between agricultural and aquatic ecosystems. For example, water-quality conditions during baseflow have been correlated with the presence or absence of riparian forests as have increased sediment levels, suspended solids, turbidity, and phosphorus during runoff conditions (Schlosser and Karr, 1981). Coastal Plain riparian forests located in agricultural watersheds of the southeastern and mid-Atlantic states—areas similar to the study areas described herein—have been reported to be effective nutrient filters that could potentially buffer streams from excess nutrient runoff due to upland agriculture (Lowrance and others, 1984a, 1984b; Peterjohn and Correll, 1984; Hill, 1996; Puckett, 2004; Puckett and Hughes, 2005). Nutrient uptake capacity of soils and vegetation, the presence of organic carbon sources, and the presence of anoxic conditions in these Coastal Plain riparian forests have also been shown to decrease nutrient fluxes into the stream, but only if runoff and shallow subsurface flow moved through the biologically-active root zone of the riparian forest (Lowrance, 1992; Puckett and Hughes, 2005). Hydrogeologic controls, certain soil characteristics such as grain size, texture, and absence of organic material, and the ditching and draining of fields have been shown to render intact riparian zones incapable of nutrient mitigation. This effect can occur through the limitation of denitrification, the bypassing of the riparian zone, and ground-water runoff that flows more directly to surface waters (Böhlke and Denver, 1995; Hill, 1996; Puckett, 2004). Results from these and other studies indicate, with caveats, strong relations between water-quality properties—such as nutrient concentration—and the riparian zone and provide a scientific basis for the development of best management practices (BMPs) for forestry and agriculture involving conservation of riparian zone vegetation (Welsch, 1991). Although the body of literature concerning riparian zone ecology is large, few studies have attempted to address the relative influence of riparian zone alteration on nutrient conditions simultaneously at the watershed, segment, and reach scales. Many studies have indicated adverse effects on stream invertebrate and algal communities from agriculture and elevated nutrient conditions (Winter and Duthie, 2000; Munn and others, 2002; Davis and others, 2003; Black and others, 2004; Muenz and others, 2006). Few studies, however, have attempted to address these effects at multiple scales of land use and riparian condition (Black and others, 2004; Rios and Bailey, 2006).

The USEPA has developed ecoregion-based nutrient criteria to serve as recommendations to States and Indian Tribes for use in developing local water-quality standards for instream nutrients. These standards will serve to assist States and Tribes in assessing attainment of uses, developing water-quality-based permit limits, and establishing targets for total maximum daily loads (U.S. Environmental Protection Agency, 1998). Mueller and Spahr (2006) reported that annual flow-weighted concentrations of total nitrogen and total phosphorus nutrient levels in streams and rivers classified as draining agricultural, urban, or mixed land uses frequently exceeded these ecoregion-based nutrient criteria at modest degrees of development. Understanding how biological communities respond to nutrient enrichment is a critical link in developing effective nutrient-management strategies.

Purpose and Scope

This report describes the physical, chemical, and biological responses to increasing agricultural land use over multiple scales in streams on the Delmarva Peninsula (DP) and in the Georgia Upper Coastal Plain (GCP). Multivariate techniques are utilized to enable the comparison of (1) stream benthic communities (algae and invertebrates), (2) the two study areas, (3) the role of scale in the relation between land use and biota, and (4) the influence of individual measured variables on stream community compositions.

This study was conducted as part of the USGS NAWQA Program (Gilliom and others, 1995) and was designed to determine how biological communities and processes respond to varying levels of nutrient enrichment in agricultural streams in contrasting environmental settings (Munn and Hamilton, 2003). This information will be relevant to ongoing efforts by the USEPA and States to develop regional nutrient criteria for rivers and streams and to further describe nutrient processing and transport in riparian zones in agricultural landscapes.

Study objectives were to determine the association of algal and invertebrate communities to nutrient conditions in streams from two distinct regions of the eastern U.S. Coastal Plain and to determine the extent to which biota and nutrient relations can be regionalized using environmental factors. The objectives of the study were to (1) describe the stream habitat, nutrient conditions, and basin characteristics of the two study areas, (2) establish if stream nutrient levels would be reflective of the estimated manure and fertilizer application rates in their respective watersheds, (3) determine if near-stream and reach-scale land use would be more strongly linked to biological community composition than would segment-scale and basin-scale land use while assessing similar relations to nutrient chemistry, and (4) identify environmental variables best corresponding to changes in biological community composition.

Study Areas

Fifty-six wadeable streams were selected for study from agriculturally dominated areas on the DP and from the GCP (fig. 1). The study on the DP involved 27 streams located within the USGS NAWQA Potomac–Delmarva study unit (PODL) and the Middle Atlantic Coastal Plain Level 3 Ecoregion (Omernik, 1987). The study in the GCP involved 14 streams located within the Apalachicola–Chattahoochee–Flint (ACFB) NAWQA study unit and 15 streams within the Georgia–Florida (GAFL) NAWQA study unit—all 29 of which are in the Southeastern Plains Level 3 Ecoregion (Omernik, 1987).

The area investigated on the DP lies in the Delmarva Uplands (Level 4) ecoregion (Omernik, 1995) and the Eastern Coastal Plain nutrient ecoregion (nutrient ecoregion 14) (U.S. Environmental Protection Agency, 1998) and includes most of the State of Delaware, and parts of the State of Maryland between Delaware Bay on the east and Chesapeake Bay

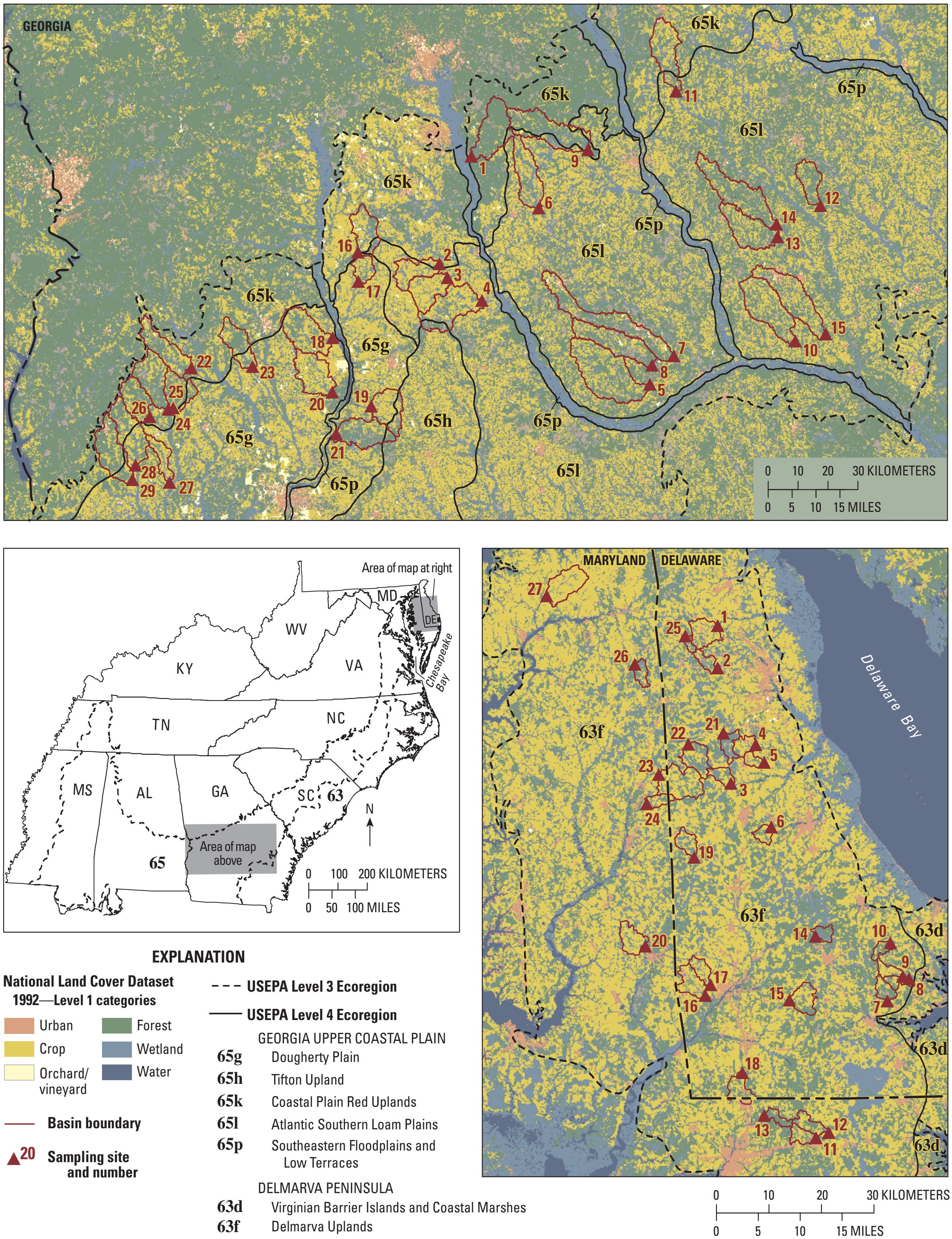

Figure 1. Locations of study areas, sampling locations, and watershed boundaries in the Georgia Upper Coastal Plain and the Delmarva Peninsula including portions of Delaware and Maryland. Indicated are Level 3 and 4 ecoregions (Omernik, 1995) and land use from the 1992 National Land Cover Dataset (U.S. Geological Survey, 2000).

on the west (nutrient ecoregions not shown in figure 1). This area is characterized by level to gently rolling upland areas with altitude ranging from about 6 meters (m) to less than 30 m, a humid subtropical climate receiving an average annual rainfall of 112 centimeters (cm), and a growing season of approximately 200 days. DP hydrogeology is characterized by multiple aquifers and confining units dipping to the south and varying in sediment depth from 0 to 2400 m (Denver and others, 2004). The surficial aquifer is shallow and irregular in the north and generally deepens to the south. Streams of the DP typically originate in the central uplands, are low gradient, and are tidally influenced near the coast. Many of the streams have been artificially straightened and deepened to improve drainage. Watersheds are relatively small—based on geographic controls—usually less than 26 square kilometers (km^2) in area. Land use is predominantly rural, about half (48 percent) of the peninsula is used for agriculture (e.g., corn, soybeans and pasture), one-third is forested, less than 10 percent is urban, and the remaining land surface is either wetlands or open water (Denver and others, 2004). Much of the corn and soybean crop is used locally for poultry feed for the more than 570 million broiler chickens that are produced annually on the peninsula (Delmarva Poultry Industry, Inc., 2006).

Streams selected for the DP study range in basin size from 4 to 41 km^2. Stream conditions range from areas with well-drained soils containing natural stream channels and intact deciduous riparian corridors to areas of poorly drained soils that have been ditched or modified to improve drainage with crop or grass riparian buffers. Basins in the central core of the peninsula in Delaware and Maryland are generally poorly drained, whereas basins in the northern portion and outer edges of the peninsula are well drained. Streams in well-drained areas have sandy substrates with little silt and large amounts of woody debris and other organic matter. Streams located in poorly drained areas have sandy substrates but have large amounts of silt and other fine-grained sediments and minimal amounts of woody debris and leaf litter. Denver and others (2004) provide an extensive description of the environmental setting of the DP.

The area investigated in the GCP lies mainly within the Southeastern Temperate Forested Plains and Hills nutrient ecoregion (nutrient ecoregion 12) (U.S. Environmental Protection Agency, 1998) (nutrient ecoregions not shown in figure 1) and within three separate Level 4 ecoregions: the Dougherty Plain, the Atlantic Southern Loam Plains, and the Coastal Plain Red Uplands (Griffith and others, 2001). This area is characterized by a humid subtropical climate receiving an average annual rainfall of 120 cm and a growing season of approximately 240 days. GCP hydrogeology generally varies in a northwest to southeast direction characterized by multiple aquifers (sand, clay, sandstone, dolomite, and limestone) and confining units (silts and clays) dipping to the southeast. Thickness and depth of individual units also increase to the southeast. Land use in the study area consists of approximately 36 percent cropland, 18 percent pastureland, 40 percent woodland, 4 percent water, and 2 percent residential (Sheridan, 1997). The majority of all recent expansions in broiler chicken production within the State have been in the GCP, based on the availability of croplands for the application of the chicken waste as a source of nitrogen and phosphorus fertilizer (Gascho and Hubbard, 2006). The U.S. Department of Agriculture estimates that peanut and upland cotton production in Georgia accounts for approximately one-half and one-tenth, respectively, of that grown in surveyed States (U.S. Department of Agriculture, 2005, 2006). Of the 2,500 km^2 in peanut production, approximately 50 percent receives application of nitrogen fertilizer and 60 percent receives phosphate. Of the 4,800 km^2 in cotton production, 97 percent receives nitrogen fertilizer and 88 percent receives phosphate. In headwater streams of the GCP, riparian buffers are commonly narrow, and farming is conducted near the streams. Extensive forested wetlands are present along the higher order streams creating broad riparian buffers commonly exceeding a width of 1 km.

Streams selected in the GCP study area typically are low gradient and sandy bottomed, draining watersheds with varied intensity of agricultural production. Drainage patterns generally are dendritic with high drainage densities. Selected streams range in basin size from 55 to 300 km^2 with an average altitude of 107 m (range 65 to 147 m). Streams originating in the western section of the GCP study area drain to the Gulf of Mexico through the Apalachicola River drainage, whereas streams originating in the eastern half of this study area are part of the Altamaha River drainage and flow to the Atlantic Ocean. Streams of the extreme northern part of the western region of the study area first flow through the Coastal Plain Red Uplands, an area distinguished mainly by the appearance of red clay subsoils. Streams originating in the more southwestern part of the GCP study area drain the Dougherty Plain, which is somewhat flatter and is characterized by intermittent surficial karstic terrain. In the eastern part of the GCP, streams primarily flow through the Atlantic Southern Loam Plains ecoregion, an area characterized by excessively drained dunal sand ridges. Extensive descriptions of the environmental setting of the GCP can be found in other reports (Couch and others, 1996; Berndt and others, 1998; Frick and others, 1998).

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Site Selections

A geographic information system (GIS) was used to delineate watersheds of appropriate minimum sizes in each region that contained potential sampling reaches with perennial flow. Study sites were selected from the population of potential sites using methods designed to construct a gradient of nutrient conditions in each study area. Sites were selected by evaluating the estimated nutrient loading to the watersheds (atmospheric and land-applied fertilizer and manure) (Ruddy and others, 2006), by evaluating percentage of row-crop land use within the basin, and by using actual nutrient concentration data ascertained from reconnaissance site visits using a portable nutrient analyzer (HACH™ model DREL/2010). The stream sites selected in both areas and associated riparian-zone and basin characteristics are presented in table 1.

Data Collection and Processing

Data collection followed published USGS and NAWQA methods and protocols for physical habitat (Fitzpatrick and others, 1998), water quality (U.S. Geological Survey, 1997 to present), and algal and invertebrate communities (Moulton and others, 2002). These data, combined with results from spatial analyses, were statistically analyzed using multiple methods to determine correlative patterns.

Habitat

Habitat conditions at all stream reaches were assessed during summer 2004 using a protocol designed to balance qualitative and quantitative measures of habitat integrity (Fitzpatrick and others, 1998). Reach lengths were designated as 20 times the mean wetted-channel width, with a minimum reach length of 150 m and a maximum of 300 m. All but 1 of the 56 streams assessed had reach lengths of 150 m. Measures of instream habitat were made along 11 equally spaced transects perpendicular to the direction of streamflow and included assessment of geomorphic unit type (riffle, run, pool), water velocity, depth, dominant substrate, substrate size, substrate embeddedness, and instream cover. Features outside of the channel such as streambank angles, bank heights, and estimates of bank stability were noted. Estimates of canopy closure were made using a spherical densitometer, measuring the canopy closure at each bank and at two points facing upstream and downstream at the midpoint of the channel. Estimates of the potential solar radiation reaching the streams' surfaces were made using Solar Pathfinders™ at 5 of the 11 transects at each reach. Instream measurements were adjusted for season and indexed to nearby reference stations that were at similar latitudes with data published by National Renewable Energy Laboratory (*http://www.nrel.gov*). Instantaneous discharge was measured either at the time of data collection or taken from established rating curves at co-located USGS gages. Channel gradients (percent slope) were determined using a TOPCON™ GTS-211D Total Station. Digital photographs were taken at each habitat transect to document habitat conditions during collection. All habitat data were recorded on standardized data sheets and summarized at the reach level prior to analysis.

Water Chemistry

Water samples were collected twice at all sites during synoptic surveys conducted in the spring of 2004, a period that was hydrologically typical for both study areas and was little interrupted by high-flow runoff events. Samples were collected using isokinetic depth-integrated equal-width increment (EWI) sampling methods unless the stream was too shallow or water velocity was insufficient, in which case, samples were collected as multi-vertical grab samples (U.S. Geological Survey, 1997 to present). Field water-quality properties were measured at each sampling event and included water temperature, dissolved oxygen, specific conductance, and pH using a multiparameter sonde that was calibrated daily prior to use. Water-chemistry analysis included: nutrients (nitrogen and phosphorus as total and dissolved species), dissolved and particulate organic carbon, and particulate nitrogen. Samples were collected for the determination of suspended sediment, and instantaneous stream discharge and turbidity were measured during site visits. All laboratory analyses for chemical constituents were conducted at the USGS National Water-Quality Laboratory (NWQL) in Denver, Colorado, using methods by Fishman (1993) and Patton and Kryskalla (2003).

Benthic Algae

Algal community composition was assessed at each stream from episammic (sand-substrate) habitats using standardized protocols (Moulton and others, 2002) during spring 2004. Episammic samples were collected from shallow depositional areas with the lowest velocities, usually along the margins of streams, using a 5-cm-diameter petri dish cover to stabilize bottom material (coarse sands) while lifting the top 2 cm of substrate with a spatula and placing into a container. Five to 10 depositional algal samples were collected and composited into a single sample at each site. The composite sample was mixed thoroughly and preserved with full-strength buffered formaldehyde for taxonomic identifications, cell counts, and biovolume estimates conducted by the Philadelphia Academy of Natural Science in Philadelphia, Pennsylvania, using protocols by Charles and others (2002). Area estimates were made by using the surface area sampled in the depositional habitats.

Table 1. Selected site information for Delmarva Peninsula and Georgia Upper Coastal Plain study areas, sorted by USGS station code. Riparian land use delineated from the basin-wide National Hydrography Dataset derived network and a buffer width of 75–105 meters. Land use data were derived from the 1992 National Land Cover Dataset.

[km^2, square kilometer; km/km^2, kilometer per square kilometer]

Site number (location shown in figure 1)	USGS station code	Drainage area (km^2)	Stream density (km/km^2)	Percent land use					
				Riparian			Basin		
				Forested	Agriculture	Wetland	Forested	Agriculture	Wetland
Delmarva Peninsula									
1	01483500	23.7	0.99	28	35	34	15	69	10
2	01483666	9.8	1.17	22	28	39	24	43	23
3	01483990	11.0	0.66	40	28	32	28	53	19
4	01484036	12.6	0.94	22	54	22	15	67	10
5	01484050	7.2	0.85	25	29	38	13	67	7
6	01484100	9.0	0.43	62	5	33	34	60	6
7	01484534	11.4	0.37	52	22	19	57	29	5
8	01484640	9.3	0.59	24	52	25	16	77	7
9	01484645	8.4	0.31	35	29	36	43	53	4
10	01484652	7.7	0.66	48	29	23	49	40	11
11	01485025	14.4	0.46	65	23	12	45	49	6
12	01485030	11.9	0.40	34	32	13	35	48	4
13	01486100	8.4	0.55	64	1	35	82	11	7
14	01487060	8.6	0.56	58	2	40	58	10	32
15	01487116	20.2	1.14	46	41	12	39	53	8
16	01487250	12.2	1.18	23	47	29	16	73	10
17	01487300	21.4	0.91	19	45	32	17	69	10
18	01487910	18.0	0.54	45	36	9	36	52	2
19	01488530	13.9	1.01	27	34	39	21	58	20
20	01489000	20.6	0.59	16	46	38	12	74	14
21	01490590	12.2	0.74	18	7	75	29	22	48
22	01490600	22.9	0.98	33	41	26	32	45	23
23	01491020	41.1	1.07	33	48	18	23	67	10
24	01491050	9.5	0.77	40	30	30	22	69	9
25	01492900	4.5	1.06	23	20	57	21	53	26
26	01492995	10.1	0.50	42	28	30	34	37	28
27	01493500	33.0	0.58	12	47	36	4	90	5

Table 1. Selected site information for Delmarva Peninsula and Georgia Upper Coastal Plain study areas, sorted by USGS station code. Riparian land use delineated from the basin-wide National Hydrography Dataset derived network and a buffer width of 75–105 meters. Land use data were derived from the 1992 National Land Cover Dataset.—Continued

[km^2, square kilometer; km/km^2, kilometer per square kilometer]

Site number (location shown in figure 1)	USGS station code	Drainage area (km^2)	Stream density (km/km^2)	Percent land use					
				Riparian			Basin		
				Forested	Agriculture	Wetland	Forested	Agriculture	Wetland
Georgia Upper Coastal Plain									
1	02214315	158.9	0.82	90	2	6	84	7	3
2	02215090	90.8	0.61	28	33	34	12	71	13
3	02215120	110.7	0.73	26	45	24	13	76	8
4	02215295	179.8	0.72	39	24	27	29	51	9
5	02215375	299.8	0.86	69	13	9	49	38	4
6	02215656	139.8	0.74	39	13	43	29	48	17
7	02216170	236.3	0.87	58	11	17	49	28	7
8	02216185	174.6	0.95	60	15	11	49	32	4
9	02223900	213.4	0.70	60	8	29	59	25	11
10	02225105	200.6	1.05	61	18	16	44	43	6
11	02225148	145.7	0.99	52	18	25	40	44	9
12	02225317	78.0	0.94	76	9	11	61	29	4
13	02225353	205.7	1.01	75	9	8	60	26	3
14	02225365	152.1	1.01	68	13	11	47	39	5
15	02225600	194.4	1.02	59	17	16	40	43	5
16	02349685	76.6	0.83	28	33	37	24	58	17
17	02349900	122.9	0.82	31	31	33	18	65	12
18	02350080	161.5	0.84	36	26	33	30	54	12
19	02350360	110.3	0.84	42	32	15	28	58	5
20	02350470	132.7	0.74	34	35	27	19	67	10
21	02350509	135.5	0.48	50	21	22	54	25	10
22	02350798	124.5	0.63	51	5	38	45	42	8
23	02351790	99.8	0.80	49	16	32	47	40	9
24	02353097	134.8	0.70	58	3	35	53	31	8
25	02353098	94.5	0.61	44	8	41	54	34	6
26	02353190	98.2	0.57	61	4	31	56	33	5
27	02353245	54.8	0.62	16	21	49	13	71	12
28	02353330	151.7	0.65	54	9	31	46	42	6
29	02353360	151.6	0.71	63	6	25	59	29	5

Invertebrates

Invertebrates were sampled from stable pieces of woody debris at each stream with a semi-quantitative sampling method using modified surber samplers (Slack sampler) with 500-micron mesh nets (Moulton and others, 2002). Samples were collected by selecting approximately 10 small, medium, and large pieces of conditioned, native woody debris from a variety of current velocities within the reach. Small- and medium-sized pieces either were collected whole, or were carefully cut with shears or a small handsaw while an assistant positioned the modified d-frame net directly downstream from the piece of wood. Smaller pieces and the cut pieces of woody debris were brushed while in the bucket and washed with filtered native water to remove all epidendric material. Larger pieces of woody debris were sampled in place by positioning a Slack sampler directly downstream from the piece of woody debris and by vigorously brushing the epidendric material into the net with a large brush. Woody debris also was examined and any remaining invertebrates were hand-picked with forceps. All material collected in the net and from examination of the pieces of wood was composited into a 5-gallon container. Composited materials were elutriated to remove sediment and heavier material and then sieved through a 500-micron sieve where larger pieces of detritus were removed. Large or fragile individual invertebrates were removed and placed in separate containers to avoid damage to specimens. The remaining material was placed into 1-liter bottles, preserved with 10 percent buffered formalin and shipped to the NWQL where identifications and enumerations were conducted by the Biological Group using standard protocols (Moulton and others, 2000).

Basin and Riparian Land-Use Analysis

Land use within the study basins and over the basin-wide riparian networks (buffer 75–105 m) was characterized using the National Hydrography Dataset (NHD 100K) (U.S. Geological Survey, 2005) and the National Land Cover Dataset (NLCD 1992) (U.S. Geological Survey, 2000). Minor land-use categories were aggregated into three major categories: forested, agriculture (including orchards), and wetlands (table 1). The minor categories were retained for subsequent analyses. The near-stream riparian land-use dataset was generated using a protocol recently developed for mapping and characterizing land use and land cover in riparian zones (Johnson and Zelt, 2005). This method involved the delineation and characterization of land use and land cover within various distances from the stream and along several lengths within the stream drainage network with a maximum distance from the stream of 250 m and a maximum segment extent proportional to the log base 10 of the watershed area. Segment lengths for the DP and GCP sites averaged 1.1 and 2.1 km, respectively. Delineating land uses within these areas was conducted by using digital orthophoto quarter quadrangles onto which nine classes of land use and land cover were delineated using onscreen digitization. These nine land-use classes were the same as land-use classes used for the basin-wide analysis and were ultimately reduced to five major classes. From this process, six separate datasets were produced. Two of the six were at the 150-m reach scale, and the lateral extents were 25 m and 50 m. Four of the six were at the segment scale with lateral extents of 50 m, 100 m, 150 m, and 250 m on each side of the stream. These approaches were designed to produce ecologically relevant scales of land-use and land-cover data to test hypotheses related to nutrient conditions and ecological community compositions.

Statistical Analyses

Invertebrate and algal data were processed using the Invertebrate Data Analysis System (IDAS) and the Algal Data Analysis System (ADAS) software programs, which systematically and consistently adjust the entire dataset in terms of handling multiple levels of taxonomic resolution or “ambiguous” taxonomic data (Cuffney, 2003). Community and tolerance metrics were calculated using an attribute file of published values. IDAS was used to calculate a suite of 7 functional, 12 tolerance, and 20 community invertebrate metrics. Invertebrate tolerance metrics were calculated using published tolerance data (Barbour and others, 1999). ADAS was used to calculate 30 algal community and tolerance metrics also using an attribute file of published values (Stephen Porter, U.S. Geological Survey, written commun., 2006). The primary algal metrics used for this study were those commonly used to indicate trophic preferences (Van Dam and others, 1994) and pollution tolerance (Lange-Bertalot, 1979). All classified algal taxa were included in the generation of metrics including soft algae and diatoms; however, this approach was limited to only those taxa that had defined attributes. Spearman rank correlations (r_s) between indices and abiotic variables were used to identify potential relations for further investigation. The same approach was used to identify correlations within the environmental dataset, specifically between the land use and nutrient chemistry of the respective study areas.

The comparison of algal and invertebrate community compositions to gradients of environmental conditions was accomplished using multiple statistical approaches. PRIMER software (version 6, Plymouth, United Kingdom) was used to construct and test resemblance matrices of the biotic and the abiotic environmental variables. This allowed for various types of community analyses including nonmetric multidimensional scaling (MDS) ordination, which is considered a highly effective ordination method for ecological community analysis (Clarke, 1993; Clarke and Warwick, 2001; McCune and Grace, 2002). Square-root transformed relative abundance measures of the various biotic communities consistently achieved the lowest ordination stress—a measure of ordination reliability—while preventing overtransformation. For the purposes of the multivariate analyses in this study, algal communities were pretreated by removing nondiatom species, again, because diatom-only ordinations achieved the lowest multivariate stress for both study areas. Bray-Curtis resemblance matrices were constructed from the transformed species datasets for subsequent analysis. For the environmental datasets, variable distributions were inspected and appropriate transformations were conducted from no transformation to square root to fourth root; no variables required a log-based transformation to approach normality. All environmental data were standardized to the same scale by subtracting the mean and dividing by the standard deviation (normalization in PRIMER) within each variable. Euclidean-based resemblance matrices were then constructed to analyze each dataset (e.g., land use and nutrient chemistry).

The RELATE procedure in PRIMER was used to test for the relative strength of rank-based relations between basin and riparian land use and nutrient condition as well as between nutrient conditions and algal and invertebrate communities. This allowed for the testing of the presence of gradients in species and environmental space. This nonparametric Mantel-type procedure conducts a multivariate regression on two independently collected datasets and tests the hypothesis that no relation exists between the resemblance matrix of the biological community and that of an environmental dataset by calculating a test statistic rho (ρ) between the community and environmental matrices (Clarke and Ainsworth, 1993; Clarke and Warwick, 2001). Under the null hypothesis (no relation between environmental and ecological datasets), ρ values will be near 0 or negative; however, if this permutation-based procedure determines that the datasets are near perfectly related, ρ values will be close to 1, and the null hypothesis is rejected. For datasets with sample sizes between 24 and 29, statistical significance of $p<0.05$ was achieved at ρ values in the range of 0.18 to 0.20; $p<0.01$ was achieved where ρ exceeded 0.30.

Data reduction techniques are commonly used in ecological community analyses to manage the large abiotic datasets that may be available and to limit analyses to those variables that have the best potential for explaining patterns in abiotic and biotic structure. The BEST-BVSTEP procedure in PRIMER (Clarke and Ainsworth, 1993; Clarke and Warwick, 2001) was used on each of the separate datasets—water physicochemistry, basin and riparian land use, multiple measures of stream geomorphic and habitat characteristics, geology and soils, and estimates of nutrient loadings to the basin—after root (or fourth root) transformation and normalization. The stepwise-nonparametric procedure identifies the most influential variable combinations that account for the multivariate patterns across the Euclidean (in this case) resemblance matrices of the sites studied. This was done separately for the two study areas. Spearman rank correlations (r_s) were calculated within the two compilations of the selected variables, and variables were removed that were intercorrelated at a level $|r_s|\geq 0.8$ and loaded least on a separately run Principal Components Analysis (PCA) axis 1. Several sites in the study required removal from the analysis based on incompleteness of data upon pooling of the final variable sets. For the DP study, the excluded sites were 6, 13, 14, and for the Georgia study, site 26 (fig. 1).

The reduced abiotic variable sets for the GCP and DP study areas were compiled and then analyzed in terms of the invertebrate and algal community Bray-Curtis similarity matrices to determine the subset of influential variables that best explained the biotic structure. The same method was applied as detailed above, except the procedure was modified to use the more exhaustive BEST-BIOENV procedure in PRIMER that assesses all possible combinations of supplied variables. The resulting set of variables was used in a nonparametric nonlinear multivariate analogue to classification regression tree analysis termed linkage tree analysis in PRIMER—LINKTREE—that sequentially identifies non a priori subsets of samples from the Bray-Curtis similarity matrices that are most attributable to break points in specific variables within the abiotic dataset. Maximization of the multivariate R statistic is used by PRIMER to assure adequate separation of assigned groups (Clarke and Gorley, 2006). The splits between groups were constrained by a permutation-based significance test (alpha$<$0.05). Once a group of sites is split based on the biotic information and appropriate environmental variable thresholds, that group is removed from the pool of sites, and the routine moves on to assess the remaining sites once again in terms of the original biotic and abiotic data. One major benefit of the LINKTREE analysis is that it can identify variables and values of those variables that can explain local variability in an ordination that potentially would not emerge from a traditional linear direct or indirect gradient analysis. One drawback, however, is that once a group of samples is removed by the procedure, further interpretation of that group may not occur based on the selection criteria, leading to possible misconceptions about the importance of other variables in shaping those biotic assemblages.

Graphical illustration of the relations between Bray-Curtis similarity matrices of species compositions from this study are presented through MDS ordinations with sites coded by the above mentioned LINKTREE groups. This was done to illustrate between site similarities and the environmental variables that may be most influencing the biotic compositions.

Finally, biological indices and nutrient chemistry values were pooled for the two study areas and related with Spearman rank correlation. This was done to determine if any indicators could be useful in a regional manner to describe or infer nutrient condition.

Stream Habitat, Nutrients, and Community Composition in Agricultural Streams

The two areas studied were found to be dissimilar in terms of some aspects of stream habitat condition, water chemistry, and biological compositions. No attempts are made to statistically test these differences between the respective study areas. These characteristics are presented; however, the primary focus of these results are to highlight the mechanisms that influence the variability within the study area as well as to place these sites within the context of regional nutrient conditions.

Habitat

The GCP and DP study areas were originally chosen for comparison based on general similarities in basin land use, stream types, and underlying hydrogeology. Analysis of habitat data showed that streams in these areas are similar in other ways as well. For example, investigated streams in both study areas were low-gradient, run-dominated systems. Benthic materials were composed mainly of sand and silt, with woody debris and root wads providing the majority of hard semi-stable instream substrates. Median stream gradients were lower in GCP streams (0.0008), which also had more pool habitat than did the streams in the DP (0.0012). Median stream velocities were similar in both study areas (GCP, 0.11 meter per second [m/s]; DP, 0.12 m/s). Major differences between DP and GCP streams were primarily based on differences in watershed size, in the amount of riparian vegetation, and in the availability of solar radiation. For example, GCP basins were an order of magnitude larger (146 km^2 compared with 15 km^2), and stream widths were wider (5.6 m compared with 3.6 m) than those in the DP study. Due to the lack of dense riparian forest adjacent to the DP streams, they received approximately 28 percent of the potential energy available at 39 degrees latitude, whereas GCP streams received only about 10 percent of the solar energy available at 32 degrees latitude. Even after correcting for differences in latitude, the DP streams studied still received about two times more solar energy than the GCP streams. This result is reflected in habitat measurements such as within channel vegetation cover, which was twice as high in the DP streams (56 percent compared with 21 percent), and macrophyte cover, which was 20 times higher in the DP streams than in the GCP streams.

Nutrients

One of the primary interests of this study was the degree to which instream nutrient chemistry could be related to basin-wide and riparian land use, specifically agricultural land use, and to estimates of nutrient loading to watersheds. Two synoptic chemistry samples were taken approximately 1 month apart, the second just prior to the biological sample. Individual nutrient concentrations remained relatively constant—with some exceptions—between the two samplings even though streamflow had decreased by approximately one-half to one-third over the month-long period. Specific conductance (SC) also remained essentially the same for both study areas. Spearman rank correlations (r_s) exceeded 0.70 in both study areas for total Kjeldahl nitrogen (TKN), nitrate-nitrite nitrogen (NOx), total phosphorus (TP), dissolved inorganic nitrogen (DIN), and total nitrogen (TN). The highest correlations between the two samplings were NOx (0.95), TP (0.83), and TN (0.94) for the DP, and TKN (0.86) and NOx (0.92) for the GCP. Lower correlations were seen for ammonium (NH_4) (DP, 0.49; GCP, 0.62) and orthophosphate (OP) (DP, 0.65; GCP, 0.50).

Nutrient conditions in the streams of the two respective study areas were considerably different for most properties assessed. Smith (1982) provided evidence from lentic systems that TN to TP ratios of less than 10 indicated nitrogen limitation and ratios greater than 17 indicated phosphorus limitation. Using this definition and averaging the two sampling periods, DP streams were nitrogen (N) limited at 3 to 5 sites and phosphorus (P) limited at 17 to 18 sites. No GCP sites were N limited, and 21 to 25 sites were P limited. On average, DP streams had three times the TP and TN concentrations (0.112 milligram per liter [mg/L] and 3.43 mg/L; table 2) as GCP streams (0.035 mg/L and 0.983 mg/L) and five times the DIN (3.02 mg/L for the DP and 0.598 mg/L for the GCP). The DP streams also had five times the NOx and OP (2.93 and 0.540 mg/L; 0.021 and <0.006 mg/L, respectively). Average nutrient amounts that were comparable across the study areas were TKN (0.5 mg/L) and NH_4 (0.08 mg/L). The DP basins also had three times the nitrogen and twice the phosphorus loadings (kilograms per square kilometer) than GCP basins, based on the estimates of land application and atmospheric sources.

Nutrient chemistry was correlated with basin characteristics in a variety of ways. In the DP study area, pasture/hay land use (NLCD81) in the basin and in the riparian zone (basin wide) was the best predictor of DIN and TN ($r_s = 0.76$ and 0.75, respectively). Row-crop agriculture in the riparian zone (basin wide) also correlated with DIN ($r_s = 0.69$). OP was negatively correlated with percentage of grassland within 250 m of the stream ($r_s = -0.69$). Nutrient chemistry in the GCP showed contrasting patterns to those seen in the DP. Concentrations of NOx were negatively correlated with conifer forest land use throughout the basin and the extent of the basin-wide riparian corridor (−0.69 and −0.65). Much of the land not used for row crop agriculture in the Coastal Plain is used for silviculture. As a general rule, the more specified the land-use data (that is, based on lowest classification levels available), the stronger correlations were for both study areas at the basin and riparian scales.

Nutrient Loadings

Estimates of nutrient loadings (table 2) to watersheds based on 2001 data (Ruddy and others, 2006) did not yield any strong relations for the DP study for either nitrogen or phosphorus species. This may have been due to a number of factors including limitations in the estimates of applied nutrients, previously mentioned hydrogeologic controls over ground-water linkages to streams, variability across sites of instream nutrient processing, interbasin transfer of applied animal manure, and/or the fact that samples were taken only during base-flow conditions, decreasing the likelihood for characterizing potential phosphorus inputs. In contrast, nitrogen loading estimates were correlated to instream NOx and DIN ($r_s = 0.70$) in the GCP.

Invertebrate and Algal Communities

Based on interpretation of the multivariate RELATE analysis of the aquatic community compositions' association to land use, algal diatom communities in the two study areas were more strongly related to land use at almost all scales than were invertebrate communities (fig. 2). Strongest relations occurred at the segment scales within riparian buffer widths of between 100 and 250 m from the stream margins of the DP study sites, although significant relations ($p < 0.05$) were observed at all of the scales analyzed. The association of land use to the algal diatom communities in the GCP were weaker albeit more consistent than those seen in the DP. The strongest relation was observed with land use in the riparian zone at the segment scale of 150 m or less to the stream margins. However, land use within the 100-m segment and the 50-m and 25-m reaches corresponded similarly to the algal community composition. Land use at the watershed scale (basin wide and riparian network) was least associated with the algal community in the GCP study area. In contrast, watershed-scale land use from the DP produced relations that were approximately equivalent to those derived from areas near a stream.

In contrast to diatom communities in the DP, invertebrates were more associated with land use closer to the streams and near the sampled reach than with basin-scale datasets. The same pattern, although with less significance, was observed for invertebrate communities in the streams sampled in the GCP study area. In the GCP, significant ($p < 0.05$) relations were observed only in terms of land use within 50 m of the stream reach. In all comparisons, relations between land-use datasets and invertebrate communities were more strongly linked in the DP than in the GCP.

Correlations between combined nutrient chemistry for the two synoptic samples and the biotic communities also were assessed and, in most cases, equaled or exceeded the relations between land use and biota. Rho statistics were highest for the nutrient samples taken temporally closest to the biological sample for algae in both areas and for invertebrates in the GCP, whereas nutrient chemistry from the early spring sample showed slightly more explanatory potential for invertebrate communities in the DP. Nutrient chemistry ρ statistics for comparisons between invertebrate and algae samples were as follows: early spring nutrients and invertebrates and algae in the DP (0.41, 0.37), in the GCP (0.31, 0.48); late spring nutrients and invertebrates and algae in the DP (0.35, 0.48), in the GCP (0.47, 0.52). All nutrient comparisons were significant at $p < 0.01$.

Table 2. Summary statistics for nutrient samples collected in the Delmarva Peninsula and Georgia Upper Coastal Plain studies. All nutrient values in milligrams per liter.

[NH_4, ammonium; NO_2, nitrite; TKN, total Kjeldahl nitrogen; NOx, nitrate-nitrite nitrogen; DIN, dissolved inorganic nitrogen; TN, total nitrogen; TP, total phosphorus; OP, orthophosphate; N, nitrogen; P, phosphorus; <, less than; values preceded by less than symbol are the laboratory reporting levels (LRL). Some statistics reported are below LRL but were detected above the method detection levels (MDL). Estimates of watershed nutrient loadings (Ruddy and others, 2006) are in kilogram per square kilometer (kg/km^2)]

Statistic	NH_4	NO_2	TKN	NOx	DIN	TN	TP	OP	N Loading	P Loading
Delmarva Peninsula										
Minimum	<0.040	<0.008	<0.10	<0.060	0.042	0.447	0.011	<0.006	2,090	230
Maximum	.535	.081	1.80	11.4	11.4	11.7	.961	.105	13,400	3,000
Mean	.084	.019	.51	2.93	3.02	3.43	.112	.021	7,350	1,400
Median	.043	.011	.44	2.49	2.62	2.94	.066	.015	6,760	1,100
25th percentile	.023	.006	.28	.870	.926	1.61	.042	<.006	5,050	580
Georgia Upper Coastal Plain										
Minimum	<0.040	<0.008	<0.10	<0.060	<0.060	0.376	0.010	<0.006	482	67
Maximum	.633	.015	.99	3.66	3.67	3.52	.074	.011	4,870	1,300
Mean	.083	.005	.45	.540	.598	.983	.035	<.006	2,420	690
Median	.050	.005	.38	.348	.406	.930	.037	<.006	2,120	600
25th percentile	.033	.004	.24	.082	.214	.688	.024	<.006	1,530	410

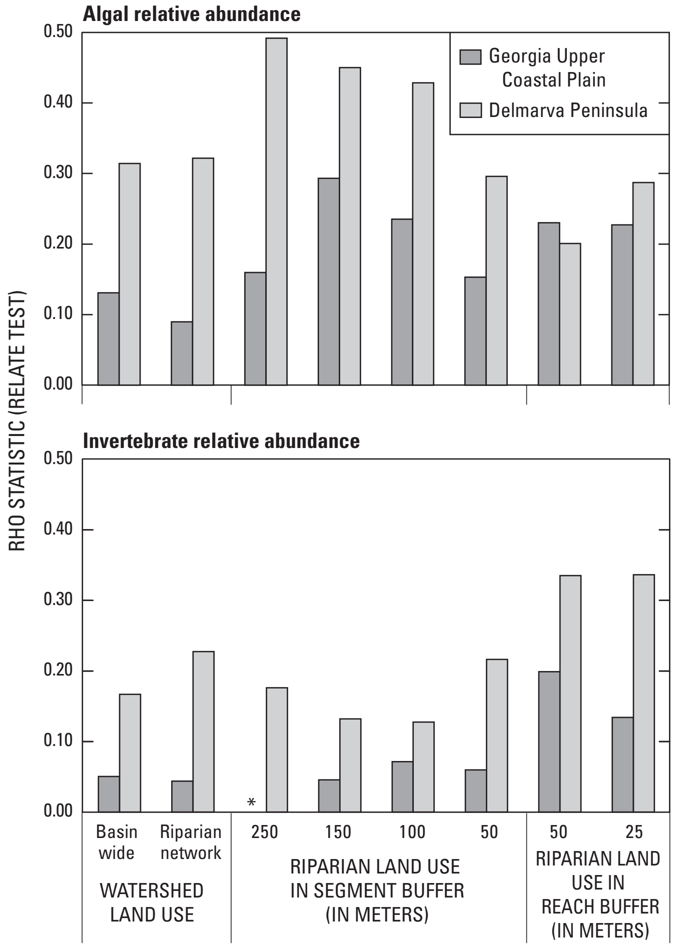

Figure 2. Determination of the multivariate regression between biotic community relative abundance and land use at multiple scales. Scales consist of land use over the basin, the riparian corridor of the stream network from the National Hydrography Dataset (1:24,000 scale) with a 75- to 105-meter buffer, the riparian segment (length defined as log10 drainage area) with the denoted buffer widths, and the sampled riparian reach (150 meters long) with buffer widths of 50 and 25 meters. Statistical significance of $p < 0.05$ is reached where the sample statistic (Rho) for these analyses reached 0.20 and $p < 0.01$ at 0.30. [*, indicates value slightly less than zero; <, less than]

Linking Environmental Variables to Biological Communities

Variable reduction and processing with the BEST routine yielded a combined dataset of 36 variables for each of the two study areas that represented basin and riparian land use, nutrient chemistry, soil classification, geomorphic variables, and habitat conditions (table 3). When analyzed in relation to the four community resemblance matrices (algae and invertebrates for the DP and the GCP), variables were further reduced to those best explaining the variability in the algal and invertebrate community structure.

MDS ordinations of the algal and invertebrate communities from the two study areas are illustrated in figures 3–6. The ordination stress is indicated on each figure and ranged from a high of 0.17 to a low of 0.08. Stress levels above 0.20 have been cited to be suspect for further two-dimensional interpretation because unique solutions can no longer be assured through excessive distortion of the information in the resemblance matrix (Clarke and Warwick, 2001; McCune and Grace, 2002). Using symbols, the sites are coded by the statistically significant, derived linkage tree analysis separations based on environmental variables identified through the data reduction process (table 3). Surrounding the central figures are bubble plots of the original ordination with bubble sizes proportional in Euclidean space to the values of environmental variables and invertebrate and algal metrics. Environmental variables and biological metrics shown are combinations that exhibited the highest Spearman rank correlations between abiotic and biotic variables. The correlated variables are those subplots in vertical opposition to one another throughout figures 3–6, for example, TN/TP ratio and abundance of intolerant invertebrates (fig. 3). All variables listed in the figures and all LINKTREE designations are included in the Appendix and in table 3.

From the MDS ordination, invertebrates in the sampled DP streams appear to fall into two definable groups (central graph in figure 3). The first LINKTREE split highlights near-stream conditions that may have affected the sites' biotic composition. Where the percentage of woody vegetation within 25 m of the sampled reach fell below 80 percent (average 33 percent for the group and 97 percent for the remaining sites), the communities were distinct. Near this split was a group of three sites characterized by agricultural activity within 50 m of the stream segment of greater than 15 percent (average 27 percent for the group and 4 percent for the remaining sites). Of the sites that remained, dissolved organic carbon (DOC) greater than 4.2 mg/L (average 5.1 mg/L for the group and 2.3 mg/L for the remaining sites) along with dissolved inorganic nitrogen (DIN) greater than or less than approximately 4 mg/L (average 5.7 mg/L for the group and 2.4 mg/L for the remaining sites) best explained differences in the communities. DOC was positively correlated with TP for all of the sites ($r_s = 0.84$), and three of the four DOC selected sites were in excess of 0.18 mg/L TP. This is noted because TP was not included in the analysis as indicated in table 3. The separation of the invertebrate communities by the initial two splits coincided with low total nitrogen to total phosphorus ratios (TN/TP)—less than approximately 30—by a diminished abundance of intolerant invertebrate taxa, by lower percentages of omnivores, and by an increase in the average tolerance scores of the invertebrate abundance indicating a more tolerant community. The scale for invertebrate tolerance is from 0 to 10, with higher scores indicating highly tolerant taxa. The sites with lower tolerance values also were sites with relatively high concentrations of DIN ($r_s = -0.67$). The percentage of omnivores was highly and negatively correlated to the percentage of the basin that was forested ($r_s = -0.80$), and TN/TP was moderately reflected in an abundance of intolerant invertebrates ($r_s = 0.65$) at sites with elevated TN/TP.

Table 3. Variables selected through variable reduction process and PRIMER BEST routine for the Delmarva Peninsula and Georgia Upper Coastal Plain studies.

[Variables indicated with X were used in the LINKTREE analyses. Multivariate correlation statistics are included in parentheses below community type. Test statistics with asterisk (*) indicate significance of $p < 0.01$; variables in bold indicate primary variable selection by community type. Variable categories are sequentially separated (vertically) based on general categories of land use, water quality, soils data, habitat characterization, and geographic position]

Delmarva Peninsula

Variable abbreviation	Variable description	Invertebrates (0.69*)	Algae (0.70*)
	Land use		
RZ2	Percent riparian zone developed		
BA1	Percent basin water		
BA4	Percent basin forested		X
BA9	Percent basin wetland		
RZ81	Percent riparian zone composed of pasture/hay		X
BA21	Percent basin composed of low intensity residential		
g150sp	Grassland (in percent) within 150-meter buffer along the segment		
o150sp	Open water (in percent) within 150-meter buffer along the segment		
u150sp	Urban/built-up land (in percent) within 150-meter buffer along the segment		
w150sp	Wetland (in percent) within 150-meter buffer along the segment		
wv150sp	Woody vegetation (in percent) within 150-meter buffer along the segment		
f100sp	Farmstead (in percent) within 100-meter buffer along the segment		
c050sp	Cropland (in percent) within 50-meter buffer along the segment	X	
wv025rp	Woody vegetation (in square meters) with in 25-meter buffer along the reach	X	

Georgia Upper Coastal Plain

Variable abbreviation	Variable description	Invertebrates (0.67*)	Algae (0.60*)
	Land use		
RZ1	Percent riparian zone water		
RZ2	Percent riparian zone developed		
RZ4	Percent riparian zone forested		
RZ8	Percent riparian zone agriculture		X
RZ9	Percent riparian zone wetland		
BA3	Percent basin barren		
BA41	Percent basin composed of deciduous forest		
wv150sp	Woody vegetation (in percent) within 150-meter buffer along the segment		
o100sp	Open water (in percent) within 100-meter buffer along the segment		
u100sp	Urban/built-up land (in percent) within 100-meter buffer along the segment		
w050sp	Wetland (in percent) within 50-meter buffer along the segment		
wmrallp	Woody vegetation (in meters) at the 30-meter margin line along the reach		

Table 3. Variables selected through variable reduction process and PRIMER BEST routine for the Delmarva Peninsula and Georgia Upper Coastal Plain studies.—Continued

[Variables indicated with X were used in the LINKTREE analyses. Multivariate correlation statistics are included in parentheses below community type. Test statistics with asterisk (*) indicate significance of $p < 0.01$; variables in bold indicate primary variable selection by community type. Variable categories are sequentially separated (vertically) based on general categories of land use, water quality, soils data, habitat characterization, and geographic position]

Delmarva Peninsula				Georgia Upper Coastal Plain			
Variable abbreviation	Variable description	Invertebrates (0.69*)	Algae (0.70*)	Variable abbreviation	Variable description	Invertebrates (0.67*)	Algae (0.60*)
Water quality				Water quality			
SC	Specific conductance, in microsiemens per centimeter			**SC**	**Specific conductance, in microsiemens per centimeter**	X	**X**
SSC	Suspended sediment concentration, in milligrams per liter			Turb	Turbidity, in nephelometric turbidity units		
DOC	Dissolved organic carbon, in milligrams per liter	X		TPN	Total particulate nitrogen, in milligrams per liter		
TPN	Total particulate nitrogen, in milligrams per liter			**NOx**	**Nitrate nitrogen, as nitrite + nitrate, in milligrams per liter**	**X**	X
OP	Orthophosphate, in milligrams per liter		X	OP	Orthophosphate, in milligrams per liter		
DIN	Dissolved inorganic nitrogen, in milligrams per liter	X	X	TN	Total nitrogen, in milligrams per liter		X
NH_4	Nitrogen as ammonia, in milligrams per liter			DIN_OP	Dissolved inorganic nitrogen to orthophosphate ratio		
TP	**Total phosphorus, in milligrams per liter**		**X**				
TN_TP	Total nitrogen to total phosphorus ratio		X				
Soils				Soils			
Wtdepl	Minimum value of depth of soil to seasonally high water table (feet)			Omh	Maximum value of organic matter content (percent by weight)		
No200h	Maximum value of percent by weight of soil material less than 3 inches in size and passing a No. 200 sieve (.074 mm)			Slopeh	Maximum value of land surface slope (percent)		
Cech	Maximum value of cation exchange capacity of soils			Sandave	Sand content of soil (percent)		
Flow_perm	Flow permanence, ratio of two measured discharges in early and late spring						

Table 3. Variables selected through variable reduction process and PRIMER BEST routine for the Delmarva Peninsula and Georgia Upper Coastal Plain studies.—Continued

[Variables indicated with X were used in the LINKTREE analyses. Multivariate correlation statistics are included in parentheses below community type. Test statistics with asterisk (*) indicate significance of p < 0.01; variables in bold indicate primary variable selection by community type. Variable categories are sequentially separated (vertically) based on general categories of land use, water quality, soils data, habitat characterization, and geographic position]

Delmarva Peninsula			
Variable abbreviation	Variable description	Invertebrates (0.69*)	Algae (0.70*)
	Habitat		
IQ	Instantaneous discharge, in cubic feet per second		
Width_Cv	Coefficient of variation of wetted channel width (percent)		
WD_Min	Minimum wetted channel width-depth ratio		
BFWidth_Avg	Average bankfull channel width, in meters	X	
Vel_Avg	**Average flow velocity, meters per second**	**X**	
P_Mphy_Avg	Average percent macrophyte cover		X
WetXArea_Avg	Average cross-sectional area of wetted channel, in square meters		
	Geographic position		
LATDEC	Latitude, in decimal degrees		
LONDEC	Longitude, in decimal degrees		

Georgia Upper Coastal Plain			
Variable abbreviation	Variable description	Invertebrates (0.67*)	Algae (0.60*)
	Habitat		
P_Run	Relative proportion of the total length of all geomorphic channel units that are composed of runs (percent)		
Width_Cv	Coefficient of variation of wetted channel width (percent)		
WD_Avg	Average wetted channel width-depth ratio	X	
BFDepth_Avg	Average bankfull channel depth, in meters		
Vel_Avg	Average flow velocity, meters per second	X	
PBankCov_Avg	Average percent bank vegetative cover		
P_SiltClay	Percent occurrence of transect points where silt and clay layer was observed on streambed		
P_SiltCov	Percent occurrence of transect points where silt layer was observed on streambed		
OCAngle_Avg	Average open canopy angle, in degrees		
WetPerm_Avg	Average perimeter of wetted channel, in meters		
WetShape_Min	Minimum wetted channel shape		
FlowStbl_Min	Minimum flow stability ratio		
	Geographic position		
LATDEC	Latitude, in decimal degrees		
LONDEC	Longitude, in decimal degrees	X	

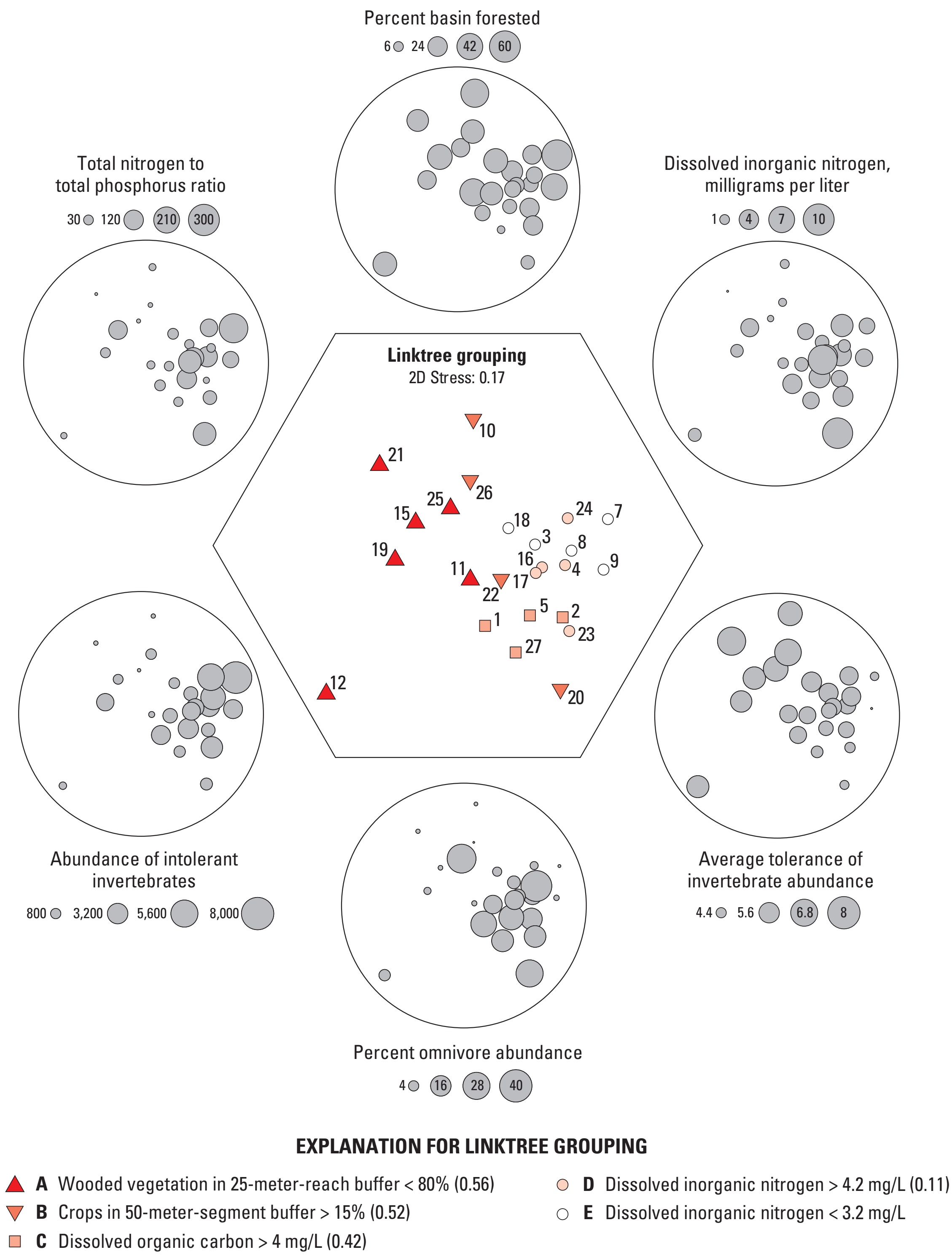

Figure 3. Plot series illustrating nonmetric multidimensional scaling (MDS) ordinations of Delmarva Peninsula invertebrate community composition from Bray-Curtis similarity matrices. The central plot is grouped by successive linkage tree breaks as controlled by the environmental variables indicated and a maximization of local separation of sample resemblances (SIMPROF Test; $p < 0.05$). Symbols are identified in the figure explanation and in the appendixes; values in parentheses are the test statistic for that split. Numbers in the central plot refer to sampling sites listed in figure 1, table 1, and in the appendixes. Two-dimensional stress of the ordination is included. Bordering plots are based on the same ordination; sites are represented by bubbles indicating relative values of the denoted variables. Top graphs are environmental variables, and bottom graphs are invertebrate community indices. Concept for figures 3 through 6 adapted from Edgerly and Rooks (2004). [<, less than; %, percent; >, greater than; mg/L, milligram per liter]

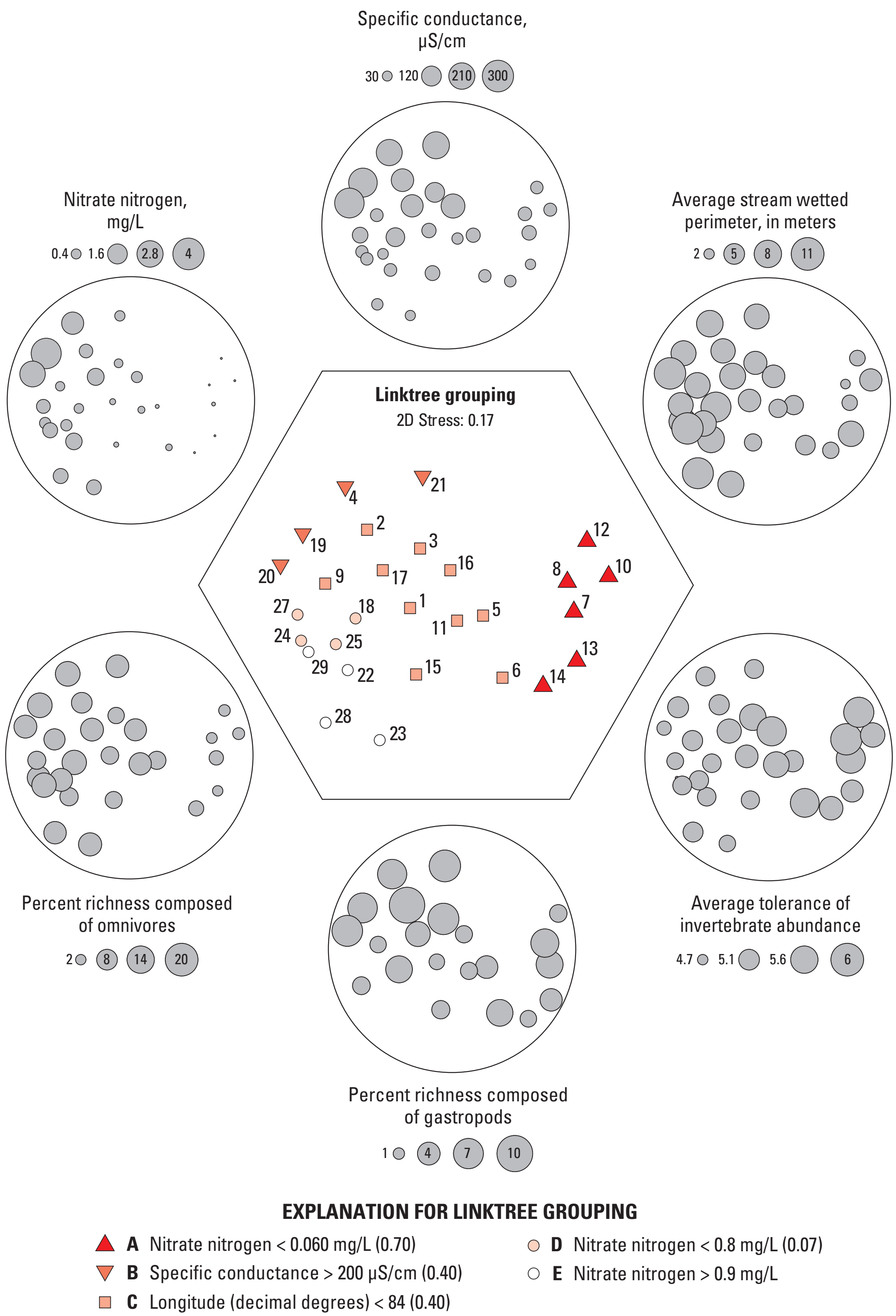

Figure 4. Plot series illustrating nonmetric multidimensional scaling (MDS) ordinations of Georgia Upper Coastal Plain invertebrate community composition from Bray-Curtis similarity matrices. For description of central plot see figure 3 caption. Top graphs are environmental variables, and bottom graphs are invertebrate community indices. [µS/cm, microsiemens per centimeter; <, less than; mg/L, milligram per liter; >, greater than]

The LINKTREE analysis of the GCP invertebrate communities was driven primarily by variables that had an interpretable geographic component and is illustrated in figure 4. Six sites where NOx was detected at low levels (less than 0.06 mg/L) were first split from the remaining communities (average 0.85 mg/L for the remaining sites). Sites with relatively elevated dissolved chemical composition—as estimated by SC—were split (average 236 microsiemens per centimeter [µS/cm] for the group and 75 µS/cm for the remaining sites), and this was followed by longitudinal position, which identified the central portion of the ordination. The sites with low NOx are located in the eastern section of the study area, the four sites with SC greater than 200 µS/cm are located in the upper portions of the Dougherty Plain of Georgia where karstic features interact with surface water, and the similarity of the centrally located sites clearly separated them from the remaining sites. The final separation in the analysis was based on NOx values less than 0.80 mg/L (average 0.55 mg/L for the group and 0.96 mg/L for the remaining sites). Corresponding indicators of the composition of the invertebrate communities showed shifts in the percent richness of omnivores toward the sites with higher NOx ($r_s = 0.81$), an increase in the percent richness of gastropods toward the sites with elevated SC ($r_s = 0.66$), and overall increase in the average tolerance scores as the stream channels became smaller as defined by the wetted perimeter ($r_s = -0.71$). Chironomids were a large portion of the composition at many of these sites and are likely to be more able to tolerate the less stable hydrologic regime inherent in the streams to the east of the study area. A decrease in Ephemeroptera, Plecoptera, and Trichoptera (EPT) taxa and an increase in Odonate richness also characterized many of these streams, although the relations were not significant.

The primary LINKTREE designation with regard to the algal communities in the DP involved those sites defined as having 33 percent or higher benthic macrophyte cover (average 39 percent for the group and 4 percent for the remaining sites) (fig. 5). These are many of the same sites with little intact forest in the near-reach riparian buffer that helped characterize the invertebrate communities. The second split was defined by sites with a concentration of TP of greater than 0.19 mg/L (average 0.38 mg/L for the group and 0.06 mg/L for the remaining sites). The third split defined sites with a percentage of the basin with forest cover of greater than 36 percent (average 46 percent for the group and 19 percent for the remaining sites). The proportion of the riparian zone used as pasture (RZ81) then defined the remaining sites at a split of between 8 and 9 percent (average 6 percent for the group and 13 percent for the remaining sites). The bubble plots of the abiotic variables (specifically TN/TP ratios, RZ81, and TP) were well reflective of the community composition and corresponded to a shift in the communities from a majority of beta-mesosaprobic diatoms ($r_s = 0.70$)—less tolerant to anthropogenic alterations in water chemistry—to increases in the percent eutrophic diatoms ($r_s = 0.60$), and ultimately to dominance by alpha-mesosaprobic diatoms, which are defined as more tolerant ($r_s = 0.69$).

In contrast to the invertebrate communities in the GCP study, the algal ordination resulted in a more linear arrangement of sites, along with some similar but less-defined geographic components (fig. 6). The sites with elevated SC (left side of the ordination) (average 236 µS/cm for the group and 69 µS/cm for the remaining sites) and those with relatively more riparian agriculture (just to the right side of the ordination first split) (average 34 percent for the group and 11 percent for the remaining sites) were initially split in the LINKTREE analysis. Next, the sites with levels of NOx below the laboratory reporting limit (less than 0.06 mg/L for the group and 0.48 mg/L for the remaining sites) defined the right side of the ordination. Subsequently, sites were split where TN was less than 0.76 mg/L (average 0.64 mg/L for the group and 1.1 mg/L for the remaining sites). Site splits where agriculture in the basin-wide riparian zone composed less than 10 percent (average 7 percent for the group and 16 percent for the remaining sites) resolved the remaining communities. The bubble plots of the trophic status of the diatom communities reveal clear gradients based on percentage of the taxa defined as eutrophic or oligotrophic and corresponded well to gradients defined by SC ($r_s = 0.74$) and nitrogen water chemistry in the form of TKN ($r_s = 0.76$). Percent mesotrophic diatoms showed some correlation with the percentage of the riparian zone in agricultural land use ($r_s = -0.65$) indicating an intermediate association between the aforementioned two endpoints.

An effort was made to determine if regional biological indicators of nutrient enrichment could be found in a combined dataset for the eastern Coastal Plain (DP and GCP data pooled). Traditional invertebrate indicators such as EPT and tolerance scores were of limited value in interpreting nutrient condition. The main problem stemmed from the regionally grouped combined dataset where nutrient conditions were much higher in the streams of the DP than in streams of the GCP. Indicators that did show promise in at least responding to this difference (or another not assessed) were abundances of tolerant species and TN ($r_s = 0.66$), percent Ephemeroptera (mayflies) and OP ($r_s = -0.70$), and the abundance of Coleopterans and TN ($r_s = 0.66$). As stated, these data were not normally distributed, and caution should be used to prevent over-generalizations. The combination of algal indices from both studies also provided roughly two endpoints when compared with nutrient chemistry where relations existed in the combined dataset, but where they may not have existed in the individual analyses. The percentages of diatoms identified as nitrogen heterotrophs (Van Dam and others, 1994) correlated with TP and OP ($r_s = 0.71, 0.74$), and the percentage of oligotrophic diatoms was never above 20 when TP levels exceeded 0.06 mg/L. Diatoms identified as oligosaprobic—or highly intolerant—(Lange-Bertalot, 1979) showed the strongest relation to nitrogen as DIN ($r_s = -0.69$). The strongest correlation was seen in the algal composition and stream ionic concentration as measured by SC. Values of SC were well distributed between the study areas, and both the percentages of oligotrophic and eutrophic diatoms correlated strongly to this gradient ($r_s = -0.81, 0.80$).

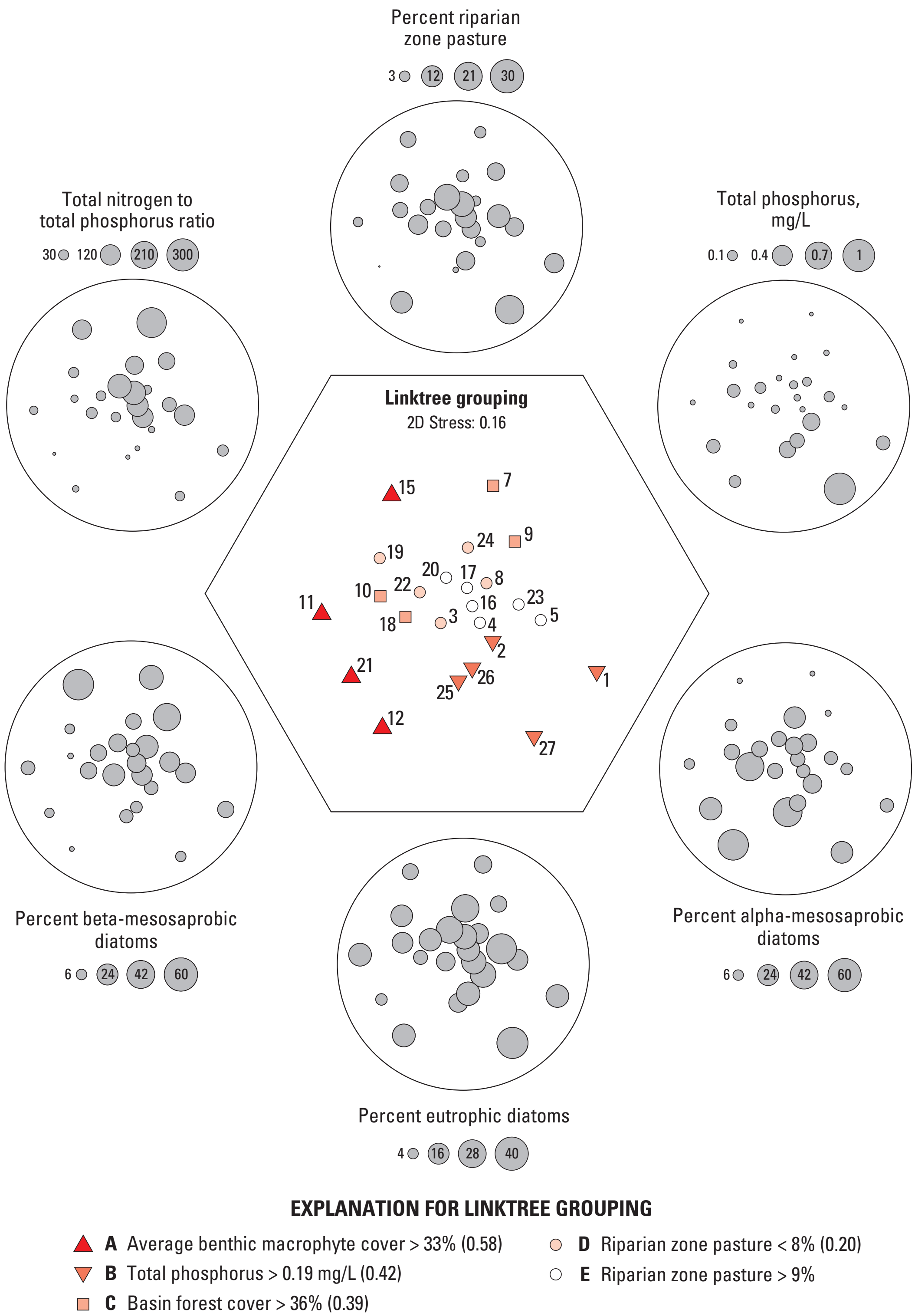

Figure 5. Plot series illustrating nonmetric multidimensional scaling (MDS) ordinations of Delmarva Peninsula algal community composition from Bray-Curtis similarity matrices. For description of central plot see figure 3 caption. Top graphs are environmental variables, and bottom graphs are autecological algal indicators. [mg/L, milligram per liter; >, greater than; %, percent; <, less than]

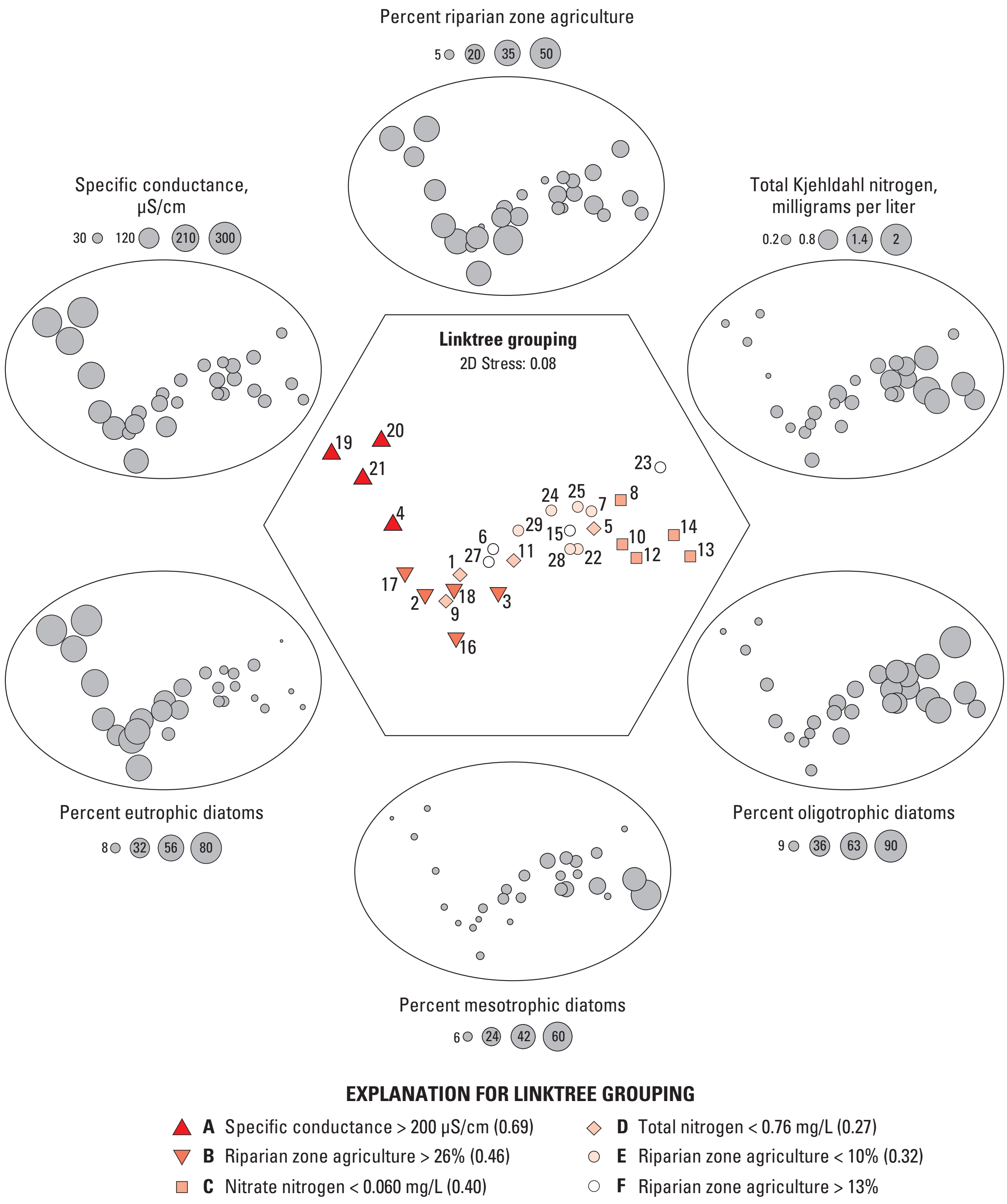

Figure 6. Plot series illustrating nonmetric multidimensional scaling (MDS) ordinations of Georgia Upper Coastal Plain algal community composition from Bray-Curtis similarity matrices. For description of central plot see figure 3 caption. Top graphs are environmental variables, and bottom graphs are autecological algal indicators. [µS/cm, microsiemens per centimeter; >, greater than; %, percent; <, less than; mg/L, milligram per liter]

Table 4 contains summary calculations for the combined nutrient chemistry samples collected in this study. Also included are the values of selected nutrients provided by the USEPA to the States and Indian Tribes to begin the process of nutrient criteria development. These values are based on the 25th percentile of the combined datasets available at the ecoregion level and are termed “reference condition” given the lack of sufficient data for a more direct designation (U.S. Environmental Protection Agency, 2000a, 2000b). Comparisons between the mean and median values of the data from this study are provided. A percent difference between the USEPA values and those of the 25th percentile of the data from this study also is shown in table 4. Because this study was designed to represent a gradient of nutrient conditions in the respective regions, the 25th percentile of phosphorus and nitrogen constituents was taken from the combined two synoptic samples and compared to the published preliminary nutrient criteria values. Within the GCP study, this assumption appears to hold where TKN, NOx, TP, and TN were fairly close to the USEPA values. If nutrient conditions in the 25th percentile of these streams were to meet the criteria, a reduction of TP and TN of approximately 10 percent would be needed. A more significant reduction in nutrient levels, however, would be needed for the median condition of the streams to be consistent with the established reference condition. In the DP streams studied, significant reductions in NOx and TN would be needed to be near the published reference values. With some notable exceptions, TP values were within levels expected at the 25th percentile, although a 30-percent reduction in TP would be required to bring the median levels in the streams to the published reference values. In addition, if these samples had been taken during rainfall-runoff conditions, then a different picture may have emerged where sediment-bound phosphorus likely would have had a significant effect on observed values (Novak and others, 2003). Also included in table 4 is the percentage of samples collected that exceeded the preliminary nutrient criteria for the four published properties. In the GCP, 75 and 84 percent of the samples exceeded criteria for TP and TN, respectively; the DP samples exceeded criteria 65 and 90 percent of the time for TP and TN, respectively, during the base-flow conditions assessed.

Table 4. Summary data for nutrient samples obtained during study.

[TKN, total Kjeldahl nitrogen; NOx, nitrate-nitrite nitrogen; TP, total phosphorus; TN, total nitrogen. Included are: USEPA Level III Ecoregion (ER) reference conditions, ratios between combined sample means and medians for the Delmarva Peninsula and Georgia Upper Coastal Plain studies to the reference condition, the percentage of samples above the reference condition, and the percent reduction in the 25th percentile of the sampled sites to meet the reference condition. Positive values indicate that the 25th percentile was higher than the reference condition and negative values indicate the reverse]

Summary type	TKN	NOx	TP	TN
Delmarva Peninsula				
USEPA Level III ER 63 reference condition	0.510	0.040	0.053	0.870
Ratio of mean to reference	1.0	73.4	2.1	3.9
Ratio of median to reference	0.9	62.3	1.3	3.4
Percent exceedence from reference	38	92	65	90
Percent difference of the 25th percentile from the USEPA reference condition	–46	2,000	–19	85
Georgia Upper Coastal Plain				
USEPA Level III ER 65 reference condition	0.300	0.095	0.023	0.618
Ratio of mean to reference	1.5	5.7	1.5	1.6
Ratio of median to reference	1.3	3.7	1.6	1.5
Percent exceedence from reference	66	73	75	84
Percent difference of the 25th percentile from the USEPA reference condition	–20	–14	8	11

Summary and Conclusions

Differing nutrient levels found between these two study areas, without regard to watershed land use, may be due in part to the sizes of the respective watersheds, the proximity of agricultural activities to the sampling locations, and the inherent differences in hydrogeology. For example, although watershed land use was similar in terms of percent agriculture and percent forest, the Georgia Upper Coastal Plain (GCP) watersheds were on average 10 times larger than the streams in the Delmarva Peninsula (DP) study area. The investigated stream reaches in the DP study also were narrower, shallower, and located higher in the watershed where the effects of agriculture and farming practices would be expected to have a larger influence on stream conditions. Farming practices such as the degree to which manure applications are more concentrated in these watersheds and presumably nearer to streams may be a significant factor in the DP study area. With increasing poultry production in the GCP, this may become more of an issue in the future, although the generally intact and wide riparian areas in the GCP provide an effective means for nutrient uptake and processing. Limited riparian denitrification and shallow subsoils with more direct hyporheic connection to surface waters (Hamilton and others, 1993; Lowrance and others, 1997) likely played a larger role in shaping the nutrient condition in the DP streams. In regards to the estimates of fertilizer and manure application rates used in this study, a more refined quantification at a more applicable scale may be required to elucidate the connections between watershed loadings and instream nutrient concentrations.

Invertebrate communities in the studied streams appear to be more influenced by differences in near-stream habitat and by landscape position than by gradients in nutrient condition. The multivariate analysis indicated clear influences of near-stream riparian cover on the habitat availability of invertebrates and communities separated out prior to any influence of nutrient condition. LINKTREE groupings selected near-stream riparian land use as being the most influential variable for the invertebrate communities in DP streams, although GCP invertebrate communities also grouped with respect to agriculture in the riparian zone even in these somewhat larger systems. This finding is consistent with research directly focused on determining this effect (Lammert and Allan, 1999; Death and Joy, 2004; Rios and Bailey, 2006). Classic indicators of species intolerant to disturbance were not useful in determining nutrient conditions, and in some cases, sensitive taxa were more abundant in streams with elevated nutrient levels (e.g., many in the DP area). Research has indicated that invertebrate assemblages in Coastal Plain streams can vary widely based on time of year and flow conditions, leading to a shift to species more tolerant of higher temperatures and low dissolved oxygen in the low-flow summer period regardless of other potential stressors (Gregory, 1996; Davis and others, 2003). Furthermore, invertebrate communities in low-gradient Coastal Plain streams are likely to be pre-adapted to harsh environmental conditions that are typical even under relatively undisturbed conditions. Factors such as low dissolved oxygen levels, high stream temperatures, and intermittent flow conditions commonly occur in these study areas and potentially select for invertebrate assemblages that are more tolerate of environmental stresses (Clifford, 1966; Feminella, 1996). Further study in Coastal Plain streams with multiple collections spread throughout the year under differing flow conditions is needed to better clarify these relations. Total nitrogen (TN) to total phosphorus (TP) ratios were moderately linked to abundances of intolerant species. The RELATE analysis indicated that nutrient chemistry, taken as a whole, was strongly related to the observed assemblages. Also, additional studies of relations of invertebrate community compositions of nitrogen (N) to phosphorus (P) ratios would be required to elucidate potential controlling mechanisms and to determine if N and P limitation has functional implications in these study areas.

Algal community compositions in both study areas were most correlated with watershed and segment-scale land use and nutrient condition. Algal autecological indices were better suited as indicators of nutrient condition than were invertebrate autecological indices. Percentages of the communities that were composed of diatoms associated with trophic status and pollution tolerance were associated with elevated levels of TP and N in forms other than dissolved inorganic nitrogen (DIN) and TN (e.g., total Kjeldahl nitrogen [TKN]). Algal indicators have been shown to adequately respond to nutrient conditions (Charles and others, 2006; Rier and Stevenson, 2006; Sgro and others, 2006), and considerations should be made to include these in bioassessments of streams with environmental settings similar to those in this study. Indications of agricultural activity in the riparian zone delineated some of the patterns in the multivariate ordinations of community composition, and levels above 25 percent appeared to adversely affect sensitive taxa. Dissolved ionic composition as measured by specific conductance (SC) appears to play a strong role in shaping algal communities not only at the regional scale as previously noted (Potapova and Charles, 2003; Charles and others, 2006), but also within regions at smaller scales such as the Level IV ecoregion. The role of agricultural chemicals and animal waste in increasing stream dissolved-ion concentrations is well established (Lowrance and others, 1984a), and a connection to diatom trophic status and pollution tolerance in agricultural streams may be worthy of further study.

One major difference between the two study areas that was not addressed in this analysis, but is of potential management concern, is the relative importance of instream processing of nutrients. In the GCP, most streams draining areas of intense agriculture drain into progressively larger, low-gradient creek swamp systems (Wharton, 1978) where the potential for further processing of nutrients is increased. In contrast, DP streams, located on a peninsula, flow directly to the Chesapeake and Delaware Bays where nutrient enrichment problems have been documented.

The increased use of managed riparian forest buffers has been suggested as a potential tool to mitigate effects of agriculturally derived, nonpoint-source pollution loading into the Chesapeake Bay (Lowrance and others, 1997). However, these suggestions would only be useful where hydrogeologic and human controls do not prevent the functional riparian zone from playing a role in denitrification. Intact riparian zones in any setting will ameliorate inputs of sediment-associated phosphorus and organic forms of nitrogen although nutrient management schemes would be necessary at the watershed level to fully reduce nutrient inputs when the riparian zone is bypassed. This may primarily be the case regarding streams in the DP (J.M. Denver, U.S. Geological Survey, written commun., 2007).

Results from this study provide additional evidence of the important role that riparian zones have at multiple spatial scales, provide evidence of the role that riparian ecosystems can play at the interface between agricultural and aquatic ecosystems, and show examples of where riparian-zone management alone may not be an effective nutrient reduction scheme. Multivariate tools that are relatively new to these types of studies were used to reach these conclusions. This approach enabled the interpretation of subsets of ecological datasets to identify individual relevant variables influencing the communities of sites and groups of sites.

From 2000 to 2005, more than 10 percent of Delaware's agricultural land was taken out of active production, with much of it converted to development. During this same period in Georgia, more than 5 percent of agricultural land was converted (Corporation for Enterprise Development, 2007). This placed Delaware first and Georgia sixth in conversion of agricultural lands nationally (Corporation for Enterprise Development, 2007). Many of the streams studied on the DP are in areas close to growing towns and cities where this conversion is most likely to take place. It could be assumed that many of the influences that agricultural land use has had on these streams' physical habitats, water chemistries, and biological communities will be replaced by the influences of urban development in some areas and become more concentrated on the remaining agricultural lands. Riparian buffers have been shown to mitigate numerous stressors and, when combined with watershed-wide management of nutrients and stormwater runoff, may prevent detrimental effects of existing and converted land use from occurring. Implementing nutrient criteria and monitoring the effectiveness of watershed and streamside management will be needed for mitigation to be ensured.

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Appendix A. Variables Determined by LINKTREE Procedures, Algal and Invertebrate Indices, and Abiotic Variables for the Delmarva Peninsula Study as Illustrated in Figures 3 and 5.

Appendix A. LINKTREE split variables, algal and invertebrate indices, and abiotic variables for the Delmarva Peninsula study as illustrated in figures 3 and 5. Abiotic variables presented in algal analyses indicated by subscript 1, those for invertebrates by subscript 2. Variable abbreviations can be found in table 3.

[<, less than; >, greater than]

Site number (location shown in figure 1)	Invertebrate LINKTREE group	Algal LINKTREE group	Abiotic variables								
			$BA4_2$	$RZ81_1$	$c050sp_2$	$wv025rp_2$	TP_1	$TN\_TP_{1,2}$	DOC_2	DIN_2	$P\_Mphy\_Avg_1$
1	DOC>4	TP>0.19	14.8	10.1	4.8	100	0.189	37	4.89	3.86	0
2	DOC>4	TP>0.19	23.5	2.7	2.1	100	.293	12	5.73	2.79	0
3	DIN<3.2	RZ81<8	27.5	6.7	1.4	100	.065	27	2.64	1.97	0
4	DIN>4.2	RZ81>9	14.9	9.0	5.0	90	.029	127	2.16	4.99	0
5	DOC>4	RZ81>9	12.6	9.1	3.5	100	.027	121	4.08	4.61	4
7	DIN<3.2	BA4>36	57.4	3.5	0.8	100	.014	259	2.06	2.61	0
8	DIN<3.2	RZ81<8	16.4	2.7	5.0	100	.078	24	3.15	1.60	1
9	DIN<3.2	BA4>36	42.7	8.2	4.1	95	.030	83	2.32	3.15	0
10	c050sp>15	BA4>36	48.9	6.3	17.4	100	.163	16	7.32	0.845	0
11	wv025rp<82	P_Mphy_ Avg>33	45.3	2.4	15.2	12	.025	21	3.28	1.15	42
12	wv025rp<82	P_Mphy_ Avg>33	35.4	12.7	47.5	31	.136	13	11.7	1.75	55
15	wv025rp<82	P_Mphy_ Avg>33	38.5	6.8	33.5	35	.016	109	2.09	3.37	58
16	DIN>4.2	RZ81>9	15.5	12.4	2.3	100	.043	135	2.39	5.39	14
17	DIN>4.2	RZ81>9	16.6	16.5	7.2	100	.067	161	2.09	8.79	0
18	DIN<3.2	BA4>36	36.2	10.1	11.8	83	.034	36	1.35	2.66	16
19	wv025rp<82	RZ81<8	21.3	8.2	26.4	0	.058	31	1.82	1.65	10
20	c050sp>15	RZ81>9	11.5	18.1	15.3	91	.031	163	2.65	9.51	7
21	wv025rp<82	P_Mphy_ Avg>33	29.3	0.1	.745	36	.181	3	3.31	.042	33
22	c050sp>15	RZ81<8	32.0	6.4	54.8	100	.120	28	3.01	1.94	0
23	DIN>4.2	RZ81>9	23.2	14.1	0	100	.105	58	2.92	4.17	1
24	DIN>4.2	RZ81<8	22.0	3.9	5.3	100	.026	96	1.38	5.08	0
25	wv025rp<82	TP>0.19	20.9	0.8	23.8	82	.265	5	6.71	.424	6
26	c050sp>15	TP>0.19	33.8	9.2	19.6	92	.210	7	5.59	.672	21
27	DOC>4	TP > 0.19	3.7	21.9	1.9	100	.961	27	5.70	2.86	0

Appendix A. LINKTREE split variables, algal and invertebrate indices, and abiotic variables for the Delmarva Peninsula study as illustrated in Figures 3 and 5. Abiotic variables presented in algal analyses indicated by 1, those for invertebrates by 2. Variable abbreviations can be found in Table 3.—Continued

[<, less than; >, greater than]

Invertebrate indices			Algal indices		
Percent abundance of omnivores[a]	Average tolerance of invertebrate abundance[a]	Abundance of intolerant invertebrates	Percent alpha-mesosaprobic diatoms[b]	Percent beta-mesosaprobic diatoms[b]	Percent eutrophic diatoms[c]
25.2	5.1	2,769	9.8	14.4	18.5
17.7	5.0	1,398	19.1	10.1	23.7
6.3	5.4	1,487	12.4	26.0	12.6
14.0	5.1	3,240	9.7	22.6	21.6
23.6	4.8	3,157	7.4	21.1	16.0
0.3	4.3	7,656	1.3	31.2	11.0
36.3	5.4	4,635	16.3	28.8	14.2
1.1	4.0	2,734	1.9	40.3	9.5
0.5	6.3	142	13.0	1.7	15.5
1.0	5.2	275	5.9	10.4	18.6
4.9	5.9	426	50.3	1.2	18.7
0.8	6.0	737	1.5	50.4	9.1
8.0	5.2	2,397	11.3	19.2	18.6
13.9	4.9	2,469	17.0	9.5	21.9
3.2	5.6	872	42.7	14.7	7.1
2.0	5.8	2,302	7.4	5.1	16.9
28.1	4.3	1,093	12.1	17.1	25.7
0.7	7.1	146	27.2	4.8	4.8
13.0	5.1	1,594	13.6	14.6	17.9
17.8	4.4	3,524	9.3	21.7	32.5
2.5	5.3	5,449	24.8	13.1	27.1
31.8	6.4	86	44.9	9.5	12.8
0.1	6.8	853	14.6	7.3	19.8
18.4	4.5	984	25.5	5.8	34.9

[a] Barbour and others, 1999
[b] Lange-Bertalot, 1979
[c] Van Dam and others, 1994

Appendix B. Variables Determined by LINKTREE Procedures, Algal and Invertebrate Indices, and Abiotic Variables for the Georgia Upper Coastal Plain Study as Illustrated in Figures 4 and 6.

Appendix B. LINKTREE split variables, algal and invertebrate indices, and abiotic variables for the Georgia Upper Coastal Plain study as illustrated in figures 4 and 6. Abiotic variables presented in algal analyses indicated by subscript 1, those for invertebrates by subscript 2. Variable abbreviations can be found in table 3.

[<, less than; >, greater than]

Site number (location shown in figure 1)	Invertebrate LINKTREE group	Algal LINKTREE group	Abiotic variables						
			$RZ8_1$	$SC_{1,2}$	$NOx_{1,2}$	TN_1	TKN_1	$LONGDEC_2$	$WetPerm\_Avg_2$
1	LONG-DEC<84	TN<0.756	1.9	63	0.149	0.531	0.382	83.49833	5.7
2	LONG-DEC<84	RZ8>26	33.2	155	.749	.940	.191	83.61306	6.5
3	LONG-DEC<84	RZ8>26	44.9	118	.311	.631	.320	83.58417	6.0
4	SC>200	SC>200	23.9	207	2.00	2.05	.054	83.46194	7.6
5	LONG-DEC<84	TN<0.756	13.3	61	.062	.756	.694	82.86556	4.5
6	LONG-DEC<84	RZ8>13	13.3	47	.179	.843	.664	83.25806	4.6
7	NOx<0.060	RZ8<10.4	10.4	64	< .060	.787	.729	82.78083	3.2
8	NOx<0.060	NOx<0.060	13.9	56	< .060	1.18	1.17	82.85611	1.9
9	LONG-DEC<84	TN<0.756	7.7	50	.353	.653	.300	83.08083	7.1
10	NOx<0.060	NOx<0.060	18.0	51	< .060	1.55	1.54	82.34639	5.6
11	LONG-DEC<84	TN<0.756	17.8	39	.203	.617	.414	82.76444	4.1
12	NOx<0.060	NOx<0.060	8.8	48	< .060	1.30	1.29	82.25111	3.3
13	NOx<0.060	NOx<0.060	9.1	33	< .060	.836	.821	82.40889	7.1
14	NOx<0.060	NOx<0.060	13.0	41	< .060	1.16	1.14	82.40639	3.5
15	LONG-DEC<84	RZ8>13	16.7	67	.103	1.02	.916	82.23833	3.8
16	LONG-DEC<84	RZ8>26	32.3	175	.500	.925	.425	83.89194	5.8
17	LONG-DEC<84	RZ8>26	30.8	150	1.17	1.48	.309	83.90222	7.3
18	NOx<0.8	RZ8>26	26.0	107	.376	.617	.241	83.99639	9.4
19	SC>200	SC>200	32.3	252	3.72	3.85	.131	83.85500	6.9
20	SC>200	SC>200	34.6	267	2.67	2.82	.153	83.99278	10.3
21	SC>200	SC>200	20.9	219	.433	.603	.170	83.97833	6.8
22	NOx>0.9	RZ8<10.4	4.9	49	1.11	1.61	.501	84.49389	7.7
23	NOx>0.9	RZ8>13	15.4	32	.896	1.13	.235	84.27611	7.4
24	NOx<0.8	RZ8<10.4	2.9	46	.520	1.01	.489	84.56750	7.8
25	NOx<0.8	RZ8<10.4	8.2	35	.522	.963	.441	84.56472	7.1
27	NOx<0.8	RZ8>13	19.9	75	.781	.992	.211	84.56667	6.8
28	NOx>0.9	RZ8<10.4	8.6	40	.912	1.26	.351	84.68750	10.0
29	NOx>0.9	RZ8<10.4	5.7	47	.916	1.13	.214	84.69833	10.2

Appendix B. LINKTREE split variables, algal and invertebrate indices, and abiotic variables for the Georgia Upper Coastal Plain study as illustrated in Figures 4 and 6. Abiotic variables presented in algal analyses indicated by 1, those for invertebrates by 2. Variable abbreviations can be found in Table 3.—Continued

[<, less than; >, greater than]

Invertebrate indices			Algal indices		
Percent richness composed of omnivores[a]	Average tolerance of invertebrate abundance[a]	Percent richness composed of gastropods[a]	Percent oligotrophic diatoms[b]	Percent mesotrophic diatoms[b]	Percent eutrophic diatoms[b]
5.8	5.2	1.9	15.8	4.7	45.8
8.0	5.1	9.8	7.9	1.8	35.1
5.0	5.5	7.5	23.6	1.9	13.7
11.4	5.0	6.7	15.0	2.8	60.3
6.1	5.1	3.9	44.1	4.6	8.0
0	5.7	5.5	24.7	5.9	39.1
3.8	5.7	5.7	49.1	8.4	9.9
2.2	5.9	6.1	52.2	6.9	12.1
8.3	5.1	2.1	8.5	2.7	58.2
2.6	5.3	0	52.3	16.2	3.8
9.3	5.4	2.3	18.3	5.5	28.7
2.5	5.8	2.4	57.0	2.5	6.5
2.0	5.2	3.8	26.9	52.4	2.3
4.3	5.3	2.1	43.3	32.0	2.3
5.4	5.0	2.6	42.6	5.2	10.8
9.1	5.7	2.3	11.1	3.5	55.7
9.8	5.3	4.8	15.8	2.4	42.1
11.1	5.0	5.6	9.7	1.9	53.9
11.4	5.1	6.8	3.0	0.8	77.9
9.8	4.8	7.3	4.9	2.1	79.4
9.6	5.0	7.7	8.8	2.5	56.7
6.3	5.0	0	36.8	13.6	9.1
10.0	4.9	0	85.1	2.0	0.6
9.8	4.5	2.4	28.6	13.3	12.1
9.8	5.0	0	45.0	9.2	6.1
6.1	4.9	0	18.5	6.9	36.5
9.4	5.1	0	32.1	9.0	9.9
11.1	5.0	0	23.5	7.0	25.6

[a] Barbour and others, 1999

[b] Van Dam and others, 1994

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