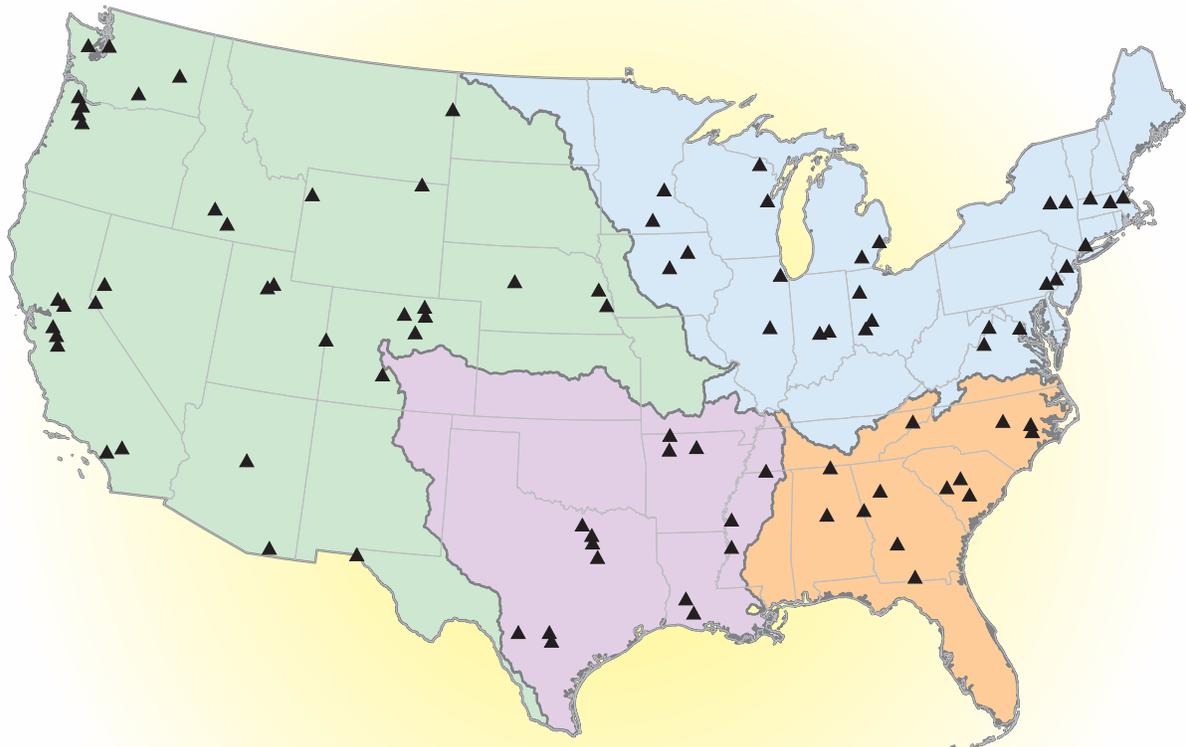


**National Water-Quality Assessment Program**

**Multi-Regional Synthesis of Temporal Trends in Biotic Assemblages in Streams and Rivers of the Continental United States**



Scientific Investigations Report 2013–5046

**Cover.** (Top) Four geographic regions described in this report and locations of 91 long-term surface-water status and trend sites. (Bottom, left to right) Photographs showing *Achnanthydium minutissimum*, a monoraphid diatom (Potapova, 2009); *Neophylax*, (Autumn Mottled Sedges) Caddisfly Larva (Photograph by Steven Fend, USGS); *Oncorhynchus mykiss*, Rainbow Trout (Photograph by Terry Maret, USGS).

# **Multi-Regional Synthesis of Temporal Trends in Biotic Assemblages in Streams and Rivers of the Continental United States**

By Matthew P. Miller, Anne M.D. Brasher, and Jonathan G. Kennen

National Water-Quality Assessment Program

Scientific Investigations Report 2013–5046

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

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**U.S. Geological Survey**  
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## Foreword

The U.S. Geological Survey (USGS) is committed to providing the Nation with reliable 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, measured in terms of quantity and quality, even more essential to the long-term sustainability of our communities and ecosystems.

The USGS implemented the National Water-Quality Assessment (NAWQA) Program in 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 quality of our Nation's streams and groundwater? How are conditions changing over time? How do natural features and human activities affect the quality of streams and groundwater, 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. From 1991 to 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/studies/study\\_units.html](http://water.usgs.gov/nawqa/studies/study_units.html)).

In the second decade of the Program (2001–2012), a major focus is on regional assessments of water-quality conditions and trends. These regional assessments are based on major river basins and principal aquifers, which encompass larger regions of the country than the Study Units. Regional assessments extend the findings in the Study Units by filling critical gaps in characterizing the quality of surface water and groundwater, and by determining water-quality status and trends at sites that have been consistently monitored for more than a decade. In addition, the regional assessments continue to build an understanding of how natural features and human activities affect water quality. Many of the regional assessments employ modeling and other scientific tools, developed on the basis of data collected at individual sites, to help extend knowledge of water quality to unmonitored, yet comparable areas within the regions. The models thereby enhance the value of our existing data and our understanding of the hydrologic system. In addition, the models are useful in evaluating various resource-management scenarios and in predicting how our actions, such as reducing or managing nonpoint and point sources of contamination, land conversion, and altering flow and (or) pumping regimes, are likely to affect water conditions within a region.

Other activities planned during the second decade include continuing national syntheses of information on pesticides, volatile organic compounds (VOCs), nutrients, trace elements, and aquatic ecology; and continuing national topical studies on the fate of agricultural chemicals, effects of urbanization on stream ecosystems, bioaccumulation of mercury in stream ecosystems, effects of nutrient enrichment on stream ecosystems, and transport of contaminants to public-supply wells.

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. We hope this NAWQA publication will provide you with 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.

William H. Werkheiser  
USGS Associate Director for Water

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## Conversion Factors and Abbreviations

Inch/Pound to SI

<b>Multiply</b>	<b>By</b>	<b>To obtain</b>
	Length	
mile (mi)	1.609	kilometer (km)

SI to Inch/Pound

<b>Multiply</b>	<b>By</b>	<b>To obtain</b>
	Length	
kilometer (km)	0.6214	mile (mi)
meter (m)	1.094	yard (yd)
	Area	
square meter (m <sup>2</sup> )	0.0002471	acre
square kilometer (km <sup>2</sup> )	247.1	acre
square meter (m <sup>2</sup> )	10.76	square foot (ft <sup>2</sup> )
square kilometer (km <sup>2</sup> )	0.3861	square mile (mi <sup>2</sup> )

## Abbreviations

ADAS	Algal Data Analysis System
IDAS	Invertebrate Data Analysis System
NARS	National Aquatic Resource Survey
NAWQA	National Water-Quality Assessment
NMDS	Non-metric multidimensional scaling
NWIS	National Water Information System
NWQL	National Water-Quality Laboratory
PRIMER	Plymouth Routines In Multivariate Ecological Research
PRISM	Parameter elevation Regressions on Independent Slopes Model
PTI	Pesticide toxicity index
USGS	U.S. Geological Survey

# Multi-Regional Synthesis of Temporal Trends in Biotic Assemblages in Streams and Rivers of the Continental United States

By Matthew P. Miller, Anne M.D. Brasher, and Jonathan G. Kennen

## Abstract

Biotic assemblages in aquatic ecosystems are excellent integrators and indicators of changing environmental conditions within a watershed. Therefore, temporal changes in abiotic environmental variables often can be inferred from temporal changes in biotic assemblages. Algae, macroinvertebrate, and fish assemblage data were collected from 91 sampling sites in 4 geographic regions (northeastern/north-central, southeastern, south-central, and western), collectively encompassing the continental United States, from 1993 to 2009 as part of the U.S. Geological Survey National Water-Quality Assessment Program. This report uses a multivariate approach to synthesize temporal trends in biotic assemblages and correlations with relevant abiotic parameters as a function of biotic assemblage, geographic region, and land use. Of the three groups of biota, algal assemblages had temporal trends at the greatest percentage of sites. Of the regions, a greater percentage of sites in the northeastern/north-central and western regions had temporal trends in biotic assemblages. In terms of land use, a greater percentage of watersheds draining agricultural, urban, and undeveloped areas had significant temporal changes in biota, as compared to watersheds with mixed use. Correlations between biotic assemblages and abiotic variables indicate that, in general, macroinvertebrate assemblages correlated with water quality and fish assemblages correlated with physical habitat. Taken together, results indicate that there are regional differences in how individual biotic assemblages (algae, macroinvertebrates, and fish) respond to different abiotic drivers of change.

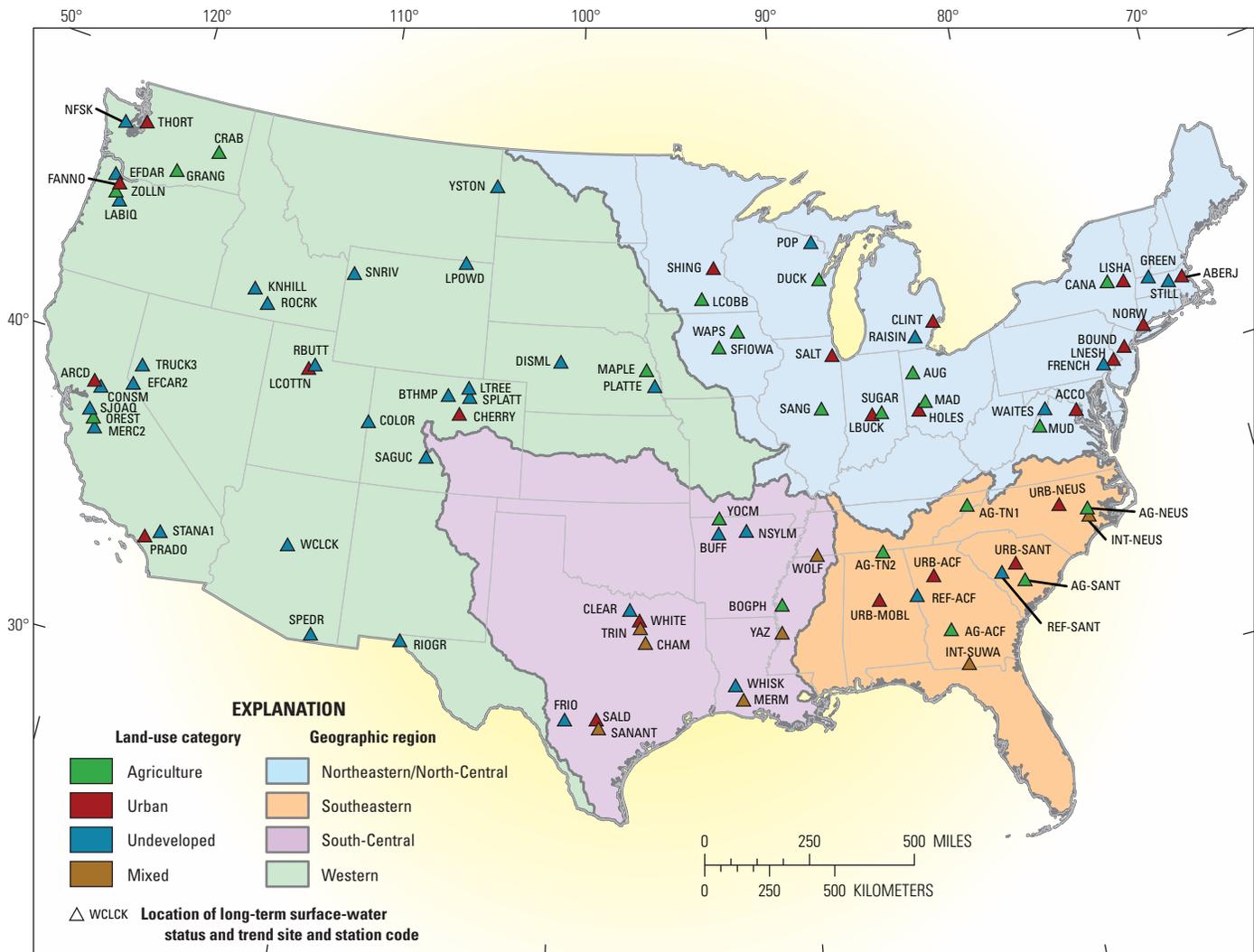
## Introduction

Biotic assemblages (algae, macroinvertebrates, and fish) in aquatic ecosystems are influenced by hydrologic, physical-habitat, water-quality, and land-use conditions in the stream and watershed. Consequently, temporal changes taking place

in the abiotic environment often can be inferred from observed temporal changes in the composition of biotic assemblages. Monitoring temporal change in biotic assemblages can provide an early indication of abiotic environmental change; can complement information on hydrologic, physical, and water-quality conditions; and is important to understanding the long-term incremental effects of human and natural effects on ecosystems. Additionally, understanding how biotic assemblages change over time is important for characterizing biological integrity, which is a major focus of the Clean Water Act (Cairns, 1975; Frey, 1975; Karr, 1981; Karr and Chu, 1997). While numerous studies have investigated temporal trends in biotic metrics and (or) assemblages (Jackson and Füreder, 2006 and references therein), broad multi-regional temporal changes in biotic assemblages have been poorly documented because of a lack of long-term datasets and an inability to identify a common approach for evaluating trends. Synthesizing common temporal patterns at large geographic scales (for example, the regional scale) is one way to better understand how environmental and anthropogenic conditions are more broadly affecting aquatic ecosystems.

The U.S. Geological Survey (USGS) National Water-Quality Assessment (NAWQA) Program evaluated ecological trends in each of four geographic regions (northeastern/north-central, southeastern, south-central, and western) in order to document temporal change and the processes responsible for change in each region (fig. 1). Within each region, temporal change in biotic assemblages has been assessed among a broad range of abiotic environmental conditions and in diverse land-use settings. These region-specific studies have identified biotic metrics that are representative of, and abiotic metrics that may be responsible for, observed temporal change in biotic assemblages. In addition to the region-specific findings described in these studies, the data collected as part of these efforts provide a foundation from which among-region differences in trends in biotic assemblages can be synthesized and compared. Specifically, data presented in the region-specific reports provide an opportunity to identify—at a broad spatial scale—how the percentage of sites with temporal

## 2 Multi-Regional Synthesis of Temporal Trends in Biotic Assemblages in Streams and Rivers of the United States



Base modified from U.S. Geological Survey  
1:2,000,000-scale digital data

**Figure 1.** The spatial distribution of the 91 long-term surface-water status and trend sites in the continental United States. The four geographic regions described in the report are shown, and dominant land use at each site is indicated by the color of the site symbols.

trends in biotic assemblages vary among (1) biotic assemblages (for example, if one biotic assemblage—algae—tends to change at a greater percentage of sites than another biotic assemblage—fish); (2) geographic regions; and (3) land-use categories. Such a multi-regional analysis is relevant to the management of stream ecosystems at the regional and national scale and can provide information that may be useful in developing public policy necessary for land-use and resource development decisions.

### Purpose and Scope

The purpose of this report is to summarize and synthesize the findings of four regional studies of temporal trends in biotic assemblages (algae, macroinvertebrates, and fish) collected as part of the NAWQA Program. These reports

include published journal articles describing trends in the combined northeastern and north-central United States (U.S.) (Kennen and others, 2012) and the south-central U.S. (Miller and others, 2012), a journal article that is currently in review describing trends in the southeastern U.S. (Daniel Calhoun, USGS, unpub. data, October 5, 2011) and a USGS Open-File Report describing trends in the western U.S. (Wiele and others, 2012). Regional differences are examined in the percentage of sites that have been identified as having statistically significant temporal trends in one or more of the three biotic assemblages. The synthesis of biologic data collected from the four regions has resulted in a dataset that includes many sites that span a gradient of land use, and the potential role of land use in determining biotic trends is explored. Finally, generalized abiotic environmental variables that are correlated with, and are possible drivers of, the biotic assemblages are examined.

## Approach and Methods

This section describes the general approach used for summarizing and synthesizing temporal trends in biotic assemblages as well as correlated abiotic environmental variables among regions, sites included in the four trends reports, sampling and data-processing methodology, and statistical approaches to data analysis. Given the differences in the methodological approaches among the four regions, the general approach used for the present study was to summarize the major commonalities and differences in the methods applied in each of the region-specific reports. In turn, by focusing on the commonalities and accounting for the differences in methodology among regions, a quantitative comparison of the findings among regions was possible. Details on the frequency and dates of sample collection and lists of final environmental variables selected for analysis in each region are available in the region-specific reports.

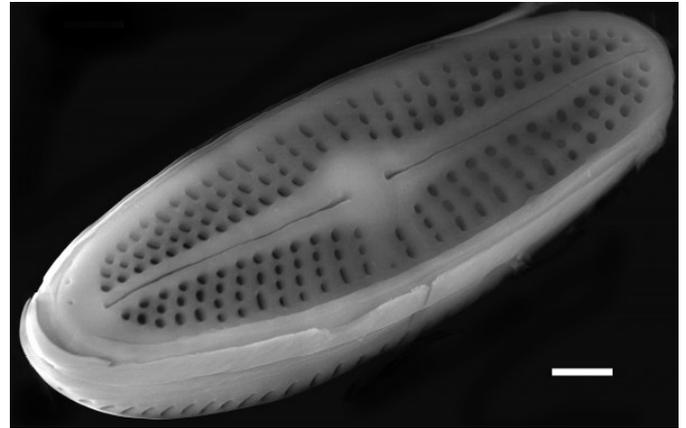
### Sites

A total of 91 sampling sites, located in four geographic regions, sampled from 1993 to 2009 are included in this report (table 1, at end of report; fig. 1). Twenty-seven sites are located in northeastern and north-central United States, 13 sites are located in southeastern U.S., 15 sites are located in south-central U.S., and 36 sites are located in western U.S. Drainage areas range from 19 square kilometers (km<sup>2</sup>) at Red Butte Creek at Fort Douglas, near Salt Lake City, UT, to 220,908 km<sup>2</sup> at the Platte River at Louisville, NE, with an average drainage area of 8,585 km<sup>2</sup>. Sites were classified based on dominant land use in the watershed. Land-use categories include agricultural, urban, and undeveloped. Additionally, some sites in the southeastern and south-central U.S. were categorized as having mixed land use (urban plus agriculture).

### Sample Collection and Data Processing

#### Biota

Algae, macroinvertebrates, and fish were collected using standard methods as part of the NAWQA Program (Cuffney and others, 1993; Meador and others, 1993a; Porter and others, 1993; Moulton and others, 2002). It is important to note that not all biotic assemblages were analyzed for temporal trends at all sites (table 1). Algae (benthic periphyton) were collected by scraping five rocks or snags within each stream reach, composited into a single sample, and the area sampled was recorded (Porter and others, 1993; Moulton and others, 2002). Algae were preserved in 5-percent formalin and identified/enumerated to the lowest practical taxonomic level at the Philadelphia Academy of Sciences (Charles and others, 2002).



*Achnanthisdium minutissimum*, a monoraphid diatom; scale bar = 1 micrometer (μm). (Potapova, 2009).

Aquatic macroinvertebrates were collected from an area of 0.25 square meter (m<sup>2</sup>) in each of five riffle habitats within each stream reach using a slack sampler (500 μm mesh) and composited into a single sample (Cuffney and others, 1993; Moulton and others, 2002). At sites where riffles were not present, macroinvertebrates were collected from five snags, composited into a single sample, and the area sampled was recorded. Samples were preserved in 10-percent formalin and sent to the USGS National Water-Quality Laboratory (NWQL) in Denver, Colorado, for identification (Moulton and others, 2000). In the laboratory, a quantitative fixed-count processing method was used to identify and estimate the abundance of each taxon sorted in the samples.

Fish were collected from all habitat types in the stream reach (20 times the wetted width, a minimum of 150 meters (m)) using backpack, towed barge, or boat mounted electrofishing units and regularly supplemented with three seine hauls, following standard NAWQA protocols (Meador and others, 1993a; Moulton and others, 2002). Fish were identified



*Neophylax* (Autumn Mottled Sedges) Caddisfly Larva. (Photograph by Steven Fend, USGS).



*Oncorhynchus mykiss*, Rainbow Trout. (Photograph by Terry Maret, USGS).

to species, enumerated, weighed, and measured in the field before being released back into the stream.

Algal and macroinvertebrate density (abundance per unit area) data as well as fish abundance data were used to calculate a variety of biotic metrics. The methods for metric calculation and final metric selection varied by region. However, in addition to the use of some region-specific metrics, the USGS Algal Data Analysis System (ADAS; <ftp://ftpext.usgs.gov/pub/er/nc/raleigh/tfc/ADAS/Manual/>) and Invertebrate Data Analysis System (IDAS; Cuffney, 2003) software packages were used to generate a common subset of algal and macroinvertebrate metrics, respectively, at all sites where algae and macroinvertebrates were analyzed (table 1). Algal metrics include a range of indicators for selected water-quality variables including nitrogen tolerance, pollution tolerance, salinity tolerance, and oxygen tolerance (Porter, 2008). Macroinvertebrate metrics include those based on community composition, life history, mobility, morphology, and ecology (Cummins, 1973; Barbour and others, 1999; Cuffney, 2003; Poff and others, 2006). Fish metrics include status (native, endemic, or introduced), tolerance, trophic ecology, and reproductive strategy (Barbour and others, 1999; Meador and others, 1993a; Goldstein and Meador, 2004; Whittier and others, 2007a, b; Frimpong and Angermeier, 2009; Froese and Pauly, 2009).

## Environmental Variables

The specific environmental variables/metrics tested for correlations with biotic assemblages varied by region. Therefore, it was not possible to quantitatively compare and contrast specific environmental variables identified as being significantly correlated with biotic assemblages among regions. To address this limitation, information regarding generalized abiotic environmental variables (for example, the general category of “water quality” as opposed to the specific category of “nitrate concentrations”) correlated with biotic assemblages was compared and contrasted among regions. The specific environmental variables/metrics assessed in all regions fall into

one of three general categories: hydrology, physical habitat, and water quality (including precipitation and air temperature). Additionally, biotic trends were synthesized in the context of dominant land-use type (table 1). While environmental variables were compiled in the USGS Open-File Report describing temporal trends in biotic assemblages in the western U.S. (Wiele and others, 2012), a report identifying the correlations between environmental variables and biotic assemblages for sites in the western U.S. has not been published. Therefore, correlations between environmental variables and biotic assemblages at sites in the western U.S. are not discussed.

Hydrologic metrics (magnitude, frequency, duration, timing, and rate of change) were calculated using data acquired from the USGS National Water Information System (NWIS, <http://waterdata.usgs.gov/nwis/sw>), and a variety of approaches for calculating hydrologic metrics was applied (Richter and others, 1996; McMahon and others, 2003; Henriksen and others, 2006; The Nature Conservancy, 2009). Physical habitat data were acquired following standard USGS methods (Meador and others, 1993b; Fitzpatrick and others, 1998), and water-quality data (nitrogen and phosphorous, pH, dissolved oxygen, specific conductance, water temperature, major ions, suspended sediment, and pesticides) were acquired from NWIS and the NAWQA Data Waterhouse (<http://infotrek.er.usgs.gov/nawqa>). Climate (precipitation and air temperature) metrics were calculated using data acquired from the Parameter elevation Regressions on Independent Slopes Model (PRISM, <http://prism.oregonstate.edu>) and National Oceanic and Atmospheric Administration weather observation stations (<http://www.ncdc.noaa.gov/oa/climateresearch.html>).

## Statistical Analyses

Temporal trends in biotic assemblages were investigated at all sites using a multivariate statistical approach. This approach allows for temporal change in the entire biotic assemblage in question (algae, macroinvertebrates, or fish) at a given site to be quantitatively assessed by accounting for changes in the abundance of all species, as opposed to, for example, quantifying temporal change in the abundance of a single species. At all sites and on all sample dates, abundance or density data were standardized by total abundance or density, respectively, and either square root- or fourth root-transformed prior to generation of Bray-Curtis similarity resemblance matrixes using the Plymouth Routines In Multivariate Ecological Research (PRIMER) program (Clarke and Gorley, 2006). The type of data (abundance or density) and type of transformation (square root or fourth root) varied by region and biotic assemblage. PRIMER was then used to generate non-metric multidimensional scaling (NMDS) ordination plots that included data from all sample dates for each site. NMDS plots are graphical representations of the Bray-Curtis similarity matrixes, with points (representing biotic assemblages on a given sample date) that have more similar biotic assemblages plotting closer to one another than those with more dissimilar

biotic assemblages. The statistical significance of temporal change in biotic assemblages at each site was tested using PRIMER's RELATE procedure, which is a non-parametric seriation procedure (Clarke and Gorley, 2006; Clarke and others, 2006). For the present study, a statistically significant temporal trend at a given site was defined as having  $p < 0.05$ . Fisher's exact test was used to identify whether the percentage of sites with significant temporal trends was significantly different among biotic groups, regions, and land-use categories (Fisher, 1922). Fisher's exact test generally is used to determine if there are non-random associations between categorical variables. This test is appropriate to use when dealing with small sample sizes because, rather than approximating the significance of deviation from the null hypothesis (as is done with other tests that can be used to analyze contingency tables, such as a chi-square test), Fisher's exact test calculates the exact significance of deviation from the null hypothesis. This distinction means that Fisher's exact test provides greater confidence than other significance tests, especially when dealing with small sample sizes.

In contrast to the consistent approach used for the identification of temporal trends described above, the approaches used to identify subsets of environmental variables and biotic metrics that are strongly correlated with the biotic assemblages varied among regions. At sites where a significant temporal trend was identified in the biotic assemblage, subsets of environmental variables and biotic metrics were identified that strongly correlated with the biotic assemblage. This approach provides insights into which environmental variables are likely abiotic drivers of change in the biotic assemblage, and which subsets (that is, metrics) of the broader biotic assemblage are related to the overall temporal change in the biotic assemblage. In the northeastern/north-central and south-central regions (Kennen and others, 2012; Miller and others, 2012), the general approach used to identify the aforementioned subsets was to use the PRIMER routines BIOENV (for environmental variables; Clarke and Ainsworth, 1993) and BVSTEP (for biotic metrics; Clarke and Warwick, 1998). Both BIOENV and BVSTEP use Spearman rank correlation coefficients ( $\rho$ ) to compare the biotic-assemblage resemblance matrix at a given site with the environmental-variable and biotic-metric resemblance matrixes (based on Euclidean distance), respectively. The subset of environmental variables and biotic metrics found to have the highest correlation ( $\rho$ ) to the biotic assemblage were then identified. BIOENV compares the biotic-assemblage matrix with all possible subsets of environmental-variable matrixes, whereas BVSTEP uses a stepwise approach to compare the biotic-assemblage matrix with the biotic-metrics matrix. At sites in the southeastern region (Daniel Calhoun, USGS, unpub. data, October 5, 2011) with significant temporal trends in biotic assemblages (as identified by RELATE), non-parametric Spearman correlation coefficients were calculated to compare Euclidean-resemblance matrixes for environmental variables and biotic metrics with the biotic-assemblage matrix. This allowed for the identification of the subset of environmental variables and biotic metrics

that were most strongly correlated with the biotic assemblage. Additionally, Kendall's tau-b correlation coefficients were calculated and used to identify temporal trends in environmental variables and biotic metrics at each site. Fisher's exact test was used to identify whether the percentage of sites with significant correlations between a given biotic assemblage and a given environmental-variable category was significantly different among environmental-variable categories and regions.

## Multi-Regional Comparisons of Biotic Trends and Drivers of Trends

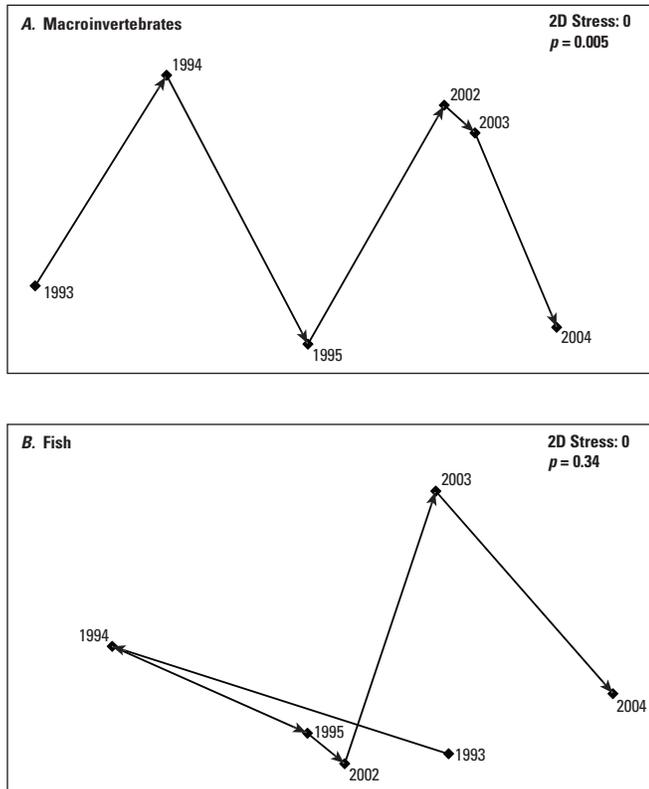
The percentage of sites identified as having significant temporal trends in biotic assemblages as a function of biotic assemblage, region, or land use are presented in the following sections. The percentage of sites within each region that were identified as having both significant temporal trends in biota and significant correlations among the biota and environmental variables also are discussed. These results provide a context for making generalizations about temporal change in biotic assemblages and environmental drivers of that change across broad geographic regions and place ecosystem trends in a national context.

### Trends in Biotic Assemblages

The multivariate approach differentiated between sites with and without significant trends in biotic assemblages. For example, NMDS seriation plots for the macroinvertebrate and fish assemblages from the Buffalo River near Boxley, Arkansas (fig. 2A and B, respectively), provide a contrast between an assemblage with a significant temporal trend (macroinvertebrates) and an assemblage identified as not having a significant temporal trend (fish). In the macroinvertebrate NMDS plot (fig. 2A), the points representing the biotic assemblage for a given year changed position in multivariate space in a unidirectional manner (from left to right in this plot), and the assemblage had a significant change over time ( $p = 0.005$ ). In the fish NMDS plot (fig. 2B), the points indicating the earlier sampling times folded back upon themselves, indicating little directional change in the fish assemblage from 1993 to 2004. Subsequently, a significant temporal trend in the fish assemblage was not identified ( $p = 0.34$ ).

### Trends as a Function of Biotic Assemblage

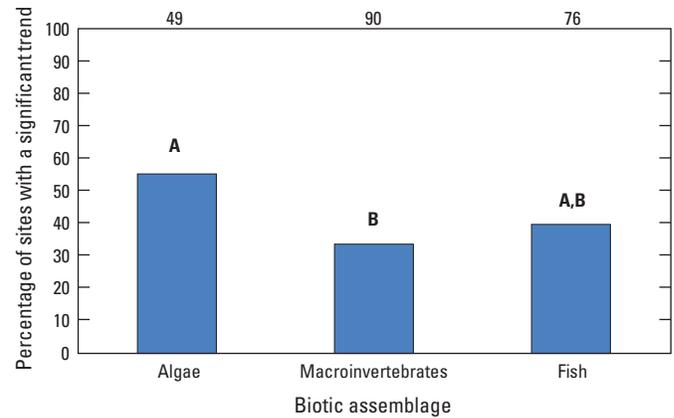
With data from all regions combined, significant temporal trends in algal assemblages were identified at 27 of the 49 sites (55 percent) at which temporal trends in algae were investigated (table 1; fig. 3). A significantly smaller percentage of sites had significant trends in the macroinvertebrate assemblages (30 of 90 sites, 33 percent), and an intermediate percentage of sites (30 of 76, 39 percent) had significant



**Figure 2.** Non-metric multidimensional scaling (NMDS) seriation plots for the Buffalo River near Boxley, Arkansas, for *A*, macroinvertebrate assemblage and *B*, fish assemblage. Points that plot closer together represent biotic assemblages that are more similar to one another, whereas those that plot further apart are more dissimilar. The macroinvertebrate assemblage changed in such a way that in each progressive sampling year the assemblage was more different than any of the previous years, and a significant temporal trend in the macroinvertebrate assemblage was identified. A significant temporal trend in the fish assemblage was not found (that is, the trajectory of the assemblage folded back upon itself).

temporal trends in fish assemblages. The percentage of sites identified as having trends in fish assemblages, however, was not significantly different from the percentage of sites with temporal trends in algae or macroinvertebrate assemblages.

The finding that temporal trends are more frequently identified in algal assemblages as compared to macroinvertebrate or fish assemblages may indicate that algae are, in general, more sensitive to and (or) respond more quickly to environmental change than macroinvertebrates or fish. This idea is further supported by the findings of previous studies (McCormick and Cairns, 1994; Barbour and others, 1999; Coles and others, 2009). The difference in the sensitivity of response of different biotic assemblages to environmental change may have implications for the design of continued/future monitoring programs. For example, if the goal of a monitoring

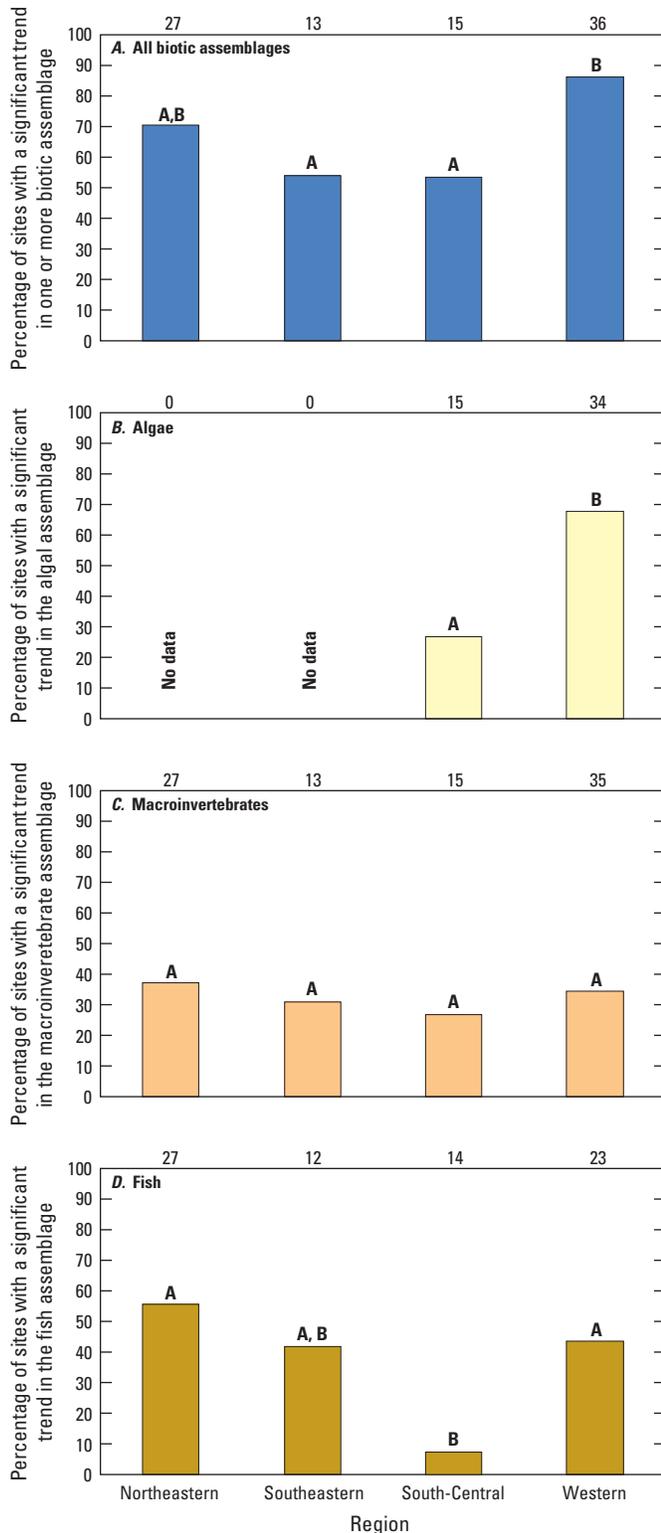


**Figure 3.** Percentage of sites where significant temporal trends were identified for each biotic assemblage when data from all regions were combined. The numbers of samples (*n*) are shown at the top of the figure. Letters indicate significant differences among biotic assemblages. For example, A is significantly different than B, but neither A nor B are significantly different from AB.

program were to identify short-term responses to environmental change, it may be beneficial to put greater resources into monitoring algal assemblages, whereas programs interested in longer term responses may want to place more resources into monitoring macroinvertebrate or fish assemblages. Regardless of the monitoring program objectives, understanding the relative sensitivity of various biotic assemblages over multiple time frames (after 5, 10, and 20 years of monitoring) will aid in identification of the time scales at which different stressors affect biota.

## Trends as a Function of Region

Regional differences were identified in the percentage of sites with significant temporal trends in one or more biotic assemblages, and for each biotic assemblage individually. Significant temporal trends in one or more biotic assemblages were identified at 65 of the 91 (71 percent) assessment sites (table 1). Significant temporal trends were identified in one or more biotic assemblages at 19 of 27 (70 percent) sites in the northeastern/north-central region, 7 of 13 (54 percent) sites in the southeastern region, 8 of 15 (53 percent) sites in the south-central region, and 31 of 36 (86 percent) sites in the western region (fig. 4*A*). The western region had a significantly greater percentage of sites with significant temporal trends in one or more biotic assemblages as compared with the southeastern or south-central regions. In the western region, 23 of 34 (68 percent) sites had significant temporal trends in algal assemblages, which was significantly more than the 4 of 15 (27 percent) sites with significant temporal trends in algal assemblage in the south-central U.S. (fig. 4*B*). Ten of 27 (37 percent), 4 of 13 (31 percent), 4 of 15 (27 percent), and



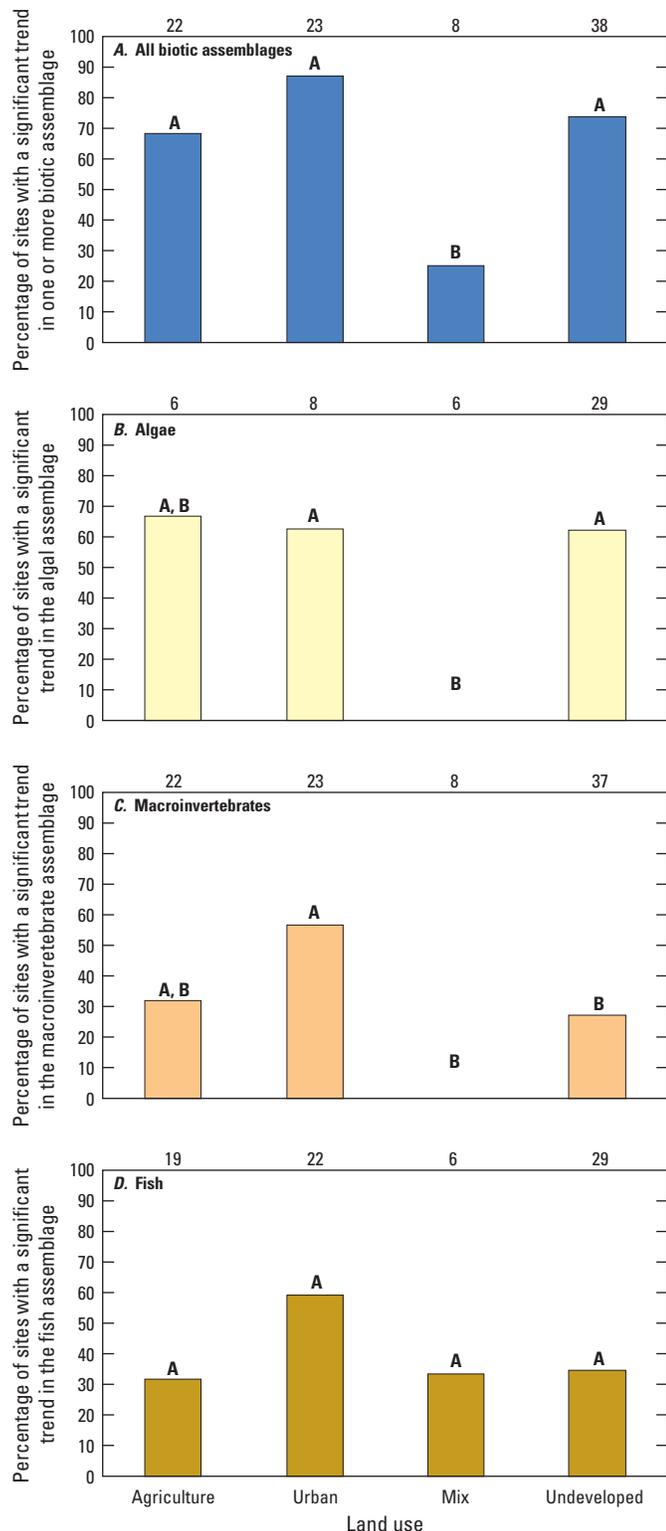
**Figure 4.** Percentage of sites in each region where significant temporal trends were identified for *A*, one or more of the biotic assemblages, *B*, algal assemblages, *C*, macroinvertebrate assemblages, and *D*, fish assemblages. The numbers of samples (*n*) are shown at the top of each figure. Letters indicate significant differences among regions.

12 of 35 (34 percent) sites were identified as having significant temporal trends in macroinvertebrate assemblages in the northeastern/north-central, southeastern, south-central, and western regions, respectively. However, no significant differences were identified among regions in the percentage of sites with trends in macroinvertebrate assemblages (fig. 4C). There was no significant difference in the percentage of sites with significant temporal trends in fish assemblages among the northeastern/north-central (15 of 27, 56 percent), southeastern (5 of 12, 42 percent), or western (10 of 23, 43 percent) regions (fig. 4D). However, only 1 of 14 (7 percent) sites in the south-central U.S. had significant temporal trends in the fish assemblage, which is significantly fewer than in the northeastern/north-central or western regions.

In general, the northeastern/north-central region and to a greater extent the western region was identified as having proportionally more sites with significant temporal trends in biotic assemblages than in the southeastern and south-central regions (fig. 4A–D). That is, significant temporal changes in biotic assemblages were more common in the northeastern/north-central and western regions. To the best of our knowledge, the finding that there were regional differences in the percentage of sites with significant temporal trends for all biotic assemblages combined, as well as individually for the algal and fish assemblages, has not been reported. Interestingly, the finding that there were no significant inter-regional differences in macroinvertebrate trends (fig. 4C) was surprising given the significant differences found for algae (fig. 4B) and fish (fig. 4D). This result may be, at least in part, an artifact of the possibility that greater uncertainty exists in defining the “true” algal and fish assemblages because of smaller sample sizes (49 and 76, respectively, as compared with 90 macroinvertebrate samples). Taken together, the results generated from this comparison provide ecological information at a spatial scale that is relevant to national monitoring programs such as NAWQA.

## Trends as a Function of Land Use

Land use was identified as an important determinant of the percentage of sites with significant temporal trends in one or more biotic assemblages and for each biotic assemblage individually. Significant temporal trends were found in one or more biotic assemblages at 15 of 22 (68 percent) of the agricultural sites, 20 of 23 (87 percent) of the urban sites, 2 of 8 (25 percent) of the mixed land-use sites, and 28 of 38 (74 percent) of the undeveloped sites (fig. 5A). The percentage of sites with significant temporal trends in one or more biotic assemblages was significantly greater at agricultural, urban, and undeveloped sites than at mixed land-use sites. However, the few sites available for analysis in the mixed category (*n* = 8) relative to the other land-use categories, coupled with the fact that the mixed land-use designation was applied to sites in only two of the four regions, may be driving that finding. The percentage of sites with significant temporal trends in algal assemblages differed little among agricultural (4 of 6,



**Figure 5.** Percentage of sites in each land-use category where significant temporal trends were identified for *A*, all biotic assemblages combined, *B*, algal assemblages, *C*, macroinvertebrate assemblages, and *D*, fish assemblages. The numbers of samples (*n*) are shown at the top of each figure. Letters indicate significant differences among land-use categories.

67 percent), urban (5 of 8, 63 percent), and undeveloped (18 of 29, 62 percent) sites (fig. 5*B*). There were no significant trends in algal assemblages at the mixed land-use sites (0 of 6).

A statistical comparison of trends in algae among land-use categories showed mixed land-use sites to be significantly different from urban and undeveloped sites, but not agricultural sites (owing to the small sample sizes ( $n = 6$ ) for both the agricultural and mixed land-use sites). Significant temporal trends in macroinvertebrate assemblages were identified at 7 of 22 (32 percent) agricultural sites, 13 of 23 (57 percent) urban sites, 0 of 8 mixed land-use sites, and 10 of 37 (27 percent) undeveloped sites (fig. 5*C*). The percentage of urban sites with temporal trends in macroinvertebrate assemblages was significantly greater than the percentage of mixed land use and undeveloped sites with temporal trends. No significant differences in the percentage of sites with temporal trends in fish assemblages were found among land-use categories (fig. 5*D*). Fish assemblages showed trends at 6 of 19 (32 percent) agricultural sites, 13 of 22 (59 percent) urban sites, 2 of 6 (33 percent) mixed land-use sites, and 10 of 29 (34 percent) undeveloped sites.

Observed differences in the percentage of sites with significant temporal trends among land-use categories (fig. 5*A*) provides some insight into the potential role of land use as a determinant of change in assemblage composition. Sixty eight and 87 percent of agricultural and urban sites, respectively, had temporal trends in one or more biotic assemblages, indicating that some physical or chemical characteristics of these anthropogenically impacted systems may have changed during the course of the study. Numerous other studies have identified agricultural and urban land uses as impacting biotic assemblages (Lenat and Crawford, 1994; Paul and Meyer, 2001; Roy and others, 2003; Brasher and others, 2004; Coles and others, 2004; Cuffney and others, 2005; Meyer and others, 2005; Kennen and others, 2005; Wang and others, 2008; Cuffney and others, 2010; and many others).

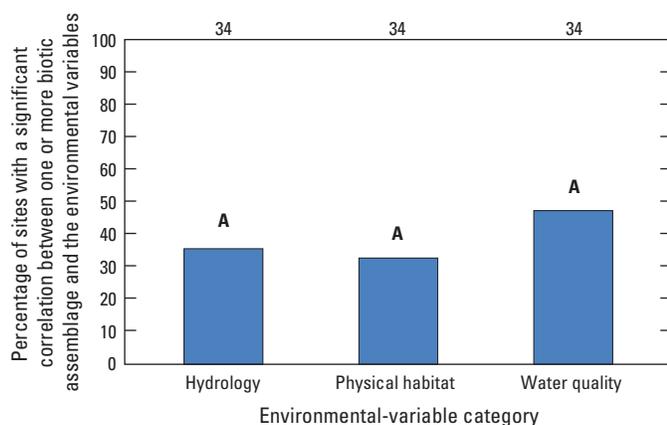
For all biotic assemblages combined, 74 percent of undeveloped sites, where direct anthropogenic impacts are limited, showed significant temporal trends in one or more biotic assemblages (fig. 5*A*). This finding may indicate that climate-related processes are driving these trends. This finding (based on all four regions combined) supports similar findings to that of the northeastern/north-central and south-central region reports, that the use of undeveloped sites as an ecological baseline for monitoring programs requires careful evaluation (Kennen and others, 2012; Miller and others, 2012). It also is interesting to note that the percentage of mixed land-use sites with temporal trends in biotic assemblages was lower than for all other land-use categories. Watersheds categorized as having mixed land use are, by definition, draining a large area consisting of multiple land uses. Therefore, it is conceivable that this diversity in land uses could result in more heterogeneous abiotic environmental conditions, thereby dampening temporal trends in biotic assemblages. These findings may require additional evaluation because the number of mixed land-use sites in the analysis was small ( $n = 8$ ) relative to the

number of urban ( $n = 23$ ), agriculture ( $n = 22$ ), and undeveloped sites ( $n = 38$ ). The observed differences in response to land use among biotic assemblages provide further support for the concept that different stressors, including land use, act on different time scales for different biotic assemblages, which indicates the need for continued support of long-term monitoring programs for more effective identification of biotic assemblage-specific response times to various stressors.

## Environmental Drivers of Trends

The general categories of environmental drivers—hydrology, physical habitat, and water quality—correlated with biotic assemblages are discussed in this section. Correlations between biotic assemblages and environmental-variable categories were investigated only at sites where significant temporal trends in biotic assemblages were identified (table 1). However, correlations between biotic assemblages and general environmental-variable categories for the western geographic region were not included in this comparative analysis because final results are still pending. Biotic assemblages at 26 of the 34 (76 percent) sites in the northeastern/north-central, southeastern, and south-central regions were significantly correlated with one or more environmental-variable categories (table 2). For all three regions combined, one or more of the biotic assemblages were found to be significantly correlated with hydrology at 12 sites (35 percent), physical habitat at 11 sites (32 percent), and water quality at 16 sites (47 percent) (fig. 6). However, there were no significant differences among the percentages of correlations with biotic assemblages for the three environmental-variable categories (fig. 6).

Comparison of the percentage of sites identified as having significant correlations with hydrologic, physical habitat, and (or) water-quality variables provides insight into the



**Figure 6.** Percentage of sites with significant correlations between one or more biotic assemblages and environmental-variable category. The numbers of samples ( $n$ ) are shown at the top of the figure. Letters indicate that there were no significant differences among environmental-variable categories.

relative importance of these environmental drivers both among and within regions. Figure 7 compares among-region differences for each environmental-variable category (for example, differences between the percentage of sites in the northeastern/north-central region with correlations between biotic assemblages and hydrology and the percentage of sites in the southeastern region with correlations between biotic assemblages and hydrology). Figure 7 also compares within-region differences among environmental-variable categories (for example, differences between the percentage of sites in the south-central region with correlations between biotic assemblages and hydrology and the percentage of sites in the south-central region with correlations between biotic assemblages and physical habitat). Among regions, when considering all biotic assemblages combined, there were no significant differences in the percentage of sites with correlations between environmental-variable categories and one or more biotic assemblages (fig. 7A). Within regions, no differences in the percentage of sites with significant assemblage and environmental correlations were identified in the northeastern/north-central or south-central regions (fig. 7A). Within the southeastern region, a significantly greater percentage of sites had biotic assemblages that were correlated with water quality (6 of 7, 86 percent) than those correlated with hydrology (1 of 7, 14 percent). However, physical habitat (4 of 7, 57 percent) was not significantly different from either hydrology or water quality (fig. 7A).

The south-central region was the only region for which temporal trends in algal assemblages and correlations between algal assemblages and environmental-variable categories were evaluated owing to an absence of algal information in the other two regions (fig. 7B). Four sites were identified as having significant temporal trends in algal assemblages within the south-central region (table 2). Two of the four sites (50 percent) were found to be correlated with each of the environmental variable-categories (fig. 7B). Interestingly, the two sites correlated with physical habitat variables were undeveloped sites, whereas the two sites correlated with hydrology and water quality were developed (agriculture and urban) sites (table 2).

The southeastern region had a greater percentage (3 of 4, 75 percent) of sites with significant correlations between the macroinvertebrate assemblage and water quality than the northeastern/north-central region (1 of 10, 10 percent, fig. 7C). No significant differences were observed among regions in the percentage of sites with correlations between macroinvertebrate assemblages and hydrology, or between macroinvertebrate assemblages and physical habitat (fig. 7C). Within regions, there were no significant differences in the percentage of sites with correlations between macroinvertebrate assemblages and the different environmental-variable categories.

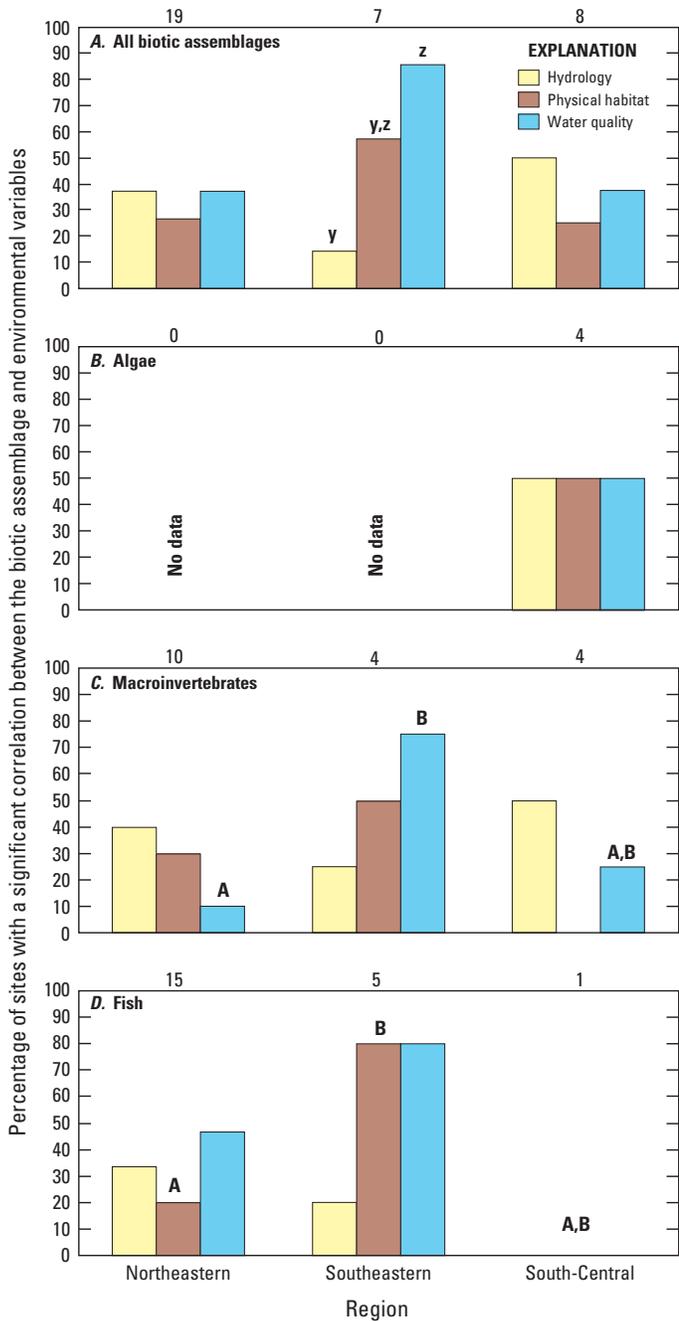
Physical habitat was the only environmental-variable category correlated with trends in fish assemblage that showed significant differences among regions (fig. 7D). In the southeastern region, 4 of 5 sites (80 percent) had significant correlations between fish assemblages and physical habitat,

**Table 2.** Environmental-variable categories found to be significantly correlated with the biotic assemblage for sites/biotic assemblages that were identified as having significant temporal trends in the northeastern/north-central, southeastern, and south-central regions.

[AG, Agricultural land use; URB, Urban land use; UNDEV, Undeveloped land use, MIX, Mixed land use; NA, sites for which temporal trends in biotic assemblages were not analyzed; --, sites for which a significant temporal trend was not identified; NS, sites for which a significant temporal trend was identified but no environmental variables/metrics were found to be significantly related to the biotic assemblage; **HYD**, Hydrologic variables/metrics; **HAB**, Physical habitat variables/metrics; **WQ**, Water-quality variables/metrics]

Station code	Land-use category	Algae	Macroinvertebrates	Fish
Northeastern/North-Central U.S.				
CANA	AG	NA	--	<b>HYD</b>
DUCK	AG	NA	<b>NS</b>	--
MAD	AG	NA	--	<b>HYD, HAB</b>
MUD	AG	NA	--	<b>WQ</b>
SFIOWA	AG	NA	<b>HAB</b>	--
SUGAR	AG	NA	--	<b>WQ</b>
BOUND	URB	NA	--	<b>NS</b>
CLINT	URB	NA	<b>HYD, HAB, WQ</b>	<b>HYD, HAB, WQ</b>
HOLES	URB	NA	<b>NS</b>	<b>WQ</b>
LISHA	URB	NA	<b>HYD, HAB</b>	--
LBUCK	URB	NA	<b>NS</b>	--
LNESH	URB	NA	<b>HYD</b>	<b>WQ</b>
NORW	URB	NA	<b>NS</b>	<b>NS</b>
SALT	URB	NA	--	<b>HYD, WQ</b>
SHING	URB	NA	--	<b>NS</b>
FRENCH	UNDEV	NA	--	<b>HAB</b>
GREEN	UNDEV	NA	<b>NS</b>	<b>WQ</b>
RAISIN	UNDEV	NA	--	<b>NS</b>
WAITES	UNDEV	NA	<b>HYD</b>	<b>HYD</b>
Southeastern U.S.				
AG-TN1	AG	NA	<b>WQ</b>	--
AG-NEUS	AG	NA	<b>WQ</b>	--
AG-TN2	AG	NA	--	<b>WQ</b>
URB-MOBL	URB	NA	<b>HYD, HAB</b>	<b>HYD, HAB</b>
URB-ACF	URB	NA	<b>HAB, WQ</b>	<b>HAB, WQ</b>
URB-NEUS	URB	NA	--	<b>HAB, WQ</b>
INT-NEUS	MIX	NA	--	<b>HAB, WQ</b>
South-Central U.S.				
YOCM	AG	<b>HYD, WQ</b>	--	--
SALD	URB	<b>HYD, WQ</b>	--	--
WHITE	URB	--	<b>HYD</b>	--
BUFF <sup>1</sup>	UNDEV	--	<b>NS</b>	--
CLEAR	UNDEV	--	<b>HYD, WQ</b>	--
FRIO	UNDEV	<b>HAB</b>	--	--
NSYLM	UNDEV	<b>HAB</b>	<b>NS</b>	--
YAZ	MIX	--	--	<b>NS</b>

<sup>1</sup>Correlations between the macroinvertebrate assemblage and physical habitat variables at BUFF were not tested because fewer than 5 years of physical habitat data were available (see Miller and others, 2012).



**Figure 7.** Percentage of sites in each region with significant correlations between the biotic assemblage and environmental-variable categories for *A*, all biotic assemblages combined, *B*, algal assemblages, *C*, macroinvertebrate assemblages, and *D*, fish assemblages. The numbers of samples (*n*) are shown at the top of each figure. Capital letters (A, B) indicate significant differences among regions for a given environmental-variable category. Lowercase letters (y, z) indicate significant differences among environmental-variable categories within a given region. Note that letters indicating significance are shown only for the among-region or among-environmental-variable category comparisons for which significant differences were identified.

which was significantly greater than in the northeastern/north-central region (3 of 15 sites, 20 percent). Similar to what was observed among regions for the combined biotic assemblages (fig. 7A) and the macroinvertebrate assemblage (fig. 7C), water quality—while not statistically significant—was more commonly correlated with fish assemblages in the southeastern region than either the northeastern/north-central or south-central regions. In contrast, hydrology—while not statistically significant—was more commonly an environmental driver in the northeastern/north-central region than it was in the southeastern or south-central regions. Within regions there were no significant differences in the percentage of sites with correlations between fish assemblages and environmental-variable categories.

## Study Limitations and Future Directions

The standardized field and laboratory methods used as part of the NAWQA Program enable analyses at broad spatial and geographic scales, such as those presented herein. However, limitations may arise when combining large datasets that have been analyzed by scientists in different regions. This section reviews some of the data compilation and analysis limitations faced when synthesizing and interpreting data collected over multiple decades at the continental scale. Further, suggestions for future work that could build upon the results presented herein are discussed.

The collection of a large number of biotic samples for quantification of biotic assemblages is often limited by the high cost of sample processing. Given this constraint, it is not surprising that a relatively small number of samples was available for analyses in this study. Of the published papers upon which our analyses were based, not all reports included data for specific biotic assemblages. This lack of data in certain geographic regions limited our ability to identify differences in environmental drivers of temporal trends among biotic assemblages in a multi-regional context. For example, the lack of data on algal assemblages in the northeastern/north-central and southeastern regions greatly limited the broader regional comparison for that taxonomic group. However, analysis of temporal trends in algal assemblages and correlations of those assemblages with environmental variables is currently (2013) underway in the northeastern/north-central, southeastern, and western regions and should be available soon for a more comprehensive comparative analysis. Once completed, it will be possible to derive a more complete understanding of the differences in important environmental drivers of temporal change in algal assemblages among regions.

Among-region and among-land-use comparisons of the percentage of sites with significant trends in biotic assemblages also are limited by data availability. Specifically, the southeastern and south-central regions as well as the mixed land-use category have fewer sites than other regions or land-use categories, respectively. Collection of additional data

and consistent categorization of land-use categories among regions, would provide the opportunity to further define among-region or land-use differences in the percentage of sites with significant trends in biotic assemblages. These limitations highlight the importance of maintaining a spatially complex and numerically robust monitoring program. This finding is particularly pertinent as programs like NAWQA transition into cycles of reduced funding and a greatly restricted spatial sampling framework. However, as the NAWQA Program and others like it (for example, The U.S. Environmental Protection Agency National Aquatic Resource Survey (NARS) Program) continue to collect biological samples across broad spatial scales, the extent of valuable trend datasets will be expanded and a better understanding of ecosystem response to environmental change will be developed. Further, continued collection of ecological data will provide programs such as NAWQA and NARS with the opportunity to examine a number of topics of management concern including, for example, species-specific temporal change and temporal change in biodiversity.

The approach of comparing and contrasting the percentage of correlations between biotic assemblages and general categories of environmental variables (hydrology, physical habitat, and water quality) was adopted because the suite of environmental variables investigated varied among regions. While this approach does provide some insight into differences in environmental drivers among regions, a comprehensive analysis that begins with a consistent set of environmental variables among regions would provide a broader understanding of how water quality and watershed conditions are changing across the country. For example, the greater percentage of sites in the southeastern region with significant correlations between water quality and macroinvertebrate and fish assemblages, as compared to the northeastern/north-central and south-central regions, may be owing to the fact that pesticides were included in the water-quality category in the southeastern region but not in the other regions. Specifically, a pesticide toxicity index (PTI; Munn and Gilliom, 2001; Munn and others, 2006) was negatively correlated with macroinvertebrate-assemblage metrics indicative of “good” water-quality conditions (for example, percent Ephemeroptera, Plecoptera, and Trichoptera) at all three of the sites with significant correlations between macroinvertebrate assemblages and water quality in the southeastern region (Daniel Calhoun, USGS, unpub. data, October 5, 2011). While this points to the PTI as a potentially important water-quality variable with respect to understanding water-quality drivers of trends in biotic assemblages, it is not possible to thoroughly evaluate the importance of the correlation between biotic assemblages and the PTI among regions because the PTI was only included as a potential driver of change in the report from the southeastern region. It is suggested that future ecological-trend analyses include the calculation of a common set of environmental variables for all the study sites. This would undoubtedly provide an opportunity for a more scientifically rigorous approach to identifying

and understanding how water-quality and watershed conditions are changing at the multi-regional scale and, thereby, providing a more robust basis of comparison.

## Summary and Conclusions

Temporal trends in biotic assemblages (algae, macroinvertebrates, and fish) from 91 streams and rivers sampled as part of the U.S. Geological Survey National Water-Quality Assessment (NAWQA) Program, which were reported in four region-specific reports and collectively encompass the continental U.S., were summarized and synthesized. The percentages of sites with significant temporal trends in biotic assemblages were compared among biotic assemblages (algae, macroinvertebrates, and fish), geographic regions, and land-use categories. Correlations between biotic assemblages and three general environmental-variable categories (hydrology, physical habitat, and water quality) were compared among and within three geographic regions (northeastern/north central, southeastern, and south central). Finally, limitations to the study approach and suggestions for future work were discussed, which could avoid such limitations and (or) build upon the present study.

Sites with significant temporal trends in algae, macroinvertebrate, and (or) fish assemblages were successfully identified using a multivariate statistical approach that allows for a quantitative assessment of temporal change for the entire ecological community. Synthesis of the region-specific reports (Kennen and others, 2012; Miller and others, 2012; Daniel Calhoun, USGS, unpub. data, October 5, 2011; and Wiele and others, 2012) indicates that significant temporal trends in algal assemblages were identified at a greater percentage of sites (55 percent) than macroinvertebrate (33 percent) or fish (39 percent) assemblages. This finding may indicate that algae respond more quickly to environmental change than either macroinvertebrates or fish. Such findings could be used to more accurately identify the amount of time or number of samples required to detect temporal trends in different biotic assemblages and also may be used to better inform the allocation of resources for a more effective and efficient design of future monitoring or synoptic-sampling efforts.

In general, a greater percentage of sites with significant temporal trends were identified in the northeastern/north-central (70 percent) and western (86 percent) regions than in the southeastern (54 percent) or south-central (53 percent) regions. The finding that there are among-region differences is a novel result, and while the results presented herein are limited by data availability and different methodological approaches, as previously described, this finding does provide a foundation from which future multi-regional analyses can better assess how environmental and anthropogenic conditions are affecting aquatic ecosystems.

Results also indicate that there was a greater percent-

age of sites with temporal trends in agricultural (68 percent), urban (87 percent), and undeveloped (74 percent) land uses than of sites draining mixed (25 percent) land uses. A greater percentage of temporal change at the agricultural and urban sites, which generally are exposed to a high degree of human alteration of the landscape, raises the possibility that there may have been changes in the abiotic environment at these sites that resulted in temporal change in biotic assemblages. The large percentage of sites draining basins with undeveloped land use with significant temporal trends in biotic assemblages may indicate that climate-related impacts are influencing the more sensitive taxa, which tend to be more abundant at undeveloped sites. This finding, which is based on data from all four regions combined, is consistent with those of previous region-specific reports. Results of this comparative analysis also may indicate that the few temporal trends identified in mixed land-use basins may be a result of dampening of assemblage response owing to the heterogeneous abiotic environmental conditions commonly found in mixed land-use basins.

Results have identified differences in hydrology, physical habitat, and water quality as potential drivers of trends in biotic assemblages among regions. Specifically, results appear to indicate that physical habitat and water quality may be more important drivers of temporal trends in biotic assemblages in the southeastern region than in the northeastern/north-central or south-central regions. Macroinvertebrate and fish assemblages were more commonly correlated with water quality and physical habitat variables, respectively, in the southeastern region than in the northeastern/north-central or south-central regions. These results indicate that multiple interacting stressors likely are involved in determining trends in biotic assemblages. Therefore, additional data compilation and analysis is warranted to support any conclusions regarding potential environmental drivers of change in biotic assemblages among regions. Taken together, the region-specific studies and the multi-regional synthesis presented herein make evident the potential importance of site- and region-specific management approaches aimed at mitigating anthropogenic changes in the environment to manage and protect ecological resources.

These results highlight the importance of continued long-term monitoring of biotic assemblages similar to what was previously accomplished as part of the NAWQA Program Surface Water Status and Trends network. Datasets that include samples collected over a broader time scale provide greater certainty for understanding long-term temporal change. The insights into potential environmental drivers of temporal trends provided here, however, could be strengthened by future analyses that use a consistent analytical methodology and common subsets of environmental variables. Ultimately, such studies would provide a more detailed understanding of how water quality and watershed conditions are changing at the multi-regional scale.

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**Table 1.** General site characteristics for the 91 U.S. Geological Survey (USGS) National Water-Quality Assessment (NAWQA) Program long-term surface-water status and trend sites.

[USGS, U.S. Geological Survey; km<sup>2</sup>, square kilometer; AG, Agricultural land use; URB, Urban land use; UNDEV, Undeveloped land use; MIX, Mixed land use; YES, sites for which temporal trends in biotic assemblages were analyzed; No, sites for which temporal trends in biotic assemblages were not analyzed; **Bold** type indicates sites and biotic assemblages with significant temporal trends; \*,  $p < 0.05$ ; \*\*,  $p < 0.01$ ]

USGS station identification number	Station name	Station code	Land-use category	Drainage area (km <sup>2</sup> )	Algae analyzed <sup>1</sup>	Macroinvertebrates analyzed	Fish analyzed
Northeastern/North-Central United States							
04186500	Auglaize River NR Fort Jennings, OH	AUG	AG	860	No	YES	YES
01349150	Canajoharie Creek NR Canajoharie, NY	CANA	AG	155	No	YES	<b>YES**</b>
04072050	Duck Creek at Seminary Road NR Oneida, WI	DUCK	AG	280	No	<b>YES*</b>	YES
05320270	Little Cobb River NR Beauford, MN	LCOBB	AG	337	No	YES	YES
0395650083504400	Mad River NR highway 41 NR Springfield, OH	MAD	AG	803	No	YES	<b>YES*</b>
01621050	Muddy Creek at Mount Clinton, VA	MUD	AG	37	No	YES	<b>YES**</b>
05572000	Sangamon River at Monticello, IL	SANG	AG	1,425	No	YES	YES
05451210	South Fork Iowa River Northeast of New Providence, IA	SFIOWA	AG	73	No	<b>YES*</b>	YES
0394340085524601	Sugar Creek at County Road 400 South at New Palestine, IN	SUGAR	AG	243	No	YES	<b>YES**</b>
05420680	Wapsipicon River NR Tripoli, IA	WAPS	AG	896	No	YES	YES
01102500	Aberjona River at Winchester, MA	ABERJ	URB	64	No	YES	YES
01654000	Accotink Creek NR Annandale, VA	ACCO	URB	62	No	YES	YES
01403900	Bound Brook at Middlesex, NJ	BOUND	URB	125	No	YES	<b>YES*</b>
04161820	Clinton River at Sterling Heights, MI	CLINT	URB	800	No	<b>YES**</b>	<b>YES**</b>
0393944084120700	Holes Creek in Huffman Park at Kettering, OH	HOLES	URB	48	No	<b>YES*</b>	<b>YES*</b>
01356190	Lisha Kill Northwest of Niskayuna, NY	LISHA	URB	40	No	<b>YES**</b>	YES
03353637	Little Buck Creek NR Indianapolis, IN	LBUCK	URB	44	No	<b>YES**</b>	YES
01464907	Little Neshaminy Creek at Valley Road nr Neshaminy, PA	LNESH	URB	69	No	<b>YES*</b>	<b>YES**</b>
01209700	Norwalk River at South Wilton, CT	NORW	URB	78	No	<b>YES**</b>	<b>YES*</b>
05531500	Salt Creek at Western Springs, IL	SALT	URB	298	No	YES	<b>YES**</b>
05288705	Shingle Creek at Queen Ave, Minneapolis, MN	SHING	URB	580	No	YES	<b>YES**</b>
01472157	French Creek NR Phoenixville, PA	FRENCH	UNDEV	153	No	YES	<b>YES**</b>
01170095	Green River at Stewartville, MA	GREEN	UNDEV	107	No	<b>YES**</b>	<b>YES**</b>
04063700	Popple River NR Fence, WI	POP	UNDEV	360	No	YES	YES
04175600	River Raisin NR Manchester, MI	RAISIN	UNDEV	342	No	YES	<b>YES**</b>
01095220	Stillwater River NR Sterling, MA	STILL	UNDEV	82	No	YES	YES
01610400	Waites Run NR Wardensville, WV	WAITES	UNDEV	33	No	<b>YES*</b>	<b>YES*</b>

**Table 1.** General site characteristics for the 91 U.S. Geological Survey (USGS) National Water-Quality Assessment (NAWQA) Program long-term surface-water status and trend sites.—Continued

[USGS, U.S. Geological Survey; km<sup>2</sup>, square kilometer; AG, Agricultural land use; URB, Urban land use; UNDEV, Undeveloped land use; MIX, Mixed land use; YES, sites for which temporal trends in biotic assemblages were analyzed; No, sites for which temporal trends in biotic assemblages were not analyzed; **Bold** type indicates sites and biotic assemblages with significant temporal trends; \*,  $p < 0.05$ ; \*\*,  $p < 0.01$ ]

USGS station identification number	Station name	Station code	Land-use category	Drainage area (km <sup>2</sup> )	Algae analyzed <sup>1</sup>	Macroinvertebrates analyzed	Fish analyzed
Southeastern United States							
03466208	Big Limestone Creek NR Limestone, TN	AG-TN1	AG	205	No	<b>YES*</b>	YES
02091500	Contentnea Creek at Hookerton, NC	AG-NEUS	AG	1,898	No	<b>YES**</b>	YES
02174250	Cow Castle Creek NR Bowman, SC	AG-SANT	AG	61	No	YES	YES
0357479650	Hester Creek at Buddy Williamson Rd NR Plevna, AL	AG-TN2	AG	85	No	YES	<b>YES*</b>
02350080	Lime Creek NR Cobb, GA	AG-ACF	AG	159	No	YES	YES
0242354750	Cahaba Valley Creek at Cross Cr Rd at Pelham, AL	URB-MOBL	URB	66	No	<b>YES*</b>	<b>YES*</b>
02169570	Gills Creek at Columbia, SC	URB-SANT	URB	154	No	YES	YES
02335870	Sope Creek NR Marietta, GA	URB-ACF	URB	76	No	<b>YES**</b>	<b>YES*</b>
02087580	Swift Creek NR Apex, NC	URB-NEUS	URB	54	No	YES	<b>YES**</b>
02338523	Hillabatchee Creek at Thaxton Rd, NR Frankling, GA	REF-ACF	UNDEV	44	No	YES	YES
02172300	McTier Creek (RD 209) NR Monetta, SC	REF-SANT	UNDEV	40	No	YES	YES
02089500	Neuse River at Kingston, NC	INT-NEUS	MIX	6,972	No	YES	<b>YES*</b>
02318500	Withlacoochee River at US 84, NR Quitman, GA	INT-SUWA	MIX	3,833	No	YES	No
South-Central United States							
07288650	Bogue Phalia BLW Leland, MS	BOGPH	AG	1,301	YES	YES	YES
07053250	Yocum Creek NR Oak Grove, AR	YOCM	AG	134	<b>YES**</b>	YES	YES
08178800	Salado Creek at Loop 13 San Antonio, TX	SALD	URB	506	<b>YES*</b>	YES	YES
08057200	White Rock Creek at Greenville Ave Dallas, TX	WHITE	URB	173	YES	<b>YES*</b>	YES
07055646	Buffalo River NR Boxley, AR	BUFF	UNDEV	153	YES	<b>YES**</b>	YES
08051500	Clear Creek NR Sanger, TX	CLEAR	UNDEV	763	YES	<b>YES*</b>	YES
08195000	Frio River at Concan, TX	FRIO	UNDEV	1,028	<b>YES*</b>	YES	YES
07060710	North Sylamore Creek NR Fifty Six, AR	NSYLM	UNDEV	150	<b>YES**</b>	<b>YES**</b>	YES
08014500	Whiskey Chitto Creek NR Oberlin, LA	WHISK	UNDEV	1,305	YES	YES	YES
08064100	Chambers Creek NR Rice, TX	CHAM	MIX	2,136	YES	YES	No
08012150	Mermentau River at Mermentau, LA	MERM	MIX	3,576	YES	YES	YES
08181800	San Antonio River NR Elmendorf, TX	SANANT	MIX	4,528	YES	YES	YES
08057410	Trinity River BLW Dallas, TX	TRIN	MIX	16,227	YES	YES	YES
07030392	Wolf River at Lagrange, TN	WOLF	MIX	543	YES	YES	YES
07288955	Yazoo River BLW Steele Bayou NR Long Lake, MS	YAZ	MIX	34,850	YES	YES	<b>YES*</b>

**Table 1. General site characteristics for the 91 U.S. Geological Survey (USGS) National Water-Quality Assessment (NAWQA) Program long-term surface-water status and trend sites.—Continued**

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USGS station identification number	Station name	Station code	Land-use category	Drainage area (km <sup>2</sup> )	Algae analyzed <sup>1</sup>	Macroinvertebrates analyzed	Fish analyzed
Western United States							
12464770	Crab Creek at Rocky Ford Road NR Ritzville, WA	CRAB	AG	1,188	<b>YES*</b>	YES	<b>YES**</b>
12505450	Granger Drain at Granger, WA	GRANG	AG	160	No	<b>YES**</b>	No
06800000	Maple Creek NR Nickerson, NE	MAPLE	AG	954	<b>YES**</b>	YES	YES
11274538	Orestimba Creek at River RD NR Crows Landing CA	OREST	AG	28	<b>YES*</b>	<b>YES**</b>	No
14201300	Zollner Creek NR Mt Angel, OR	ZOLLN	AG	39	YES	<b>YES*</b>	No
11447360	Arcade Creek NR Del Paso Heights CA	ARCD	URB	82	<b>YES*</b>	<b>YES**</b>	No
06713500	Cherry Creek at Denver, CO	CHERRY	URB	1,063	<b>YES**</b>	YES	YES
14206950	Fanno Creek at Durham, OR	FANNO	URB	81	<b>YES**</b>	YES	YES
10168000	Little Cottonwood Creek at Jordan River NR SLC, UT	LCOTTN	URB	117	YES	<b>YES*</b>	<b>YES**</b>
11074000	Santa Ana R BLW Prado Dam CA	PRADO	URB	3,727	<b>YES**</b>	<b>YES**</b>	<b>YES**</b>
12128000	Thornton Creek NR Seattle, WA	THORT	URB	29	YES	<b>YES*</b>	<b>YES*</b>
402114105350101	Big Thompson BLW Moraine Park NR Estes Park, CO	BTHMP	UNDEV	103	YES	YES	<b>YES*</b>
09163500	Colorado River NR Colorado-Utah State Line	COLOR	UNDEV	46,274	<b>YES**</b>	<b>YES*</b>	YES
11335000	Cosumnes River ABV Michigan Bar, CA	CONSM	UNDEV	1,389	<b>YES**</b>	YES	No
06775900	Dismal River NR Thedford, NE	DISML	UNDEV	2,503	<b>YES*</b>	YES	YES
10309010	E FK Carson River NR Dresslerville, NV	EFCAR2	UNDEV	970	<b>YES*</b>	YES	No
14205400	East Fork Dairy Creek NR Meacham Corner, OR	EFDAR	UNDEV	88	<b>YES*</b>	<b>YES**</b>	YES
14200400	Little Abiqua Creek NR Scotts Mills, OR	LABIQ	UNDEV	25	YES	<b>YES*</b>	YES
06324970	Little Powder River ABV Dry Creek, NR Weston, WY	LPOWD	UNDEV	3,204	YES	YES	<b>YES*</b>
06753990	Lonetere Creek NR Greeley, CO	LTREE	UNDEV	1,478	<b>YES*</b>	YES	No
11273500	Merced River ABV River Road Bridge NR Newman CA	MERC2	UNDEV	3,621	<b>YES*</b>	YES	YES
12056500	NF Skokomish River BLW Staircase RPDS NR Hoodspport, WA	NFSK	UNDEV	147	<b>YES*</b>	YES	No
06805500	Platte River at Louisville, NE	PLATTE	UNDEV	220,908	YES	No	No
10172200	Red Butte Creek at Fort Douglas, NR SLC, UT	RBUTT	UNDEV	19	YES	YES	No
08364000	Rio Grande at El Paso, TX	RIOGR	UNDEV	77,556	<b>YES**</b>	YES	No
13092747	Rock Creek ABV Hwy30/93 Xing at Twin Falls ID	ROCRK	UNDEV	623	<b>YES**</b>	YES	<b>YES**</b>
08227000	Saguache Creek NR Saguache, CO	SAGUC	UNDEV	1,327	YES	YES	YES
11303500	San Joaquin River NR Vernalis CA	SJOAQ	UNDEV	19,153	No	YES	<b>YES*</b>

**Table 1.** General site characteristics for the 91 U.S. Geological Survey (USGS) National Water-Quality Assessment (NAWQA) Program long-term surface-water status and trend sites.—Continued

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Western United States—Continued							
09471000	San Pedro River at Charleston, AZ	SPEDR	UNDEV	3,257	YES	YES	YES
340843117032501	Santa Ana River ABV Upper PH NR Running Springs CA	STANA1	UNDEV	398	<b>YES**</b>	YES	YES
13010065	Snake River ABV Jackson Lake at Flagg Ranch WY	SNRIV	UNDEV	1,324	<b>YES*</b>	YES	YES
13154500	Snake River at King Hill ID	KNHILL	UNDEV	92,942	<b>YES**</b>	<b>YES**</b>	YES
06754000	South Platte River NR Kersey, CO	SPLATT	UNDEV	25,016	<b>YES*</b>	YES	<b>YES**</b>
10350500	Truckee River at Clark, NV	TRUCK3	UNDEV	4,310	<b>YES*</b>	YES	No
09505800	West Clear Creek NR Camp Verde, AZ	WCLCK	UNDEV	615	<b>YES**</b>	<b>YES*</b>	<b>YES**</b>
06329500	Yellowstone River NR Sidney, MT	YSTON	UNDEV	177,139	YES	YES	No

<sup>1</sup> Algae samples collected in the northeastern/north-central and southeastern regions currently (2013) are being analyzed as part of a separate study.

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