



Contents lists available at [ScienceDirect](#)

## Ecological Indicators

journal homepage: [www.elsevier.com/locate/ecolind](http://www.elsevier.com/locate/ecolind)



# A comparison of algal, macroinvertebrate, and fish assemblage indices for assessing low-level nutrient enrichment in wadeable Ozark streams

B.G. Justus<sup>a,\*</sup>, James C. Petersen<sup>a</sup>, Suzanne R. Femmer<sup>b</sup>, Jerri V. Davis<sup>b</sup>, J.E. Wallace<sup>a</sup>

<sup>a</sup> USGS Arkansas Water Science Center, 401 Hardin Road, Little Rock, AR 72211, United States

<sup>b</sup> USGS Missouri Water Science Center, 1400 Independence Road MS 100, Rolla, MO 65401, United States

### ARTICLE INFO

#### Article history:

Received 23 June 2009

Received in revised form 8 October 2009

Accepted 13 October 2009

#### Keywords:

Nutrient

Algae

Macroinvertebrate

Fish

Metric

Biotic index

Cattle

Poultry

### ABSTRACT

Biotic indices for algae, macroinvertebrates, and fish assemblages can be effective for monitoring stream enrichment, but little is known regarding the value of the three assemblages for detecting perturbation as a consequence of low-level nutrient enrichment. In the summer of 2006, we collected nutrient and biotic samples from 30 wadeable Ozark streams that spanned a nutrient-concentration gradient from reference to moderately enriched conditions. Seventy-three algal metrics, 62 macroinvertebrate metrics, and 60 fish metrics were evaluated for each of the three biotic indices. After a group of candidate metrics had been identified with multivariate analysis, correlation procedures and scatter plots were used to identify the four metrics having strongest relations to a nutrient index calculated from log transformed and normalized total nitrogen and total phosphorus concentrations. The four metrics selected for each of the three biotic indices were: algae—the relative abundance of most tolerant diatoms, the combined relative abundance of three species of *Cymbella*, mesosaprobic algae percent taxa richness, and the relative abundance of diatoms that are obligate nitrogen heterotrophs; macroinvertebrate—the relative abundance of intolerant organisms, Baetidae relative abundance, moderately tolerant taxa richness, and insect biomass; fish—herbivore and detritivore taxa richness, pool species relative abundance, fish catch per unit effort, and black bass (*Micropterus* spp.) relative abundance.

All three biotic indices were negatively correlated to nutrient concentrations but the algal index had a higher correlation ( $\rho = -0.89$ ) than did the macroinvertebrate and fish indices ( $\rho = -0.63$  and  $-0.58$ , respectively). Biotic index scores were lowest and nutrient concentrations were highest for streams with basins having the highest poultry and cattle production. Because of the availability of litter for fertilizer and associated increases in grass and hay production, cattle feeding capacity increases with poultry production. Studies are needed that address the synergistic effect of poultry and cattle production on Ozark streams in high production areas before ecological risks can be adequately addressed.

Published by Elsevier Ltd.

## 1. Introduction

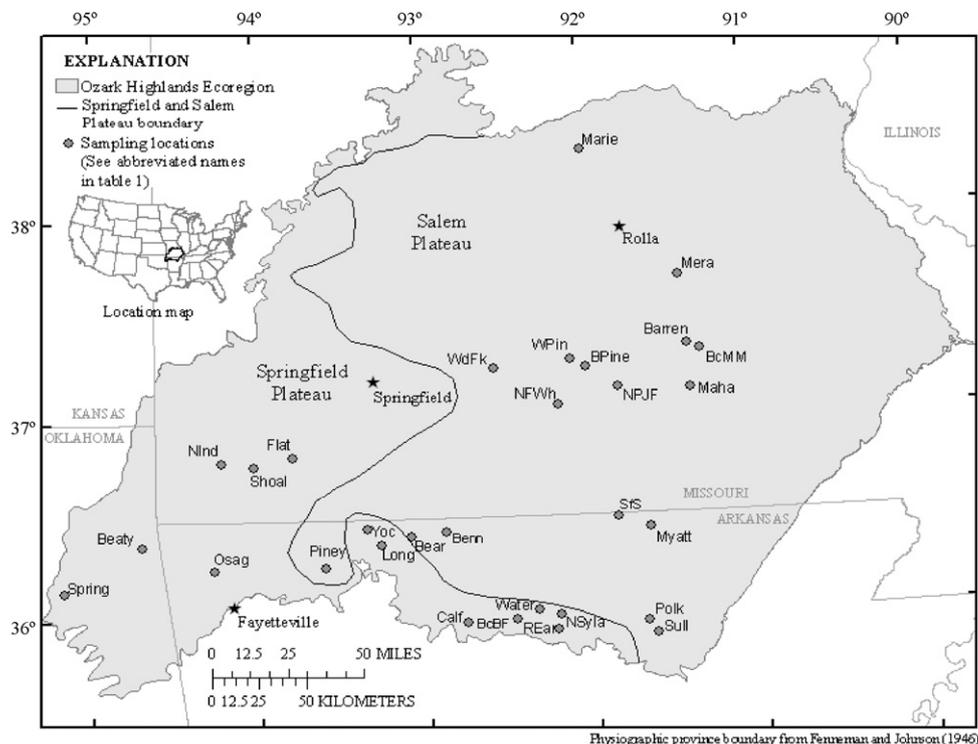
In 2003, the U.S. Geological Survey (USGS) initiated several studies to evaluate the effects of nutrient enrichment on stream ecosystems in agricultural basins (Munn and Hamilton, 2003). These studies were initiated after the U.S. Environmental Protection Agency (USEPA) reported that nutrient enrichment was the cause of 40% of reported water-quality impairments (USEPA, 1998) and after results from studies conducted in the 1990s by the USGS National Water-Quality Assessment (NAWQA) Program demonstrated that high concentrations of both nitrogen (N) and phosphorus (P) were common in streams draining agricultural areas (Fuhrer et al., 1999). More recent USGS studies have indicated that agricultural streams can

transport up to 50% of the N and 20% of the P applied annually to the land (Mueller and Spahr, 2006). USGS models indicate that manure may be a larger source of P to the Gulf of Mexico than are row-crop sources (Alexander et al., 2008), and USGS data indicate that manure sources of total nitrogen (TN) and total phosphorus (TP) are increasing in the Ozarks (Rebich and Demcheck, 2007).

Confined poultry and loosely confined beef cattle are often produced on the same or adjacent farms in the Ozarks and increases in animal production have resulted in increased nutrient runoff to streams. However, nutrient concentrations in most Ozark streams are relatively low compared to concentrations in other regions of the United States. Herlihy and Sifneos (2008) compared nutrient concentrations for wadeable streams across the United States and determined that TP and TN concentrations for reference streams in the nutrient ecoregion containing the Ozarks were typically lowest and second lowest (respectively) of the 11 nutrient ecoregions evaluated.

\* Corresponding author. Tel.: +1 501 228 3626.

E-mail address: [bjustus@usgs.gov](mailto:bjustus@usgs.gov) (B.G. Justus).



**Fig. 1.** Locations of 30 wadeable stream sites sampled in the Ozark Highlands in 2006 with a general border for the Springfield and Salem Plateaus.

Interassemblage response to nutrients can vary because of differences related to trophic structure, mobility, and longevity, and the biotic assemblage that is best suited for monitoring nutrients and other forms of ecological disturbance is frequently debated (Griffith et al., 2005; Hering et al., 2006; Resh, 2008). Algal indices have been shown to be effective for monitoring well-established nutrient gradients (Lavoie et al., 2004; Potapova and Charles, 2007; Porter et al., 2008), but indices using macroinvertebrate (King and Richardson, 2007; Haase and Nolte, 2008) or fish assemblages (Wang et al., 2007) have also been successful. Few, if any, studies, however, have compared the value of the three assemblages for detecting perturbation as a consequence of low-level nutrient enrichment.

Conducting biotic assessments when nutrient levels are low can be challenging because effects are often subtle and can appear to be positive in nature (Biggs and Smith, 2002; Stevenson et al., 2008), but also because low-level nutrient enrichment may influence biota less than other water-quality and habitat variables. It is important that relations between nutrient concentrations and biotic assemblages be investigated in this setting to ensure that assessment methods are capable of detecting ecosystem perturbation as a consequence of nutrient enrichment in areas that are relatively undisturbed.

The objectives of this paper are to (1) assess the value of algal, macroinvertebrate, and fish assemblage metrics and indices for assessing low-level nutrient enrichment, and (2) characterize relations between agricultural land use (livestock production) and the three biotic indices.

### 1.1. Study area

We sampled 30 wadeable streams along a nutrient-concentration gradient in the Ozarks. Sites were divided between the Springfield and Salem Plateau physiographic areas (Fig. 1), which contain most of northern Arkansas, southern Missouri, and extreme eastern Oklahoma, and overlap much of the Ozark Highlands Ecoregion. Topography of the Springfield and Salem

Plateaus varies to some degree with gently rolling hills dominating the former and rugged hills dominating the latter; elevation above sea level ranges between 425 and 520 m (Fenneman and Johnson, 1946). The 30 streams generally are clear, with pool, riffle, and run sequences, and have moderate gradients with dominant substrates ranging in size from medium gravel to bedrock. Basin size ranges from 50 to 483 km<sup>2</sup> and streamflow measured at the time of sampling ranged from 0.01 to 0.55 m<sup>3</sup>/s (Table S1 in Supplementary Material).

Land use in the 30 basins (Table 1) represented a gradient for pasture; urban land use was usually less than 5%, and no wastewater-treatment plants discharged into the streams. Poultry were produced in 17 of the 30 stream basins and cattle were produced in all basins. Agricultural intensity was greatest in basins of extreme northwestern Arkansas and southwestern Missouri, which have the highest poultry and cattle production of counties within the two states and Oklahoma (NASS, 2008a,b).

## 2. Methods

### 2.1. Site selection

Geographic information system analysis and field reconnaissance were the primary methods used to select 30 streams that maximized the nutrient gradient across Ozark streams. Potential stream reaches were identified using the Elevation Derivatives for National Applications (USGS, 2005). Field reconnaissance was conducted at 54 candidate stream reaches that were selected from a larger group of reaches that met the basin size criterion (initially 90–300 km<sup>2</sup>, however, 5 streams with basins outside this range but with a streamflow characteristic of the remaining streams were included). Nutrient concentrations were measured using a portable nutrient analyzer (Hach™ model DREL/2010) and dissolved oxygen, pH, specific conductance, temperature, and turbidity were measured in the field with water-quality monitors. Field forms were completed that documented observations for habitat quality and flow characteristics. Land use, geographic

**Table 1**

Nutrient and land-use characteristics for 30 Wadeable streams sampled in the Ozark Highlands, 2006.

Site name	Abbreviated name (Fig. 1)	Physiographic section	Mean total nitrogen (mg/L)	Mean total phosphorus (mg/L)	Nutrient index score	Pasture (percent)	Cattle produced (number per km <sup>2</sup> )	Poultry (houses per km <sup>2</sup> )
Barren Fork near Timber, Missouri	Barren	Salem	0.07	0.003	0.00	7	12	0.0
Big Creek near Big Flat, Arkansas	BcBF	Springfield	0.29	0.027	0.93	33	75	0.2
Big Creek at Mauser Mill, Missouri	BcMM	Salem	0.14	0.002	0.05	4	6	0.0
Bear Creek near Omaha, Arkansas	Bear	Salem	0.14	0.005	0.14	35	86	2.0
Beaty Creek near Sycamore, Oklahoma	Beaty	Springfield	1.56	0.047	2.27	71	259	9.0
Bennetts River near Vidette, Arkansas	Benn	Salem	0.37	0.010	0.47	56	80	0.0
Big Piney River at Simmons, Missouri	BPine	Salem	0.25	0.024	0.78	42	106	0.0
Calf Creek near Silver Hill, Arkansas	Calf	Springfield	0.41	0.029	1.08	32	73	0.0
Little Flat Creek near McDowell, Missouri	Flat	Springfield	2.51	0.031	2.15	58	184	3.3
Long Creek southeast of Denver, Arkansas	Long	Springfield	0.72	0.038	1.55	37	98	1.8
Mahans Creek at West Eminence, Missouri	Maha	Salem	0.39	0.011	0.53	7	11	0.0
Maries River Near Freeburg, Missouri	Marie	Salem	0.56	0.035	1.35	41	104	0.1
Meramec River above Cook Station, Missouri	Mera	Salem	0.10	0.004	0.05	17	29	0.0
Myatt Creek east of Salem, Arkansas	Myatt	Salem	0.39	0.011	0.54	42	52	0.0
North Fork White River near Cabool, Missouri	NFWH	Salem	0.23	0.007	0.27	32	80	0.0
North Indian Creek near Wanda, Missouri	NInd	Springfield	4.71	0.052	3.30	81	265	11.7
North Prong Jacks Fork below Arroll, Missouri	NPJF	Salem	0.22	0.006	0.24	21	52	0.0
North Sylamore Creek near Fifty Six, Arkansas	NSyla	Springfield	0.10	0.005	0.08	2	5	0.2
Little Osage Creek at Healing Springs, Arkansas	Osag	Springfield	3.33	0.051	2.95	76	284	8.5
Piney Creek near Cabanol, Missouri	Piney	Salem	0.56	0.009	0.61	31	94	4.0
Poke Bayou near Sidney, Arkansas	Poke	Salem	0.58	0.025	1.10	47	84	0.0
Roasting Ear Creek near Newnata, Arkansas	REar	Springfield	0.51	0.016	0.77	20	46	0.7
South Fork Spring River north of Moko, Arkansas	SFS	Salem	0.43	0.013	0.63	45	42	0.0
Shoal Creek near Wheaton, Missouri	Shoal	Springfield	2.02	0.062	2.88	81	258	10.9
Spring Creek near Locust Grove, Oklahoma	Spring	Springfield	0.25	0.010	0.38	44	93	2.6
Sullivan Creek near Sandtown, Arkansas	Sull	Salem	0.54	0.018	0.85	31	73	2.2
Water Creek near Evening Star, Arkansas	Water	Springfield	0.14	0.004	0.10	18	71	0.3
Woods Fork near Hartville, Missouri	WdFk	Salem	0.27	0.035	1.12	55	142	0.2
West Piney Creek at Bado, Missouri	WPin	Salem	0.33	0.015	0.60	48	122	0.0
Yocum Creek near Oak Grove, Arkansas	Yoc	Springfield	2.37	0.047	2.57	71	217	8.4

coverage, and spatial distribution were other factors considered as sites were selected.

## 2.2. Water-quality sampling

Water-quality samples were collected during base-flow conditions at the 30 sites in late June 2006 and again in July–August 2006 with the following exceptions. Flooding delayed the second round of water-quality sampling until early September at one site and drought conditions in the summer of 2006 resulted in 5 of the original 30 sites sampled in June being replaced for the July–August sampling effort. At the 25 sites sampled twice, nutrient concentrations for the two samples were averaged to indicate nutrient enrichment for the month prior to biotic sampling; at the 5 remaining sites, the concentration from the single sample was used.

Standard USGS methods were used to collect and process water-quality samples. Water-quality samples were grabbed (because water velocities were <0.46 m/s) and were composited from three points that were equally distributed along the stream cross-section. Streamflow and field properties were measured at each site using a current meter (Rantz et al., 1982). Samples were analyzed for nutrient or nutrient-related (e.g. chlorophyll *a* and total organic carbon) constituents and all analyses were performed by the USGS National Water Quality Laboratory (NWQL) in Lakewood, Colorado (Patton and Kryskalla, 2003; Fishman, 1993). Total nitrogen was determined by summing nitrogen species. For purposes of statistical analysis, all nondetect values were assigned one-half of the reporting limit. Quality-control

samples were collected to assess bias and variability in the field and laboratory (Brightbill and Munn, 2008). The maximum difference between TP concentrations and TN concentrations in replicate samples was 0.0011 and 0.0260 mg/L, respectively. One of five blank samples had detections of TP (0.0029 mg/L) and TN (0.0350 mg/L).

## 2.3. Land use

Cattle density on pasture was estimated for each county contained in the stream basin by multiplying the amount of pasture in the county by county-level cattle density (the number of cattle produced in 2005 divided by the area of the county, NASS, 2008a). Cattle density on pasture then was combined for all counties in the stream basin, and that sum was divided by basin area to obtain an estimate of cattle density across the stream basin. Poultry production information was not available for 2005 (NASS, 2008a) and was not available for all counties in other years (NASS, 2008b). Consequently, poultry house density was used as a surrogate for poultry density. Poultry houses in each stream basin were counted using aerial photography (Center for Advanced Spatial Technologies, 2008) and were divided by the stream basin size to estimate the poultry houses per square kilometer of basin (Table 1).

## 2.4. Biotic sampling

Biotic sampling was conducted concurrently with the second water-quality sampling effort using NAWQA protocols (Moulton

et al., 2002). Biotic samples were collected from a reach length that measured approximately 20 times the mean wetted channel width, with a minimum reach length of 150 m and a maximum of 300 m.

Algal assemblages were sampled using a cylinder surface-area method. A quantitative algal subsample was collected from five cobbles at each of the five riffle locations (i.e. 25 subsamples were composited). The method involved placing a short cross-section of PVC pipe (2.8- or 3.3-cm diameter) on each cobble, dislodging all algae outside of the pipe template with a wire brush or small knife, and rinsing the dislodged algae from the cobble with native water. Algae remaining inside the pipe template was dislodged with a wire brush or (scraped free) with a knife and rinsed into a sample bottle as the subsample. Sample area and total sample volume were recorded, and the sample was preserved with buffered formalin. Taxa were identified and enumerated at the Academy of Natural Sciences of Philadelphia (ANSP) Phycology Section in Philadelphia, Pennsylvania. The ANSP also determined cell density for each algal species using methods described in Charles et al. (2002). Chlorophyll *a* was determined at the USGS NWQL using methods described in Arar and Collins (1997).

A disturbance-removal process was used to collect macroinvertebrate samples from coarse-grained riffle substrates that were adjacent to locations where algal samples were collected. Five discrete samples were collected with a Slack sampler (50-cm × 33-cm net frame, 500- $\mu$ m Nitex™ net, and retrofitted with a 0.25-m<sup>2</sup> template) from riffles located throughout the reach. Macroinvertebrates were sampled from within the template as it was positioned on the stream bottom and immediately upstream from the Slack sampler. Substrate within the template was thoroughly disturbed using a small hand rake (or brushed if large cobble) and dislodged organisms were transported into the net by water current. All sample material was composited into a 20-L container and elutriated to remove sediment and larger particles. The material remaining on a 500- $\mu$ m sieve after elutriation was preserved in 10% formalin and shipped to the USGS NWQL for identification and enumeration.

Fish were sampled at 29 sites using electrofishing and seining methods (fish were not sampled at Maries River because of potential occurrence of a federally listed threatened species). A backpack unit (Smith-Root model 12B) was used to electrofish all sites, and one pass was made along each bank. Electrofishing passes progressed from the downstream boundary of the sampling reach to the upstream boundary. Riffle habitats also were sampled by kick seining in conjunction with electrofishing. Most fish were identified and counted in the field and then were released. Fish that could not be positively identified in the field were preserved for laboratory identification. Fish were identified using taxonomic keys for Arkansas (Robison and Buchanan, 1988), Missouri (Pflieger, 1997), and Oklahoma (Miller and Robison, 2004), however, nomenclature follows Robins et al. (2004).

## 2.5. Metric sources

Two USGS software programs—the Macroinvertebrate Data Analysis System (IDAS; Cuffney, 2003) and the Algal Data Analysis System (ADAS; a derivative of the IDAS program)—were the primary means for calculating algal and macroinvertebrate metrics. Both programs process multiple levels of taxonomic resolution, resolve taxonomic ambiguities, and use attribute files to calculate assemblage and tolerance metrics common to the literature (Barbour et al., 1999; Porter, 2008). Also, some macroinvertebrate metrics used by local natural resource agencies were considered as potential metrics, as were all species—order level taxa for the macroinvertebrate and fish assemblages.

ADAS was used to calculate algal metrics using an attribute file of published values (Porter, 2008). A total of 73 algal metrics was calculated for soft algae and diatoms (Table S2 in Supplementary Material). Algal metrics were primarily indicative of trophic preferences (Van Dam et al., 1994) and pollution tolerance (Lange-Bertalot, 1979).

A total of 62 macroinvertebrate metrics was calculated (Table S3 in Supplementary Material) using data specific to the south-eastern (Barbour et al., 1999; Lenat, 1993) and midwestern (Hilsenhoff, 1987) United States. Values for richness, percent richness, abundance, and percent relative abundance were evaluated for all but a few metrics where percentages were not beneficial to the analysis (e.g. diversity indices).

A total of 60 fish metrics used by local natural resource agencies or obtained from biotic indices developed for use in the Ozarks or adjacent areas (Dauwalter et al., 2003; Justus, 2003; Dauwalter and Jackson, 2004) were considered as candidates for the fish index (Table S4 in Supplementary Material). Fish metrics were calculated using fish traits from several sources (Robison and Buchanan, 1988; Pflieger, 1997; Petersen et al., 2008; USGS, 2008).

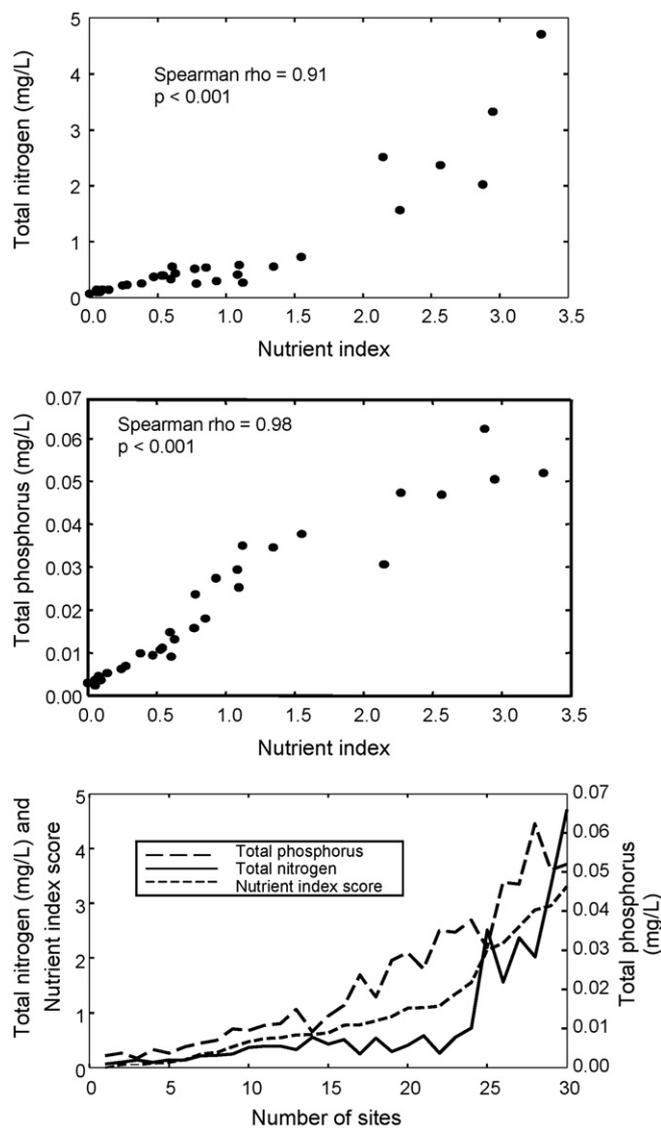
## 2.6. Statistical analysis

TN and TP were combined into a nutrient index to facilitate comparisons of nutrient enrichment and biotic metrics. TN and TP are commonly used by State monitoring agencies to characterize nutrient enrichment in the Ozarks and typically have close relations to livestock production in the Ozarks (Davis and Bell, 1998) and much of the United States (Alexander et al., 2008). Chlorophyll *a* also is used by State monitoring agencies to characterize nutrient enrichment and also was considered for the nutrient index but relations between chlorophyll *a* and TN and TP were poor (Spearman  $\rho = 0.14$  and  $0.30$ , respectively).

A three-step process was used to calculate the nutrient index. First, mean values for TN and TP were normalized to a mean of 0 and a standard deviation of 1. Second, normalized values for TN and TP were averaged, and third, all normalized (average) values were standardized to positive numbers by adding the difference between the minimum value and zero. The resulting nutrient index ranged from 0 to 3.3 (Table 1, Fig. 2).

For each of the biotic indices, four nonredundant metrics were selected from the initial 195 (73 algal, 62 macroinvertebrate, and 60 fish) metrics aggregated for this study. Index robustness may sometimes be associated with increasing metric number, however, a decision was made to limit the number of metrics (to four) after preliminary analyses indicated that, for one or more assemblages, relations between the next best candidate metric(s) and the nutrient index were nonexistent. The decision to select a relatively small number of metrics for each index also reduced the risk that redundant metrics were included in the final indices.

Metrics that were the best candidates for the three biotic indices were identified with a process that included a combination of univariate and nonparametric multivariate methods. Prior to analysis, metrics were separated by guild (e.g. tolerance, behavior, feeding, or nesting traits) and scoring method (e.g. relative abundance, relative density, and richness). Pairs of metrics from respective metric guilds initially were evaluated using Spearman rank correlation to identify and eliminate redundant metrics. When two metrics that had taxa in common had  $\rho > 0.70$ , the metrics were considered to be redundant and one metric was eliminated to avoid index bias and error. Scatter plot matrices also were used to visually identify outlying values or spurious correlations. Metric relevancy to nutrient enrichment (e.g. increasing biomass, a decrease in organisms intolerant of organic pollution, an increase in organisms tolerant of organic pollution)



**Fig. 2.** Scatter plots and a line graph demonstrate relations of a nutrient index to total nitrogen and total phosphorus concentrations at 30 Wadeable Ozark streams.

was the primary consideration that determined which of the redundant metrics was retained for further analysis.

Once redundant metrics had been eliminated, BVSTEP, a nonparametric screening procedure in PRIMER v6 (Clarke and Warwick, 2001), was used to identify candidate metrics that “best” represented each of the three biotic assemblages. First, BVSTEP was used to compare the similarity matrices for an individual metric to the similarity matrix of all metrics in the same guild (group). This step helped identify individual metrics and metric combinations with the highest similarity to the metric guild (i.e. a multivariate sample pattern that matched that of the entire guild) and greatly reduced the number of metrics to be considered in further data-reduction steps. The similarity matrix of the metric with the highest correlation to the similarity matrix of the entire guild was retained for further analysis. This step was repeated using an  $n - 1$  approach (once identified as an index candidate the metric was removed from the guild) until all metrics having a similarity matrix that was correlated ( $\rho \geq 0.25$ ) to the similarity matrix of the parent guild had been identified. A  $\rho$  value of 0.25 was selected because matrix correlations occur over a lower range than simple univariate correlations.

Metrics identified with the analytical step, above, were combined into a final “candidate metric subset” (generally 10–15 metrics). The BVSTEP process was used again, but on this occasion, similarity matrices of the candidate metrics were compared to the similarity matrix of the nutrient index. The candidate metrics that had similarity matrices with the highest correlations to the similarity matrix of the nutrient index were retained. Spearman  $\rho$  was used again to evaluate for metric redundancy but this time for the small group of candidate metrics identified with the second round of BVSTEP. When pairs of redundant metrics with similar correlations to the nutrient index were identified, scatter plots were evaluated to determine which of the two redundant metrics had the best relation to nutrients and, ultimately, to identify the four candidate metrics that were selected for the respective assemblage index.

Scores for each of the three biotic indices were calculated by combining values for the four respective metrics using a centering method (Justus, 2003). An advantage of the centering method is that it is more robust than other scoring methods (e.g. scores range from 0 to 100 rather than tiered, preassigned metric classes of 1, 3, or 5). A disadvantage of the centering method is that it does not facilitate comparison of sites from independent data sets because metric scores are based on the range of sampling conditions that may not include least- or most-impaired sites. The centering method uses one of two scoring procedures depending if high or low metric values represent least-degraded conditions. If a high metric value indicated least-degraded conditions, the metric value was first divided by the maximum metric value (for all 30 sites), and the resulting quotient was multiplied by 100 to obtain a metric score. To obtain a metric score if low metric values indicated least-degraded conditions, the metric value was again divided by the maximum metric value, but the resulting quotient was subtracted from 1 before being multiplied by 100. Scores for the four metrics were averaged to obtain an index score. Sites having the highest biotic index scores had the least-degraded conditions. Relations between the three biotic indices and the nutrient index and TP and TN also were evaluated with correlation procedures and scatter plots. Scatter plots also were used to determine how poultry (houses) and cattle production varied for the 30 basins and to evaluate relations between the three biotic indices and the two forms of livestock production.

### 3. Results

#### 3.1. Biotic metric/nutrient relations

Median concentrations of TN and TP were 0.393 mg/L (0.07–4.71 mg/L) and 0.015 mg/L (0.002–0.062 mg/L), respectively. Values for the nutrient index ranged from 0 to 3.3 and were highly correlated to TN and TP concentrations ( $\rho = 0.91$  and  $0.98$ , respectively; Fig. 2). The 30 sites were equally divided above and below an index score of 0.75 (because TN and TP concentrations associated with that index score,  $\sim 0.40$  and  $0.018$  mg/L, respectively, are comparable to median concentrations).

Although, the four metrics selected for each of the three assemblage indices had the strongest relations to the nutrient index of all metrics evaluated for that assemblage, relations between a few of the 12 metrics and the nutrient index were weak ( $\rho \leq 0.36$  and  $p > 0.05$ ). In most cases, however, metric values above and below the nutrient index score of 0.75 had different distributions. The four biotic metrics selected for each index are reported in the order of the correlation of the metric to the nutrient

**Table 2**  
Algae, macroinvertebrate, and fish metrics selected for three indices, their expected response to nutrient exposure, correlation to a nutrient index, and a comparison of values above and below a median concentration.

Assemblage	Metric description	Expected response to nutrients	Rho	Distinction for sites above and below median concentrations
Algae	Most tolerant diatoms, relative abundance (percent)	Positive (Bahls, 1993)	0.80	Percent RA $\geq 3\%$ at 3 of 15 sites; percent RA $\geq 3\%$ at 12 of 15 sites
Algae	<i>Cymbella affinis</i> , <i>C. delicatula</i> , and <i>C. hustedtii</i> relative abundance (percent)	Negative (Potapova and Charles, 2007)	-0.71	Percent RA $> 10\%$ at 11 of 15 sites; percent RA $> 10\%$ at 2 of 15 sites
Algae	Mesosaprobic algae taxa richness (percent)	Positive (Lange-Bertalot, 1979)	0.65	Percent TR $> 10\%$ at 5 of 15 sites; percent TR $> 10\%$ at 11 of 15 sites
Algae	Obligate nitrogen heterotroph relative abundance (percent)	Positive (Leland, 1995)	0.57	Percent RA $> 1\%$ at 1 of 15 sites; percent RA $> 1\%$ at 8 of 15 sites
Macroinvertebrate	Intolerant relative abundance (percent)	Negative (Barbour et al., 1999)	-0.50	Percent RA $> 85\%$ at 14 of 15 sites; percent RA $> 85\%$ at 9 of 15 sites
Macroinvertebrate	Baetidae relative abundance (percent)	Positive (USEPA, 2008)	0.48	Percent RA $> 10\%$ at 2 of 15 sites; percent RA $> 10\%$ at 9 of 15 sites
Macroinvertebrate	Insect biomass (grams)	Positive (King and Richardson, 2007)	0.47	$> 2$ g at 1 of 15 sites; $> 2$ g at 7 of 15 sites
Macroinvertebrate	Moderately tolerant taxa richness	Positive (Barbour et al., 1999)	0.30	$\geq 20$ taxa at 6 of 15 sites; $\geq 20$ taxa at 10 of 15 sites
Fish	Herbivore/detritivore taxa richness	Positive (Rashleigh, 2004)	0.41	$\geq 4$ taxa at 7 of 15 sites; $\geq 4$ taxa at 10 of 14 sites
Fish	Pool species relative abundance (percent)	Indirect	-0.38	Percent RA $> 50\%$ at 11 of 15 sites; percent RA $> 50\%$ at 7 of 14 sites
Fish	Fish collected per meter	Positive (Pilati et al., 2009)	0.36	$> 2.5$ fish/m at 5 of 15 sites; $> 2.5$ fish/m at 7 of 14 sites
Fish	Black bass relative abundance (percent)	Indirect	-0.35	Percent RA $> 1\%$ at 8 of 15 sites; percent RA $> 1\%$ at 4 of 14 sites

Notes: Rho, Spearman correlation to a nutrient index; RA, relative abundance; TR, taxa richness.

index, which may also reflect or approximate each metric's relevance to nutrients (Table 2).

### 3.2. Algae metric and index performance

All four metrics selected for the algal index were associated with nutrient tolerance or dependence (Table 2). The four metrics were: relative abundance of most tolerant diatoms, a metric associated with tolerance to elevated nutrient concentrations; the combined relative abundance of *Cymbella delicatula*, *C. affinis*, and *C. hustedtii*, three species of diatoms that respond to low to moderate nutrient concentrations; mesosaprobic algae percent taxa richness, a metric associated with tolerance to moderately elevated nutrients; and lastly, the relative abundance of diatoms that are obligate nitrogen heterotrophs, a metric associated with nitrogen dependence. All but the second metric would be expected to have a positive relation to nutrient concentrations.

The algal index, calculated with the four metrics above, ranged from 20.9 to 94.7 (Table S5 in Supplementary Material) and had a high correlation to the nutrient index ( $\rho = -0.89$ , Fig. 3). Correlations between the algal index and TP ( $\rho = -0.91$ ) were much higher than between the algal index and TN ( $\rho = -0.72$ , Fig. 4).

### 3.3. Macroinvertebrate metric and index performance

The four metrics selected for the macroinvertebrate index included three metrics associated with organisms that are intolerant or moderately tolerant of organic pollution, and a fourth metric associated with productivity. The three metrics evaluating tolerance included: the relative abundance of intolerant organisms, Baetidae (a family with several species that are moderately tolerant of nutrients) relative abundance, and moderately tolerant taxa richness. The fourth macroinvertebrate metric, and the metric related to productivity, was insect biomass. All but the first metric would be expected to have a positive relation to nutrient concentrations.

The macroinvertebrate index ranged from 36.3 to 85.7 (Table S6 in Supplementary Material) and decreased in relation to the nutrient index scores ( $\rho = -0.63$ , Fig. 3). Correlations between the macroinvertebrate index and TN and TP concentrations were similar ( $-0.64$  and  $-0.60$ , respectively; Fig. 4).

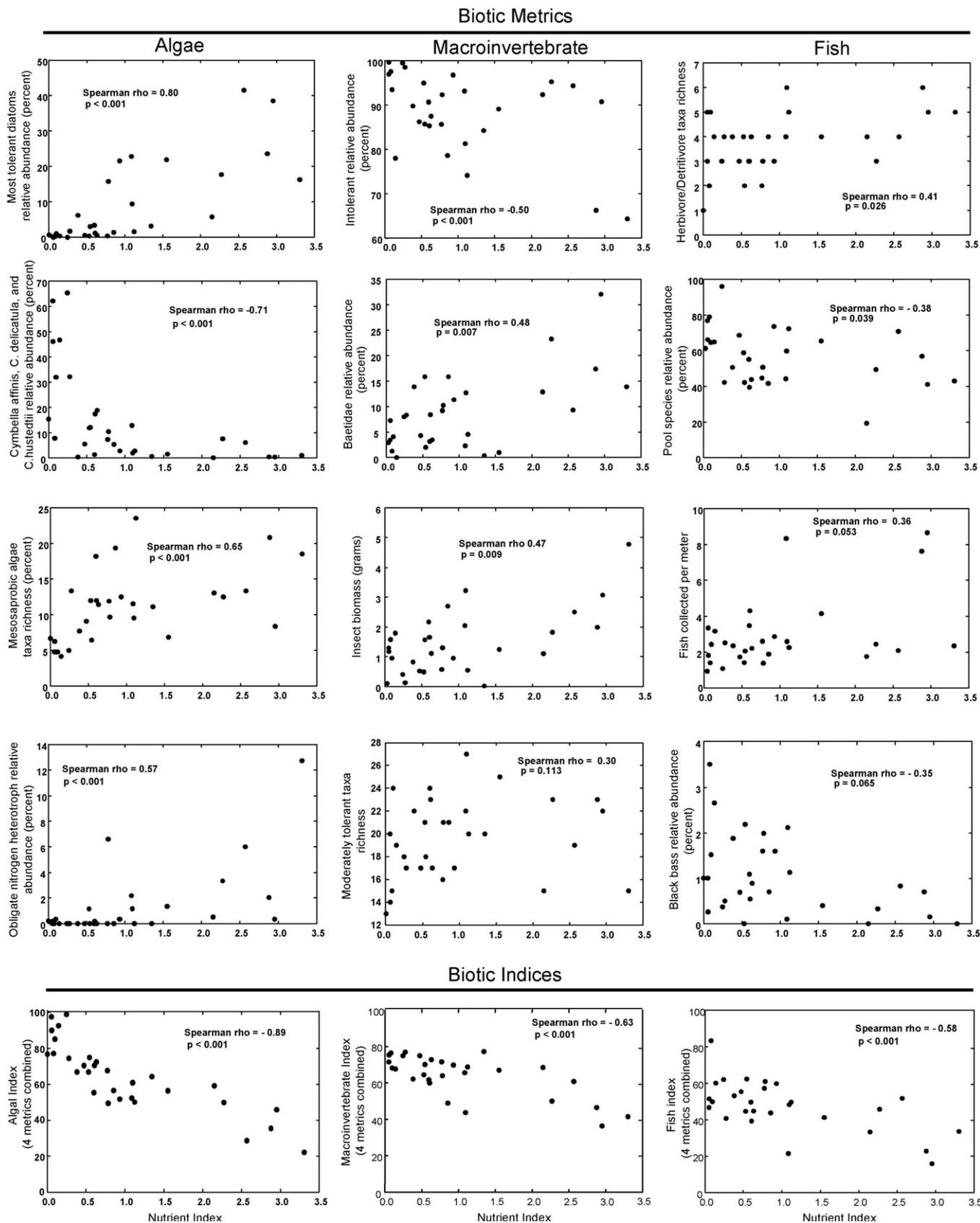
### 3.4. Fish metric and index performance

The four fish metrics selected for the fish assemblage index were: herbivore and detritivore taxa richness, pool species relative abundance, fish catch per unit effort, and black bass (*Micropterus dolomieu*, *M. punctatus*, and *M. salmoides*) relative abundance. Two of the metrics—herbivore and detritivore taxa richness and fish catch per unit effort would be expected to have a positive relation to nutrient concentrations; however, the two remaining metrics—pool species relative abundance and black bass relative abundance—probably have indirect relations to nutrients.

The fish index ranged from 15.9 to 83.7 (Table S7 in Supplementary Material) and also decreased with increasing nutrient index scores ( $\rho = -0.58$ , Fig. 3). The fish index had a stronger correlation to TN than to TP ( $\rho = -0.68$  and  $-0.54$ , respectively; Fig. 4).

### 3.5. Indices comparison

Of the three biotic indices, the algal index had a much higher correlation to the nutrient index (i.e. a  $\rho$  of  $-0.89$ , compared to  $-0.63$  and  $-0.58$ ). Correlations to the nutrient index, for the algal, macroinvertebrate, and fish metrics ranged from 0.57 to 0.80, 0.30 to 0.50, and 0.35 to 0.41 (reported as absolute values, Fig. 3), respectively. All relations among the four algal metrics and the nutrient index were statistically significant ( $p \leq 0.05$ ); however, relations for only 3 of 4 macroinvertebrate, and only 2 of 4 fish metrics were statistically significant to the nutrient index. Correlations of the three biotic indices to TN were similar (a range between  $-0.64$  and  $-0.72$ , Fig. 4) but the algal index had a much higher correlation to TP ( $\rho = -0.91$ ) than did the



**Fig. 3.** Scatter plots and correlations comparing 12 biotic metrics and 3 biotic indices to a nutrient index (representing total nitrogen and total phosphorus concentrations) at 30 Wadeable Ozark streams.

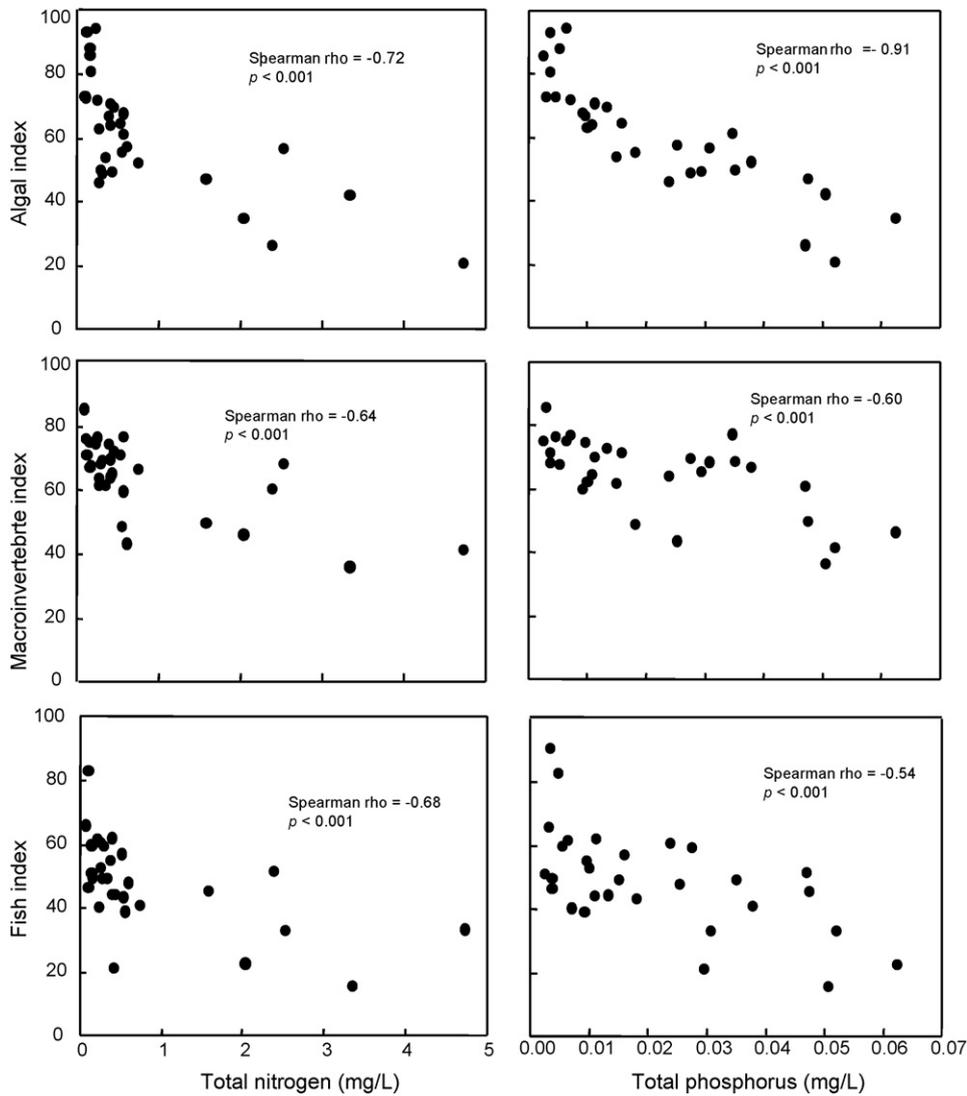


Fig. 4. Scatter plots and correlations comparing relations between three biotic indices and total nitrogen and total phosphorus concentrations at 30 wadeable Ozark streams.

macroinvertebrate and fish indices ( $\rho = -0.60$  and  $-0.54$ , respectively).

3.6. Land use

Cattle were produced in all basins (a range of 5–284 cattle per km<sup>2</sup> of basin), but poultry were produced in only 17 of the 30 basins (the number of poultry houses ranged from 0 to 11.7 per km<sup>2</sup> of basin, Table 1). Cattle production generally was much higher in basins where poultry were produced than in basins where poultry were not produced, and was highest in basins with the highest poultry production (Fig. 5). The three biotic indices were negatively related to cattle production; correlations ranged from  $-0.46$  to  $-0.76$  (Fig. 6).

4. Discussion

4.1. Metric performance

Ten of the 12 metrics selected for the three biotic indices were measures of tolerance, biomass, or density that are known to fluctuate in response to stream productivity (e.g. Porter et al., 2008; Ortiz and Puig, 2007), and, thus, have an ecological relevance to nutrients. Correlations between metrics and the nutrient index

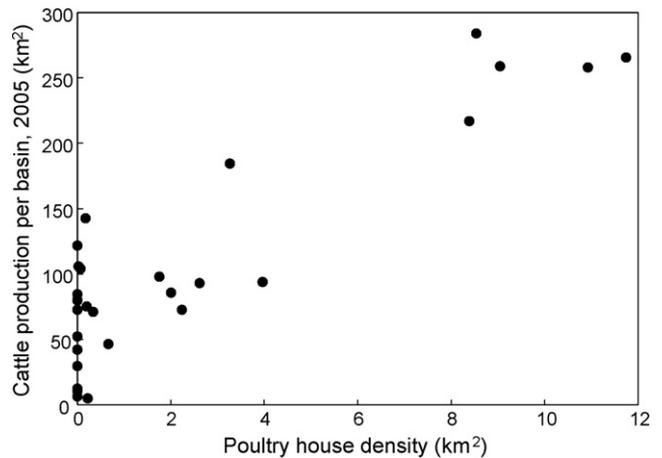


Fig. 5. A scatter plot comparing relations between cattle production and the number of poultry houses in 30 Ozark stream basins. Cattle production in the basins ranged from 5 to 125 cattle/km<sup>2</sup> when no poultry were produced but generally exceeded 75 cattle per km<sup>2</sup> when there was one or more poultry house in the basin.

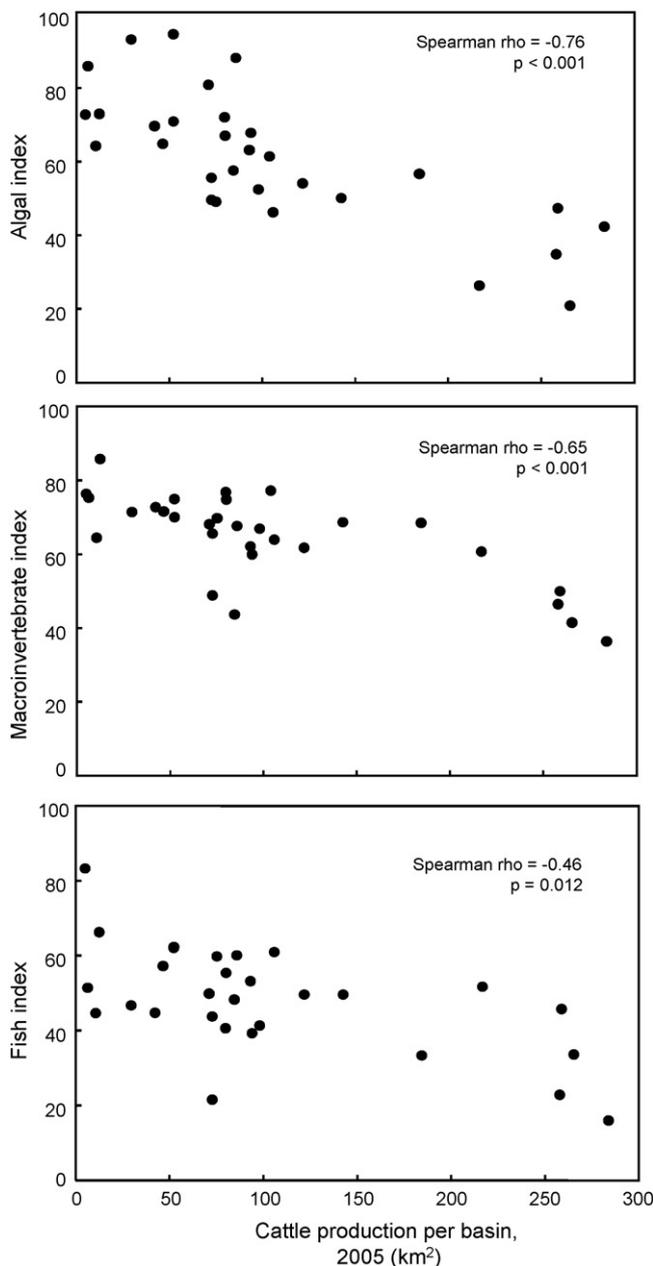


Fig. 6. Relations of three biotic indices to cattle density in 30 Ozark stream basins.

generally declined across assemblages (from algae to macroinvertebrates to fish)—a probable consequence of the trophic level of the taxa targeted by the metrics and an associated decrease in dependence on inorganic nutrients. For the relative abundances of pool species and black bass, two fish metrics that are comprised of species of Centrarchidae which are known to be moderately tolerant of nutrients (Maceina and Bayne, 2001), relations may have been equal or stronger to variables associated with habitat quality than to nutrients.

Relations between the three biotic indices and the nutrient index were stronger than relations between the biotic metrics and the nutrient index, indication that even metrics that had weak relations to the nutrient index were beneficial to biotic indices. However, weak relations are to be expected between biotic metrics and nutrient enrichment when concentrations at some sites are below a threshold for which a biotic response occurs. Terrel et al. (1996) noted that wedge-shaped scatter plots are characteristic of the relation between a dependent variable and an independent

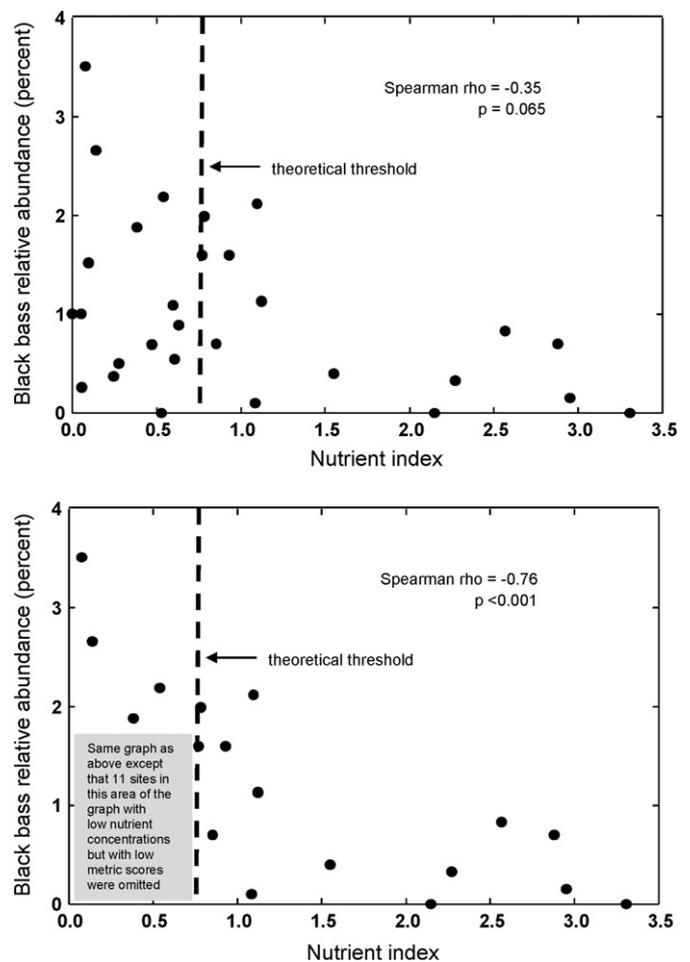


Fig. 7. Relations of black bass relative abundance to the nutrient index emphasize the relevance of the wedge-shaped scatter pattern. The correlations in the second plot doubles that of the previous plot after sites with low nutrient concentrations but with poor metric scores were omitted.

[test] variable when some values for the independent variable are below the threshold for which a response occurs and when other unknown or unmeasured independent variables are influencing the dependent variable (see example in Fig. 7). Of the 12 metrics selected for the three indices, wedge-shaped scatter plots are most apparent for the relative abundance of three *Cymbella* species and black bass relative abundance.

The small size of the data set limits our ability to identify thresholds for TN and TP, however, some literature indicate that TN and TP concentrations near median values for this study are near threshold concentrations that distinguish between reference streams and streams that are slightly enriched (i.e. near background, Table 3). Biotic metric scores were inversely related to nutrients and were generally highest when TN and TP concentrations were less than about 0.40 mg/L and about 0.018 mg/L (respectively), but were generally lowest when concentrations were higher. These TN and TP concentrations are comparable to background concentrations from sites across the United States (Clark et al., 2000; Smith et al., 2003; Herlihy and Sifneos, 2008). Other studies have indicated that substantial changes in macroinvertebrate assemblage structure (Smith et al., 2007) and algal biomass (Stevenson et al., 2006) may occur near these concentrations (Table 3).

#### 4.2. Index/nutrient relations

Of the three assemblages evaluated, the algal assemblage seems to be most appropriate for assessing effects of low-level nutrient

**Table 3**

A comparison of median total nitrogen (TN) and total phosphorus (TP) concentrations at 30 Wadeable Ozark streams to TN and TP concentrations that are equivalent to a nutrient index score of 0.75, and to concentrations suspected of distinguishing between reference streams and slightly enriched streams.

Description or data source	Total nitrogen (mg/L)	Total phosphorus (mg/L)
Median concentrations	0.39	0.015
Concentrations equivalent to a nutrient index score of 0.75	0.40	0.018
Dodds et al. (1998) <sup>a</sup>	0.70	0.025
Clark et al. (2000) <sup>b</sup>	0.26	0.022
Smith et al. (2003) <sup>c</sup>	0.26	0.020
Smith et al. (2007)	0.29	0.020
Herlihy and Sifneos (2008) <sup>d</sup>	0.31	0.017

<sup>a</sup> Concentrations are based on differences in chlorophyll *a* for oligotrophic and mesotrophic stream categories.

<sup>b</sup> Flow-weighted concentrations.

<sup>c</sup> Modeled values (not measured).

<sup>d</sup> 75th percentile of least-impaired sites sampled as part of the Environmental Protection Agency Wadeable Stream Assessment.

enrichment in Wadeable Ozark streams. These results are consistent with those of Lavoie et al. (2008) who found that algal diatoms were effective for monitoring low-level TN and TP concentrations similar to those observed in this study. Algae are primary producers and nutrient availability may be the most important variable influencing algae (Lowe and Pan, 1996; Borchardt, 1996; Porter, 2008). By contrast, variables other than nutrients may be of equal or greater importance to macroinvertebrates and fish because they are primary and secondary consumers. Other reasons why algae are effective for assessing low-level nutrient enrichment are related to motility and longevity. Most algae are sessile organisms that have a short life cycle that is completed in the sampling area (Lowe and Pan, 1996) and algae may be more resistant to hydrologic disturbance than macroinvertebrates or fish when benthic habitats are armored as they are in Ozark streams (Riseng et al., 2004). Even though algae seem to be well suited for assessing low-level nutrient enrichment, the increased assurance of an accurate assessment (Hering et al., 2006; Griffith et al., 2005) and public perception regarding the economic importance of macroinvertebrates and fish may justify costs associated with sampling multiple assemblages for some monitoring programs.

Algal indices may be an alternative to chlorophyll *a* for assessing the effects of nutrient enrichment in some regions. Relations between chlorophyll *a* and TN and TP were poor for our data set and have been found to be poor in the Midwest United States (Morgan et al., 2006; Lowe et al., 2008), possibly because of confounding factors (i.e. light intensity, degree of nutrient limitation, and habitat quality, Miltner and Rankin, 1998).

#### 4.3. Biotic index/land-use relations

Poultry litter applications are a concern in the Ozarks and elsewhere because N and P application rates are difficult to quantify and because litter application rates may exceed commercial fertilizing rates when an abundance of litter is available (Knowlton et al., 2004). Ozark land-use data also indicate that because of the availability of litter for fertilizer and associated increases in grass and hay production, cattle feeding capacity is increased in areas where poultry are produced.

Although the TN and TP contribution to Ozark streams from manure seems to be increasing in high poultry and cattle production areas (Rebich and Demcheck, 2007), we found no studies that have been designed to address the ecological risks to streams when high poultry and cattle production dominate basin land use. The combined influence of poultry litter and cattle

manure on nutrient runoff has been simulated in field experiments (Sauer et al., 1999; Vadas et al., 2007), and several studies have addressed runoff loss from poultry litter (Pierson et al., 2001; Butler et al., 2008; Sistani et al., 2008) or cattle manure (Edwards et al., 2000; Capece et al., 2007; Butler et al., 2008) under various conditions (i.e. different application rates, precipitation rates, soil saturations, and grazing intensities), but the effects of cattle and litter applications are rarely considered in combination.

Cattle production can increase nutrient runoff to streams directly (i.e. fecal deposition) or indirectly (i.e. habitat alteration). Unrestricted cattle generally will spend a large part of the day in the riparian zone regardless of the season or the availability of water elsewhere (Zuo and Miller-Goodman, 2004; Bagshaw et al., 2008), and James et al. (2007) observed that fecal deposition was significantly higher near streams than in other areas of the pasture. Cattle influence habitat variables that have indirect relations to nutrients and can confound relations between biotic integrity and nutrients (Miltner and Rankin, 1998; Maret et al., 2008). Nutrient runoff potential increases when the grass filter in the riparian zone is over grazed (Sistani et al., 2008) and can increase as much as 90% when cattle trample and compact soils (Nguyen et al., 1998). Streambank stability also declines when cattle graze banks and access streams which, in turn, can increase nutrient runoff, particularly for TP (Vidon et al., 2008; Zaines et al., 2008).

#### 4.4. Conclusions

Biotic assessment methods used to evaluate areas with little or no disturbance should be sensitive to low-level nutrient enrichment because changes in land use and associated effects on water-quality and ecological condition often occur slowly and over extended periods. Some biotic metrics selected for the three indices had weak relations to nutrient enrichment probably because TN and TP concentrations were below a threshold to which a biological response occurs. Relations of the three biotic indices to nutrient enrichment, however, were much stronger than relations between the biotic metrics and nutrient enrichment. This observation indicates that metrics selected for the indices were beneficial to index development and provides some validation for the index approach.

The algal index had a much stronger relation to low- to moderate-level nutrient enrichment than did the macroinvertebrate or fish index but all three indices were negatively correlated to nutrient enrichment. Biotic index scores were lowest and nutrient concentrations were highest for streams with basins having the highest poultry and cattle production. Because of the availability of litter for fertilizer and associated increases in grass and hay production, cattle feeding capacity increases with poultry production. The synergistic effect of poultry and cattle production on Ozark streams in high production areas has not been evaluated and additional studies are needed before ecological risks are adequately assessed.

#### Acknowledgments

This is one of the several studies supported by the USGS NAWQA program to evaluate the effects of nutrient enrichment on stream ecosystems. The project would not have been possible had it not been for a number of USGS employees who assisted with reconnaissance, sampling, data organization and compilation, maps and figures, and manuscript review. Special thanks are extended to Amy Beck, Kelly Brady, Brian Clark, Jimmy Clark, Rheannon Hart, Shannon Kelly, Dwight Lasker, and Dan Yeatts. Thanks are extended to Greg Kloxin, who provided metrics that originated from the Oklahoma Conservation Commission. Thanks are also extended to two students from the University of Arkansas

at Pine Bluff, Avian Wright and Byron Burns, who assisted with biotic sampling. Colleague reviews by Terry Maret, Mark Munn, Ian Waite, and two anonymous reviewers improved the quality of the report.

## Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at doi:10.1016/j.ecolind.2009.10.007.

## References

- Alexander, R.B., Smith, R.A., Schwarz, G.E., Boyer, E.W., Nolan, J.V., Brakebill, J.W., 2008. Differences in phosphorus and nitrogen delivery to the Gulf of Mexico from the Mississippi River Basin. *Environ. Sci. Technol.* 42, 822–830.
- Arar, E.J., Collins, G.B., 1997. In Vitro Determination of Chlorophyll *a* and Pheophytin *a* in Marine and Freshwater Algae by Fluorescence, Method 445.0, Revision 1.2. U.S. Environmental Protection Agency, National Exposure Research Laboratory, Office of Research and Development, Cincinnati, OH.
- Bagshaw, C.S., Thorrold, B., Davison, M., Duncan, I.J.H., Matthews, L.R., 2008. The influence of season and of providing a water trough on stream use by beef cattle grazing hill-country in New Zealand. *Appl. Animal Behav. Sci.* 109, 155–166.
- Bahls, L.L., 1993. Periphyton Bioassessment Protocols for Montana Streams. Montana Department of Health and Environmental Sciences, Water Quality Bureau, Helena, Montana.
- Barbour, M.T., Gerritsen, J., Snyder, B.D., Stribling, J.B., 1999. Rapid Bioassessment Protocols for Use in Streams and Wadeable Rivers: Algal, Benthic Macroinvertebrates, and Fish, EPA 841-B-99-002, 2nd ed. U.S. Environmental Protection Agency, Office of Water.
- Biggs, B.J.F., Smith, R.A., 2002. Taxonomic richness of stream benthic algae: effects of flood disturbance and nutrients. *Limnol. Oceanogr.* 47, 1175–1186.
- Borchardt, M.A., 1996. Algal ecology. In: Stevens, R.J., Bothwell, M.L., Lowe, R.L. (Eds.), *Nutrients*. Academic Press, San Diego, pp. 183–227.
- Brightbill, R.A., Munn, M.D., 2008. Environmental and biological data of the nutrient enrichment effects on stream ecosystems project of the National Water Quality Assessment Program, 2003–04: U.S. Geological Survey Data Series 345, Tacoma, Washington, 12 p., <http://wa.water.usgs.gov/neet/products.html> (accessed April 2009).
- Butler, D.M., Franklin, D.H., Cabrera, M.L., Tasistro, A.S., Xia, K., West, L.T., 2008. Evaluating aeration techniques for decreasing phosphorus export from grasslands receiving manure. *J. Environ. Qual.* 37, 1279–1287.
- Capece, J.C., Campbell, K.L., Bohlen, P.J., Graetz, D.A., Portier, K.M., 2007. Soil phosphorus, cattle stocking rates, and water quality in subtropical pastures in Florida, USA. *Rangeland Ecol. Manag.* 60 (1), 19–30.
- Center for Advanced Spatial Technologies, 2008. University of Arkansas Spatial Library, <http://watersheds.cast.uark.edu> (accessed November 2008).
- Charles, D.F., Knowles, C., Davis, R.S. (Eds.), 2002. Protocols for the analysis of algal samples collected as part of the U.S. Geological Survey National Water-Quality Assessment Program. Report No. 02–06. Patrick Center for Environmental Research, The Acad. of Natural Sci., Philadelphia, PA, 124 pp.
- Clark, G.M., Mueller, D.K., Mast, M.A., 2000. Nutrient concentrations and yields in undeveloped stream basins of the United States. *J. Am. Water Res. Assoc.* 36, 849–860.
- Clarke, K.R., Warwick, R.M., 2001. *Changes in Marine Communities: An Approach to Statistical Analysis and Interpretation*. PRIMER\_E Ltd., Plymouth, UK, 174 pp.
- Cuffney, T.F., 2003. User's manual for the National Water-Quality Assessment Program macroinvertebrate data analysis system (IDAS) software: Version 3. U.S. Geological Survey Open-File Report 03-172, 103 pp.
- Dauwalter, D.C., Jackson, J.R., 2004. A provisional fish index of biotic integrity for assessing Ouachita Mountains streams in Arkansas, USA. *Environ. Monit. Assess.* 91, 27–57.
- Dauwalter, D.C., Pert, E.J., Keith, W.E., 2003. An index of biotic integrity for fish communities in Ozark Highland streams of Arkansas. *Southeastern Nat.* 2, 447–468.
- Davis, J.V., Bell, R.W., 1998. Water-quality assessment of the Ozark Plateaus study unit, Arkansas, Kansas, Missouri, and Oklahoma—analysis of information on nutrients, suspended sediment, and suspended solids, 1970–92. U.S. Geological Survey Water-Resources Investigations Report 95-4042, 112 pp.
- Dodds, W.K., Jones, J.R., Welch, E.B., 1998. Suggested classification of stream trophic state: distributions of temperate stream types by chlorophyll, total nitrogen, and phosphorus. *Water Res.* 32 (5), 1455–1462.
- Edwards, D.R., Hutchens, T.K., Rhodes, R.W., Larson, B.T., Dunn, L., 2000. Quality of runoff from plots with simulated grazing. *J. Am. Res. Assoc.* 36 (5), 1063–1073.
- Fenneman, N.M., Johnson, D.W., 1946. *Physical Divisions of the United States* (Map). U.S. Geological Survey, Washington, DC.
- Fishman, M.J., 1993. Methods of analysis by the U.S. Geological Survey National Water Quality Laboratory—determination of inorganic and organic constituents in water and fluvial sediments. U.S. Geological Survey Open-File Report 93-125, 217 pp.
- Fuhrer, G.J., Gilliom, R.J., Hamilton, P.A., Morace, J.L., Nowell, L.H., Rinella, J.F., Stoner, J.D., Wentz, D.A., 1999. The quality of our nation's water: nutrients and pesticides. U.S. Geological Survey Circular 1225, 82 pp.
- Griffith, M.B., Hill, B.H., McCormick, F.H., Kaufmann, P.R., Herlihy, A.T., Selle, A.R., 2005. Comparative applications of indices of biotic integrity based on periphyton, macroinvertebrates, and fish to southern Rocky Mountain streams. *Ecol. Indicators* 5, 117–136.
- Haase, R., Nolte, U., 2008. The invertebrate species index (ISI) for streams in southeast Queensland. *Aust. Ecol. Indicators* 8, 599–613.
- Hering, D., Johnson, R.K., Dram, S., Schmutz, S., Szoszkiewicz, K., Verdonschot, P.F.M., 2006. Assessment of European streams with diatoms, macrophytes, macroinvertebrates, and fish: a comparative metric-based analysis of organism response to stress. *Freshwater Biol.* 51, 1757–1785.
- Herlihy, A.T., Sifneos, J.C., 2008. Developing nutrient criteria and classification schemes for Wadeable streams in the conterminous US. *J. North Am. Benthological Soc.* 27, 932–948.
- Hilsenhoff, W.L., 1987. An improved biotic index of organic stream pollution. *Great Lakes Entomol.* 20, 31–39.
- James, E., Kleinman, P., Veith, T., Stedman, R., Sharpley, A., 2007. Phosphorus contributions from pastured dairy cattle to streams of the Cannonsville Watershed. *J. Soil Water Conserv.* 62, 40–47.
- Justus, B.G., 2003. An index of ecological integrity for the Mississippi Alluvial Plain Ecoregion: index development and relations to selected landscape variables. U.S. Geological Survey Water-Resources Investigations Report 03-4110, 32 pp.
- King, R.S., Richardson, C.J., 2007. Subsidy-stress response of macroinvertebrate community biomass to a phosphorus gradient in an oligotrophic wetland ecosystem. *J. North Am. Benthological Soc.* 26, 491–508.
- Knowlton, K.F., Radcliffe, J.S., Novak, C.L., Emmerson, D.A., 2004. Animal management to reduce phosphorus losses to the environment. *J. Anim. Sci.* 82, 173–195.
- Lange-Bertalot, H., 1979. Pollution tolerance of diatoms as a criterion for water quality estimation. *Nova Hedwigia* 64, 285–304.
- Lavoie, I., Vincent, W.F., Pienitz, R., Painchaud, J., 2004. Benthic algae as bioindicators of agricultural pollution in the streams and rivers of southern Québec (Canada). *Aquat. Ecosyst. Health Manag.* 7, 43–58.
- Lavoie, I., Campeau, S., Darchambeau, F., Cabana, G., Dillon, P.J., 2008. Are diatoms good integrators of temporal variability in stream water quality. *Freshwater Biol.* 53, 827–841.
- Leland, H.V., 1995. Distribution of phyto-benthos in the Yakima River basin, Washington, in relation to geology, land use, and other environmental factors. *Can. J. Fish. Aquat. Sci.* 52, 1108–1129.
- Lenat, D.R., 1993. A biotic index for the southeastern United States: Derivation and list of tolerance values, with criteria for assigning water quality ratings. *J. North Am. Benthological Soc.* 12, 279–290.
- Lowe, R.L., Pan, Y., 1996. Benthic algal communities as biological monitors. In: Stevens, R.J., Bothwell, M.L., Lowe, R.L. (Eds.), *Algal Ecology—Freshwater Benthic Ecosystems*. Academic Press, San Diego, pp. 705–739.
- Lowe, B.S., Leer, D.R., Frey, J.W., Caskey, B.J., 2008. Occurrence and distribution of algal biomass and its relation to nutrients and basin characteristics in Indiana streams. U.S. Geological Survey Scientific Investigations Report 2008-5203, 146 pp.
- Maceina, M.J., Bayne, D.R., 2001. Changes in the black bass community and fishery with oligotrophication in West Point Reservoir, Georgia. *North Am. J. Fish. Manag.* 21 (4), 745–755.
- Maret, T.R., McCoy, D.E., Carlisle, D.M., 2008. Long-term water quality and biological responses to multiple best management practices in Rock Creek, Idaho. *J. Am. Water Res. Assoc.* 44, 1248–1269.
- Miller, R.J., Robison, H.W., 2004. *Fishes of Oklahoma*. University of Oklahoma Press, Norman, p. 450.
- Miltner, R.J., Rankin, E.T., 1998. Primary nutrients and the biotic integrity of rivers and streams. *Freshwater Biol.* 40, 145–158.
- Morgan, A.M., Royer, T.V., David, M.B., Gentry, L.E., 2006. Relationships among nutrients, chlorophyll-*a*, and dissolved oxygen in agricultural streams in Illinois. *J. Environ. Qual.* 35 (4), 1110–1117.
- Moulton, S.R., II, Carter, J.L., Grotheer, S.A., Cuffney, T.F., Short, T.M., 2002. Revised protocols for sampling algal, macroinvertebrate, and fish communities as part of the National Water-Quality Assessment Program. U.S. Geological Survey Open-File Report 02-150, 87 pp.
- Mueller, D.K., Spahr, N.E., 2006. Nutrients in streams and rivers across the Nation—1992–2001. U.S. Geological Survey Scientific Investigations Report 2006-5107, 44 pp.
- Munn, M.D., Hamilton, P.A., 2003. New studies initiated by the U.S. Geological Survey—effects of nutrient enrichment on stream ecosystems. U.S. Geological Survey Fact Sheet FS-118-03, 4 pp.
- National Agriculture Statistical Service, 2008. <http://www.nass.usda.gov/QuickStats> (accessed 30 April 2008).
- National Agriculture Statistical Service, 2008. 2002 Census of Agriculture, <http://www.nass.usda.gov> (accessed May 2008).
- Nguyen, M.L., Sheath, G.W., Smith, C.M., Cooper, A.B., 1998. Impact of cattle treading on hill land. 2. Soil physical properties and contaminant runoff. *N. Z. J. Agric. Res.* 41 (2), 279–290.
- Ortiz, J.D., Puig, M.A., 2007. Point source effects on density, biomass, and diversity of benthic macroinvertebrates in a Mediterranean stream. *River Res. Appl.* 23, 155–170.
- Patton, C.J., Kryskalla, J.R., 2003. Methods of analysis by the U.S. Geological Survey National Water Quality Laboratory—evaluation of alkaline persulfate digestion as an alternative to Kjeldahl digestion for determination of total and dissolved nitrogen and phosphorus in water. Water-Resources Investigations Report 03-4174, 33 pp.

- Petersen, J.C., Justus, B.G., Dodd, H.R., Bowles, D.E., Morrison, L.W., Williams, M.H., Rowell, G.A., 2008. Methods for monitoring fish communities of Buffalo National River and Ozark National Scenic Riverways in the Ozark Plateaus of Arkansas and Missouri: Version 1. U.S. Geological Survey Open-File Report 2007-1302, 94 pp.
- Pflieger, W.L., 1997. The Fishes of Missouri, revised ed. Missouri Department of Conservation, Jefferson City, Missouri, 372 pp.
- Pierson, S.T., Cabrera, M.L., Evanylo, G.K., Kuykendall, H.A., Hoveland, C.S., McCann, M.A., West, L.T., 2001. Phosphorus and ammonium concentrations in surface runoff from grasslands fertilized with broiler litter. *J. Environ. Qual.* 30 (5), 1784–1789.
- Pilati, A., Vanni, M.J., González, M.J., Gaulke, A.K., 2009. Effects of agricultural subsidies of nutrients and detritus on fish and plankton of shallow-reservoir ecosystems. *Ecol. Appl.* 19 (4), 942–960.
- Porter, S.D., 2008. Algal attributes: an autecological classification of algal taxa collected by the National Water-Quality Assessment Program. U.S. Geological Survey Data Series 329, <http://pubs.usgs.gov/ds/ds329/> (accessed May 2009).
- Porter, S.D., Mueller, D.K., Spahr, N.E., Munn, M.D., Dubrovsky, N.M., 2008. Efficacy of algal metrics for assessing nutrient and organic enrichment in flowing waters. *Freshwater Biol.* 53, 1036–1054.
- Potapova, M., Charles, D.F., 2007. Diatom metrics for monitoring eutrophication in rivers of the United States. *Ecol. Indicators* 7, 48–70.
- Rantz et al., 1982. Measurement and computation of streamflow: volume 1. Measurement of stage and discharge. U.S. Geological Survey. Water Supply Paper 2175, v.1, 284 pp.
- Rashleigh, B., 2004. Relation of environmental characteristics to fish assemblages in the upper French Broad River basin, North Carolina. *Environ. Monit. Assess.* 93 (1–3), 139–156.
- Rebich R.A., Demcheck, D.K., 2007. Trends in nutrient and sediment concentrations and loads in major river basins of the South-Central United States, 1993–2004. U.S. Geological Survey Scientific Investigations Report 2007-5090, 112 pp.
- Resh, V.H., 2008. Which group is best? Attributes of different biological assemblages used in freshwater biomonitoring programs. *Environ. Monit. Assess.* 138, 131–138.
- Riseng, C.M., Wiley, M.J., Stevenson, R.J., 2004. Hydrologic disturbance and nutrient effects on benthic community structure in midwestern US streams: a covariance structure analysis. *J. North Am. Benthol. Soc.* 23, 309–326.
- Robins, C.R., Bailey, R.M., Bond, C.E., Brooker, J.R., Lachner, E.A., Lea, R.N., Scott, W.B., 2004. Common and Scientific Names of Fishes from the United States, Canada and Mexico, vol. 29. American Fisheries Society, Bethesda, MD, 386 pp. (Special Publication).
- Robison, H.W., Buchanan, T.M., 1988. Fishes of Arkansas, 5th ed. The University of Arkansas Press, Fayetteville, Arkansas, p. 536.
- Sauer, T.J., Daniel, T.C., Moore Jr., P.A., Coffey, K.P., Nichols, D.J., West, C.P., 1999. Poultry litter and grazing animal waste effects on runoff water quality. *J. Environ. Qual.* 28 (3), 860–865.
- Sistani, K.R., Brink, G.E., Oldham, J.L., 2008. Managing broiler litter application rate and grazing to decrease watershed runoff losses. *J. Environ. Qual.* 37 (2), 718–724.
- Smith, R.A., Alexander, R.B., Schwarz, G.E., 2003. Natural background concentrations of nutrient in streams and river of the conterminous United States. *Environ. Sci. Technol.* 37, 3039–3047.
- Smith, A.J., Bode, R.W., Kleppel, G.S., 2007. A nutrient biotic index (NBI) for use with benthic macroinvertebrate communities. *Ecol. Indicators* 7, 371–386.
- Stevenson, R.J., Rier, S.T., Riseng, C.M., Schultz, R.E., Wiley, M.J., 2006. Comparing effects of nutrients on algal biomass in streams in two regions with different disturbance regimes and with applications for developing nutrient criteria. *Hydrobiologia* 561, 149–165.
- Stevenson, R.J., Hill, B.H., Herlihy, A.T., Yuan, L.L., Norton, S.B., 2008. Algae–P relationships, thresholds, and frequency distributions guide nutrient criterion development. *J. North Am. Benthol. Soc.* 27, 783–799.
- Terrel, J.W., Cade, B.S., Carpenter, J., Thompson, J.M., 1996. Modeling stream fish habitat limitations from wedge-shaped patterns of variation in standing stock. *Trans. Am. Fish. Soc.* 125, 104–117.
- U.S. Environmental Protection Agency, 1998. National strategy for the development of regional nutrient criteria. U.S. Environmental Protection Agency Office of Water Fact Sheet EPA-822-F-98-002, <http://epa.gov/waterscience/criteria/nutrient/> (accessed February 2009).
- U.S. Environmental Protection Agency, 2008. Biological Indicators of Watershed Health, <http://www.epa.gov/bioiweb/html/invertclass.html> (accessed October, 8, 2008).
- U.S. Geological Survey, 2005. Elevation Derivatives for National Applications, <http://edna.usgs.gov> (accessed June 2009).
- U.S. Geological Survey, 2008. National Water Quality Assessment Program, Ecological National Synthesis Project, Fish Traits and Tolerances, U.S. Geological Survey, [http://water.usgs.gov/nawqa/ecology/pubs/FishSppTraits\\_PublicWebpage\\_011404](http://water.usgs.gov/nawqa/ecology/pubs/FishSppTraits_PublicWebpage_011404) (accessed February 2009).
- Vadas, P.A., Harmel, R.D., Kleinman, P.J.A., 2007. Transformations of soil and manure phosphorus after surface application of manure to field plots. *Nutrient Cycling Agroecosyst.* 77 (1), 83–99.
- Van Dam, H., Mertens, A., Sinkeldam, J., 1994. A coded checklist and ecological indicator values of freshwater diatoms from The Netherlands. *Neth. J. Aquat. Ecol.* 28, 117–133.
- Vidon, P., Cambell, M.A., Gray, M., 2008. Unrestricted cattle access to streams and water quality in till landscape of the Midwest. *Agric. Water Manag.* 95, 322–330.
- Wang, W., Robertson, A.E., Garrison, P.J., 2007. Linkages between nutrients and assemblages of macroinvertebrates and fish in Wadeable streams: implication to nutrient criteria development. *Environ. Manag.* 39, 194–212.
- Zaimes, G.N., Schultz, R.C., Isenhardt, T.M., 2008. Total phosphorus concentrations and compaction in riparian areas under different riparian land-uses of Iowa. *Agric. Ecosyst. Environ.* 127, 22–30.
- Zuo, M.H., Miller-Goodman, M.S., 2004. Landscape use by cattle affected by pasture developments and season. *J. Range Manag.* 57, 426–434.