

# DEVELOPMENT AND EVALUATION OF THE LAKE MACROINVERTEBRATE INTEGRITY INDEX (LMII) FOR NEW JERSEY LAKES AND RESERVOIRS\*

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**Abstract.** In response to the recent focus by the U.S. Environmental Protection Agency on bioassessment of lakes, a multimetric index was developed for New Jersey lakes and reservoirs using benthic macroinvertebrates. Benthic samples were collected from reference and impaired lakes with muck and intermediate sediments in central and northern New Jersey during summer 1997. We used a stepwise process to evaluate properties of candidate metrics and selected five for the Lake Macroinvertebrate Integrity Index (LMII): Hilsenhoff Biotic Index (HBI), percent chironomid individuals, percent collector-gatherer taxa, percent oligochaetes/leeches, and number of Diptera taxa. We scored metrics as the fraction of the best expected value (based on all sites) achieved at a site and summed them into the LMII. Evaluation of the LMII showed that it discriminated well between reference and impaired lakes and was strongly related to several potential stressors. Chemical and physical gradients distinguished between reference and impaired lakes, and the LMII summarized these gradients well. The LMII corresponded strongly with land use, but some lakes with more urban land use still achieved high scores. Based on a power analysis, the ability of the LMII to detect differences in condition was sensitive to the number of samples from each lake.

**Keywords:** biotic integrity, lakes, macroinvertebrates, multimetric index, New Jersey

## 1. Introduction

In response to the Clean Water Act of 1972, many methods have been developed and standardized for the assessment of biological condition of water bodies, primarily streams and rivers, based on benthic macroinvertebrates (OEPA, 1987; Resh and Jackson, 1993; DeShon, 1995; Barbour *et al.*, 1996; FDEP, 1996; USEPA, 1998a, b). Many states have developed or are in the process of developing appropriate biocriteria for streams and rivers based on data from bioassessment programs (Davies *et al.*, 1995; Southerland and Stribling, 1995). Bioassessment and biocriteria development allow individual states to evaluate the condition of their water

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resources (e.g., streams) and report the results to Congress. However, only recently has the U.S. Environmental Protection Agency (USEPA) focused on the development of bioassessment protocols and biocriteria for lakes and reservoirs (USEPA, 1998b).

Florida (FDEP, 2000), Maine, Vermont, and Wisconsin (USEPA, 1998b) are among the states developing bioassessment programs for lakes and reservoirs. These states are developing multimetric indices (USEPA, 1998b), which are often used in bioassessment protocols, because they integrate information on diverse biological attributes into a single measure of condition (Barbour *et al.*, 1995). Individual biological metrics are measures of responsive characteristics of a particular assemblage of organisms (i.e., macroinvertebrates, fish, zooplankton, or periphyton), such as pollution tolerance and species richness. Metrics are then standardized and combined into a single index score. In the development of a biotic index for lakes, many natural factors that may affect specific metrics, some of which are not important in streams, should be considered. Some of these factors are water color, ecoregion, impoundments, drainage area, maximum depth, and surface area (USEPA, 1998b).

We developed a multimetric index of biotic integrity for lakes and reservoirs in New Jersey based on benthic macroinvertebrates. The New Jersey Department of Environmental Protection's (NJDEP) biomonitoring program, the Ambient Biological Network (AMNET), includes 763 stream sites throughout the state and uses benthic macroinvertebrates to assess biological condition at these sites (NJDEP, 1998). In contrast, the NJDEP does not routinely monitor its lakes. Most of the existing data on lakes came from the Clean Lakes Program, which is no longer funded. The Clean Lakes Program focused on compiling data generated for specific lakes as part of Clean Lakes Restoration projects, but did not involve statewide routine monitoring (NJDEP, 1998). The lake-specific restoration projects focused on the assessment of lake trophic status and recreational water quality impairment based on total phosphorus, chlorophyll a, and secchi disk transparency. This information was sometimes supplemented with data on excessive macrophytes, bacterial contamination, or sedimentation (NJDEP, 1998). Based on these data, the majority of lakes surveyed in New Jersey are influenced by human activities, and most are considered eutrophic (NJDEP, 1998). The index presented here was developed specifically to assist the NJDEP in developing a routine biomonitoring program and provide a tool to measure the biological condition of lakes throughout New Jersey.

To develop the benthic macroinvertebrate index, we followed a stepwise process using statistical analyses to select a subset of metrics from a large set of candidate metrics. This index was created specifically for lakes and reservoirs dominated by muck or intermediate sediments. In addition, it assumes sampling of the water body in the sublittoral zone, the area just beyond the littoral zone.

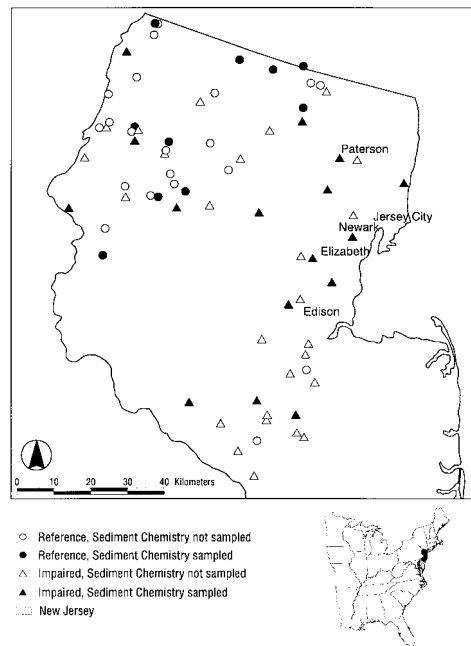


Figure 1. Reference and impaired lakes sampled for benthic macroinvertebrates in New Jersey. Sediment chemistry was sampled in a subset of lakes.

## 2. Methods

### 2.1. STUDY LAKES AND RESERVOIRS

Biologists from USEPA Region II selected the 58 lakes used in this study and designated each as representing either reference or impaired conditions based on best professional judgement. Twenty-one of the lakes had minimal levels of disturbance and were considered reference lakes, while 37 were judged to be impaired, with significant anthropogenic stressors. All of the lakes were in central and northern New Jersey (Figure 1). Most of the reference lakes were in the far northern part of the state, primarily in forested watersheds with low human population densities. Historical monitoring data and field reconnaissance of land use were used to verify the reference and impaired designations. The study lakes ranged in size from approximately 25 to 813 ha. All were considered eutrophic.

Each lake was classified as dominated by muck or intermediate (a mixture of sand and muck) sediments found in the sublittoral zone. Although there are lakes with sandy sediments in New Jersey, only lakes with muck or intermediate sediment types were used in this study because sandy lakes were very different in benthic community composition and only 14 were sampled. There were eight intermediate and 13 muck lakes designated as reference. Fourteen intermediate

and 23 muck lakes were designated as impaired. Due to small sample sizes, data were not further stratified by sediment type for this analysis.

## 2.2. SAMPLE COLLECTION AND ANALYSIS

All lakes were sampled between 23 June and 3 September 1997. At each lake, petite Ponar grab samples (USEPA, 1990, 1997; ASTM, 1998a, b) were collected from the sublittoral zone of ten randomly selected sites and composited into a single benthic macroinvertebrate sample. Each composited benthic sample was wet sieved in the field through a 595  $\mu\text{m}$  mesh screen, and specimens were preserved in 10% formalin. In the laboratory, samples were sorted, and a minimum of 100 organisms was subsampled and identified to the lowest taxonomic level possible, usually to species. Recounting and re-identification was conducted on 10% of macroinvertebrate samples as a quality assurance measure. In September 1997, two lakes were sampled at three locations each to assess spatial and temporal variability.

Dissolved oxygen (DO) and temperature were measured at the surface and bottom, and conductivity, pH, water depth, and secchi depth readings were attempted at or near the deepest point in each lake. However, in some lakes, no accurate measure of secchi depth could be obtained because the secchi disk was visible all the way to the bottom. At other lakes, conductivity, bottom DO, and/or bottom temperature were not measured due to instrument failure. At 22 of the lakes (Figure 1), sediment samples were collected in September 1997 at or near the deepest location in the lake using a hand-operated Ponar dredge. Five grabs at each lake were composited into a single sample per lake. The sediment samples were analyzed for cadmium (Cd), chromium (Cr), silver (Ag), aluminum (Al), lead (Pb), copper (Cu), nickel (Ni), zinc (Zn), iron (Fe), and magnesium (Mg) using inductively coupled plasma emission spectroscopy (USEPA, 1994b). Selenium (Se), arsenic (As), and antimony (Sb) were analyzed using hybrid generation flow injection atomic absorption (USEPA, 1995), and mercury was analyzed using a cold vapor technique (USEPA, 1994a). Collection of sediment chemistry data was limited to a subset of lakes due to cost constraints. At these same 22 lakes, water samples were collected at the time of other limnological and macroinvertebrate sampling to assess sulfate concentrations (USEPA, 2000c) and total suspended solids (TSS) (USEPA, 2000b). Quality assurance and control procedures were based on those in Region II laboratory QA/QC plans and included analysis of one blank, two duplicates, and two spiked samples for every 20 or less samples (USEPA, 1993, 2000a).

## 2.3. CHARACTERIZATION OF REFERENCE AND IMPAIRED LAKES

The distributions of limnological and sediment chemistry variables were examined to identify those potential stressors that differed between reference and impaired lakes. The chi-square approximation to the nonparametric Mann-Whitney test was

used to test for statistically significant differences in characteristics between reference and impaired lakes.

#### 2.4. METRIC EVALUATION

We calculated 33 candidate metrics based on species richness, species composition, pollution tolerance, and functional feeding groups (Table I). Candidate metrics were chosen by reviewing the literature for those that would be appropriate for lakes (Resh and Jackson, 1993; Barbour *et al.*, 1996; USEPA, 1998b; Lewis *et al.*, 2001). Richness metrics can be calculated based on either distinct taxa only or all taxa identified. We used only distinct taxa to calculate richness metrics. Functional feeding groups were determined for taxa based on literature (Green, 1990; Merritt and Cummins, 1996; USEPA, 1999).

Metrics describing pollution tolerance characteristics were based on pollution tolerance values (PTV) for each taxon. These PTVs were determined by finding from the literature (Green, 1990; Lewis and Klemm, 1990; Lenat, 1993; USEPA, 1999) and/or professional observation and judgement, the tolerances of taxa to different types of stressors (acidity, domestic waste, organics, heavy metals, agriculture, and sedimentation) and averaging the values (USEPA NERL-Cincinnati unpublished report). One pollution tolerance metric we considered, the Hilsenhoff Biotic Index (HBI), was a modification of the original HBI. While the original HBI was based on the tolerances of different taxa to organic pollution (Hilsenhoff, 1987; Lenat, 1993), the version of the metric that we evaluated gauges the community tolerance to several types of pollutants because the PTVs were measures of average tolerance across a variety of pollutant types.

Several metrics were eliminated immediately due to sparseness of data (i.e., most of the values were zero) (Table II). A stepwise process based on USEPA technical guidance (1998) for establishing biocriteria for lakes and reservoirs was used to evaluate the remaining metrics for use in a multimetric index. Four characteristics were evaluated for each metric: 1) discriminatory power, 2) relative scope of impairment, 3) redundancy, and 4) relationship with stressors.

##### 2.4.1. *Discriminatory Power*

We defined discriminatory power of a metric as the ability of that metric to distinguish between reference and impaired lakes and evaluated metrics by examining their distributions using box-and-whisker plots. The degree of overlap between interquartile (IQ) ranges (the box) of reference and impaired lakes was considered a signal of the discriminatory capability of the metric. Using the system developed by Barbour *et al.* (1996), metrics with no overlap of IQ ranges were assigned a score of '3', those with both medians outside the other's IQ range received a score of '2', those with only 1 median outside the other's IQ range received a score of '1', and those with overlap of each median with the other's IQ range scored a '0'.

TABLE I

Definitions and expected direction of response under stress for candidate metrics for New Jersey lakes

Metric type	Metric	Definition	Response to stress
Richness measures	Total number of taxa	Number of distinct taxa in sample	decrease
	Simpson's Diversity Index	Probability that 2 randomly chosen individuals will belong to the same taxon	increase
	Shannon Diversity Index	Incorporates diversity and evenness	decrease
	No. of chironomid taxa	Number of taxa of Chironomidae (midge) larvae	decrease
	No. of Diptera taxa	Number of Diptera (fly) taxa	decrease
	No. of ETO taxa	Number of taxa in the orders Ephemeroptera (mayflies), Trichoptera (caddisflies) and Odonata (dragonflies, damselflies)	decrease
Tolerance measures	No. intolerant taxa	Number of taxa with Pollution Tolerance Value (PTV) < 4	decrease
	No. facultative taxa	Number of taxa with $PTV \geq 4$ and $\leq 6$	decrease
	No. tolerant taxa	Number of taxa with $PTV > 6$	increase
	HBI	$\sum_i p_i t_i$ , where $p_i$ is the proportion of individuals in taxon $i$ and $t_i$ is the PTV for taxon $i$	increase
	% Intolerant taxa	Percentage of taxa with $PTV < 4$	decrease
	% Facultative taxa	Percentage of taxa with $PTV \geq 4$ and $\leq 6$	decrease
	% Tolerant taxa	Percentage of taxa with $PTV > 6$	increase
	% Intolerant individuals	Percentage of individuals in taxa with $PTV < 4$	decrease
	% Facultative individuals	Percentage of individuals in taxa with $PTV \geq 4$ and $\leq 6$	decrease
% Tolerant individuals	Percentage of individuals in taxa with $PTV > 6$	increase	
Composition measures	% Dominant taxon	Percentage of individuals in the single most dominant taxon	increase
	No. individuals per taxon	(Total number of individuals)/(Total number of distinct taxa)	increase
	% Diptera individuals	Percentage of individuals in Diptera taxa	decrease
	% Chironomid individuals	Percentage of individuals in Chironomidae	decrease

TABLE I  
(continued)

Metric type	Metric	Definition	Response to stress
Composition measures	% Non-insects	Percentage of individuals in non-insect taxa	increase
	% Oligochaetes and leeches	Percentage of individuals in Oligochaeta and Hirudinea	increase
	% Amphipoda	Percentage of individuals in Amphipoda	decrease
	% Crustacea and Mollusca	Percentage of individuals in Crustacea and Mollusca	decrease
	% Chironomid taxa	Percentage of taxa in Chironomidae	decrease
	% Diptera taxa	Percentage of taxa in Diptera	decrease
Trophic measures	No. of predator taxa	Number of taxa as predators	variable
	No. of collector-gatherer taxa	Number of taxa as collector-gatherers	decrease
	% Predator taxa	Percentage of taxa as predators	variable
	% Collector-gatherer taxa	Percentage of taxa as collector-gatherers	decrease
	% Predator individuals	Percentage of individuals in predator taxa	variable
	% Collector-gatherer individuals	Percentage of individuals in collector-gatherer taxa	decrease
	% Collector-filterer individuals	Percentage of individuals in collector-filterer taxa	decrease

Thus, metrics scoring 2 or 3 showed the ability to discriminate between reference and impaired lakes, and only these were retained for further analysis.

#### 2.4.2. Relative Scope of Impairment

Relative scope of impairment is a measure of the ease of detecting impairment compared to some ideal condition (USEPA, 1998b). Using only values from reference lakes, we evaluated the relative scope of impairment using the IQ coefficient for each metric as

$$(q^{0.75} - q^{0.25}) / (q^{\alpha} - q^{\beta}),$$

where  $q^{\alpha}$  and  $q^{\beta}$  are the  $\alpha$ th and  $\beta$ th percentiles of the distribution among reference lakes and  $q^{0.75} - q^{0.25}$  represents the IQ range among reference lakes (USEPA, 1998b; Barbour *et al.*, 1996). For metrics that decrease with stress (e.g., total number of taxa),  $\alpha = 0.25$  and  $\beta = 0$  (minimum). For metrics that increase with

TABLE II

Step in metric evaluation process at which each metric failed to meet criteria, if any. Blank values indicate all metric evaluation criteria were met

Metric	Reason for elimination
Total number of taxa	Poor discriminatory power
Simpson's Diversity Index	Poor discriminatory power
Shannon Diversity Index	Poor discriminatory power
No. of chironomid taxa	
No. of Diptera taxa	
No. of ETO taxa	Data too sparse
No. intolerant taxa	Data too sparse
No. facultative taxa	Poor relative scope of impairment
No. tolerant taxa	Poor discriminatory power
<b>HBI</b>	
% Intolerant taxa	Data too sparse
% Facultative taxa	
% Tolerant taxa	
% Intolerant individuals	Data too sparse
% Facultative individuals	
% Tolerant individuals	
% Dominant taxon	Poor discriminatory power
No. individuals per taxon	Poor discriminatory power
% Diptera individuals	
% Chironomid individuals	
% Non-insects	
% Oligochaetes and leeches	
% Amphipoda	Data too sparse
% Crustacea and Mollusca	Data too sparse
% Chironomid taxa	Poor discriminatory power
% Diptera taxa	
No. of predator taxa	Poor discriminatory power
No. of collector-gatherer taxa	
% Predator taxa	Poor discriminatory power
% Collector-gatherer taxa	
% Predator individuals	Poor relative scope of impairment
% Collector-gatherer individuals	Poor relative scope of impairment
% Collector-filterer individuals	Data too sparse

stress (e.g., HBI),  $\alpha = 1.00$  (maximum) and  $\beta = 0.75$  (USEPA, 1998b; Barbour *et al.*, 1996). Metrics with an IQ coefficient greater than 1 have a small relative scope of impairment, such that variability among reference sites is greater than the potential range of impairment, and these were excluded from further consideration.

#### 2.4.3. *Relationships to Stressors*

The remaining metrics were evaluated to find those related to potential stressors. The potential stressors included limnological variables, such as secchi depth, DO, and TSS, and sedimentary metals (Table III). Significant Spearman rank correlations ( $p < 0.01$ ) and scatter plots were used to identify important stressor-metric relationships. Only metrics showing a relationship with at least one potential stressor were retained.

#### 2.4.4. *Redundancy*

We evaluated redundancy among metrics to ensure that each metric in the final index provides new information. Using the remaining metrics, Pearson correlation coefficients and scatter plots were used to identify highly correlated metrics. A simple correlation alone is not considered sufficient to regard two metrics as redundant (USEPA, 1998b). The USEPA guidance (1998) suggests that usually a tight correlation ( $r > 0.9$ ) and a linear relationship is necessary to consider two metrics redundant. Pairs of metrics with lower correlation coefficients usually showed enough scatter or nonlinearity in scatter plots to indicate that each metric provided some new information. We selected one metric from each group of redundant metrics. We retained the one that had the highest stressor correlations and the best discriminatory power of the group for the scoring and final selection steps.

### 2.5. SCORING OF METRICS

Two general methods of scoring metrics have been developed: 1) those based on metric distributions under reference conditions and 2) those based on metric distributions across all sites (Simon and Lyons, 1995). Because lakes classified as reference did not represent true reference conditions, but were rather only least disturbed for the region at this point in time, we followed guidance on the creation of biocriteria for lakes that suggests setting expectations for scoring metrics based on the distribution across all lakes (USEPA, 1998b).

Rather than using the maximum or minimum values as the best expected value, we used the 95th or 5th percentile value because this method avoids using anomalously high or low outliers as the best expected value (USEPA, 1999). The 95th percentile value was set as the best expected value for metrics that decreased in response to stress (e.g., percent collector-gatherer taxa). The score of the metric was calculated as the fraction of 95th percentile value (metric value/95th percentile value). For metrics that increased in response to stress (e.g., percent oligochaetes and leeches), the 5th percentile value was used as the best expected value (USEPA,

TABLE III  
Limnological and sediment chemistry characteristics of reference and impaired lakes

Variable (units)	Reference			Impaired		
	Median	Range	Interquartile range	Median	Range	Interquartile range
Maximum depth (m) <sup>a</sup>	3.9	1-28	3.2-5.7	2.4	1-8.1	1.7-3.3
Surface temp. (degrees C)	25.5	22-29	25-27	25.5	18-32	24-27
Bottom temp. (degrees C)	23	5.5-27	21-24.5	21.5	14.5-27.5	18.75-24
Surface DO (mg L <sup>-1</sup> ) <sup>a</sup>	8	6.4-10.1	7.8-8.5	8.5	4.6-18.2	7.6-10.6
Bottom DO (mg L <sup>-1</sup> )	4.5	0.7-9.2	0.9-7.5	2.4	0.6-18.3	0.8-6.35
Secchi depth (m) <sup>a</sup>	2	1-3.5	2-3	1	0.5-2	0.5-1.5
pH	7.1	5.8-8.7	6.2-8.1	7.6	4.1-9.3	6.5-8.5
Conductivity ( $\mu$ mhos) <sup>a</sup>	140	25-390	50-250	220	30-788	135-400
Total alkalinity (mg L <sup>-1</sup> )	32	17-204	24-64	52	8-157	36-120
Sulfate (mg L <sup>-1</sup> ) <sup>a</sup>	7.8	6.8-16.2	7.1-13.1	19.3	4-61.2	9.2-30
Total suspended solids (mg L <sup>-1</sup> ) <sup>a</sup>	2	2-2.6	2-2.2	7.6	3.6-23.7	4.8-13.2
Silver (mg kg <sup>-1</sup> ) <sup>a</sup>	0.3	0.1-0.8	0.2-0.5	1.4	0.1-9.3	0.4-5.3
Aluminum (mg kg <sup>-1</sup> ) <sup>a</sup>	23300	13600-37800	16500-26000	30200	17500-58200	21600-42100
Arsenic (mg kg <sup>-1</sup> )	14	4-31	7-18	9	2-27	5-14
Cadmium (mg kg <sup>-1</sup> )	1.4	0.4-4.3	0.6-1.6	1.4	0.3-14.6	0.8-1.8
Chromium (mg kg <sup>-1</sup> ) <sup>a</sup>	26	12-34	25-30	47	19-68	33-65
Copper (mg kg <sup>-1</sup> )	40	17-129	34-74	87	14-679	36-141
Iron (mg kg <sup>-1</sup> )	26100	17700-36300	20900-33700	29000	9360-48200	22700-39200
Mercury (mg kg <sup>-1</sup> )	0.21	0.14-0.48	0.17-0.22	0.26	0.1-0.84	0.18-0.31
Manganese (mg kg <sup>-1</sup> )	502	128-1550	271-708	479	191-884	437-774
Nickel (mg kg <sup>-1</sup> ) <sup>a</sup>	22	9-29	19-27	28	13-41	23-36
Lead (mg kg <sup>-1</sup> )	122	2-429	25-216	106	29-390	36-240
Antimony (mg kg <sup>-1</sup> )	1.7	0.7-4	1-2.4	1.1	0.4-3	0.8-2
Selenium (mg kg <sup>-1</sup> ) <sup>a</sup>	3	2-6	2.2-5	0.8	0.2-5	0.5-2
Zinc (mg kg <sup>-1</sup> )	229	91-519	197-289	319	103-604	194-409

<sup>a</sup> Reference and impaired lakes differed significantly ( $p$ -value based on chi-squared approximation of Mann-Whitney test was  $< 0.10$ ).

1999). For those metrics, the score was calculated as the ratio of the difference between the maximum value and the value for a given lake and the difference between the maximum value (100% for all percentage metrics) and the 5th percentile value (i.e.,  $(\text{max} - \text{metric value})/(\text{max} - 5\text{th percentile value})$ ). Thus, a higher metric score was indicative of better biological condition. Metric scores were truncated to a range between 0 and 1. The 95th and 5th percentile values were rounded to the nearest 0.1 for use in the index, except for the 5th percentile for the HBI, which was rounded to the nearest 0.05.

## 2.6. FINAL SELECTION OF METRICS

For the final selection of metrics, we initially included all with the highest discriminatory power (no overlap of interquartile ranges between impaired and reference lakes). We further examined the types of metrics retained by this step to avoid over-representation of a single group of organisms in the index. From among the remaining metrics, we selected those that increased discriminatory power the most. We first added each remaining metric score to the sum of those already selected. We then used a Wilcoxon 2-sample test with a normal approximation and continuity correction (SAS Institute, 1990) to compare sums from reference and impaired lakes. Because all tests were already highly significant, we selected the metric that increased the z-score the largest amount beyond that for previously selected metrics. The sum of those five selected metrics was then calculated and the process repeated until further metrics failed to increase the discriminatory power of the index.

A final multimetric index of biotic integrity was created by summing selected metric scores into the Lake Macroinvertebrate Integrity Index (LMII), which could range from zero to a value equal to the number of metrics used. We then used Spearman rank correlations to determine the relationship between the LMII and potential stressors.

## 2.7. RELATIONSHIP WITH LAND USE

Land cover data were derived from leaves-on and leaves-off Landsat satellite thematic mapper (TM) scenes acquired from 1991 to 1993, which were projected to Lambert Azimuthal coordinates (Vogelmann *et al.*, 1998). The 30 m<sup>2</sup> picture elements (pixels) of the Landsat scenes were clustered into 100 spectrally distinct classes using an unsupervised clustering algorithm (Kelly and White, 1993). Aerial photographs and supplemental data were used to interpret the classes and assign them one of 15 land cover category labels. Using Global Positioning System (GPS) latitude and longitude data, each lake was placed into the appropriate 14-digit hydrological unit code (HUC) unit for New Jersey. Then, Geographic Information System (GIS) overlay techniques were used to determine the percentages of agricultural, urban (i.e., commercial and residential), and forested land use in each

HUC containing one of the lakes sampled. We then ran a correlation analysis of land use data with the LMII and its component metric scores.

## 2.8. MULTIVARIATE ANALYSES

A Principal Components Analysis (PCA) (Johnson and Wichern, 1992) was run to assess the effectiveness of the LMII in capturing variability in the benthic community. Using the power transformation given by the Box-Cox solution (Johnson and Wichern, 1992), we achieved approximate normality for each metric included in the analysis. The full set of metrics, which included all candidate metrics remaining after those with sparse data were removed, was used to run PCAs on correlation matrices. A correlation analysis was run using the first two principal component axes and the LMII scores. This analysis was intended to indicate how well the LMII represents the variability in the data.

A second PCA was run using limnological and sediment chemistry data, transformed to approximate normality, to determine the relationship between physical and chemical parameters and lake designations (i.e., reference or impaired). Complete data were available from only 18 lakes for this analysis. The first two principal component axes were used in a simple linear regression to predict the LMII scores. The results of this regression served as an indication of how well the LMII reflects chemical and physical gradients.

## 2.9. POWER ANALYSIS

To assess the ability of the final index to distinguish sites or conditions, we ran power analyses based on estimates of temporal and spatial variability. There were only two lakes that were visited twice, with only one sample collected during the first visit and three samples collected during the second visit. Therefore, we estimated temporal variability for each of the two lakes by calculating the variance of the first visit with each replicate of the second visit for each lake. We calculated the average variance for each lake and used the highest average variance. To estimate spatial variability, we calculated the variance for each of the two lakes that had three replicate samples each. We used the higher of the two variance values as our estimate. We then calculated the minimum significant difference (MSD) in scores required using each variance estimate, assuming a type I error rate of 0.05 and a power of 0.80 (Neter *et al.*, 1996). We calculated the MSD using sample sizes of four (two samples per lake) and six (three samples per lake). The MSD indicates how different in scores two lakes need to be to conclude they are different from one another in condition.

### 3. Results

#### 3.1. CHARACTERIZATION OF REFERENCE AND IMPAIRED LAKES

Several limnological and sediment chemistry variables showed differences in distribution between reference and impaired lakes (Table III). Among the limnological variables, maximum depth, secchi depth, and TSS differed the most between reference and impaired lakes (all  $p$ -values  $< 0.01$ , Table III). The most discriminating chemistry variables were Cr and Se, with no overlap of IQ ranges between impaired and reference lakes (Table III).

#### 3.2. METRIC EVALUATION

##### 3.2.1. *Discriminatory Power*

Several metrics distinguished between reference and impaired lakes (box plot scores of 2 or 3) for both muck and intermediate sediment lakes. Only a few of the metrics with good discriminatory power differed significantly ( $p < 0.05$ ) between intermediate and muck reference lakes (percent tolerant taxa, percent facultative taxa). Because data by sediment type were limited, we decided not to treat intermediate and muck lakes separately. Combining data from muck and intermediate sediment lakes, 17 metrics scored a 2 or 3 in discriminatory power between reference and impaired lakes (Table II).

##### 3.2.2. *Relative Scope of Impairment*

Of the remaining metrics, only three had IQ coefficients over 1, indicating poor relative scope of impairment (Table II). We eliminated these metrics, and retained 14 metrics for further evaluation.

##### 3.2.3. *Relationships to Stressors*

All of the 13 remaining metrics were highly correlated with secchi depth, and most were correlated with Cr and Se concentrations (Table IV). A few were correlated with conductivity, total alkalinity, TSS, hardness, and sulfate. Percent and number of collector-gatherer taxa were correlated with the largest number of chemistry and limnological variables. No metrics were eliminated at this step.

##### 3.2.4. *Redundancy*

Among these 14 metrics, several pairs or groups were highly correlated and considered redundant, including HBI, percent facultative individuals, and percent tolerant individuals (all  $|r| \geq 0.932$ ,  $p < 0.001$ ). Percent tolerant taxa and percent facultative taxa were redundant ( $r = -0.985$ ,  $p < 0.001$ ) as were percent oligochaetes and leeches, percent dipteran individuals, and percent non-insects (all  $|r| > 0.95$ ,  $p < 0.001$ ). Finally, number of chironomid taxa was redundant with number of dipteran taxa ( $r = 0.983$ ,  $p < 0.001$ ), as expected.

TABLE IV

Highest 3 significant ( $p < 0.01$ ) correlations between each metric and limnological and sediment chemistry variables

Metric	Highest correlations (r, n)		
% Chironomid indiv.	Se (0.768, 22)	Cr (-0.702, 22)	Secchi (0.584, 51)
% Diptera indiv.	Se (0.791, 22)	Secchi (0.702, 51)	Cr (-0.637, 22)
No. chironomid taxa	Se (0.638, 22)	Cr (-0.605, 22)	TSS (-0.575, 22)
No. Diptera taxa	Se (0.651, 22)	Cr (-0.621, 22)	TSS (-0.612, 22)
% Diptera taxa	Se (0.711, 22)	Cr (-0.599, 22)	Secchi (0.588, 51)
% Non-insect indiv.	Se (-0.800, 22)	Secchi (-0.708, 51)	Cr (0.656, 22)
% Oligochaetes and leeches	Se (-0.838, 22)	Cr (0.714, 22)	Secchi (-0.697, 51)
HBI	Se (-0.820, 22)	Cr (0.766, 22)	Secchi (-0.693, 51)
% Facultative taxa	Se (0.762, 22)	Cr (-0.528, 22)	Fe (-0.523, 22)
% Facultative indiv.	Se (0.832, 22)	Cr (-0.690, 22)	Secchi (0.626, 51)
% Tolerant taxa	Se (-0.789, 22)	Cr (0.560, 22)	Fe (0.537, 22)
% Tolerant indiv.	Se (-0.832, 22)	Cr (0.690, 22)	Secchi (-0.633, 51)
No. collector-gath. taxa	TSS (-0.774, 22)	Secchi (0.705, 51)	Sulfate (-0.616, 22)
% Collector-gath. taxa	Sulfate (-0.686, 22)	TSS (-0.648, 22)	Secchi (0.646, 51)

We selected the HBI, number of Diptera taxa, percent oligochaetes and leeches, and percent tolerant taxa from the four groups of redundant metrics. These metrics were chosen as candidates for final selection because they had the best discriminatory power and the highest stressor correlations within each group. Four other metrics that were not redundant with any metrics were also candidates for final selection, leaving a total of eight metrics as candidates for the index.

### 3.3. SCORING OF METRICS

The rounded values of the 5th and 95th percentiles are provided in Table V. For the HBI, the true maximum among lakes in this analysis, 8.75, was used as the maximum value for scoring purposes. Thus, any lakes with HBI values above this value would receive a score of 0 for that metric.

### 3.4. FINAL SELECTION OF METRICS

Only eight metrics remained at this step. We first considered the five metrics with the highest overall discriminatory power. We excluded number of collector-gatherer taxa, which was among the five metrics, for several reasons. Although this metric qualified in terms of discriminatory power, relative scope of impairment, and stressor relationships, it had a 95th percentile of only six taxa, resulting in a limited

TABLE V

Rounded 95th and 5th percentiles used in calculating metric scores. Value is 95th percentile unless otherwise noted

Metric	Target value
Percent Diptera taxa	100
Number of Diptera taxa	15
Percent chironomid indiv.	90.7
Percent oligochaetes/leeches	0 <sup>a</sup>
Number of collector-gatherer taxa	6
Percent collector-gatherer taxa	50.0
Percent tolerant taxa	36.4 <sup>a</sup>
HBI	4.65 <sup>a</sup>

<sup>a</sup> 5th percentile value.

TABLE VI

Formulas for calculation of LMII metric scores, based on values in Table V. Calculated scores greater than 1 were truncated to 1

Metric	Equations for scores
Number of Diptera taxa	No. Diptera taxa/15
Percent chironomid individuals	% Chironomid indiv./90.7
Percent oligochaetes/leeches	(100 – % Oligochaetes/leeches)/100
Percent collector-gatherer taxa	(% Collector-gatherer taxa)/50
HBI	(8.75 – HBI)/(8.75 – 4.65)

ability to score it effectively. In addition, percent collector-gatherer taxa, representing the same population component, was preferred because it was the only metric correlated with conductivity and alkalinity and had a smaller IQ coefficient (larger relative scope of impairment). Number of Diptera taxa was added to the other four selected metrics because only this metric increased the discriminatory power of the index ( $\Delta z$ -score = +0.15). Formulas for calculation of the LMII are provided in Table VI.

Because of a difference in maximum depth between reference and impaired lakes, we also examined the relationship between depth and the scores for the LMII and its metrics. Again, we found no apparent relationship between scores and depth. For this reason and because the benthic samples were not collected from the deepest part of a lake but rather from randomly selected sites in the sublittoral zone, we felt that adjusting scoring for maximum depth was not necessary.

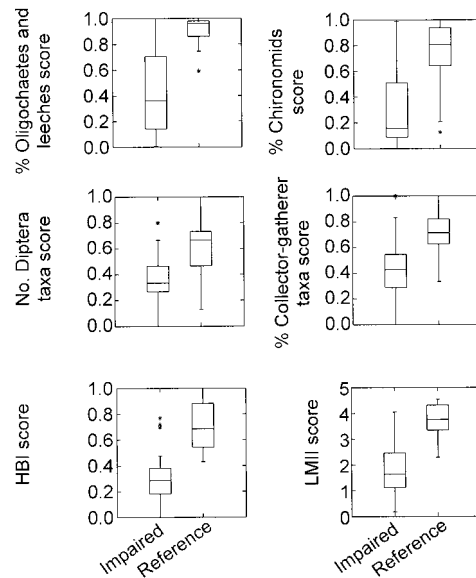


Figure 2. Discriminatory capability of the LMII and its component metrics.

The scores for each selected metric generally had good to excellent discriminatory power (2 or 3) (Figure 2). The LMII itself had excellent discriminatory power between reference and impaired lakes. The LMII was correlated strongly with secchi depth ( $r = 0.744$ ), Cr ( $r = -0.755$ ), and Se ( $r = 0.813$ ), moderately with TSS ( $r = -0.568$ ), and weakly with conductivity ( $r = -0.311$ ) and total alkalinity ( $r = -0.298$ ).

### 3.5. RELATIONSHIP WITH LAND USE

Over 75% of lake watersheds examined contained less than 5% agricultural land use, while percent urban land use was less sparse and strongly negatively correlated with percent forested watershed ( $r = -0.85$ ). The LMII was significantly correlated with percent urban area (Figure 3) and percent forested area in the watershed ( $r = -0.578$  and  $r = 0.672$ , respectively; both  $p$ -values  $< 0.001$ ) but not with percent agriculture. All of the metric scores were significantly correlated with both percent urban and percent forested watershed ( $p < 0.005$ ) except percent collector-gatherer taxa, which was slightly less correlated with urban and forested land use ( $p = 0.008$  and  $p = 0.027$ , respectively).

### 3.6. MULTIVARIATE ANALYSES

The first principal component axis based on candidate metrics, representing 45.2% of the variation in the data, had relatively large loadings ( $|\text{loading}| > 0.22$ ) for most of the metrics selected for the index. Only percent collector-gatherer taxa had

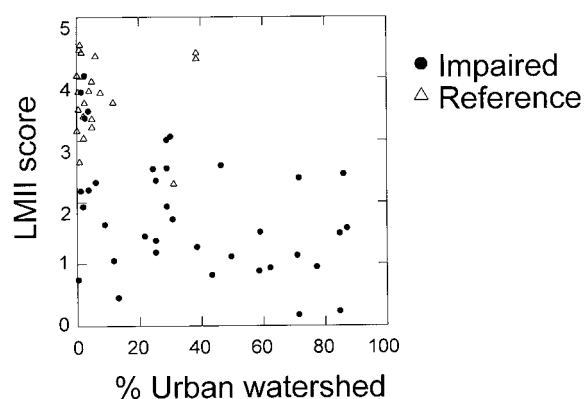


Figure 3. Relationship between percent urban land use in watershed and LMII scores.

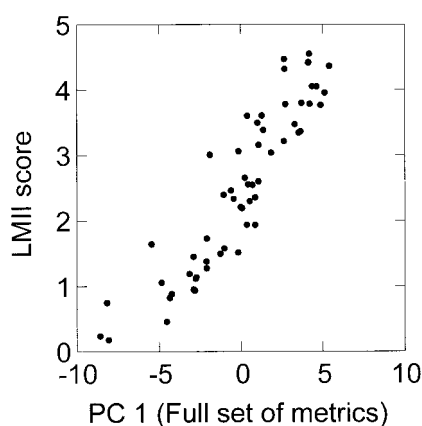


Figure 4. Relationship between the first principal component for the full set of candidate metrics and the LMII scores.

a slightly lower loading than the other index metrics (0.17). The second principal component axis represented approximately 20% of the variation, and the highest loadings all consisted of metrics with little or no discriminatory power, such as total number of taxa, Simpson's Diversity Index, and percent dominant taxon. The LMII scores were highly correlated with the first principal component ( $r = 0.909$ , Figure 4), though not with the second principal component ( $r = -0.309$ ). Thus, the LMII seems to represent the largest portion of variation in the benthic community well.

The PCA on limnological and chemical variables revealed that two axes could explain approximately 50% of the variation. The axes were complex, with a separation along the first axis based on secchi depth, TSS, sulfate, Ni, Cr, and Zn. The second axis was primarily related to pH, total alkalinity, As, and Sb. However, the reference and impaired lakes clearly separated based on these two gradients

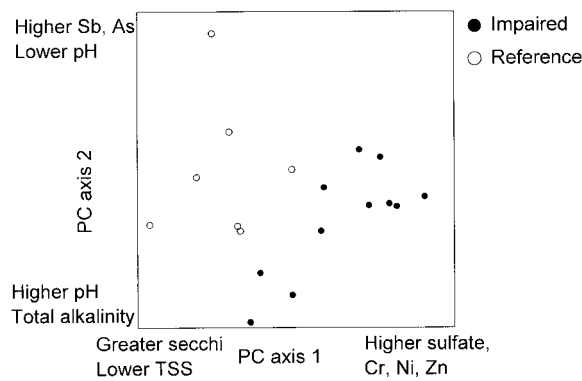


Figure 5. Separation of lakes along first 2 principal component axes based on chemical and limnological variables.

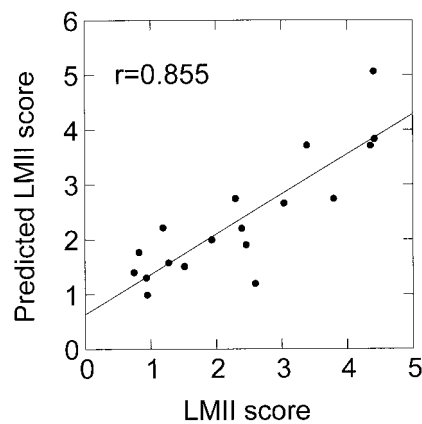


Figure 6. Relationship between predicted LMII score based on physical and chemical gradients (PCA axes) and actual LMII score.

(Figure 5). In addition, these two chemical and physical gradients were able to predict the LMII score reasonably well ( $R^2 = 0.694$ ) (Figure 6).

### 3.7. POWER ANALYSIS

The estimate of temporal variance (0.117) was about 25% larger than that for spatial variance (0.0935). The calculated MSD values based on two samples per lake were about 50% larger than those based on three samples per lake (Table VII).

## 4. Discussion

Based on existing data, the developed index performs well for New Jersey lakes and reservoirs with muck and intermediate sediments that have eutrophication as

TABLE VII

Results of power analysis based on a type I error rate ( $\alpha$ ) of 0.05 and a power of 0.80

	Variance estimate	Samples/ lake	d.f.	Minimum significant difference (MSD)
Temporal	0.117	2	2	2.05
	0.117	3	4	1.37
Spatial	0.094	2	2	1.84
	0.094	3	4	1.23

the primary stressor. This index is a starting point in terms of lakes bioassessment and biocriteria development for New Jersey, but additional data are necessary to determine the long-term reliability and usefulness of the LMII. In the short term, this index will provide resource managers with a tool to assess the biological condition of reservoirs and lakes in New Jersey. Meanwhile, the data they gather can be used to test repeatability of the index and refine the scoring criteria. The index is currently calibrated for use only in New Jersey lakes and focuses on expectations set by the best conditions existing among this set of lakes. A comparison of reference conditions among New Jersey lakes and reservoirs with those among other potentially more pristine lakes in the northeast United States is desirable and necessary to gauge the overall condition of lakes in New Jersey. However, data collected in a similar fashion from other lakes in the northeast are currently not available for comparison.

The results of the power analysis indicate that the LMII does have the ability to detect changes in condition at a given lake through time or differences between two lakes. However, this ability is sensitive to the number of samples collected from a given lake. Increasing the number of samples from two to three per lake or per year at a given lake greatly increases the ability of the LMII to detect differences. Thus, sampling frequency and replication are important issues to consider with the use of this index.

This index also relies on identification of specimens to the lowest possible taxon (species level whenever possible). This may present an obstacle for use of the LMII among state agencies that currently identify organisms with lower resolution or lack personnel with the appropriate expertise to identify organisms to genus and species. More time per sample and additional training would likely be required. However, identification of most specimens to a higher taxonomic level (i.e., family or class) would require redevelopment of the index because many of the metric characteristics evaluated (i.e., discriminatory power) would probably change,

and some of the metrics would no longer be appropriate (i.e., number of chironomid taxa). Without further testing, we cannot determine just how genus-level identification might affect this index, if at all.

The LMII scores were most highly correlated with Se concentrations in the sediments, and this correlation was positive. Concentrations among reference sites were at levels presenting a high hazard to fish and aquatic birds from accumulation into the benthic food chain (Lemly, 1996). These results seem to indicate that reference lakes tended to be contaminated with Se but were otherwise relatively undisturbed. However, the soils in the region of the state containing most of the reference sites are somewhat high in Se naturally (Jonathon Kennen, USGS, pers. comm.), so the Se measured may not actually be available to the benthos. Because our methods of measuring total Se did not separate different forms, we do not know if the Se may have been present in the form of inorganic selenides, which are insoluble and not considered a hazard (Maier and Knight, 1993), or in some other form. In addition, sediment samples were collected near the deepest part of each lake or reservoir, not where the biological samples were collected. Thus, the sedimentary Se concentrations may not represent concentrations where the majority of biota reside.

Sediment type appears to be an important classification factor for the purpose of biocriteria development. The LMII is intended only for lakes and reservoirs with a sublittoral zone dominated by muck and intermediate sediment types. In Florida lakes, early data suggested that species composition in lakes was strongly related to sediment type (FDEP, 2000). Thus, the most responsive set of metrics might differ among lakes of different sediment types. Too few reference and impaired lakes in New Jersey with sand-dominated sediments were sampled to be included in this analysis. Thus, additional data would be required to create a separate index for sandy lakes.

Further refinement of the existing LMII incorporating classification of lakes by muck and intermediate sediment types could be examined with more data, but there are potential problems with this approach. Although the benthic community was related to sediment type in Florida lakes, the FDEP ultimately decided not to classify lakes by dominant sediment (FDEP, 2000). The primary concern was that impaired lakes might have a higher percentage of muck sediments as a result of anthropogenic eutrophication and not due to natural factors (FDEP, 2000). Thus, in New Jersey lakes, this division may not be a natural one either.

Finally, this index does not take into account lake origin, but this factor may affect calibration and performance of the index. There can be no truly reference reservoirs, and age of a reservoir may be very influential on the biological condition of even relatively undisturbed reservoirs (USEPA, 1998b). This data set was relatively small and not suited to development of an index using a classification of lakes based on origin, but this could be the focus of future studies.

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