Development of Bioassessment Criteria for Headwater Streams, Phase III

ANS Project Number 566

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Summary

The Academy of Natural Sciences of Drexel University (ANS) and the New Jersey Department of Environmental Protection (NJDEP) have been developing an Index of Biotic Integrity for headwater streams since 2004. Past studies have identified several aspects of amphibian, crayfish, and fish assemblages which were correlated with environmental variables. To further this work, in 2009, ANS set out to sample additional reference streams; determine the range and applicability of metrics with respect to low gradient streams and streams with upstream ponds; refine salamander sampling techniques; and validate and refine previously proposed candidate metrics.

Sites were selected based on environmental variables: land use, land cover, reach gradient, pond/lake presence, and drainage area (typically <12.95 km² [5 mi²]). Field and fish sampling procedures generally followed those of the NJ DEP Bioassessment Unit. New methods for sampling amphibians were developed and tested. Canonical Correspondence Analysis (CCA) was used to identify species or groups of species whose abundance was correlated to differences in environmental variables and to guide metric development. Pond presence and gradient were identified as confounding factors and controlled by stratification. Metrics able to distinguish stressed from reference sites were developed for three strata: high gradient streams with ponds, high gradient streams without ponds, and low gradient streams. Pond presence was not a confounding factor in low gradient streams.

Backpack electrofishing collected the most amphibian species per unit sample and the timed search technique collected the most per unit time. Both methods were recommended for determining amphibian richness since using both methods would result in sampling the greatest diversity of habitats and add minimal effort to existing procedures. In addition, many amphibian species are semi-aquatic and use terrestrial habitats at different times of the year and during different life stages. Repeat sampling was recommended to evaluate precision and seasonal changes in catchability/gear recruitment.

Eleven metrics were proposed. Salamander and Sensitive Frog Richness was the most responsive, applicable, and had the greatest range. Other amphibian, crayfish, and fish based metrics were developed, each providing discriminatory power and/or making ecological sense in the absence of reference conditions. The resulting multimetric indices were successful at distinguishing reference from stressed sites (when reference sites were available). More sampling of low gradient streams is needed to determine reference conditions and improve metrics for low gradient watersheds. Additional sampling would be useful to understand the influence of pond type and pond proximity on reach fauna. Developing new metrics and/or testing proposed (EPA) metrics for intermittent streams with drainage areas less than 1 km² is needed. Currently, these streams are not adequately assessed.

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Introduction

In 2004, The Academy of Natural Sciences of Drexel University (ANS) and the New Jersey Department of Environmental Protection (NJDEP) conducted a pilot study (phase I) on the feasibility of developing an Index of Biotic Integrity (IBI) for headwater streams, i.e., those streams with drainage areas smaller than the minimum 12.95 km² (5 mi²) used for the State's Ambient Fish IBI Monitoring for streams and rivers. For phase I, ANS and NJDEP sampled fish, crayfish and amphibians in a small number of headwater streams, ranging from those in undisturbed, forested watersheds to those in highly urban watersheds. Phase I found several aspects of fish, crayfish, salamander and frog assemblages which were correlated with watershed disturbance and that could become potential IBI metrics. In 2005, ANS developed a proposal for additional studies to test sampling methods and to include a larger number of sites, with a greater range of magnitudes and types of watershed disturbance. The ANS proposed this study as a two-year study, to be funded as two consecutive annual grants. The first year of study was funded and implemented, but the second year of the Phase II study was not funded and therefore not implemented.

The completed first year Phase II work successfully identified a number of candidate metrics and helped develop appropriate sampling techniques. The samples were taken in streams covering a range of watershed conditions, with particular attention to streams with intermediate urban development and/or substantial amounts of agricultural development. Additional work necessary before implementation of the IBI was identified in the report and by reviewers of the report. This included work originally designated for the second year of Phase II.

In 2009, Phase III was started. This work incorporated work originally proposed for the second year of the Phase II study, including sampling of at least 24 sites and additional data analysis. Specific areas addressed in this study included sampling additional reference streams; determining range and applicability of metrics with respect to low gradient streams and streams with upstream ponds; refinement of salamander sampling techniques; validation and refinement of previously proposed candidate metrics by application to new sites.

Methods

Site Selection

Sites were selected based on land use, land cover, reach gradient, pond/lake presence, and drainage area (typically <12.95 km² [5 mi²]). Unusual conditions such as tributaries entering the sample reach, nearby juncture of sample reaches with larger tributaries, dams, and other large in-stream obstructions were avoided when choosing reaches. Adventive streams (small streams flowing into major tributaries) were not sampled. Adventive tributary streams were defined as follows: let the Horton stream order of a stream be j and the Horton stream order of the tributary stream be I; if (j-I) is greater than

2, the tributary stream is adventive. For this study, tributary streams $I \ge 3$ entering stream $j \ge 5$ were not considered adventive.

Except where specifically defined otherwise, field sampling procedures followed those of the NJ DEP Bioassessment Unit so as to provide comparable data to those collected for regular monitoring. These procedures follow USEPA's Rapid Bioassessment protocol (Barbour et al. 1999) with region-specific modifications (Kurtenbach 1994).

All streams were sampled from May through November. Temporal variability was not addressed. Sampling took place in periods after spawning of most species (excepting trout) when consistent fauna was expected and after spawning of white sucker, the primary migratory species likely to occur in the area. Fish sampling was not done in trout production waters after September 1.

Fish Sampling

Reaches for fish sampling were 150 m in length. Reaches were divided into 100 m and 50 m subreaches. Each of these subreaches was blocked at the upper and lower end using nets (1/8 in. delta mesh), unless natural barriers sufficient to prevent fish escapement were present. Samples were taken when the ability to see and capture stunned fish was not compromised by transient site conditions. In particular, samples were not taken immediately following precipitation if turbidity and water levels were deemed an impediment to sampling.

Fish sampling was conducted using a Smith-Root model 15-D backpack electrofisher equipped with a 1.8 m (6 ft.) long anode pole and standard 27.9 cm (11 in.) diameter anode ring. Typically, 100-300 volts were used to output 0.3 amps of pulsed DC (50Hz, 6 ms). Higher voltages were used in streams with conductivities less than 100 μ s/cm. Sampling was done during daylight with a two to five person crew. Stunned fish were collected using one or more dip nets (mesh size 1/8 in. [3.18 mm]). The standard dip net used had a 42 x 27 cm rectangular opening. At times, this net was supplemented by smaller nets (e.g., aquarium dip nets) to capture small target taxa on the bottom. Polarized sunglasses were worn to reduce sun glare and increase capture rates (sunglasses were not worn when they decreased visibility, e.g., in dense shade). All fishes, lampreys, salamanders, and adult crayfishes were captured or documented. Notes on frogs captured were taken.

Fish, salamanders, and frogs were identified to species, measured for length, and evaluated for condition (anomalies and disease excluding blackspot disease). Hatchery fish were not distinguished from wild fish because occurrence was expected to be rare (which proved to be the case since hatchery markings were not observed: eroded fins, clipped fins, and tags). All fish over 25 mm total length were counted. Crayfish were typically identified to species. Individuals not identifiable to species (mainly juveniles) were proportionally allocated to the species documented at the reach. Some fishes, salamanders, frogs, and crayfishes were preserved (10% buffered formalin or 95% ethanol) and identified in the laboratory. Taxa observed but not captured were identified to the lowest practical level and included in species totals when identified to species level.

Salamander – Crayfish Sampling

In past headwater projects (Phases I and II), targeted salamander and crayfish sampling was done within the 150 m reach electrofished for fish. In this study both techniques were done within a 30 m length of stream ending 5 m downstream of the 150 m electrofished reach. The first ten meters (0-10 m) of this area was used for a timed sample, the next 5 m (10-15 m) provided a buffer zone, and the following 15 m (15-30 m) was used for one 15 x 2 m transect sample (1 m on land and 1 m in the stream). A 5 m buffer zone was left between the salamander and electrofishing reaches.

In addition to collecting salamanders and crayfish during fish sampling, one timed (hereafter referred to as timed sample) and one area-based technique (hereafter referred to as transect sample) were used. The timed sample was taken in 10 meters of stream by a crew of two for 10 minutes. Available cover (greater than or equal to 7.62 x 12.7 cm [3 x 5 in.]) was turned by hand both in the stream and within 1 m of the stream. Aquarium dip nets were used to aid salamander and crayfish capture. The transect sample was started 5 m upstream of the timed sample and ended 5 m downstream of the reach designated for fish sampling. Sampling was done in a band 1 m laterally on each side of the water's edge for 15 m, i.e., a 15 x 1 m band on shore and a 15 x 1 m band in the water. Sampling was done on a randomly selected side of the stream, unless that side was unsuitable for sampling (e.g., cliff or wall). All movable cover (rocks, logs, debris) was turned, and all salamanders and crayfish were captured or documented. The sampling procedure in the one-meter band in the water depended on water depth and substrate. Kick-netting was used when water depth was sufficient and substrate did not impede collecting. Dipnetting was used when the water depth was too shallow or the substrate (e.g., large boulders) impeded kick-netting. Identification of salamander and crayfish were done the same way as done for electrofishing samples. If the 1-m band on shore contained few or no movable objects, separate samples were taken to turn objects located within the floodplain of the sampling site.

Habitat Assessment

At all sites, habitat assessments were conducted using forms similar to those used by NJ DEP Bioassessment unit. For all assessments epifaunal substrate, embeddedness, velocity/depth regimes, sediment deposition, channel flow status, channel alteration, frequency of riffles, bank stability, bank vegetative protection, and riparian vegetative zone width were evaluated (measurement or scoring based on visual observations). In this assessment, each parameter was scored and summed to provide a total habitat quality score (used same scoring as in EPA habitat assessment form). Substrate composition, channel morphology, and canopy and riparian zone cover were also measured.

Within each reach, except for salamander-crayfish reaches, 16 transects were located at 10 m intervals within and at the edges of the reach. For salamander-crayfish reaches, transects were located at 2 m intervals. At each transect, width of the wetted surface,

bankfull width, and maximum depth were measured. Dominant substrate types, presence of cover, undercut banks, eroding banks or unusual features were noted. The dominant riparian type within 15 m of the water edge was noted at each transect. Four measurements of canopy cover were taken using a spherical crown densiometer within each fish reach (at 0 m, 75 m 100 m and 150 m); this retained consistency with measurements taken during the phase I study (taken at 0 m, 75 m and 150 m). Additionally, one measure of canopy cover was taken for each salamander-crayfish reach. Gradient was measured for the 150 m reach designated for fish sampling. Gradient was measured as the total elevation drop over the combined 150 m electrofished reach (50 and 100 m subreaches, i.e., gradient was measured relative to channel length rather than to straight-line distance between start and end points). Water temperature, dissolved oxygen concentration, conductivity, and pH were measured at least once before, during or just after fish sampling.

Data Analysis

Data from 66 sites sampled as part of phase I, II, and III (current study) were combined for all analyses except to compare salamander sampling techniques (Table 1). Metrics proposed by Horwitz (2007) were evaluated for applicability, validated, or refined resulting in new candidate metrics. Because reference conditions would differ among different types of streams, three strata were defined, with new sets of candidate metrics developed for the strata: high gradient streams without ponds, high gradient streams with ponds, and low gradient streams. Ponds were defined as lentic bodies of water and included lakes, reservoirs, and natural and artificial impoundments. Having a pond anywhere upstream of the sampled reach satisfied the pond criterion. Distance to the nearest upstream pond was not considered in stratification.

All taxa encountered during fish sampling, including amphibians and crayfish, were identified and enumerated. Catch-per-unit-effort (CPUE) was standardized as the number of individuals per 150 m reach. These data were transformed by taking the natural log of 1+CPUE and used in a Canonical Correspondence Analysis (CCA) along with land-use land-cover (LULC) data provided by NJDEP. A biplot was used to view the resulting species scores and environmental variables (LULC). The CCA biplot of species scores and environmental variables was used to identify species or groups of species whose abundance is correlated to differences in environmental variables. This information was used to guide metric development for different strata. Species groupings determined by CCA were manipulated by adding and removing species which did or did not make ecological sense. Exclusion of species typically occurred for uncommon species, where correlation with environmental factors resulted from coincidental occurrence of those environmental conditions at the few sites where the species were found. Metrics were scored as a proportion of the 95th percentile value if they decreased with stress or as a proportion of the 5th percentile value if they increased with stress, as described by Flotemersch et al. (2006). Box plots, scatter plots, and discrimination efficiencies were used to evaluate a metric's response to environmental variables and ability to distinguish stressed from minimally-impacted sites. Discriminatory metrics that were applicable to a range of impairment levels were combined into a multimetric index. Multimetric scores

were calculated for each site as the mean metric score. Minimally-impacted or reference sites were defined as those with greater than 80% forested land cover in their watersheds and with no other known stressor. Stressed sites were defined as being less than 80% forested (two tiers: 20-80% and less than 20% forested).

Salamander CPUE was standardized as number collected per minute and number collected per 1 m². Sampling techniques (timed, transect, and electrofishing) were compared using non-parametric Friedman tests with a critical value of α =0.05. Post-hoc comparisons were tested using Wilcoxon Signed Rank tests with a Bonferoni adjusted α =0.016. Statistics were performed using Statistica (StatsSoft, Inc 2001) and CANOCO Version 4 (ter Braak and Smilauer 1998). Percentiles for discrimination efficiencies were determined using linear interpolation in SigmaPlot Version 12 (Systat Software, Inc. 2010).

Nomenclature

Since Phase I (2004), frogs and toads documented in this study have been moved from the genera *Rana* and *Bufo* to *Lithobates* and *Anaxyrus*, respectively. No other species names relevant to this study were changed. Common and scientific names were used according to Collins and Taggart (2009) for amphibians, Nelson et al. (2004) for fish and Hart (1994) for crayfish. Common crayfish was used as the common name for *Cambarus bartonii*, rather than Appalachian Brook Crayfish as used by Hart (1994; Table 1).

Results

From 2009 to 2010 (Phase III), 24 sites with 0-9% gradient and 9-96% forest cover were sampled (Tables 2, 3 and 4). Ten of the 24 sites had greater than 80% forest cover (five >90%; Table 3) and were considered reference sites. Seven of these sites had ponds upstream, and one of these sites was also a low gradient reach within a high gradient watershed (Jennings Brook sites; Table 3). Nineteen of twenty-four sites had ponds upstream. After combining these data with data collected during phase I and II, a CCA biplot of species scores and environmental variables identified species that correlated with differences in environmental variables among sites (Figure 1). Seven of sixty-six sites had unique characteristics and were not used to evaluate metrics when including them greatly skewed the results (Table 5). Many metrics proposed by Horwitz (2007) and others were refined, new metrics were created, and some were not applicable (e.g., anomalies were rare and so the proportion of anomalies was not used as a metric).

Salamander Collections

Eurycea bislineata had the greatest range of any salamander species, occurring at 48 sites with land covers ranging 2-97% forested and 0-92% urban. *Lithobates clamitans* had the greatest range of any frog species, occurring at 27 sites with land covers ranging 8-96% forested and 0-92% urban. *Lithobates catesbeianus* appeared slightly less widespread than *L. clamitans*, occurring at 6 sites with land covers ranging 21-87% forested and 2-

46% urban. Other amphibians demonstrated an affinity for watersheds with higher forest cover and less urbanization. *Desmognathus fuscus* occurred at 14 sites ranging from 20-97% (13 ranging 52-97%) forested and 0-26% urban. *Lithobates palustris* occurred at 14 sites with land covers ranging 47-97 % forested and 0-38 % urban. *Pseudotriton ruber* was more sensitive to impairments and occurred at 11 sites with land covers ranging 58-97 % forested and 0-35% urban. *Eurycea longicauda* was the salamander species most sensitive to impairment, occurring at 4 sites with land covers ranging 47-94 % forested and 5-9% urban. *Lithobates sylvaticus* was the frog species most sensitive to urbanization, occurring at 6 sites with land covers ranging 57-97 % forested and 0-6% urban.

Refinement of Salamander Sampling Techniques

Salamanders were documented at 17 of 24 sites using electrofishing, timed, or transect techniques (not including kick-net collections). At these sites, electrofishing (150 m reach) documented more salamanders per sample and at more sites then the timed and transect techniques (electrofishing: 16 sites, spp. mean = 1.5; timed: 12 sites, spp. mean = 0.88; transect: 13 sites, spp. mean= 0.88). At three sites, the timed technique documented species not documented while electrofishing. The timed technique also collected more salamander species per minute then transect or electrofishing techniques (P<0.044, Figure 2). However, the transect technique collected more salamanders per m² then the timed and electrofishing techniques (P<0.020), Wilcoxon Signed Rank test showed no pairwise differences between the timed and electrofishing techniques (Figure 3). The number of salamanders per minute and number of species per m² were not significantly different among techniques.

Influence of Upstream Ponds

Although we did not evaluate the influence of pond proximity on reach fauna, it is worth noting that the influence of upstream ponds was apparent in reaches up to 1.8 km downstream. Specifically, we collected 19 *Lepomis macrochirus* and 5 *Micropterus salmoides* at Blue Brook. This reach was approximately 1.7 km (1.1 mi.) downstream of the nearest upstream pond and in a watershed 64% forested. At Bear Swamp Brook, we collected one *M. salmoides* in a reach approximately 1.8 km (1.1 mi) downstream of the nearest upstream pond and in a watershed 87% forested.

Crayfish Collections

Crayfish specific collection techniques (dip net, kick net, hand) varied between study phases and sites (due to site conditions). This was problematic when using CPUE to index all 66 sites. For this reason, we used electrofishing (used at all sites) CPUE to compare sites. *Cambarus bartonii* occurred at 24 sites with land covers ranging 5-97% forested and 0-93% urban (20 sites >50% forested; 22 sites <50% urban). One of these sites, Cresskill Brook (CK), a site mainly forested with low density residential development had high quality habitat that did not reflect the 93% urban and 5% forested land cover. *Orconectes limosus* occurred at six sites with land covers ranging 12-73%

forested and 7-71% urban. *Procambarus acutus* occurred at four sites with land covers ranging 20-93% forested (3 sites >90%; one 20% site with 52% agricultural) and 0-12% urban. *P. acutus* occurred in low gradient reaches or in reaches with low gradient areas nearby. Non-native crayfishes *Procambarus clarkii* and *Orconectes robustus* each occurred once, both in highly urbanized areas. The non-native crayfish *Orconectes virilis* occurred at 17 sites with land covers ranging 2-87% forested and 0-95% urban.

Refinement of Metrics

Data from 66 sites sampled as part of phase I, II, and III (current study) were combined and used to derive metrics able to differentiate between minimally-impacted and stressed sites (see appendices A-C for raw values per station). These metrics were combined to produce a multimetric index for each stratum (Tables 6a-c, Figures 4-6). Since a visual inspection of candidate metrics regressed on drainage area did not reveal significant correlations, candidate metrics were not adjusted for drainage area. Below are descriptions of candidate metrics for each stratum.

Refinement of Metrics: High Gradient Streams without Ponds

1. Salamander and Sensitive Frog Richness

This metric has a maximum value of nine and includes all salamanders with an affinity for flowing or impounded streams and two frog species typically found in highly forested watersheds. The highest value determined in this stratum was 4 with the 95th percentile value equaling 3. Salamander species for this metric included: *Eurycea bislineata, E. longicauda, Desmognathus fuscus, D. ochrophaeus, Notophthalmus viridescens, Gyrinophilus porphyriticus* and *Pseudotriton ruber*. Frog species included: *Lithobates sylvaticus* and *L. palustris. D. ochrophaeus* and *Gyrinophilus porphyriticus* were not collected in this study but were included in this metric because each has been collected by ANS in headwater streams of Pennsylvania and is known to occur in NJ (Schwartz and Golden 2002). This metric was highly discriminatory with a discrimination efficiency of 94 % at the 25th percentile (Figure 7, Table 7).

2. Cambarus bartonii CPUE

This metric is a measure of the total number of *Cambarus bartonii* collected in a 150 m stream reach by backpack electrofishing. The highest value determined in this stratum was 13 with the 95th percentile value equaling 9. The *C. bartonii* CPUE metric was highly discriminatory with a discrimination efficiency of 88% at the 25th percentile (Figure 8, Table 7).

3. Proportion of Sensitive Cool Headwater Fishes

This metric has a maximum value of 1.00 and includes native fishes with an affinity for flowing cool headwater streams. The highest value determined in this stratum was 1.00 with the 95th percentile value equaling 0.52. The lowest value determined was zero. Fish

species for this metric included: *Cottus cognatus* and *Salvelinus fontinalis*. This metric was highly discriminatory with a discrimination efficiency of 92% at the 25th percentile (Figure 9, Table 7).

4. Proportion of Age-0 Salvelinus fontinalis

This metric has a maximum value of 1.00 and is calculated by dividing the number of age-0 *Salvelinus fontinalis* by the total number of *S. fontinalis* + 0.001 (0.001 is added to avoid division by zero and an undefined result). The highest value determined in this stratum was 0.78 with the 95th percentile value equaling 0.74. The lowest value determined was zero. Age-0 *S. fontinalis* were defined as individuals <10 cm total length. This metric was highly discriminatory with a discrimination efficiency of 92% at the 25th percentile (Figure 10, Table 7).

Refinement of Metrics: High Gradient Streams with Ponds

1. Salamander and Sensitive Frog Richness

The same salamander and sensitive frog richness metric used for high gradient streams without ponds is also used here but with different cut-points between impairment groups. The highest value determined in this stratum was 5 with the 95th percentile value equaling 4. This metric was moderately discriminatory, with a discrimination efficiency of 62% at the 25th percentile and 77% at the 50th percentile (Figure 11, Table 7).

2. Native Crayfish CPUE (excluding *Orconectes limosus*)

This metric is a measure of the total number of *Cambarus bartonii* and *Procambarus acutus* collected in a 150 m stream reach by backpack electrofishing. The highest value determined in this stratum was 23 with the 95th percentile value equaling 16. This metric was moderately discriminatory with a discrimination efficiency of 0% at the 25th percentile and 92% at the 50th percentile (Figure 12, Table 7).

3. Proportion of Sensitive Warm and Cool Headwater Fishes

This metric has a maximum value of 1.00 and includes native fishes with an affinity for flowing or impounded, warm or cool headwater streams. The highest value determined in this stratum was 1.00 with the 95th percentile value equaling 0.96. The lowest value determined was zero. Fish species for this metric included: *Cottus cognatus, Enneacanthus spp., Esox niger, Notropis bifrenatus, Notemigonus crysoleucas, Salvelinus fontinalis,* and *Umbra pygmaea.* This metric was moderately discriminatory with a discrimination efficiency of 69% at the 25th percentile and 85% at the 50th percentile (Figure 13, Table 7).

Refinement of Metrics: Low Gradient Streams

In the combined dataset of phases I, II, and III, low gradient sites with high forest cover were under-represented when compared to high gradient sites (Figure 14). Percent gradient and percent forest appeared correlated in the CCA biplot, and species associations to these environmental variables could not be distinguished. Jennings Brook Meadow (JBM), a low gradient reach in a high gradient watershed, was the only reference site for the low gradient stratum. For this reason, discrimination efficiencies for metrics of low gradient streams were not calculated.

1. Amphibian Richness

This metric has a maximum value of 13 and includes all salamanders with an affinity for flowing or impounded headwater streams and all frogs and toads. The highest value determined was 4 with the 95th percentile value equaling 4 as well (Figures 15 and 16). Salamander species for this metric included: *Eurycea bislineata, E. longicauda, Desmognathus fuscus, D. ochrophaeus, Gyrinophilus porphyriticus, Notophthalmus viridescens,* and *Pseudotriton ruber*. Frog species included: *Anaxyrus americanus, A. woodhousii, Lithobates catesbeianus, L. clamitans, L. sylvaticus* and *L. palustris.*

2. Native Crayfish CPUE

This metric is a measure of the total number of *Cambarus bartonii*, *Orconectes limosus*, and *Procambarus acutus* collected in a 150 m stream reach by backpack electrofishing. The highest value determined for this stratum was 20 with the 95th percentile value equaling 16 (Figures 17 and 18).

3. Proportion of Tolerant Fishes

This metric has a maximum value of 1.00 and includes fishes with a tolerance for urbanized headwater streams. Because this metric increases with stress, lower values indicate better water quality and the 5th percentile value was used for scoring. The lowest value determined for this stratum was 0.01 with the 5th percentile value equaling 0.05. The highest value determined was 0.99. Fish species for this metric included: *Ameiurus natalis, A. nebulosus, Catostomus commersoni, Etheostoma olmstedi, Fundulus diaphanus, F. heteroclitus, Gambusia affinis, Lepomis cyanellus, Notropis hudsonius, Pimephales promelas, and Semotilus atromaculatus (Figures 19-21).*

4. Proportion of Sensitive Warm Headwater Fishes

This metric has a maximum value of 1.00 and includes native fishes with an affinity for flowing warm low gradient headwater streams. The highest value determined for this stratum was 0.90 with the 95th percentile value equaling 0.74. The lowest value determined was zero. Fish species for this metric included: *Enneacanthus* spp., *Esox niger, Notropis bifrenatus, Notemigonus crysoleucas,* and *Umbra pygmaea* (Figures 22 and 23).

Relationship of Metrics to Total Habitat Quality Scores

Proposed metrics were plotted against total habitat quality scores to evaluate their responsiveness to this habitat rating system (determined using the EPA habitat assessment form; Figures 24-36). Regression equations and r^2 values were presented when appropriate. In addition, total habitat quality scores were plotted against percent forest for all sites and a positive relationship was indicated by the resulting model (r^2 =0.5511; Figure 37). Although there was a correlation between habitat scores and percent forest, both variables may also be related to percent development.

Comparison to NJ's Northern and Inner Coastal Plain Indices of Biotic Integrity

High gradient streams with drainage areas greater than $5.18 \text{ km}^2 (2 \text{ mi}^2)$ were scored using both NJ's Northern Index of Biotic Integrity (NIBI) and the IBI proposed above (Table 8). Stream categories assigned by both indices were similar and differed by at most one category. Additionally, all low gradient streams were scored using both NJ's Inner Coastal Plain IBI (SIBI; draft) and the proposed IBI for low gradient streams in northern NJ (Table 9). Stream categories were similar and differed by more than one category for only one site (Ryker Lake Tributary Meadow). Although stream categories differed little, relationships between rescaled scores (for both NIBI and SIBI) and scores from the proposed IBIs were poor (NIBI and proposed IBI r²=0.0118 and SIBI and proposed IBI r²=0.0366). This suggests that agreement with the proposed IBI in assessing streams is only categorical and may be due to the broad categories being used.

Streams with Drainage Areas Less than 1 km²

Four streams with drainage areas less than 1 km² were sampled: Cresskill Brook, Demerest Brook (DEM2), Green Brook, and Stephens Brook. At Cresskill Brook, only fish (*S.fontinalis* and *Rhinichthys atratulus*) and crayfish (*C. bartonii*) occurred. At Demerest Brook, fish, amphibians, and crayfish did not occur. At Green Brook, only amphibians (*L. clamitans, Plethodon cinereus* and *P. glutinosus*) and crayfish (*C. bartonii* collected in hand sample) occurred. At Stephen's Brook, only amphibians (*P. ruber, D. fuscus, E. bislineata, P. cinereus*, and unidentified frog species) and crayfish (*C. bartonii* collected in hand sample) occurred.

All streams were sampled when holding water. Green Brook was sampled when holding water and when completely dry. While holding water on September 19, 2009, one *L. clamitans* in the electrofished reach, one *C. bartonii* in the timed salamander/crayfish and two *P. cinereus* in the salamander transect sample were captured. While dry on June 30, 2010, one *P. cinereus* and *P. glutinosus* in three salamander transect samples were collected. No electrofishing was done on this day.

Discussion

Stratification

Gradient and pond presence were identified as confounding factors that influenced biotic assemblages. To control for these factors, candidate metrics were developed for three strata: high gradient streams without ponds, high gradient streams with ponds, and low gradient streams. Low gradient sites were not stratified by pond presence for reasons discussed below.

Low Gradient Reaches

The faunal assemblages of low gradient reaches are expected to differ from those of high gradient reaches because of habitat differences. Low gradient reaches tend to have more pool habitat and less (or no) riffle habitat, with corresponding differences in other factors, such as substrate and amount of aquatic vegetation. The fauna of low gradient reaches may also be affected by the topography of the entire watershed as well as that of the reach. In this study, low gradient reaches occurred in two types of settings: 1) those within low gradient watersheds (LGR-LGW) and, 2) those within high gradient watersheds (LGR-HGW; in the valley of a high gradient watershed and/or bordered by high gradient reaches). Most low gradient reaches in low gradient watersheds occurred in piedmont valleys, especially in areas near the Coastal Plain. Two LGR-HGW sites in areas of relatively high forest cover were sampled (Jennings Brook Meadow, treated as a reference, and Ryker Lake Meadow, a reach in an area of relatively high forest cover). Both of these areas had evidence of beaver activity and were impounded or were likely to be impounded by beavers for extended periods of time. We identified one other low gradient reach in a high gradient watershed but this site was too deep for sampling because it was impounded by beavers (e.g., meadow above Russia Brook).

The biotic assemblage of a LGR-HGW may differ from a LGR-LGW in a few ways. First, a LGR-HGW may have less total low-gradient habitat and that habitat may be less accessible to colonists, leading to lower total populations of species specific to those habitats. These species would be more sensitive to local extirpation (due to natural or anthropogenic causes). As a result, the characteristic fauna of low gradient reaches may be smaller in LGR-HGW areas for any given level of development. The LGR-HGW sites sampled had a number of species characteristic of low gradient sites, such as eastern mudminnow, bridle shiner, and golden shiner. Thus, the data at end do not support this effect, but there were too few sites sampled to reject this effect. Secondly, high gradient portions of watersheds (slopes and upstream reaches) should deliver water more quickly to downstream reaches. This could reduce warming in these reaches, leading to more suitable conditions for cold or coolwater species. The geographic location of the types of low gradient reaches reinforces this pattern, since the LGR-HGW sites occur more commonly in northern New Jersey, while the LGR-LGW sites occur more commonly in central New Jersey. Thirdly, species characteristic of high gradient reaches may occur in nearby low gradient reaches due to dispersal or short-term movements. These two factors would cause LGR-HGW sites to be more diverse and more similar to high gradient

reaches (due to mixing between high gradient and low gradient species). Thus, different metrics or different metric scoring may be appropriate for LGR-HGW and LGR-LGW sites. However, too few of each kind of reach were located and sampled to support different metrics or scoring.

Determination of Reference Conditions

One of the goals of phase III studies was to provide additional information on reference conditions in headwater streams. To address this need, 10 sites with greater than 80% forest cover were sampled. This increased the total number of sites in this group from 7 to 17 and helped to establish better reference criteria for high gradient streams.

The possible differences between LGR-HGW and LGR-LGW sites discussed in the previous section affected determination of reference conditions for low gradient reaches. Most LGR-LGW sites were in highly developed watersheds, and no LGR-LGW reference sites were located. LGR-HGW sites occurred in highland meadows. These sites were highly forested, with some sites impounded by beaver. Reference conditions for all low gradient streams were estimated using the LGR-HGW site with high forest cover (Jennings Brook Meadow). As noted above, use of this as a reference for all low gradient sites could be misleading if this site has fewer (or lower abundance of) characteristic low gradient species or has some high gradient species not typical of low gradient sites in general. As noted above, the data from Jennings Brook Meadow did not suggest the former issue. Presence or abundance of characteristic high gradient species was not used in forming metrics for low gradient streams, so the latter issue is likely to be moot.

LGR-LGW sites did not have beaver activity, possibly because of paucity of tree foods for beaver, human interference, difficulty of colonization of beavers in urban areas, or because ponds have been built in appropriate sites for beaver impoundments. It is likely that beaver impoundments would be present under reference conditions for LGR-LGW sites, so absence of beaver may represent a type of impairment. As such, it may be difficult to separate various causes of impairment. More work needs to be done to determine the appropriateness of using low gradient reaches in high gradient watersheds to guide metric development for low gradient streams.

Refinement of Salamander Sampling Techniques

In phase III, three techniques were evaluated for salamander sampling: backpack electrofishing, timed search, and area-based (transect) search. Backpack electrofishing added the least amount of time to sampling because it was done while fish sampling. Per sample, electrofishing was also the most effective technique for determining species richness. The transect technique added the most time to sampling but collected the most salamanders per unit area. The timed technique added a fixed time of ten minutes to sampling and collected the most salamander species per unit time. The timed technique also allowed the collectors to cover the widest variety of habitat types in the shortest amount of time. In addition, during the timed search, the collector's attention was focused on collecting salamanders as opposed to collecting salamanders, fish, and crayfish as done during electrofishing. Although the timed search collected less salamander species per sample than electrofishing, it added little time to site assessment, added species at three sites, and included riparian habitat not sampled by electrofishing. In addition, adding the timed technique to site assessment provided a way to assess sites not suitable for electrofishing. Because the proposed amphibian metrics are richness based, we recommend that NJDEP use the timed search and backpack electrofishing techniques for collecting salamanders. We also suggest pooling all salamanders and amphibians observed and using this pooled data for calculating metrics based on amphibian richness. The accuracy of the timed search technique was not evaluated in this study and may be improved by increasing the sampling time or number of samples taken.

Thirteen species of salamanders and frogs relevant to this study occur in northern NJ. Many of these species are known to respond negatively to stream pollution (Petranka 1998). Amphibians relevant to this study occurred at 57 of 66 sites. Because of the high occurrence of amphibians, amphibian richness metrics minimize the occurrence of zero values. The sensitivity of some amphibian species and the tolerance of others allows for richness-based metrics to be developed that increase in score with decreasing impairment. The different ranges in which amphibian species occur within the spectrums of forest cover and urban land-use make amphibians good candidates for responsive metrics that are applicable to a range of impairment levels.

Influence of Upstream Ponds

Although metrics were evaluated and developed for reaches with ponds upstream, pond type and the proximity to an upstream pond were not evaluated. These factors appeared to influence downstream biotic assemblages. Two general pond categories should be considered when evaluating the influence of ponds in future studies: natural (e.g., beaver impoundments and springs) and artificial (man-made impoundments). The major difference between the two faunas occurring downstream of these pond types appears to be due to historical and/or current fish stocking. For example, Micropterus salmoides were collected in a high gradient reach on Fox Run, a station downstream of a large manmade impoundment. Conversely, Notropis bifrenatus were collected in a high gradient reach on Jennings Brook, a reach downstream and in close proximity to a beaver pond (Jennings Brook meadow). Beaver ponds on Jennings Brook, Tributary to Ryker Lake, and Russia Brook were smaller, shallower, and more vegetated than artificial ponds. Reaches downstream of these ponds had increased proportions of native lentic species. Pond type was not always uniform within drainages. Both artificial and natural ponds were found immediately upstream of the sampled reach on Russia Brook. The influence of both pond types was reflected in the reach assemblage, with *M. salmoides* and *U.* pygmaea both occurring. Reach fauna is also likely correlated to the proximity of an upstream pond. For high gradient reaches, it is hypothesized that as downstream distance between pond and reach increases, pond faunal presence decreases. This is primarily due to the inability of lentic species to persist in a high gradient, lotic environment.

Urban areas are likely to have a high proportion of small ponds due to flood control (e.g., retention basins), public recreation (e.g., fishing), and other factors associated with high population density. In this study, 10 of 16 sites with greater than 70% urban land-use had ponds. Most sites were located in low gradient areas and pond presence was not an explanatory variable for these streams. This is likely due to low gradient streams supporting many lentic species that also occur in ponds. Pond presence upstream of a high gradient reach provides a source population for lentic fishes that would otherwise not occur in a high gradient stream. Pond presence upstream of a low gradient stream does not serve the same function since lentic fishes already occur in the downstream reach.

High Gradient Sites without Ponds Upstream

Twenty-one high gradient sites without upstream ponds were sampled as part of phase I, II, and III. For these sites, salamander and sensitive frog richness was the most applicable metric, assigning zero values to only three sites. This metric was also responsive over a broad range of forest covers (5-96%) and highly discriminatory. The responsiveness of this metric was due to its composition including species that can all occur under reference conditions but are not equally tolerant of impairment. This inequality resulted in a metric responsive to a range of impairment levels.

The CPUE of *C. bartonii* was categorically responsive, highly discriminatory, and applicable to most sites, assigning zero values to ten sites. *C. bartonii* was collected at four of five reference sites for this stratum, with Stephen's Brook, a fishless reach being the only absence. Although useful, using crayfish CPUE may be problematic due to variability among investigators. To address this concern, use of a standardized protocol (e.g., equal value for crayfish and fish; defined number of netters; using consistent net types) is recommended.

The proportion of age-0 *S. fontinalis* was recommended by Horwitz to provide a measure of *S. fontinalis* reproduction and trout survival (2007). Horwitz also recommended the proportion of sensitive cool headwater fishes as a candidate metric (2007). Although these metrics each assigned zero values to 15 and 16 sites, both were strongly associated with reference conditions. Including these metrics with more applicable metrics resulted in a more responsive multimetric index with better resolution, range, and ability to distinguish reference from stressed sites.

High Gradient Sites with Ponds Upstream

Twenty-four high gradient sites with upstream ponds were sampled as part of phase I, II, and III. Although sites with ponds upstream were generally not targeted as part of phase I and II, some sites had upstream ponds. Most of these ponds were very small and considered inconsequential at the time of those studies. As part of phase III, 19 sites with upstream ponds were sampled. Salamander and sensitive frog richness was again found to be the most applicable metric, assigning zero values to only three sites while still remaining responsive across the range of forest covers.

The influence of ponds resulted in different fish and crayfish metrics for these high gradient sites when compared to high gradient sites without upstream ponds. Fauna typically found in pond systems were found persisting in high gradient reaches downstream. In some watersheds, the presence of upstream ponds appeared to reduce the ability of downstream reaches to support coolwater fishes by increasing the downstream water temperature. The presence of upstream ponds necessitated creating a metric that included both warm and cool headwater fishes. The proportion of sensitive warm and cool headwater fishes sites, assigning zero values to nine sites.

The presence of upstream ponds was also associated with the increased occurrence of *Procambarus acutus*, a crayfish species more typical of lentic water bodies and low gradient streams. The CPUE of *P. acutus* and *C. bartonii* were combined to produce a metric representative of the crayfish fauna of both high gradient and pond habitats. CPUE of native crayfish (*P. acutus* and *C. bartonii*, excluding *Orconectes limosus*) was the least applicable metric in this stratum, assigning zeros to 12 of 24 sites. However, increased native crayfish CPUE was associated with high forest cover and adding this metric to the multimetric index improved the index's ability to distinguish reference from stressed sites.

Low Gradient Streams

Twenty-one low gradient sites were sampled as part of phase I, II, and III. For these sites, the proportion of tolerant fishes and amphibian richness were metrics responsive to changes in a broad range of forest covers (2-93%) and applicable to a broad range of sites. The proportion of tolerant fishes had the greatest applicability by assigning a zero value to only one site. Amphibian richness assigned zero values to five sites, and all were impacted sites. Native crayfish and sensitive warm headwater fishes were the least applicable metrics and occurred only at sites with high forest cover. Native crayfish and sensitive warm headwater for low gradient streams to improve the range and resolution of the multimetric index. Although these metrics are based on few sites, they are thought to be indicative of good water quality.

Relationship of Metrics to Total Habitat Quality Scores

The EPA habitat assessment form was used to determine total habitat quality scores for all sites. This assessment method measures (scores) ten habitat parameters (see methods) thought to reflect habitat quality and was originally designed for streams with drainage areas greater than 12.95 km² (5 mi²). Although the EPA habitat assessment method is effective for larger streams, it may not adequately assess smaller streams. To better assess smaller streams, scoring may need to be adjusted for some habitat parameters. Specifically, certain velocity/depth regimes are less likely to occur in smaller streams (i.e. those including velocities >0.3 m/s or depths >0.5 m). Headwater streams may also be less likely to have water filling the channel and more likely to have exposed substrate (influencing the channel flow status parameter). Submerged structures may also be less

likely to occur for the reasons mentioned above (influencing epifaunal substrate/available cover parameter).

Many of the proposed metrics showed little response to increasing total habitat quality scores above 125. This suggests that the EPA habitat assessment method does not provide sufficient resolution to index habitat quality (related to these metrics) in sub-optimal and optimal streams. However, tolerant fishes and low gradient multimetric scores responded well to total habitat quality scores. Total habitat quality scores provided good resolution for indexing habitat quality related to these measures. Recalibrating habitat parameters may improve the resolution of habitat scores and the response of proposed metrics to these scores in sub-optimal and optimal streams.

Assessing Fishless Streams

Assessment of very small streams without fish is problematic, since absence of fish may be typical and not indicative of impairment in intermittent or very shallow streams. In cases where suitable habitat is available and the stream is continuously flowing (i.e., not intermittent), the proposed amphibian, crayfish, and fish metrics may be sufficient for a reliable index. In this study, four sites had drainage areas less than 1 km², and three of these were fishless. At least one (Stephens Brook) was in a highly-forested watershed with no evidence of impairment. To assess intermittent and/or streams too shallow to support fish, managers will have to either: 1) abandon the proposed fish-based metrics and use crayfish and salamander metrics, or 2) use a downstream reach to assess upstream water quality. Abandoning fish-based metrics and using only crayfish and salamander metrics will increase the variability of scores generated by the proposed multimetric indices. This will reduce the ability of these indices to distinguish impact from stressed sites and will increase the likelihood of wrongly assigning stream categories. Using a downstream reach to assess upstream water quality is done by the NJDEP to classify trout waters and is the best alternative given the current dataset. However, this approach may also result in wrongly assigning stream categories by including downstream areas not representative of the upstream reach. Another option is to develop an IBI specific for intermittent streams. Some salamander and frog species may be uniquely suited for this habitat and flow regime. Metrics targeting amphibian species such as Anaxyrus spp. should be considered.

Recommendations for Future Monitoring and Refinement.

We recommend:

1) More sampling of low gradient reaches (low gradient reaches in high gradient watersheds) to determine reference conditions and improve metrics for low gradient watersheds.

2) Determine the precision of the timed search technique (for collecting salamanders) by repeat sampling. Sites from a range of impairments should be used.

3) Some amphibian species are semi-aquatic and use terrestrial habitats at different times of the year and during different life stages. It would be valuable to determine the influence of season on amphibian metrics (e.g., late spring v. late summer).

4) Additional sampling would be useful to understand the influence of pond type and pond proximity on reach fauna.

5) Develop new metrics and/or test proposed (EPA) metrics for intermittent streams with drainage areas less than 1 km^2 . Currently, streams less than 1 km^2 are not adequately assessed.

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Table 1. Scientific and common names of amphibians (Amp), crayfishes (Cray), and fishes (Fish) collected in phases I, II, and III of the headwater IBI study.

Taxon	Scientific Name	Common Name	Code
Amp	Anaxyrus americanus	American toad	ANAME
Amp	Anaxyrus woodhousii	Fowler's Toad	ANWOO
Amp	Desmognathus fuscus	Northern dusky salamander	DEFUS
Amp	Desmognathus ochrophaeus	Mountain dusky salamander	DEOCH
Amp	Eurycea bislineata	Northern two-lined salamander	EUBIS
Amp	Eurycea longicauda	Long-tailed salamander	EULON
Amp	Lithobates catesbeianus	Bullfrog	LICAT
Amp	Lithobates clamitans	Green frog	LICLA
Amp	Lithobates palustris	Pickerel frog	LIPAL
Amp	Lithobates sylvaticus	Wood frog	LISYL
Amp	Lithobates virgatipes	Carpenter frog	LIVIR
Amp	Notophthalmus viridescens	Red-spotted newt	NOVIR
Amp	Plethodon cinereus	Redback salamander	PLCIN
Amp	Plethodon glutinosus	Northern slimy salamander	PLGLU
Amp	Pseudotriton ruber	Northern red salamander	PSRUB
Cray	Cambarus bartonii	Common crayfish	CABAR
Cray	Orconectes limosus	Spinycheek crayfish	ORLIM
Cray	Orconectes virilis	Virile crayfish	ORVIR
Cray	Procambarus acutus	White river crayfish	PRACU
Cray	Procambarus clarkii	Red-swamp crawfish	PRCLA
Fish	Ambloplites rupestris	Rock bass	AMRUP
Fish	Ameiurus natalis	Yellow bullhead	AMNAT
Fish	Ameiurus nebulosus	Brown bullhead	AMNEB
Fish	Anguilla rostrata	American eel	ANROS
Fish	Aphredoderus sayanus	Pirate perch	APSAY
Fish	Catostomus commersoni	White sucker	CACOM
Fish	Cottus cognatus	Slimy sculpin	COCOG
Fish	Cyprinella spiloptera	Spotfin shiner	CYSPI
Fish	Cyprinus carpio	Common carp	CYCAR
Fish	Enneacanthus gloriosus	Bluespotted sunfish	ENGLO
Fish	Erimyzon oblongus	Creek chubsucker	EROBL
Fish	Esox americanus	Redfin pickerel	ESAME
Fish	Esox niger	Chain pickerel	ESNIG
Fish	Etheostoma olmstedi	Tesselated darter	ETOLM
Fish	Fundulus diaphanus	Banded killifish	FUDIA
Fish	Fundulus heteroclitus	Mummichog	FUHET
Fish	Gambusia affinis	Western mosquitofish	GAAFF

Table 1 cont'd. Scientific and common names of amphibians (Amp), crayfishes (Cray), and fishes (Fish) collected in phases I, II, and III of the headwater IBI study.

Taxon	Scientific Name	Common Name	Code
Fish	Lepomis auritus	Redbreast sunfish	LEAUR
Fish	Lepomis cyanellus	Green sunfish	LECYA
Fish	Lepomis gibbosus	Pumpkinseed	LEGIB
Fish	Lepomis macrochirus	Bluegill	LEMAC
Fish	Luxilus cornutus	Common shiner	LUCOR
Fish	Micropterus dolomieu	Smallmouth bass	MIDOL
Fish	Micropterus salmoides	Largemouth bass	MISAL
Fish	Misgurnus anguillicaudatus	Oriental weatherfish	MIANG
Fish	Notemigonus crysoleucas	Golden shiner	NOCRY
Fish	Notropis bifrenatus	Bridle shiner	NOBIF
Fish	Notropis hudsonius	Spottail shiner	NOHUD
Fish	Noturus insignis	Margined madtom	NOINS
Fish	Oncorhynchus mykiss	Rainbow trout	ONMYK
Fish	Perca flavescens	Yellow perch	PEFLA
Fish	Petromyzon marinus	Sea lamprey	PEMAR
Fish	Pimephales promelas	Fathead minnow	PIPRO
Fish	Rhinichthys atratulus	Blacknose dace	RHATR
Fish	Rhinichthys cataractae	Longnose dace	RHCAT
Fish	Salmo trutta	Brown trout	SATRU
Fish	Salvelinus fontinalis	Brook trout	SAFON
Fish	Semotilus atromaculatus	Creek chub	SEATR
Fish	Semotilus corporalis	Fallfish	SECOR
Fish	Umbra pygmaea	Eastern mudminnow	UMPYG

Phase	Water Body	Station	Station Name/Location	Tributary to	Pairs	Drainage	Lat	Long
2	Bedens Brook	BB	Bedens Brook@Hopewell Park	Rock Brook		Raritan River	40.3872	-74.7614
2	Trib to Big Flat Brook	BFB	Trib to Big Flat Brook	Big Flat Brook		Delaware River	41.2408	-74.7465
2	Trib to Budd Lake	BL	Trib to Budd Lake NW	Budd Lake		Raritan River	40.8821	-74.7585
3	Blue Brook	BLB	Blue Brook	Green Brook		Raritan River	40.6750	-74.3942
2	Bear Brook	BRB	Bear Brook@Livingston	Canoe Brook		Passaic River	40.7757	-74.3072
3	Bear Creek	BRC	Bear Creek	Pequest River		Delaware River	40.9713	-74.8736
3	Bear Swamp Brook	BSB	Bear Swamp Brook	Ramapo River		Passaic River	41.0738	-74.2157
2	Cub Brook	CB	CubBrook@Livingston	Bear Brook		Passaic River	40.7723	-74.3152
2	Coles Brook	CBK	Coles Brook@Hackensack	Van Saun Mill Brook		Hackensack River	40.9106	-74.0504
3	Trib to Ramapo	CGM	Campgaw Mountain Reservation	Ramapo River		Passaic River	41.0517	-74.2121
1	Camp Harmony Brook	CHB	Camp Harmony Brook@Van Dyke Road	Stony Brook		Delaware River	40.4034	-74.8017
2	Cresskill Brook	CK*	Upper Cresskill Brook@Alpine	Tenakill Brook	U	Hackensack River	40.9413	-73.9364
3	Cooley Brook	CLC	Cooley Brook	Belcher Creek		Passaic River	41.1577	-74.3544
2	Cory's Brook	COB	Cory's Brook@Mt.Bethel	Passaic River		Passaic River	40.6387	-74.4982
3	Cherry Brook	CRL	Cherry Brook Lower	Beden Brook	D	Rartian River	40.4026	-74.6853
3	Cherry Brook	CRU	Cherry Brook Upper	Beden Brook	U	Rartian River	40.3936	-74.6772
2	Cresskill Brook	CSK	Lower Cresskill Brook@Cresskill	Tenakill Brook	D	Passaic River	40.9467	-73.9589
1	Dry Brook	DB	Dry Brook@Kymer Rd	Paulins Kill		Delaware River	41.1692	-74.7362
2	Demarest Brook	DEM	Lower Demarest Brook@Demarest	Tenakill Brook	D	Passaic River	40.9515	-73.9571
2	Demerest Brook	DEM2*	Upper Demarest Brook@Alpine	Tenakill Brook	U	Passaic River	40.9563	-73.9356
1	Dunfield Creek	DF	Dunfield Creek	Delaware River		Delaware River	40.9715	-75.1267
1	Dorotockeys Run	DOR	Dorotockeys Run	Hackensack River		Passaic River	40.9875	-73.9748
3	Duck Pond Run	DPR	Duck Pond Run	Millstone River		Raritan River	40.3062	-74.6678
3	Electric Brook	ELB	Electric Brook	S.B. Raritan River		Raritan River	40.8050	-74.7883
3	FoxRiver	FR	FoxRiver	Ramapo River		Passaic River	41.0615	-74.2315
3	Green Brook	GRB*	Green Brook	Belcher Creek		Passaic River	41.1634	-74.3738
2	Trib to 3rd Neshanic River	HQ	Trib to 3rd Neshanic River@Headquarters	3rd Neshanic River		Raritan River	40.4490	-74.9142
1	Jacksonburg Creek	JAC	Jacksonburg Creek	Delaware River		Delaware River	41.0391	-74.9649
3	Jennings Brook	JB	Jennings Brook	Wanaque River	D	Passaic River	41.1628	-74.3142
3	Jennings Brook (Meadow)	JBM	Jennings Brook (Meadow)	Wanaque River	U	Passaic River	41.1651	-74.3136
2	Loantaka Brook	LA*	Loantaka Brook@Morristown	Great Brook		Passaic River	40.7814	-74.4646
2	Ledgewood Brook	LW*	Ledgewood Brook@Ledgewood	Mill Pond		Raritan River	40.8797	-74.6620
2	Mores Creek	MC	Mores Creek@Rosell	West Brook		Arthur Kill	40.6525	-74.2695
2	Metzler Brook	MKW	Metlzer Brook@Mackay Park, Englewood	Overpeck Creek		Hackensack River	40.8904	-73.9842
1	Montana Brook	MON	Montana Brook	Millbrook		Delaware River	40.7252	-75.0855

Table 2. Stations sampled in phases I, II, and III of the headwater IBI study.

Phase	Water Body	Station	Station Name/Location	Tributary to	Pairs	Drainage	Lat	Long
2	Trib to 2nd Neshanic River	NCB	At Britton Rd.	2nd Neshanic River	D	Neshanic River	40.4696	-74.8924
2	Trib to 2nd Neshanic River	NCS	Upstream of Mason Farm Rd.	2nd Neshanic River	U	Neshanic River	40.4764	-74.9030
1	Trib to Primrose Brook	PB	Trib to Primrose Brook@Jockey Hollow NtPk	Primrose Brook		Passaic River	40.7682	-74.5340
1	Peckman River	PCK	Peckman River	Passaic River		Passaic River	40.8482	-74.2343
2	Pequannock River	PQ	Pequannock River@CR.515, Vernon	Pompton River		Passaic River	41.1480	-74.4990
2	Pleasant Run	PR	Pleasant Run@Stanton	S.B. Raritan River		Raritan River	40.5771	-74.8182
3	Ramsey Brook	RAB	Ramsey Brook	Hohokus Brook		Passaic River	41.0326	-74.1357
1	Trib to Robinsons Branch	RB	Trib Robinsons Brook	Robinsons Branch		Arthur Kill	40.6255	-74.3470
2	Rockaway Creek	RBH	Rockaway Creek@Beacon Hill	Lamington River		Raritan River	40.7380	-74.7810
1	Trib to Royce Brook	RBK	Trib to Royce Brook@Deanna Dr.	Royce Brook		Raritan River	40.5111	-74.6327
2	Rockaway Creek	RC	Rockaway Creek@Fairmount Ave	Lamington River		Raritan River	40.7254	-74.7860
1	Rock Brook	RKB	Rock Brook@Long Hill Road	Millstone River		Raritan River	40.4398	-74.7393
3	Ryker Lake Trib	RLT	Ryker Lake Trib	Ryker Lake	D	Passaic River	41.0551	-74.5529
3	Ryker Lake Trib (meadow)	RTM	Ryker Lake Trib (meadow)	Ryker Lake	U	Passaic River	41.0573	-74.5544
3	Russia Brook	RUB	Russia Brook	Pequannock River		Passaic River	41.0476	-74.5540
2	Stephen's Brook	SB*	Stephens Brook@Berkshire Valley WMA	Rockaway River		Passaic River	40.9054	-74.6255
3	Stony Brook trib.	SBT	Stony Brook trib.	Stony Brook		Delaware River	41.2064	-74.7735
3	Shabakunk Creek	SHK	Shabakunk Creek at Temple House	Assunpink Creek		Delaware River	40.2809	-74.7720
3	Shimers Brook	SHM	Shimers Brook	Delaware River		Delaware River	41.2988	-74.7451
2	Trib Rock Brook	SLM	Sourland Mountain Nature Preserve	Stony Brook		Raritan River	40.4228	-74.7848
2	Six Mile Run	SM	Sixmile@Sabilla Park	Millstone River	U	Raritan River	40.4568	-74.4971
1	Six Mile Run	SMR	Six Mile Run@Hidden Lake Drive	Millstone River	D	Raritan River	40.4553	-74.5145
3	Stony Brook	SNB	Stony Brook	Big Flat Brook		Delaware River	41.2061	-74.7741
3	Snydertown tributary	STT	Snydertown Tributary	Stony Brook		Raritan River	40.3991	-74.8332
1	Sun Valley Brook	SVB	Sun Valley Brook@Wolfe Rd	S.B. Raritan River		Raritan River	40.8514	-74.7482
2	Trib Van Saun Brook	TCB	Trib Van Saun Brook@Oradell	Hackensack River		Hackensack River	40.9445	-74.0479
3	Tillman Brook	TLB	Tillman Brook	Flat Brook		Delaware River	41.1561	-74.8699
1	Vancampens Brook	VCD	At Millbrook Village	Delaware River	D	Delaware River	41.0728	-74.9626
1	Vancampens Brook	VCU*	At Flatbrook-Spillwater Rd. Bridge	Delaware River	U	Delaware River	41.0953	-74.9294
2	West Branch Middle Brook	WBR	At Bridgewater Park off Crim Rd.	Middle Brook		Raritan River	40.6124	-74.5914
3	Woolsey Brook	WLB	Woolsey Brook, at Alliger Park	Jacob's Creek		Delaware River	40.3098	-74.8241

Table 2 cont'd. Stations sampled in phases I, II, and III of the headwater IBI study.

* = site with unique characteristic(s)

Table 3. Watershed characteristics of stations sampled in phases I, II, and III of the headwater IBI study.

Water Body	Phase	Station	Ag (%)	Barren (%)	Forest (%)	Urban (%	Water (%)	Wet- land (%)	Drain- age Area (km2)	Ln(Drain- age Area)	Pond or Lake Upstream (Y/N)
Bedens Brook	2	BB	33.88	0.28	11.38	53.38	0.31	0.78	1.38	0.32	N
Trib to Big Flat Brook	2	BFB	0.00	0.12	76.09	0.42	0.16	23.21	4.82	1.57	Y
Trib to Budd Lake	2	BL	3.78	0.00	52.87	29.39	0.00	13.96	1.31	0.27	Ν
Blue Brook	3	BLB	2.18	0.03	64.10	29.18	1.88	2.63	6.87	1.93	Y
Bear Brook	2	BRB	0.00	3.83	55.14	36.19	0.00	4.84	3.22	1.17	Ν
Bear Creek	3	BRC	23.01	0.00	56.87	5.77	4.70	9.66	3.35	1.21	Y
Bear Swamp Brook	3	BSB	0.00	0.00	87.08	2.10	2.70	8.12	7.57	2.02	Y
Cub Brook	2	CB	0.00	3.33	48.71	43.25	0.00	4.70	3.61	1.28	Ν
Coles Brook	2	CBK	0.00	1.37	4.89	92.50	0.00	1.24	2.96	1.08	Ν
Trib to Ramapo	3	CGM	0.88	0.00	82.62	9.84	0.00	6.66	1.51	0.41	Ν
Camp Harmony Brook	1	CHB	4.01	0.00	46.94	7.41	0.00	41.64	6.50	1.87	N
Cresskill Brook	2	CK	0.00	1 37	4 89	92.50	0.00	1 24	0.85	-0.16	N
Cooley Brook	3	CLC	0.00	0.00	91.76	0.10	1 47	6.52	3.92	1 37	Y
Corv's Brook	2	COB	7.80	2.36	26.51	39.93	0.46	22 94	3.47	1.37	v
Cherry Brook	3	CRI	15.05	0.25	44.02	22.60	0.10	17.91	5.64	1.21	v
Cherry Brook	3	CRU	4 50	0.25	47.78	22.00	0.15	17.94	2 38	0.87	N
Cresskill Brook	2	CSK	0.00	0.50	25.95	29.00 71.76	0.15	1 37	4.82	1.57	V
Dry Brook	1	DR	11 25	0.01	71.04	2 84	1 91	11.76	3.26	1.57	V
Dry block Demarest Brook	2	DEM	0.32	0.00	20.75	2.04	0.24	2 /1	2.48	0.01	I N
Demerest Brook	2	DEM2	0.52	0.50	20.75	71.57	0.24	0.66	0.36	-1.02	N
Dunfield Creek	1	DEWIZ	0.00	0.07	27.20	0.00	0.00	2.55	0.30	-1.02	IN V
Duilleid Cleek	1		5.27	0.00	97.34	74.00	0.11	2.33	9.20	2.25	I V
Duole Dond Dun	1	DUK	J.27	0.00	0 02	/4.99	0.07	2.45 22.76	10.96	2.40	I V
Electric Drock	2	DPK	13.83	0.57	8.83 10.07	40.52	0.09	35.70	14.15	2.05	Y V
Electric Brook	3	ELB	5.09	2.01	19.97	44.90	1.08	20.33	1.00	0.51	Y V
Fox Kiver	3	FK CDD	0.00	0.00	88.05	5.02	0.78	0.15	2.04	0.97	Y V
Green Brook	3	GKB	0.00	0.00	86.20	0.00	1.20	12.60	0.86	-0.15	Y
I rib to 3rd Neshanic River	2	HQ	52.24	0.00	19.99	12.20	0.4/	15.10	2.60	0.96	N
Jacksonburg Creek	1	JAC	0.40	0.19	83.71	4.43	1.21	10.06	6.28	1.84	Y
Jennings Brook	3	JB	0.00	0.00	93.36	0.00	0.12	6.53	6.98	1.94	Y
Jennings Brook (Meadow)	3	JBM	0.00	0.00	93.42	0.00	0.12	6.47	6.88	1.93	Y
Loantaka Brook	2	LA	0.00	0.00	11.50	86.83	0.80	0.87	1.63	0.49	N
Ledgewood Brook	2	LW	0.00	0.00	58.12	34.75	0.00	7.14	1.49	0.40	N
Mores Creek	2	MC	0.00	0.00	3.37	96.63	0.00	0.00	1.74	0.55	N
Metzler Brook	2	MKW	0.00	0.10	1.91	94.95	0.40	2.64	5.65	1.73	Y
Montana Brook	1	MON	30.38	0.00	63.50	5.88	0.08	0.17	1.56	0.44	Ν
Trib to 2nd Neshanic	2	NCB	15.67	0.00	27.94	45.64	0.19	10.56	2.25	0.81	Y
Trib to 2nd Neshanic	2	NCS	24.57	0.00	21.02	46.04	0.00	8.38	1.31	0.27	Ν
Trib to Primrose Brook	1	PB	0.00	0.00	93.52	5.27	0.00	1.21	1.45	0.37	Ν
Peckman River	1	PCK	0.11	0.22	24.32	71.81	0.51	3.04	12.03	2.49	Y
Pequannock River	2	PQ	0.00	0.00	80.41	0.49	7.32	11.78	2.61	0.96	Y
Pleasant Run	2	PR	21.88	0.00	44.32	31.27	0.08	2.45	3.48	1.25	Y
Ramsey Brook	3	RAB	0.76	0.78	10.88	78.67	2.28	6.63	5.66	1.73	Y
Trib to Robinsons Branch	1	RB	0.00	0.23	7.98	91.56	0.12	0.11	2.72	1.00	Y
Rockaway Creek	2	RBH	6.52	0.00	52.12	25.27	0.50	15.59	1.88	0.63	Ν
Trib to Royce Brook	1	RBK	4.42	8.64	12.41	71.20	0.00	3.34	4.00	1.39	Ν
Rockaway Creek	2	RC	11.87	0.15	43.38	32.55	0.37	11.68	4.00	1.39	Y
Rock Brook	1	RKB	0.50	0.99	60.44	6.82	0.07	31.18	8.43	2.13	Ν

Table 3 cont'd. Watershed characteristics of stations sampled in phases I, II, and III of the headwater IBI study.

Water Body	Phase	Station	Ag (%)	Barren (%)	Forest (%)	Urban (%	Water (%)	Wet- land (%)	Drain- age Area (km2)	Ln(Drain- age Area)	Pond or Lake Ups tream (Y/N)
Ryker Lake Trib	3	RLT	0.00	0.00	72.51	8.29	0.78	18.42	6.06	1.80	Y
Ryker Lake Trib (meadow)	3	RTM	0.00	0.00	72.69	7.36	0.82	19.13	5.74	1.75	Y
Russia Brook	3	RUB	0.07	0.00	46.76	37.67	3.81	11.69	5.33	1.67	Y
Stephen's Brook	2	SB	0.00	0.00	88.56	0.90	0.00	10.53	0.95	-0.05	Ν
Stony Brook trib.	3	SBT	0.00	0.00	88.15	0.00	0.28	11.57	1.91	0.65	Ν
Shabakunk Creek	3	SHK	4.18	3.11	14.12	67.59	0.42	10.57	7.73	2.05	Y
Shimers Brook	3	SHM	0.02	0.00	79.54	0.92	0.38	19.13	5.98	1.79	Y
Trib Rock Brook	2	SLM	10.70	0.00	51.61	7.97	0.00	29.72	1.43	0.36	Ν
Six Mile Run	2	SM	1.42	0.39	2.22	87.49	0.32	8.16	3.05	1.12	Ν
Six Mile Run	1	SMR	0.47	3.28	3.16	82.67	0.20	10.21	5.39	1.69	Ν
Stony Brook	3	SNB	0.00	0.00	91.59	0.64	1.61	6.16	4.01	1.39	Y
Snydertown tributary	3	STT	5.58	0.00	58.13	9.44	0.07	26.78	2.08	0.73	Ν
Sun Valley Brook	1	SVB	6.67	0.00	52.61	26.38	0.00	14.34	1.87	0.62	Ν
Trib Van Saun Brook	2	TCB	0.00	0.00	9.18	89.90	0.00	0.93	2.59	0.95	Ν
Tillman Brook	3	TLB	0.00	0.28	96.10	0.00	0.00	3.62	4.10	1.41	Ν
Vancampens Brook	1	VCD	0.00	1.25	96.55	0.12	1.08	2.20	9.70	2.27	Y
Vancampens Brook	1	VCU	0.00	0.00	88.63	1.47	4.54	5.37	2.90	1.06	Y
W. Branch Middle Brook	2	WBR	3.49	0.51	22.99	50.70	0.02	22.29	4.87	1.58	Ν
Woolsey Brook	3	WLB	24.98	0.53	32.41	39.43	0.34	2.31	5.39	1.68	Y

Table 4. Summary of site measurements in phases I, II, and III of the headwater IBI study.

			A = m	Decek	Comore	Ave.	Ave.
Weter Det	Dhaga	Station	Ave.	Creations	Canopy	Wetted	Thalweg
water body	Phase			Gradient	(0()	Width	Depth
			(µs/cm)	(%)	(%)	(m)	(m)
Bedens Brook	2	BB	308	0.07	95.15	3.2	0.21
Trib to Big Flat Brook	2	BFB	121	5.05	98.44	2.2	0.10
Trib to Budd Lake	2	BL	245	2.44	100	1.9	0.10
Blue Brook	3	BLB	450	0.10	88.3	2.4	0.22
Bear Brook	2	BRB	416	1.80	99.48	3.0	0.11
Bear Creek	3	BRC	463	0.06	80.5	2.9	0.13
Bear Swamp Brook	3	BSB	34	2.64	98.18	10.4	0.25
Cub Brook	2	CB	110	3.49	99.74	2.0	0.09
Coles Brook	2	CBK	639	0.52	93.76	2.2	0.10
Trib to Ramapo	3	CGM	134	1.97	98.7	3.5	0.14
Camp Harmony Brook	1	CHB	-	0.70	84.7	28.9	0.30
Cresskill Brook	2	CK	849	6.21	98.7	1.9	0.09
Cooley Brook	3	CLC	46	0.74	99.74	3.9	0.13
Cory's Brook	2	COB	238	0.69	82.58	3.2	0.22
Cherry Brook	3	CRL	305	0.23	78.68	4.3	0.29
Cherry Brook	3	CRU	279	1.91	97.66	3.2	0.10
Cresskill Brook	2	CSK	519	0.40	88.82	4.3	0.25
Dry Brook	1	DB	176	5.50	81.8	17.1	0.25
Demarest Brook	2	DEM	36	0.19	96.1	2.7	0.19
Demerest Brook	2	DEM2	-	7.14	-	1.7	0.09
Dunfield Creek	1	DF	28	3.60	82.8	9.7	0.32
Dorotockeys Run	1	DOR	707	0.26	85.9	5.0	0.38
Duck Pond Run	3	DPR	205	0.35	97.92	3.0	0.19
Electric Brook	3	ELB	34	1.63	97.92	3.1	0.20
FoxRiver	3	FR	66	3.27	100	4.4	0.17
Green Brook	3	GRB	26	9.04	-	3.4	0.12
Trib to 3rd Neshanic River	2	HQ	205	0.30	98.44	3.0	0.20
Jacksonburg Creek	1	JAC	68	1.36	92.9	6.7	0.25
Jennings Brook	3	JB	110	1.31	98.96	5.7	0.19
Jennings Brook (Meadow)	3	JBM	-	0.00	42.45	5.2	0.83
Loantaka Brook	2	LA	1427	0.23	96.62	1.6	0.10
Ledgewood Brook	2	LW	563	1.84	94.8	1.4	0.14
Mores Creek	2	MC	490	0.31	92.98	3.1	0.11
Metzler Brook	2	MKW	-	0.17	89.08	4.9	0.13
Montana Brook	1	MON	109	4.75	94.1	2.5	0.16
Trib to 2nd Neshanic River	2	NCB	181	1.23	93.24	2.3	0.20
Trib to 2nd Neshanic River	2	NCS	38	2.76	99.74	2.8	0.14
Trib to Primrose Brook	1	PB	91	2.65	94.5	6.5	0.12
Peckman River	1	PCK	740	0.78	68.3	9.4	0.36
Pequannock River	2	PQ	89	3.28	78.94	3.6	0.11
Pleasant Run	2	PR	253	2.00	65.7	2.8	0.18
Ramsey Brook	3	RAB	597	0.75	96.36	3.7	0.16
Trib to Robinsons Branch	1	RB	285	0.94	100	3.7	0.16
Rockaway Creek	2	RBH	320	0.10	96.9	2.6	0.14
Trib to Royce Brook	1	RBK	320	0.13	84.2	12.7	0.31

Table 4 cont'd. Summary of site measurements in phases I, II, and III of the headwater IBI study.

Water Body	Phase	Station	Ave. Cond. (µs/cm)	Reach Gradient (%)	Canopy Cover (%)	Ave. Wetted Width (m)	Ave. Thalweg Depth (m)
Rockaway Creek	2	RC	315	0.73	77.64	3.9	0.19
Rock Brook	1	RKB	150	1.30	66.5	19.7	0.30
Ryker Lake Trib	3	RLT	220	3.31	98.96	22.9	0.10
Ryker Lake Trib (meadow)	3	RTM	212	0.35	33.44	2.4	0.16
Russia Brook	3	RUB	510	1.43	88.04	6.4	0.17
Stephen's Brook	2	SB	67	10.29	-	1.4	0.02
Stony Brook trib.	3	SBT	33	3.57	99.48	5.8	0.12
Shabakunk Creek	3	SHK	408	0.38	97.4	5.8	0.19
Shimers Brook	3	SHM	144	5.25	92.46	1.2	0.06
Trib Rock Brook	2	SLM	50	2.29	98.18	2.1	0.26
Six Mile Run	2	SM	397	0.06	89.6	4.2	0.18
Six Mile Run	1	SMR	415	0.40	82.5	17.7	0.17
Stony Brook	3	SNB	47	2.44	98.7	5.7	0.19
Snydertown tributary	3	STT	91	3.04	96.1	3.5	0.21
Sun Valley Brook	1	SVB	165	1.60	76.9	10.8	0.21
Trib Van Saun Brook	2	TCB	874	0.12	91.68	2.0	0.10
Tillman Brook	3	TLB	39	3.26	98.96	3.5	0.14
Vancampens Brook	1	VCD	33	2.20	99.4	6.0	0.23
Vancampens Brook	1	VCU	27	3.25	100	4.8	0.15
West Branch Middle Brook	2	WBR	332	0.54	99.22	3.3	0.14
Woolsey Brook	3	WLB	377	0.45	73.22	2.3	0.22

Table 5. Stations with unique characteristics sampled in phases I, II, and III of the headwater IBI study. Data from these sites were not used in formulation of metrics when including them greatly skewed the results.

Water Body	Station	Description of unique characteristic	Fauna (all sampling combined)	
Cresskill Brook	СК	High urban land-use not reflected in site conditions	26 C. bartonii , 1 E. bislineata , 2 P. cinereus , 64 R. atratulus, 29 S. fontinalis	
Demerest Brook DEM2		Very high gradient (7%), suspected to be intermittent	No fish, salamanders, or crayfish	
Green Brook	GRB	Stream intermittent; reach dry during one of two visits	1 C. bartonii , 1 L. clamitans, 3 P. cinereus, and 1 P. glutinosus,	
Loantaka Brook	LA	Unknown cause; water quality effects	In 2005: No fish, salamanders, or crayfish. In 2008: 9 <i>L. gibbosus</i> , 3 <i>S. atromaculatus</i> , no salamanders or crayfish.	
Ledgewood Brook	LW	Water quality effects due to historical garbage disposal	1 C. bartonii , 1 E. bislineata , 1 P. ruber, 93 R. atratulus	
Stephen's Brook SB		Stream very shallow; very high gradient (10%); subterranean portions of reach	No fish, salamanders, or crayfish collected while electrofishing; 2 D. fuscus, 8 C. bartonii, 10 E. bislineata, 2 P. cinereus, 2 P. ruber	
Vancampens Brook	VCU	Acidification (3.96 pH)	70 C. bartonii , 9 L. clamitans , 2 L. palustris , 1 L. sylvaticus , 3 S. fontinalis , 5 P. cinereus , 3 P. glutinosus	

	Metric Scores							
Station	Forest (%)	Salamander & Sensitive Frog Richness	No. of C. bartonii	Prop. of Sensitive Cool HW Fishes	Prop. of S. fontinalis YOY	Multi- metric Score		
BL	52.87	0.0	0.0	0.0	0.0	0.0		
BRB	55.14	33.3	0.0	0.0	0.0	8.3		
CB	48.71	33.3	0.0	0.0	0.0	8.3		
CBK	4.89	0.0	0.0	0.0	0.0	0.0		
CGM	82.62	66.6	14.3	0.0	0.0	20.3		
CHB	46.94	66.6	0.0	0.0	0.0	16.7		
CK*	4.89	33.3	22.2	100.0	61.3	54.2		
CRU	47.78	33.3	0.0	0.0	0.0	8.3		
DEM2*	27.20	0.0	0.0	0.0	0.0	0.0		
LW*	58.12	66.6	11.1	0.0	0.0	19.4		
MON	63.50	33.3	66.7	100.0	85.6	71.4		
NCS	21.02	33.3	44.4	0.0	0.0	19.4		
PB	93.52	100.0	100.0	99.6	80.9	95.1		
RKB	60.44	66.6	22.2	0.0	0.0	22.2		
SB*	88.56	100.0	0.0	0.0	0.0	25.0		
SBT	88.15	100.0	100.0	99.0	100.0	99.7		
SLM	51.61	66.6	11.1	0.0	0.0	19.4		
STT	58.13	66.6	0.0	0.0	0.0	16.7		
SVB	52.61	100.0	22.2	2.1	0.0	31.1		
TLB	96.10	100.0	55.6	61.7	100.0	79.3		
WBR	22.99	33.3	0.0	0.0	0.0	8.3		

Table 6a. Metric scores for high gradient reaches without ponds sampled in phases I, II, and III of the headwater IBI study.

* = site with unique characteristic(s);not used to develop metrics

	Metric Scores						
Station	Forest (%)	Salamander & Sensitive Frog Richness	No. of Native Crayfish (excl. <i>O. limosus</i>)	Prop. of Sensitive Warm & Cool HW Fishes	Multi- metric Score		
BFB	76.09	75.0	25.0	61.1	53.7		
BSB	87.08	75.0	86.6	44.9	68.8		
CLC	91.76	50.0	100.0	100.0	83.3		
COB	26.51	25.0	0.0	0.0	8.3		
DB	71.04	75.0	87.5	0.0	54.2		
DF	97.34	75.0	56.3	14.6	48.6		
ELB	19.97	25.0	0.0	8.4	11.1		
FR	88.05	50.0	0.0	0.0	16.7		
GRB*	86.20	0.0	0.0	0.0	0.0		
JAC	83.71	100.0	0.0	20.1	40.0		
JB	93.36	25.0	31.3	38.9	31.7		
NCB	27.94	25.0	0.0	0.0	8.3		
PCK	24.32	25.0	0.0	0.9	8.6		
PQ	80.41	50.0	0.0	7.9	19.3		
PR	44.32	25.0	0.0	0.0	8.3		
RAB	10.88	0.0	0.0	0.0	0.0		
RB	7.98	0.0	0.0	0.0	0.0		
RC	43.38	25.0	0.0	0.0	8.3		
RLT	72.51	75.0	12.5	29.6	39.0		
RUB	46.76	50.0	6.3	14.6	23.6		
SHM	79.54	50.0	6.3	5.9	20.7		
SNB	91.59	75.0	25.0	0.4	33.5		
VCD	96.55	100.0	6.3	43.3	49.9		
VCU	88.63	50.0	100.0	100.0	83.3		

Table 6b. Metric scores for high gradient reaches with ponds sampled in phases I, II, and III of the headwater IBI study.

* = site with unique characteristic(s);not used to develop metrics

			Metric Scores					
Station	Forest	Pond or Lake	A	No. of	Prop. of Sensitive	Prop. of	Multi-	
Station	(%)	Ups tream (Y/N)	Richness	Native Crayfish	Warm HW	Tolerant Fishes	metric Score	
DD	11 20	N	25.0	62	Fisnes	42.1	19.6	
BB	11.38	IN V	25.0	0.3	0.0	45.1	18.0	
DLD	04.10	r V	100.0	100.0	0.7	49.5	04.1	
BRU	30.87	Y V	100.0	0.0	0.0	//.8	44.5	
CRL	44.02	Y	25.0	100.0	0.0	49.8	43.7	
CSK	25.95	Y	0.0	0.0	0.0	22.7	5.7	
DEM	20.75	Ν	25.0	0.0	0.0	11.4	9.1	
DOR	14.24	Y	0.0	0.0	0.0	31.4	7.8	
DPR	8.83	Y	25.0	0.0	36.3	73.0	33.6	
HQ	19.99	Ν	25.0	12.5	0.0	47.1	21.1	
JBM	93.42	Y	25.0	50.0	100.0	100.0	68.8	
LA*	11.50	Ν	0.0	0.0	0.0	78.9	19.7	
MC	3.37	Ν	0.0	0.0	0.0	0.0	0.0	
MKW	1.91	Y	0.0	0.0	0.0	20.0	5.0	
RBH	52.12	Ν	50.0	0.0	0.0	64.8	28.7	
RBK	12.41	Ν	25.0	25.0	18.6	70.4	34.8	
RTM	72.69	Y	100.0	75.0	100.0	100.0	93.8	
SHK	14.12	Y	25.0	0.0	0.0	40.9	16.5	
SM	2.22	Ν	25.0	0.0	0.0	54.2	19.8	
SMR	3.16	Ν	25.0	0.0	0.0	34.3	14.8	
TCB	9.18	Ν	0.0	0.0	0.0	67.4	16.8	
WLB	32.41	Y	75.0	0.0	0.0	56.4	32.8	

Table 6c. Metric scores for low gradient reaches sampled in phases I, II, and III of the headwater IBI study.

* = site with unique characteristic(s);not used to develop metrics

Pond(s) upstream	Metric	Reference Percentile	Reference Value	Discrimination Efficiency (%)
	Calance dan & Canaiting	25	2.750	93.8
	Salamander & Sensitive	50	3.000	93.8
	Flog Richness	75	3.250	100.0
		25	3.145	87.5
	No. of C. bartonii	50	7.000	100.0
No		75	11.000	100.0
INO	Prop of Sensitive Cool	25	0.160	92.3
	HW Fishes	50	0.418	92.3
		75	0.516	92.3
	Prop of S fontinalis	25	0.300	92.3
	VOV	50	0.671	100.0
	101	75	0.762	100.0
	Salamander & Sensitive	25	2.000	61.5
	Frog Richness	50	2.500	76.9
		75	3.000	76.9
Yes	No. of Native Cravfich	25	0.000	0.0
	(excluding O limosus)	50	4.500	92.3
	(excluding O. infosus)	75	13.850	92.3
	Pron of Sensitive Warm	25	0.075	69.2
	& Cool HW/ Fishes	50	0.282	84.6
	a courrie risiles	75	0.429	92.3

Table 7. Discrimination efficiencies and reference values for metrics developed for high gradient strata in phases I, II, and III of the headwater IBI study.

Reference values for reference percentiles were determined using linear interpolation in SigmaPlot

Table 8. High gradient sites with drainage areas greater than 2 km² scored using NJ's northern IBI for streams greater than 12.95 km² and this study's proposed IBI for smaller streams. For comparison to the northern IBI, the proposed IBI categories were used: Poor (0-25), Fair (26-50), Good (51-75), and Excellent (76-100).

Station	Forest (%)	Pond or Lak e Ups tream (Y/N)	Reach Gradient (%)	Drain- age Area (km ²)	Drain- age Area (mi ²)	NJ Northern IBI Score (10-50 scale)	NJ N IBI score rescaled (0-100)	NJ Northern IBI Category	Proposed IBI Score (% of standard)	Proposed IBI Category
PCK	24.3	Y	0.78	12.0	4.6	36	65	Fair	9	Poor
VCD	96.5	Y	2.2	9.7	3.7	34	60	Fair	50	Fair
DF	97.3	Y	3.6	9.3	3.6	32	55	Fair	49	Fair
RKB	60.4	Ν	1.3	8.4	3.3	28	45	Poor	22	Poor
BSB	87.1	Y	2.64	7.6	2.9	38	70	Good	69	Good
JB	93.4	Y	1.31	7.0	2.7	36	65	Fair	32	Fair
CHB	46.9	Ν	0.7	6.5	2.5	30	50	Fair	17	Poor
JAC	83.7	Y	1.36	6.3	2.4	28	45	Poor	40	Fair
RLT	72.5	Y	3.31	6.1	2.3	28	45	Poor	39	Fair
SHM	79.5	Y	5.25	6.0	2.3	30	50	Fair	21	Poor
RAB	10.9	Y	0.75	5.7	2.2	36	65	Fair	0	Poor
RUB	46.8	Y	1.43	5.3	2.1	30	50	Fair	24	Poor

Table 9. Low gradient sites scored using NJ's southern IBI (draft) for the inner coastal plain and this study's proposed IBI for low gradient streams ($< 12.95 \text{ km}^2$) in northern NJ. For comparison to the southern IBI, the proposed IBI categories were used: Poor (0-25), Fair (26-50), Good (51-75), and Excellent (76-100).

Station	Forest (%)	Forest + Wetland (%)	Pond or Lake Upstream (Y/N)	Reach Gradient (%)	Drain- age Area (km ²)	Drain- age Area (mi ²)	NJ Southern IBI Score (1-5 scale)	NJ Southern IBI Score rescaled (0-100)	NJ Southern IBI Category	Proposed IBIScore (% of standard)	Proposed IBI Category
DPR	8.8	42.6	Y	0.35	14.1	5.5	3.8	69	Fair	34	Fair
DOR	14.2	19.7	Y	0.26	11.0	4.2	3.0	50	Fair	8	Poor
SHK	14.1	24.7	Y	0.38	7.7	3.0	3.0	50	Fair	16	Poor
JBM	93.4	99.9	Y	0	6.9	2.7	3.0	50	Fair	69	Good
BLB	64.1	66.7	Y	0.1	6.9	2.7	3.0	50	Fair	64	Good
RTM	72.7	91.8	Y	0.35	5.7	2.2	3.5	63	Fair	94	Excellent
MKW	1.9	4.5	Y	0.17	5.7	2.2	2.5	38	Poor	5	Poor
CRL	44.0	61.9	Y	0.23	5.6	2.2	4.0	75	Good	44	Fair
SMR	3.2	13.4	Ν	0.4	5.4	2.1	3.5	63	Fair	15	Poor
WLB	32.4	34.7	Y	0.45	5.4	2.1	3.5	63	Fair	33	Fair
CSK	26.0	27.3	Y	0.4	4.8	1.9	3.5	63	Fair	6	Poor
RBK	12.4	15.7	Ν	0.13	4.0	1.5	3.8	69	Fair	35	Fair
BRC	56.9	66.5	Y	0.06	3.4	1.3	2.5	38	Poor	44	Fair
SM	2.2	10.4	Ν	0.06	3.1	1.2	4.0	75	Good	20	Poor
HQ	20.0	35.1	Ν	0.3	2.6	1.0	4.3	81	Good	21	Poor
TCB	9.2	10.1	Ν	0.12	2.6	1.0	2.3	31	Poor	17	Poor
DEM	20.7	23.2	Ν	0.19	2.5	1.0	3.5	63	Fair	9	Poor
RBH	52.1	67.7	Ν	0.1	1.9	0.7	4.0	75	Good	29	Fair
MC	3.4	3.4	Ν	0.31	1.7	0.7	1.5	13	Very Poor	0	Poor
BB	11.4	12.2	Ν	0.07	1.4	0.5	3.5	63	Fair	19	Poor



Figure 1. CCA biplot of species scores and environmental variables for taxa collected by backpack electrofishing. AG= % agriculture, BL=% barren land, DRN=drainage area, GRD= reach gradient, FST = % forest, URB = % urban, WAT=% water, and WET=% wetland. Species not included in proposed metrics are not shown.



Figure 2. Box plot of salamander collection techniques comparing the number of species collected per minute. Median, interquartile range, 95th percentile, and outliers are shown for each technique. Mean rank for timed technique was significantly greater than transect and electrofishing techniques (P<0.044). Timed N= 22, Transect N= 18, and Electrofishing N=23.



Figure 3. Box plot of salamander collection techniques comparing the number of salamanders collected per minute. Median, interquartile range, 95th percentile, and outliers are shown for each technique. Mean rank for transect technique was significantly greater than timed and electrofishing techniques (P<0.020) but no pairwise differences were significant. Timed N= 22, Transect N= 21, and Electrofishing N=23.



Figure 4. Box plot of multimetric scores for high gradient streams with ponds upstream. Median, interquartile range, 95th percentile, 5th percentile, and outliers are shown for each forest cover category when applicable. >80% N=10, 20-80% N=10, and <20% N=3.



Figure 5. Box plot of multimetric scores for high gradient streams without ponds upstream. Median, interquartile range, 95th percentile, 5th percentile, and outliers are shown for each forest cover category when applicable. >80% N=4, 20-80% N=12, and <20% N=1.



Figure 6. Box plot of multimetric scores for low gradient streams. Median, interquartile range, 95th percentile, 5th percentile, and outliers are shown for each forest cover category when applicable. >80% N=1, 20-80% N=8, and <20% N=11.



Figure 7. Box plot of salamander and sensitive frog richness for high gradient streams without ponds upstream. Median, interquartile range, 95th percentile, 5th percentile, and outliers are shown for each forest cover category when applicable. >80% N=5, 20-80% N=14, and <20% N=2.



Figure 8. Box plot of *Cambarus bartonii* CPUE for high gradient streams without ponds upstream. Median, interquartile range, 95th percentile, 5th percentile, and outliers are shown for each forest cover category when applicable. >80% N=4, 20-80% N=14, and <20% N=2.



Figure 9. Box plot of the proportion of sensitive cool headwater fishes for high gradient streams without ponds upstream. Median, interquartile range, 95th percentile, 5th percentile, and outliers are shown for each forest cover category when applicable. >80% N=4, 20-80% N=12, and <20% N=1.



Figure 10. Box plot of the proportion of age-0 *Salvelinus fontinalis* for high gradient streams without ponds upstream. Median, interquartile range, 95th percentile, 5th percentile, and outliers are shown for each forest cover category when applicable. >80% N=4, 20-80% N=12, and <20% N=1.



Figure 11. Box plot of salamander and sensitive frog richness for high gradient streams with ponds upstream. Median, interquartile range, 95th percentile, 5th percentile, and outliers are shown for each forest cover category when applicable. >80% N=10, 20-80% N=10, and <20% N=3.



Figure 12. Box plot of *native crayfish* (excluding *Orconectes limosus*) CPUE for high gradient streams with ponds upstream. Median, interquartile range, 95th percentile, 5th percentile, and outliers are shown for each forest cover category when applicable. >80% N=10, 20-80% N=10, and <20% N=3.



Figure 13. Box plot of the proportion of sensitive warm and cool headwater fishes for high gradient streams with ponds upstream. Median, interquartile range, 95th percentile, 5th percentile, and outliers are shown for each forest cover category when applicable. >80% N=10, 20-80% N=10, and <20% N=3.



Figure 14. Box plot of % forest cover for high (>0.5%) and low (<0.5%) gradient sites sampled as part of the phase I, II, and III of the headwater study. Median, interquartile range, 95th percentile, 5th percentile, and outliers are shown for each gradient grouping (>0.5 N=45, <0.5 N=21).



Figure 15. Box plot of amphibian richness for low gradient streams. Median, interquartile range, 95th percentile, 5th percentile, and outliers are shown for each forest cover category when applicable. >80% N=1, 20-80% N=8, and <20% N=12.



Figure 16. Scatter plot of amphibian richness versus percent forest cover for low gradient streams.



Figure 17. Box plot of native crayfish CPUE for low gradient streams. Median, interquartile range, 95th percentile, 5th percentile, and outliers are shown for each forest cover category when applicable. >80% N=1, 20-80% N=8, and <20% N=12.



Figure 18. Scatter plot of native crayfish CPUE versus percent forest cover for low gradient streams.



Figure 19. Box plot of tolerant fishes for low gradient streams. Median, interquartile range, 95th percentile, 5th percentile, and outliers are shown for each forest cover category when applicable. >80% N=1, 20-80% N=8, and <20% N=11.



Figure 20. Scatter plot of tolerant fishes versus percent forest cover for low gradient streams.



Figure 21. Scatter plot of tolerant fishes versus percent urban for low gradient streams.



Figure 22. Box plot of the proportion of sensitive warm headwater fishes for low gradient streams. Median, interquartile range, 95th percentile, 5th percentile, and outliers are shown for each forest cover category when applicable. >80% N=1, 20-80% N=8, and <20% N=11.



Figure 23. Scatter plot the proportion of sensitive warm headwater fishes versus percent forest cover for low gradient streams.



Figure 24. Scatter plot of the proportion of salamander and sensitive frog richness scores versus total habitat quality scores for high gradient streams (includes sites with and without ponds).



Figure 25. Scatter plot of *Cambarus bartonii* CPUE scores versus total habitat quality scores for high gradient streams without ponds.



Figure 26. Scatter plot of cool headwater fishes scores versus total habitat quality scores for high gradient streams without ponds.



Figure 27. Scatter plot of Age-0 *S. fontinalis* scores versus total habitat quality scores for high gradient streams without ponds.



Figure 28. Scatter plot of native crayfish CPUE (no *O. limosus*) scores versus total habitat quality scores for high gradient streams with ponds.



Figure 29. Scatter plot of sensitive warm and cool headwater fishes scores versus total habitat quality scores for high gradient streams with ponds.



Figure 30. Scatter plot of amphibian richness scores versus total habitat quality scores for low gradient streams.



Figure 31. Scatter plot of native crayfish CPUE scores versus total habitat quality scores for low gradient streams.



Figure 32. Scatter plot of tolerant fishes scores versus total habitat quality scores for low gradient streams.



Figure 33. Scatter plot of sensitive warm headwater fishes scores versus total habitat quality scores for low gradient streams.



Figure 34. Scatter plot of multimetric scores versus total habitat quality scores for high gradient streams without ponds.



Figure 35. Scatter plot of multimetric scores versus total habitat quality scores for high gradient streams with ponds.



Figure 36. Scatter plot of multimetric scores versus total habitat quality scores for low gradient streams.



Figure 37. Scatter plot of total habitat quality scores versus percent forest for all sites.

	Metric Values							
Station	Salamander and Sensitive Frog Richness	No. of C. bartonii	Proportion of Sensitive Cool Headwater Fishes	Proportion of Age-0 S. fontinalis				
BL	0	0	0.00	0.00				
BRB	1	0	0.00	0.00				
CB	1	0	0.00	0.00				
CBK	0	0	0.00	0.00				
CGM	2	1	0.00	0.00				
CHB	2	0	0.00	0.00				
CK	1	2	-	-				
CRU	1	0	0.00	0.00				
DEM2	0	0	-	-				
LW	2	1	-	-				
MON	1	6	1.00	63.49				
NCS	1	4	0.00	0.00				
PB	3	13	0.52	60.00				
RKB	2	2	0.00	0.00				
SB	4	-	-	-				
SBT	3	9	0.51	74.19				
SLM	2	1	0.00	0.00				
STT	2	0	0.00	0.00				
SVB	3	2	0.01	0.00				
TLB	3	5	0.32	78.26				
WBR	1	0	0.00	0.00				

Appendix A. Values used to calculate metric scores for high gradient streams without ponds upstream.

	Metric Values								
Station	Salamander and Sensitive Frog Richness	No. of Native Crayfish (no <i>O. limosus</i>)	Proportion of Sensitive Warm and Cool Headwater Fishes						
BFB	3	4	0.59						
BSB	3	14	0.43						
CLC	2	23	0.96						
COB	1	0	0.00						
DB	3	14	0.00						
DF	3	9	0.14						
ELB	1	0	0.08						
FR	2	0	0.00						
JAC	4	0	0.19						
JB	1	5	0.37						
NCB	1	0	0.00						
PCK	1	0	0.01						
PQ	2	0	0.08						
PR	1	0	0.00						
RAB	0	0	0.00						
RB	0	0	0.00						
RC	1	0	0.00						
RLT	3	2	0.28						
RUB	2	1	0.14						
SHM	2	1	0.06						
SNB	3	4	0.00						
VCD	5	1	0.41						
VCU	2	16	1.00						

Appendix B. Values used to calculate metric scores for high gradient streams with ponds upstream.

	Metric Values								
Station	Amphibian Richness	No. of Native Crayfish	Proportion of Tolerant Fishes	Proportion of Sensitive Warm Headwater Fishes					
BB	1	1	0.59	0.00					
BLB	4	20	0.53	0.05					
BRC	4	0	0.26	0.00					
CRL	1	16	0.52	0.00					
CSK	0	0	0.78	0.00					
DEM	1	0	0.89	0.00					
DOR	0	0	0.70	0.00					
DPR	1	0	0.31	0.27					
HQ	1	2	0.55	0.00					
JBM	1	8	0.01	0.90					
LA	0	0	-	-					
MC	0	0	0.99	0.00					
MKW	0	0	0.81	0.00					
RBH	2	0	0.38	0.00					
RBK	1	4	0.33	0.14					
RTM	4	12	0.05	0.74					
SHK	1	0	0.61	0.00					
SM	1	0	0.48	0.00					
SMR	1	0	0.67	0.00					
TCB	0	0	0.36	0.00					
WLB	3	0	0.46	0.00					

Appendix C. Values used to calculate metric scores for low gradient streams.