Midwest Biodiversity Institute
P.O. Box 21561
Columbus, OH 43221-0561

Development of Methods and Designs for the Assessment of the Fish Assemblages of Non-Wadeable Rivers in New England


14’ Electrofishing Raft


West River, VT


Dead River Falls, Maine


Connecticut River, Turners Falls, MA

MBI Technical Report 2015-3-3
December 31, 2015

Yoder, C.O., E.T. Rankin, and Lon E. Hersha. 2015. Development of Methods and Designs for the Assessment of the Fish Assemblages of Non-Wadeable Rivers in New England. MBI Technical Report MBI/2015-3-3. U.S. EPA Assistance Agreement RM-83379101. U.S. EPA, Office of Research and Development, Atlantic Ecology Division, Narragansett, RI and U.S. EPA, Region I, Boston, MA. 152 pp. http://www.midwestbiodiversityinst.org/.

Portions of this document were made possible by a generous grant from ESRI. The GIS elements of this report were also made possible by a grant from ESRI.

Development of Methods and Designs for the Assessment of the Fish Assemblages of Non-Wadeable Rivers in New England

December 31, 2015

MBI Technical Report MBI/2015-3-3

Submitted to:

U.S. EPA, Office of Research & Development
Atlantic Ecology Division
27 Tarzwell Drive
Narragansett, RI 02882
Donald Cobb, Project Officer

and

U.S. EPA, Region I
EPA New England Laboratory
11 Technology Drive
North Chelmsford, MA 01863-2431
Hilary Snook, Technical Contact

In Partial Fulfillment of the Work Plan for U.S. EPA Assistance Agreement RM-83379101

Submitted by:

Midwest Biodiversity Institute
P.O. Box 21561
Columbus, OH 43221-0561
Chris O. Yoder, Principal Investigator
cyoder@mwbinst.com

Table of Contents

List of Tables

List of Figures

Acknowledgements

Contributions to the report and the analyses were made by the following persons each recognized for their particular specialty. Field crew leaders were Lon Hersha (REMAP-New England and Connecticut R.), Vickie Gordon (REMAP-Maine), Bryan Apell (Connecticut River), Joshua Schering (REMAP-NRSA), and Travis Smith (REMAP-NRSA). Field sampling assistance was provided by Alex Brickett, Matt Halfhill, Benjamen Kelley, Deb Hersha, Scott Sheets, Rusty White, and Kyle Yoder. John Audet, Kleinschmidt Associates, was the first crew member for the Maine Rivers survey in 2002 through 2005 – numerous other Kleinschmidt personnel assisted as well. Rachel Day provided the maps for the fish distribution atlas (Appendix A). General logistical support was provided by Allison Boehler and Erika Lethcoe of MBI.

Walt Galloway (U.S. EPA-AED) served the U.S. EPA Project Officer beginning in 2008 and was followed in June 2012 by Donald Cobb. Hilary Snook (U.S. EPA-Region I) served as the U.S. EPA Technical Contact and provided technical advice and field assistance throughout the project. Ralph Abele (U.S. EPA-Region I) coordinated funding assistance for the Connecticut River survey of 2008-9, helped secure ESA Section 7 Biological Opinions, and provided technical assistance and advice throughout the project. Jennie Bridge, U.S. EPA, Region I provided assistance with securing the annual grants and funding that supported the Maine Rivers surveys of 2002-7.

Dave Baysley, Gordon Kramer, and Dave Boucher of the Maine DIF&W provided critical assistance with sampling and access to Maine riverine sites. We also acknowledge the many landowners who provided access to sampling sites on their property throughout New England. Rich Langdon and David Halliwell provided critical reviews of the final draft and the Atlas of New England Riverine Fish Distributions.

Finally, many thanks go to Brandon Kulik, Kleinschmidt Associates, Pittsfield, ME for providing conceptual and logistical support throughout the project. The original idea for a fish survey of Maine Rivers was his and the construction and testing of an electrofishing boat supported by Kleinschmidt in 2001 confirmed the efficacy of the primary sampling methodology. Brandon graciously offered support in many other ways including lodging at his camp on Branch Pond in Palermo, ME, boat and vehicle storage at his home in Winslow, ME, and his home for weary electrofishing crew members.

NELR REMAP PROJECT SUMMARY AND CONCLUSIONS

Project Scope & Methods

The New England Large Rivers (NELR) REMAP project scope included non-wadeable rivers in all six New England states, specifically those rivers that required a boat or raft platform to effectively sample the fish assemblages using pulsed D.C. electrofishing. Sampling sites were selected using the draw of sites from the 2008-9 National Rivers and Streams Assessment (NRSA) to form the basis of the REMAP probabilistic sampling design for a total of 149 sites. Targeted sites from an intensive pollution survey design from the 2008-9 Connecticut River and the 2002-7 Maine Rivers fish assemblage assessments were also utilized in the data analyses and assessments of fish assemblage condition for a total of 371 additional sites.

All sites were sampled with either 14’ raft or 16’ boat mounted pulsed D.C. electrofishing based on standardized equipment and sampling methods developed in Maine during 2002-3. Fish assemblage data from a 1.0 km fixed site distance were analyzed using various multivariate techniques, species-specific stressor associations, and a fish Index of Biotic Integrity (IBI) developed in Maine during 2002-7. The Maine IBI (ME IBI) is based on a cool-coldwater ecotype for moderate to high gradient non-wadeable rivers and is supplemented with a set of four diadromous metrics (DIBI) that are separate, but additive to the ME IBI. While experimental in its testing and application, the DIBI proved useful in assessing the condition of coastal rivers where diadromous fish species are expected to occur.

Potential stressors were provided by various GIS coverages including ecological connectivity (Northeast Aquatic Connectivity), thermal classification (Northeast Aquatic Habitat Classification), land cover (National Land Cover data), and Sparrow model results (nutrients). Data collected at each fish sampling location included temperature, dissolved oxygen (D.O.), conductivity, and pH recorded as instantaneous grabs. Habitat was evaluated using the Qualitative Habitat Evaluation Index (QHEI) as modified for application to non-wadeable rivers in New England (Yoder 2006a). The QHEI provides an overall habitat score and is comprised of multiple attributes (substrate, cover, channel morphology, flow characteristics, gradient). A subset of QHEI attributes termed the Hydro-QHEI served as an indicator of flow dependent habitat characteristics. All of these variables and their attributes were analyzed against the fish assemblage data to support stressor inferences and associations.

Fish Assemblage Condition Assessment

Assessing the condition of the fish assemblage in New England rivers was a primary objective of the study. The NELR REMAP probabilistic study design was intended to provide a statistically valid assessment of overall condition hence various analyses were performed to yield the assessment statistics. In addition, the results from targeted sites that were sampled in New England prior to and alongside the 2008-9 NELR REMAP study were also assessed. The ME IBI (Yoder et al. 2008) was used as the primary measure of fish assemblage condition. Because it was initially calibrated for Maine rivers and for a cool-coldwater ecotype evaluating its applicability across New England was an important sub-objective. It is the only readily available IBI that is specifically derived and calibrated for non-wadeable rivers in New England.

NELR REMAP Results

Ranges of the ME IBI were translated to their corresponding Biological Condition Gradient (BCG) Level (1-6). Using BCG Level 4 for the ME IBI as a minimum CWA goal impairment threshold, 42.3% of the non-wadeable rivers in New England performed at or above this level based on the probabilistic dataset. A slightly higher proportion of sites (48.2%) performed above this level when the targeted sites were included, with 3.2% of the sites at BCG Level 2 which was not revealed at all by the probabilistic data set. Adding the DIBI to the ME IBI resulted in a higher proportion of sites at Level 4 or higher with 64.5% of REMAP probabilistic sites and 76.8% of targeted sites meeting Level 4 or higher.

Mainstem River Case Example

Another NELR REMAP project objective was to compare the assessment outcomes produced by different spatial sampling designs in a large mainstem river. To meet this objective targeted sites were located on selected mainstem rivers in addition to the NRSA probabilistic and overdraw sites that comprised the REMAP probabilistic sampling design. For this analysis we focused on the Connecticut River mainstem based on sampling conducted in 2008-9. Probabilistic sites were selected from the 2008-9 NRSA draw of sites for two levels of coverage. Targeted sites were added to complete a longitudinal pollution survey design along the entire non-wadeable mainstem. For the latter, the probabilistic sites served as part of that more spatially intensive design. All of the ME IBI and DIBI percentiles were statistically similar between the NRSA, REMAP, and intensive survey designs except for the 95th percentile values which were highest for the intensive pollution survey design. The targeted design produced slightly higher IBI statistics than the NELR REMAP and higher than the NRSA base sites alone. It also produced a disproportionately higher 95th percentile IBI because of the inclusion of sites with IBIs that were >12 points higher than the maximums at either the NELR REMAP or NRSA sites. Differences between the DIBI and ME IBI scores were the greatest in the tidal reaches and generally becoming less upstream. However, differences occurred well upstream into New Hampshire/Vermont and enough so that a single site in the upper mainstem moved into BCG Level 2. Given the importance of diadromous species management in the Connecticut River the DIBI concept provides a way to better utilize the fish assemblage as an indicator of connectivity.

Condition Assessment Synopsis

The NELR REMAP probabilistic data set provides a randomly generated estimate of a median ME IBI score, without which we would have been uncertain about how representative of regional conditions the targeted data set actually was. The comparison provided confirmation that a high density, targeted sampling effort within a regional scope can provide more representative estimates of condition as it produced a similar median IBI, but exclusively revealed the highest quality riverine sites in New England. Key observations about differences and similarities between the probabilistic and targeted sites results include:

- **No sites in any dataset were in BCG Level 1** – it is highly doubtful it exists anywhere in a New England river and is perhaps a reflection of legacy impacts (e.g., log drives, deforestation, dams, land settlement, industrial pollution) that occurred from the late 18th to early and mid-20th centuries.
- **The median ME IBI of the pollution survey (i.e., targeted design) sites was a close approximation of the median REMAP probabilistic ME IBI**. The estimate of the median ME IBI for the targeted sites was within the confidence interval of the probabilistic sites. A similar set of results was obtained for the Connecticut River mainstem comparative assessment.
- **The targeted survey design alone produced Level 2 ME IBI scores** - the highest NRSA or REMAP probabilistic design ME IBI was Level 3.
- **A total of 19 targeted sites had higher ME IBI scores than the highest scoring REMAP probability site** - only 4 of the 27 highest scoring sites were REMAP probabilistic sites while none were NRSA sites.
- **Based on the order of remaining NRSA overdraw sites, it would have required an additional 23 site replacements to encounter a probabilistic site in a river (i.e., the Allagash R.) that would have had any likelihood of a BCG Level 2 ME IBI score**. Essentially there is little to no chance that a BCG Level 2 ME IBI would have been yielded by any probabilistic site under the 2008-9 NRSA site draw.
- **Only two of the 27 highest scoring sites occurred outside of northern Maine** and both were in the upper Connecticut River in northern New Hampshire.
- **The DIBI had 4 probabilistic sites at BCG Level 2**, but was accompanied by a corresponding increase in targeted DIBI Level 2 sites.
- **The proportion of sites that reflected the lower levels of the BCG (i.e., Levels 3 through 6) was not substantially different between the REMAP probabilistic and New England targeted results** being within ≈5% for each level.

Stressor Analysis Results

Establishing linkages between delineated stressors and fish assemblage attributes was accomplished by examining the range and distribution of selected environmental variables across New England. Readily available datasets were accessed to estimate stressor levels or sources of potential stressor types. The Qualitative Habitat Evaluation Index (QHEI) was the primary measure of habitat quality at the site-specific level. In addition to the typical attributes of structural habitat in the QHEI, attributes that are flow dependent were extracted as a measure of hydrological dependent habitat (i.e., the “Hydro-QHEI”) to describe potential impacts of flow modifications on fish. Field parameters collected during fish sampling included D.O., temperature, pH, and conductivity. We attempted to obtain additional chemical water quality data through the EPA Water Quality Exchange (WQX), but there were too few matches with our sites and within the same seasonal index period to warrant their inclusion in the analyses. We readily acknowledge that the stressor variables very likely do not represent the full range and types of stressors that affect New England riverine fish assemblages. Available GIS coverages that were employed in the stressor datasets included:

1. Ecological Connectivity – Northeast Aquatic Connectivity (NAC) information and metrics about dams, anadromous fish habitat, and other parameters.
2. Temperature Classification - Northeast Aquatic Habitat Classification (NEAHCS) system (Olivero and Anderson 2008).
3. Land Cover - consisted of land cover types in the total catchment upstream of a site and in local proximity to a site.
4. Nutrient Enrichment - Sparrow predicted nutrient concentrations for total phosphorus and nitrogen (as a surrogate for water quality data).

Multivariate Analyses

Non-metric multidimensional scaling (NMS), cluster analysis, and indicator species analysis (ISA) was used to explore fish species composition in response to natural and anthropogenic variables. Bray–Curtis dissimilarity was used as the distance measure for all analyses. A stressor variable reduction process was conducted, using principal component analysis and correlation analysis to select a reduced set of environmental and stressor variables. Canonical Correspondence Analyses (CCA) was used to identify the strength and direction of significant stressor relationships and to create an overall Human Disturbance Index (HDI). Selected stressors were plotted vs. the number of intolerant fish species to identify threshold values (by eye) in each stressor-response relationship. We then standardized each stressor on a 1-10 scale and weighted each stressor score by the average of the coefficients from the CCA analyses (1^{st} and 2^{nd} axes). The metric scores, now weighted by CCA coefficients, were then summed and standardized on a 0-100 scale. The most significant relationships existed between the total QHEI score, QHEI substrate, QHEI riffle, Hydro-QHEI current, Hydro-QHEI depth, cumulative natural land cover, Sparrow total P, Sparrow total N, and mean annual air temperature.

Weighted Stressor Values (WSVs)

Weighted stressor values (WSVs) were produced for each stressor and fish species by plotting the HDI vs. the WSV, coded by the applicable ME IBI metric (e.g., fluvial specialist or other guild assignment) to illustrate the response of each species in relation to each stressor. Both the ME IBI and number of intolerant species showed clear threshold responses to selected stressors and both forms of the HDI (i.e., with and without the connectivity variables). The HDI, however, did not account for all of the variability in the ME IBI or intolerant species as there was substantial variation in the ME IBI at low HDI levels.

Maine IBI Metric Stressor Relationships

The environmental and stressor variables were examined against the ME IBI and metrics using Pearson coefficients of determination and by a visual examination of patterns across New England. The most strongly associated environmental variables with the ME IBI were latitude and mean annual air temperature. The NAHC thermal classification was also deemed significant ($r^2 = \geq 0.10$) for this analysis. Habitat variables were significantly associated with the ME IBI and included the QHEI score, good QHEI attributes, QHEI riffle score, and QHEI channel score. Of the land use variables, locally developed land cover yielded the only significant relationship. These variables represent both natural and human disturbance gradients, the latter of which generally increases from northeast to southwest across New England. Maps of each metric indexed to its equivalent contribution to the BCG Level depicted by the ME IBI were developed and examined for general and river-specific patterns. This allowed us to highlight any site-specific or river reach responses that escaped the regional scale analyses and which could spur more detailed follow-up investigations.

ME IBI metrics that showed the most significant Pearson coefficients were native species (2 variables), %native Cyprinidae (2), white and longnose sucker biomass (1), benthic insectivores (2), fluvial specialist and dependent species (3), macrohabitat generalists (4), stenothermic species (10), non-guarding lithophils (11), and non-indigenous species (4). The DIBI metrics had significant relationships as follows; American eel (4), diadromous abundance (4), Clupeidae abundance (1), and number of diadromous species (6). Three ME IBI metrics, %native salmonids, %blackbasses, and %DELT anomalies showed no significant results in the Pearson coefficient of determination analyses. However, each of these metrics had meaningful responses based on the mapping of the BCG equivalents and the interpretation of site and reach-specific results. This result illustrates the importance of looking beyond broad regional analyses.

Effect of Survey Design on Stressor Relationships

The differences between the probabilistic and targeted survey designs were also apparent in some of the stressor analyses and included:

- Extremes in stressor variables were the most evident in the targeted sites dataset and included elevated conductivity, elevated Sparrow results (TP and TN), impassable barriers, and low natural land cover.
- Targeted pollution survey sites contributed to revealing pollution gradients in various rivers that were otherwise not revealed by the probabilistic design.
- Targeted sites actually resulted in a more equitable spatial coverage of the available river reaches in the most northern latitudes and thus were influenced by their characteristics more so than that revealed by the probabilistic sites
- Local land cover revealed site-specific impacts better than cumulative land cover and related well to indicators that revealed river reach pollution gradients.

Stressor Analysis Synopsis

The predominant influences on New England riverine fish assemblages included both fluvial and structural aspects of habitat (which includes impoundments), the presence of dams, non-native species, and land use. This was demonstrated by the multivariate analyses against the array of regional stressors that were analyzed and by examining the distribution and values of the ME IBI and the IBI metrics. From a regional perspective based on NMS ordination it appears that dams, impoundments, and flow-related habitat alterations are the predominant influences on riverine fish assemblages in New England. However, not all of the indicators emulated similar responses to all stressors. For example, impassable dams act as a discrete impact thus the typical pollution tolerance, functional, or species richness metrics may not appear to be responsive. This was reflected in the response of the ME IBI and intolerant fish species to the HDI calculated with and without the connectivity variables. Fluvial dependent and specialist species metrics tended to be more strongly associated with specific habitat niches (e.g., QHEI riffle/run attributes) and higher scores for the Hydro-QHEI as illustrated by the WSVs being skewed towards the maximum scores in most instances.

It also needs to be understood that any exploratory analysis of stressors and their apparent responses as measured by biological indicators is only as meaningful as are the actual mechanisms that are at work. In this or any other analysis a stressor is the measure of the *presence* of an impact or alteration while the biological response is an indication of the *effect* of that stressor or aggregation of stressors. The mere presence of a stressor does not equate to an effect on the biota. Simply compiling an array of stressors and then subjecting them to correlative and multivariate analyses seldom explains the majority of the variation in biological responses. Such was the case in this study as most of the relationships explained less than one-third of the variation on a regional basis. This is a tacit admission that the analyses herein either did not capture all of the stressors that are important to riverine fish assemblages in New England nor all of their responses. Instead, analyses that are keyed on “reading” the biological responses first and then diagnosing the stressors based on details about the setting in which the responses were observed are also needed to ensure a more complete approach.

It is likely that the variation in some key fish assemblage indicators (e.g., %DELT anomalies) that were most evident at the local reach and site-specific scales could be better explained if important chemical stressor data that characterizes input sources and ambient conditions was more widely available and at a sufficiently resolute spatial scale. Coupled with the regional approach taken by this study it would provide the template for developing a more monitoring dependent and data driven stressor identification process for New England rivers. A targeted design *at least as intensive* as that employed in this study would be needed to capture effects which operate along the pollution continuums in each river. Such a non-random, but spatially adequate and equitable design can provide substantial benefits particularly where there is a need to detect, characterize, and resolve reach and site-specific issues. It also needs to include sufficient site density so as to assure that the full range of resource quality is captured as was demonstrated in this study with the Level 2 BCG ME IBI scores being uniquely revealed by the targeted sampling design on both a regional and river specific basis.

The results of the stressor analyses revealed a strong latitudinal gradient that corresponds to both natural gradients and anthropogenic stress which corresponds to a general decline in the ME IBI and metric values indexed to the BCG. Separating the influence of natural and anthropogenic gradients in these observations is challenging because they are spatially correlated. The ME IBI was used in the NELR REMAP project with the understanding that it may not apply equally well to all rivers throughout New England. However, other potentially applicable IBIs simply do not exist and those that are either available or under development for the Northeastern U.S. are restricted to wadeable streams. It is doubtful, however, that refining the ME IBI would produce substantially different conclusions about the most important stressors affecting the New England riverine fish fauna.

NELR REMAP and NRSA Comparisons

One of the major objectives of the NELR REMAP project was to compare the outputs of the methods employed in this study with those used in the 2008-9 NRSA. Because of the overlapping goals of the NRSA and this project it is important to understand their comparability in terms of data characteristics, logistics, and assessment outputs. Paired fish sampling was conducted as part of the NELR REMAP project at 64 NRSA sites during 2008-9. Because the NRSA base and oversample draw of sites were used for the probabilistic aspect of the NELR REMAP assessment, data was available for each method at all NRSA sites.

Aquatic Life Condition Assessment

The comparability of the NELR REMAP and NRSA non-wadeable fish assemblage data was assessed by comparing selected data outputs such as the ME IBI, relative numbers, commonly occurring fish species, and ME IBI metrics. Other types of data and comparisons are possible, but this initial comparison of the results yielded by each method was focused first on the primary assessment outcomes in terms of aquatic life goal attainment since that would be the primary concern for the New England states and EPA Region I for determining the utility of either method. As such the ME IBI was calculated for both NRSA and NELR REMAP data and the results compared using the same thresholds as reported in Chapter 3. To determine if this modification had an effect on the utility of the NRSA data for supporting aquatic life status assessments the ME IBI based on NELR REMAP data was calculated with and without the adult white and longnose sucker biomass metric. This allowed for a comparison of the NRSA based ME IBI, the NELR REMAP based ME IBI, and the same with the adult white and longnose sucker metric removed. There were mostly similarities, but some differences existed in the distribution of the ME IBIs among the 64 sites included in the comparison. The non-adjusted NELR REMAP and NRSA based IBIs produced the same median and 75th percentile values, but the former had lower 25th percentile and minimum values. The NELR REMAP based adjusted IBI that omits the adult white and longnose sucker biomass metric had lower median and 25th percentile values than the non-adjusted ME IBI, but it yielded a higher maximum value. Omitting the adult white and longnose sucker metric "inflates" the adjusted ME IBI when adult white and longnose suckers are either absent or in low abundance in a sample. This metric was one of the least correlated with the regional stressors that were examined in chapter 4, but it does show a strong positive relationship with the deep run habitats of riverine sites and it declines in impounded or otherwise modified habitats (see Chapter 4). While some of the differences between the NELR REMAP based non-adjusted ME IBI and NRSA based IBI were due to the inherent differences between each method, some are due to the removal of this metric as evidenced by the comparisons of removing this metric within the NELR REMAP method. The contemporary approaches to developing regional IBIs (Whittier et al. 2007) would likely have omitted this metric, but it was included in the ME IBI for reasons that relied more on life history attributes, an ability to discern important impacts that are not usually included in regional stressor analyses, and taxonomic completeness. These comparisons seem to bear out the need to retain this important fish assemblage attribute.

The NELR REMAP results were necessarily used as the "standard" for agreement or disagreement with the NRSA results. The NRSA agreed about attainment at 20 of 23 sites judged as attaining this threshold by NELR REMAP and it agreed about impairment at 31 of 41 sites judged as impaired by NELR REMAP. The agreement about BCG level was less consistent with the same level assigned for only 39 of 64 sites (61%). All of the disagreements were within one BCG level for all except two sites. The results were nearly identical for a comparison of the NELR REMAP based IBI adjusted to exclude the adult white and longnose sucker biomass based metric. A scatterplot of the NRSA based IBI and NELR REMAP IBI (non-adjusted) revealed a general directional agreement, but an r^2 of only 0.53, an indication of substantial quantitative departures between each method. In this case the comparison used the NELR REMAP based IBI without adjustment as the exclusion of biomass data by the NRSA is simply one of the fundamental differences between the two methods.

Maine IBI Metric Comparisons
Comparisons between each metric of the ME IBI, fish relative abundance, and the top five species by numbers were visually examined by paired box-and-whisker plots. The two ME IBI metrics where differences were noted include %native salmonid and %DELT. The NELR REMAP method showed higher raw values and a greater frequency of those values which is an indication that the NRSA sampling method is vulnerable to under-rating these attributes at selected sites. The NELR REMAP method showed higher raw values and a greater frequency of those values which is an indication that the NRSA sampling method is vulnerable to under-rating these attributes at selected sites. Given the importance of native salmonids to discriminating between the upper BCG levels and %DELT to discriminating among lower BCG levels, the inconsistent ability to reveal such differences when they exist would not translate to a more refined IBI along the scale of the BCG. The net effect is a NRSA methodology that is limited to pass/fail assessment and a reduced capacity to support refined aquatic life uses and biocriteria.

Synopsis of NELR REMAP-NRSA Comparisons
The analyses conducted herein suggested that there are important site-specific differences, but determining these would require additional analyses that were not conducted by this study. However, and with some notable exceptions, the NELR REMAP and NRSA produced comparable results at the assemblage assessment level on a regional basis. The differences that we observed would have less influence on a pass/fail or the "good-fair-poor" level of assessment of the NRSA as the detail along the BCG gradient is less important in such truncated assessment paradigms. It would seem feasible then to use the NRSA and NELR REMAP methods interchangeably at this level of regional assessment. It also exposes the limitations of this level of comparability because critical differences that would be important at a more detailed spatial scale or under a more refined and rigorous assessment scale where such differences would have mattered.

Another key difference is that the NRSA method by virtue of the site length could mask important site-specific differences that might occur within the NRSA sampling reach especially where site lengths approach the maximum of 4 km. An example of this is in the Connecticut

River downstream from the Turners Falls dam which is affected by flow diversions to the Cabot hydropower project. Two NRSA sites were located within the 3+ miles of the bypass reach and four NELR REMAP sites were co-located within this reach. The focus of the 2008-9 Connecticut River intensive survey was to assess possible local scale effects from stressors such as the flow diversion for the Cabot hydropower project. The results between the longer NRSA sites and the shorter fixed distance of the first two NELR REMAP sites that were "embedded" within each of the two NRSA sites reveal contrasting ME IBI and DIBI scores and metric values. Metric values were much lower at the NELR REMAP site RM 67.9 which had the lowest ME IBI score in the entire Connecticut River during 2008-9 and all of New England. The comparison of the bypass reach sites showed varying BCG levels (4 to 6) from NELR REMAP with the NRSA results "homogenizing" the overall condition as Level 5 (poor). The NELR REMAP method is better suited to detecting and characterizing local reach and longitudinal scale effects than the NRSA method. Simply put the variable and longer sampling site of the NRSA method can obscure important site-specific impacts by homogenizing these effects because of the longer site distance as was shown in the Turners Falls bypass reach in the Connecticut River. In our comparison the NELR REMAP method was better suited to detecting and characterizing local reach and longitudinal scale effects than the NRSA method. Simply put the variable and longer sampling site of the NRSA method can obscure important site-specific impacts by homogenizing these effects because of the longer site distance as was shown in the Turners Falls bypass reach in the Connecticut River.

An observation reported by the NRSA field crews was that when the NRSA site included less diverse shoreline habitats (i.e., shallower inside bends, bedrock shoals, etc.) that numbers of commonly occurring, more tolerant Cyprindae and white suckers would frequently be at their highest. Such less diverse and monotonous habitats could be included in an NRSA site because the selection of the right or left shoreline was purely random and did not consider the "best habitats" as did the NELR REMAP protocol. A paired plot of the five most commonly occurring fish species (fallfish, spottail shiner, common shiner, golden shiner, and white sucker) in the NELR REMAP survey revealed significantly higher numbers of these species for the NRSA compared to lower numbers for the NELR REMAP. The differences suggest that a substantial number of NRSA sites produced samples dominated by one or more of these species. These are the most commonly occurring species in terms of the breadth of their distribution across New England rivers and they are also habitat generalists frequently found to be the predominant species in low quality habitats. As such this reflects a potential and perhaps unpredictable source of variation in the NRSA protocol by potentially irruptive species.

Biocriteria Development for New England Large Rivers

An expected outcome of this project is the description of a process for biocriteria including the development of reference condition, multimetric indices, and a template for tiered aquatic life uses for the non-wadeable rivers of New England using the Biological Condition Gradient (BCG; Davies and Jackson 2006) as a conceptual foundation. While a numeric index and interim threshold for a pass-fail framework was accomplished by the NELR REMAP project, further exploration of key issues underlying the development of biocriteria and tiered aquatic life uses is needed.

Assessment of NELR Bioassessment Protocol

The NELR fish assemblage assessment protocol was evaluated using the process described by *Biological Assessment Program Review: Assessing Level of Technical Rigor to Support Water Quality Management* (U.S. EPA 2013). It is used here to assess developments to date and to determine what steps remain to achieve the highest level of rigor for New England Large Rivers based on an examination of the development of 13 critical technical elements. The development of the current NELR protocol has fully addressed the Index Period (Element 1), Spatial Sampling Design (Element 2), Taxa and Taxonomic Resolution (Element 6), Sample Collection (Element 7), and Sample Processing (Element 8) critical technical elements *for a single assemblage.* Data management (Element 9) has been partially addressed and would require a more institutionalized process (as opposed to a project basis) to attain full development. The Stressor Association (Element 12) was also partially addressed herein, but development would be complete only when the other design and interpretation elements are more fully developed. While the NELR REMAP and Maine Rivers projects partially addressed these remaining elements by consequence of the initial BCG and ME IBI development, they remain incomplete at this time.

The NELR REMAP protocol scores at 79.8% which is consistent with a Level 2 program. This means the protocol is suited for producing large scale trend assessments, which was accomplished via the NELR REMAP assessment. Elevating the level of rigor is an important step in the development of biological criteria and tiered aquatic life uses. Attaining a Level 4 program status and having full TALU program support and implementation are *mutually inclusive.* While this ultimately includes the realities of program costs and staffing needs, the evaluation of technical capabilities and identification of technical gaps are essential first steps in this process. The steps outlined in Appendix Table D-2 would need to be completed to attain a Level 4 program. Additional exploration and development within each element is needed and if done in the appropriate sequence would be complimentary in terms of elevating the technical rigor and detail of each element. A project focused on each and guided by BCG principles would deliver a more robust and rigorous set of assessment tools and biocriteria.

Template for Biocriteria Development

As the ME IBI was applied throughout New England it became apparent that some rivers may be outside the cold water paradigm of the index, thus a better thermal classification scheme needs to be developed and tested. Such is at least conceptually available from the Northeast Aquatic Habitat Classification (NEAHCS; Olivero and Anderson 2008) that includes stream and river size, gradient, and temperature regime. One issue to resolve is the existing assessment of many sites in southern New England as being impaired when using the interim ME IBI. Without an accompanying relevant classification scheme, it is difficult to determine if the current fish assemblages are indeed impaired by the accumulation of human-induced disturbances that have substantially altered flow and thermal regimes. This begs the question about the “as naturally occurs” true natural potential of these rivers. If a cold water or transitional cold system is the “as naturally occurs” then dealing with degraded rivers becomes a different matter than if they are simply misclassified and the resulting assessment is inaccurate due to the application of the wrong baseline. The mapping of the NEAHCS classification at the NELR REMAP sites (see Ch. 3; Figure 22) shows that transitional cold water assemblages (which seem consistent with the interim ME IBI) occur as far south as mid-Connecticut and Rhode Island. There were many more sites that were classified as transitional warmwater, but this observation raises the question of whether this is a natural phenomenon or an artifact of two centuries of human-induced changes to flow and thermal regimes. All of these observations and questions point to the need to better develop a classification scheme with IBI development specific to those classes to follow.

NELR Fish Distribution Atlas

The distribution of fish species collected at the 2002-7 Maine Rivers, 2008-9 REMAP and 2008-9 NRSA sampling sites are depicted in Appendix A along with a brief narrative synopsis of each. Distribution maps showing relative numbers of a species collected at each of 502 locations were developed for the more commonly occurring species and provide a visualization and general impression of spatial occurrence throughout New England large rivers. A brief summary of the fish species according to their native and non-native status is provided by each of the six New England states. The riverine fish assemblage of New England Proper included 60 species considered as native, three (3) as introduced of intercontinental origin, and 22 as introduced of intracontinental origin (Table A-1). Eight (8) native species are also listed as introduced of interstate/intrastate origin as they have been introduced to areas outside of their original ranges within New England Proper. Of the 78 species recorded in New England Proper, more than one-fourth are introduced. Maine had the highest native species count (42) which partly owes to the cold water assemblage species that occurred in northern and western Maine. It also had the highest number of introduced species of interstate/intrastate origin, a reflection of bringing those species from southern New England to Maine. Connecticut and New Hampshire had the highest number of introduced species of intracontinental origin (14), three species more than the other states. About one-third of the present-day fish species are introduced.

NELR REMAP PROJECT RECOMMENDATIONS

1. Regional BCG Development – A formal process for developing a BCG applicable to New England rivers similar to efforts in other regions of the U.S. is recommended. The current BCG, upon which the ME IBI is based, while conceptually valid, does not span all classification strata. This would lead to a more refined and accurate classification scheme by taking into account the biogeographic differences in species distributions and natural differences in expected thermal regimes along the north to south plane. A "synthetic" model could be used to "predict" the "as naturally occurs" fish assemblages throughout New England based on simulations of historical conditions. Such an approach could also be used to better understand and cope with the influence and intractability exerted by the extensive introductions of non-native fish species in New England.
2. Connecticut River – The Connecticut River merits more focused research including attributes of the BCG and perhaps an IBI specific to the mainstem. Similar focuses have taken place elsewhere in the U.S. (e.g., Mississippi R., Missouri R., Ohio R.).
3. IBI Refinement – The BCG accomplished in #1 would be useful in further examining the observed north to south pattern in the ME IBI that coincides with the confounding influence of human disturbance which increases in magnitude along the same north to south plane. This would lend to the better development of biological criteria and applying them in tiered aquatic life uses and within a framework of determining the attainability of TALU tiers in individual rivers.
4. Diadromous IBI – Incorporate the further development and refinement of diadromous metrics as a supplemental or as part of a revised regional IBI.
5. NRSA Comparability of Regional Assessment – The comparability analyses in this study were limited by a lack of access to the NRSA multimetric index for the fish assemblage. That index is calibrated using a random forest model (Cutler et al. 2007) which is a novel approach to dealing with reference condition. It requires a large number of input variables at every sampling site in a study area. This would provide a more comprehensive comparison of the NRSA and NELR assessment and their respective outputs.
6. NRSA Methods Comparability – The comparability analyses accomplished in this study were across the sample of all NRSA sites in the NELR REMAP study area which yielded composite comparisons. Additional analyses of site by site differences that are attributable to differences in the respective methodologies need to be undertaken. These would contribute to a better understanding of the characteristics and biases of each method.
7. Revised REMAP Probabilistic Design – The array of probabilistic sites offered by the 2008-9 NRSA draw of sites compared to the coverage of the targeted sites revealed an inequitable coverage of sites in northern Maine. This seems to be the result of the NRSA using a much larger frame for allocating sites (i.e., the Northeastern Highlands) than the six New England states. It also obscured the current state of fish assemblages in that northern Maine has yet to be as affected by non-native introductions as the remainder of New England which significantly affects the understanding of what the baseline condition actually is. If a New England assessment is undertaken in the future the survey design should be modified to ensure that the resulting database is more equitable in terms of geographic coverage.

Development of Methods and Designs for the Assessment of the Fish Assemblages of Non-Wadeable Rivers in New England

Chris O. Yoder, Principal Investigator
Edward T. Rankin, Senior Research Associate
Lon E. Hersha, Research Associate
Midwest Biodiversity Institute
P.O. Box 21561
Columbus, OH 43221-0561

CHAPTER 1: INTRODUCTION

MBI responded to a request for proposals to conduct a Regional Environmental Monitoring and Assessment Program (REMAP) grant in Region I in 2007. Assistance agreement RM-83379101 was awarded to MBI in May 2008 and following the development of a detailed work plan and project quality assurance project plan (QAPP; MBI 2008) field sampling was initiated in July 2008. Field sampling took place again in 2009, and data management, analysis, and synthesis occurred in 2010-13. The analyses were supplemented with methods and data from related projects conducted on non-wadeable rivers in Maine during 2002-9 and the Connecticut River in 2008-9. The Maine project is where the methods applied in the 2008-9 New England Large Rivers (NELR) REMAP project were originally developed and refined. A probabilistic site draw from the 2008-9 National Rivers and Streams Assessment (NRSA) also provided the basis for conducting comparisons of both the fish assemblage condition assessment and sampling methods between the Maine Rivers methods that were used for the NELR REMAP project and the 2008-9 NRSA at co-occurring sites.

Scope and Purpose

The purpose of this project is to address important biological assessment methods and design issues with the non-wadeable rivers in New England that are embodied in the principal objectives of the original grant proposal. This was accomplished by producing an assessment of the fish assemblages of New England for non-wadeable or "large" rivers, a portion of which have not been included in past U.S. EPA research or by current New England state monitoring programs. Specifically these include flowing waters that are not effectively sampled by wading methods and which are labeled as being "non-wadeable". Adding operational clarification and consistency to this term is an important underlying project objective.

The principal goals of this project include:

1. Describe a standardized protocol for sampling riverine fish assemblages and habitat in the non-wadeable rivers of New England for Clean Water Act (CWA) assessment and other management support purposes;

2. Generate estimates of fish assemblage condition and stressor effects in non-wadeable rivers throughout New England;
3. Compare assessment outcomes produced by different spatial sampling designs in selected mainstem rivers;
4. Determine the transitional characteristics between wadeable and non-wadeable sites in New England;
5. Compare the outputs of two different fish sampling methodologies; and,
6. Describe a process for biocriteria development including the development of reference condition, multimetric indices, and a template for tiered aquatic life uses in the non-wadeable rivers of New England using the Biological Condition Gradient (BCG; Davies and Jackson 2006) as a conceptual foundation.

This project focused primarily on fish assemblages and physical habitat because:

- These were the most commonly assessed among the various non-wadeable river research projects that are described herein;
- Fish assemblages are responsive to the physical (flow, habitat, and temperature; Bain and Meixler 2008; Armstrong et al. 2011) and biological stressors (non-native species; Halliwell 2005; Yoder et al. 2006a, 2008) that are common to New England rivers.
- A working protocol was already established (Yoder et al. 2006b); and,
- The fish assemblage and qualitative habitat results in Maine (2002-7) were sufficient to demonstrate the merits of different sampling designs, conducting condition assessments, and meeting the objectives of the project work plan.

Macroinvertebrate and algal assemblages would have been logical choices for second and third biological assemblages, but the resources allocated to this project were insufficient for assessing more than one assemblage. One New England state, Maine, has working macro-invertebrate and algal protocols for non-wadeable rivers that include numeric biocriteria and tiered aquatic life uses (Davies and Tsomides 2002; Danielson 2006). Ultimately, state programs are expected to employ at least two assemblages in all lotic strata (U.S. EPA 2013; Davies and Yoder 2010).

Practical Issues with Non-Wadeable River Bioassessment

The biological assessment of non-wadeable rivers has been an emerging issue of importance for U.S. EPA and state biological monitoring and assessment programs since the mid-1990s and as such has been the subject of several research projects. Non-wadeable rivers were included as a distinct stratum in the National Rivers and Streams Assessment (NRSA) in 2008-9. The term non-wadeable was chosen to distinguish it from the more commonly employed bioassessment methods for wadeable streams in which sampling can be conducted while wading. Ohio EPA (1987, 1989a) defined a boat site type and Flotemersch et al. (2010) used the term boatable to describe fish sampling methods for large rivers. Simply put non-wadeable means that a floating platform is required to effectively collect an adequate sample of the fish assemblage. The term *non-wadeable* will be used for the purposes of this study recognizing that other terms may also be used to describe the same approach.

Non-wadeable rivers have not been as routinely assessed by state monitoring programs that have instead focused on smaller, wadeable streams. Likewise, early U.S. EPA bioassessment guidance (Barbour et al. 1997) focused primarily on wadeable streams. However, a few state monitoring programs have included non-wadeable rivers in their programs since the 1980s and these provide examples of sampling and assessment methodologies. U.S. EPA research on non-wadeable rivers has emphasized "great" and selected large rivers. Great rivers include the main channels of the Mississippi, Missouri, and Ohio Rivers while large rivers included the largest mainstem tributaries to the Ohio and upper Mississippi such as the Scioto, Licking, Kentucky, Wabash, Illinois, and Wisconsin Rivers (Flotemersch et al. 2001; Flotemersch and Blocksom 2004; Blocksom and Flotemersch 2004; Emery et al. 2007). Our concern herein is that thousands of miles of non-wadeable rivers and streams exist outside of the sphere of these recent research projects. In the Midwestern U.S. sites draining 150-500 $mi.^2$ and certainly >500 $mi.^2$ usually require the use of non-wadeable sampling techniques, particularly for the fish assemblage (Yoder and Smith 1999; Yoder et al. 2005). Our experiences in Maine during 2002-7 suggests that this is a relevant concern in New England (Yoder et al. 2006 a,b). Another concern is with using wadeable gear types to sample fish in non-wadeable rivers, which exposes the limited effectiveness of these gear types for assessing non-wadeable river fish assemblages as a whole. Thus an important objective of this study was to determine the "boundary" between the appropriate application of wadeable and non-wadeable fish sampling gear types as a guideline for conducting future non-wadeable river assessments.

Figure 1 illustrates the continuum of all possible stream and river size ranges and the concurrence of ongoing and in-progress research and monitoring projects that are addressing specific strata within the non-wadeable domain in selected parts of the U.S. and New England. Some of these strata require the use of specific sampling protocols which also necessitates a stratification of sampling approaches, bioassessment tools, biocriteria, logistics, and equipment. The characteristics that describe the transition from wadeable streams to non-wadeable rivers need to be defined such that future practitioners can make the most appropriate choices in terms of sampling gear and logistics.

This project was purposely coupled with other fish assessment projects in New England in order to determine the spatial scale at which non-wadeable river assessments (of status) and subsequent management applications (WQS, CWA management program support) are feasible. For non-wadeable rivers, a protocol that assesses condition at a spatial scale that is not relevant for management purposes (and visa-versa) is of limited utility and potentially cost-ineffective. Therefore, an objective of this study was to determine the spatial scale at which we should be:

- Developing standards (i.e., the broad definition of "standard" includes refining aquatic life uses and determining/setting/adopting criteria to protect those uses);
- Monitoring to assess attainment of standards in support of management goals;

- Developing and evaluating restoration efforts (e.g., TMDLs, dam modification/removal, fish passage) for impaired waters; and,
- Managing non-wadeable rivers to maximize desirable environmental outcomes.

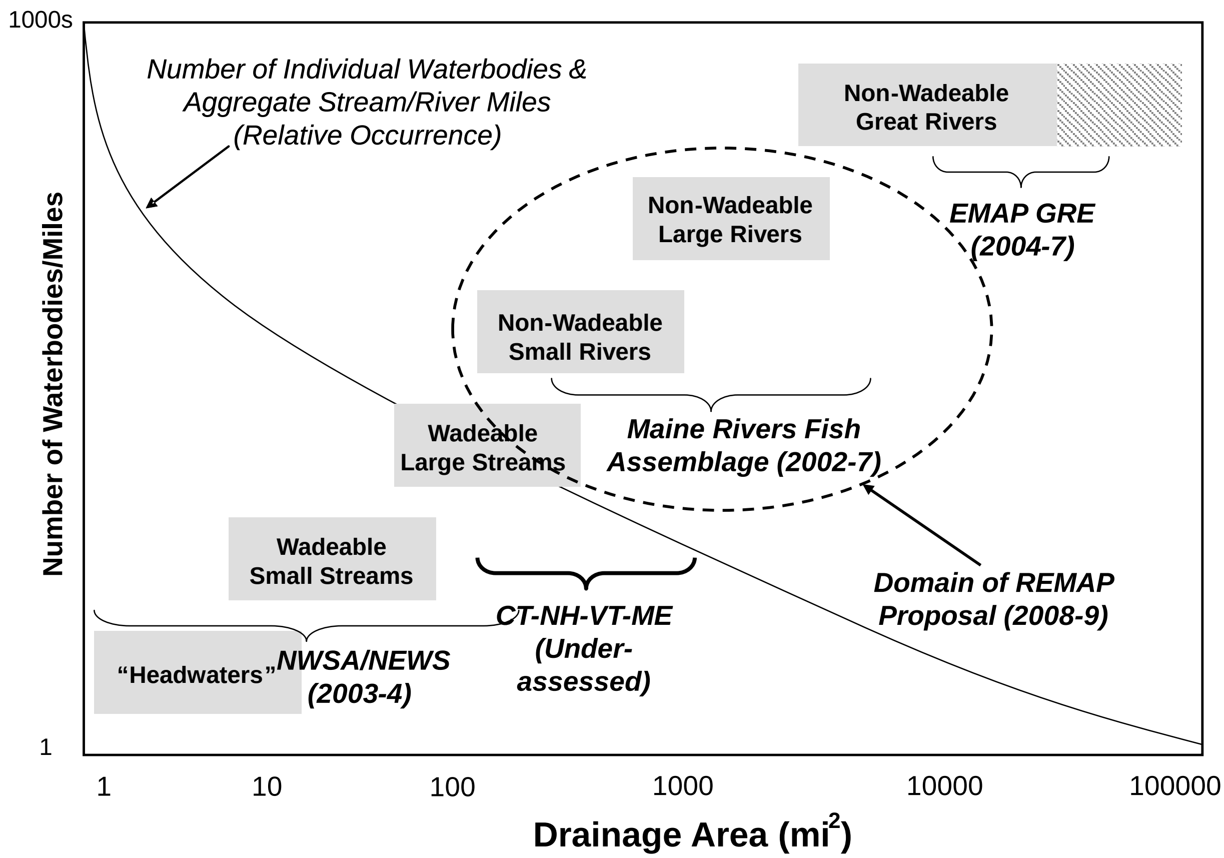


***Figure 1**. A continuum of stream and river strata that pertain to the development of sampling methods, biological assessment tools, and biological criteria for fish assemblages in New England. The number of individual waterbodies and stream and river miles is inverse to the size of the stream or river stratum (NWSA – National Wadeable Streams Survey; NEWS – New England Wadeable Streams Assessment).*

Implications for Monitoring and Assessment Programs

Only a few examples of *fully developed*[1] state-based approaches exist for non-wadeable rivers and streams in the U.S. (Ohio EPA 1987, 1989a; Yoder and Smith 1999; Lyons et al. 2001; Yoder and Kulik 2003; Yoder et al. 2005; Davies et al. 2002; Danielson 2006). Some states do not yet effectively nor routinely assess their non-wadeable rivers and streams for any biological

[1] Fully developed includes a numeric index or model that is derived using the best available methods and calibrated within a reference condition approach and consistent with the Critical Technical Elements process (U.S. EPA 2013). The assessment methodology is suitable for CWA 305[b] reporting and 303[d] listing that includes assigning causes to impairments at the river-reach scale and is capable of supporting the development and implementation of refined aquatic life uses.

assemblages. A report on the status of the U.S. EPA, Region V state bioassessment programs showed that of the six states evaluated only one had a *fully developed* biological assessment program for non-wadeable rivers and streams (MBI 2004). A similar evaluation of the Region I states likewise showed a single state with a similarly developed program (Davies and Yoder 2010). In this context a fully developed state program would include technically sound data collection methods, at least two biological assemblages, and fully calibrated indices and biological criteria in the state WQS. In New England some states may *monitor* selected rivers for one assemblage, but only Maine has developed a systematic procedure that spans wadeable and non-wadeable rivers and streams and also includes calibrated biological criteria and tiered aquatic life uses for two assemblages (macroinvertebrates and algae). We also believe that it is quite likely that wadeable fish sampling methods are at times (perhaps frequently?) extended beyond their inherent capability to sample non-wadeable river fish assemblages and this is due in large part to the lack of a comprehensive hierarchy of wadeable and non-wadeable methods and protocols. Methods that result in the under-sampling and a subsequent under-representation of a biological assemblage contribute unpredictable variability and bias in the data that makes further uses potentially unreliable. This project represents an examination of not only a critical issue in biological criteria development, but it is a unique examination of these issues on a regional scale. An example of such a hierarchy of methods has been developed for fish assemblages for large rivers in Maine (Yoder et al. 2006a) and we believe it has applicability throughout New England. This project provided an opportunity to assess that applicability and close a significant gap that currently exists among state monitoring and U.S. EPA research programs.

Status of Non-Wadeable River Bioassessment

U.S. EPA recognizes the need to better develop methods and criteria for assessing these highly visible and economically important resources. Examples of a growing emphasis on non-wadeable rivers includes the EMAP great rivers assessment (EMAP-GRE), U.S. EPA research on non-wadeable bioassessment methods (Flotemersch et al. 2001; Flotemersch and Blocksom 2004; Blocksom and Flotemersch 2004; Flotemersch et al. 2010), selected REMAP projects such as the recently completed assessment of the largest tributaries to the Ohio and upper Mississippi Rivers (Emery et al. 2007), state programs (e.g., Ohio, Maine, Wisconsin), and the 2008-9 NRSA and upcoming 2013-14 NRSA. Two other research projects within Region I that are relevant to this study include the recently completed New England Wadeable Streams (NEWS; Snook et al. 2007) assessment and the Maine rivers fish assemblage assessment and IBI development (Yoder et al. 2006 a,b, 2008).

The experience with the 2004-5 National Wadeable Streams Assessment (NWSA) is also of relevance to this project even though it was focused on wadeable streams in U.S. EPA, Region V. Our experience as a direct participant in the NWSA especially highlighted the issue concerning the transition from wadeable to non-wadeable methods (Miltner and Rankin 2009). This was readily apparent in the execution of the 2004 NWSA fish assemblage sampling in Region V where nearly one-fourth of the base sites were rejected due to non-wadeability. This resulted in a significant amount of effort being expended on non-productive field days as this determination was made on-site[2]. Better defining and resolving this issue prior to sending crews into the field is one of the principal needs prior to undertaking projects of this size.

It was the intent of this project to address the full range of non-wadeable streams and rivers that commonly occur in New England. Taken together, this study and the related research and development efforts have produced the methodologies and designs that are needed to accurately assess flowing water body types in a cost-effective manner across the continuum of all flowing water body types (e.g., the smallest wadeable streams to the largest non-wadeable rivers; Figure 1). The lessons learned by this project and collectively through related research efforts and experiences are transferable to U.S. EPA, states, tribes, and other monitoring entities in New England.

Development of NELR Fish Assemblage Assessment Procedures

An important goal of this project is to provide the underlying work to support a process for developing a cost-effective approach to assessing the non-wadeable rivers of New England. The first and most important aspect of this objective is a methodology for collecting fish assemblage samples in a manner that renders the data useful for assessing status and relative condition and for a variety of CWA and natural resource management purposes. Since a working multimetric index (i.e., an Index of Biotic Integrity for fish) was already available, we also emphasized this aspect of developing a fish assemblage assessment protocol for New England rivers. Karr et al. (1986) originally envisioned that an "IBI" type of approach would be applied to widely divergent aquatic habitat types and they provided the metric substitution concepts to accommodate the ecological realities presented by different places while preserving the consistency of the approach in an ecological sense. The guidance provided by Halliwell et al. (1999) indicates that IBI development should take into account the aquatic habitat type and resident fish assemblage characteristics which includes consideration of the relatively low native and endemic fish species richness, species origins, the status of introduced species, and the range of thermal tolerances and preferences that is inherent to New England rivers and streams. An initial delineation of taxonomic, functional, ecological role, and tolerance guilds for IBI development was accomplished for New England fish species by Halliwell et al. (1999). The Maine Rivers project extended and refined that approach for large rivers (Yoder et al. 2008). What remains is the generation and development of a New England-wide database, part of which was delivered by this NELR REMAP project.

Application to Depauperate Fish Assemblages

While many of the pioneering studies on large river fish assemblages first focused on the relatively species rich, warmwater faunas of the Midwestern U.S., later studies have focused on comparatively species depauperate cool and cold water systems. Such is the case with New England rivers that have been historically isolated from the more species rich drainages that lie south, west, and northwest of New England (Curry 2007). As such, this is a major issue for data

[2] All rejected sites were first visited by a field crew - these sites were accessible, but were rejected due to non-wadeability. The NWSA did not include a non-wadeable component.

analysis and developing an assemblage based assessment. Hughes and Gammon (1987) applied the fish assemblage assessment procedures that were originally developed in the Midwest to the Willamette River in western Oregon. They not only found that a single-gear sampling approach (pulsed D.C. boat electrofishing) could work in a relatively cool to cold water river, but that an IBI type of evaluation could be developed in a comparatively “species poor” system. Frequently, we encounter the notion that depauperate faunas are less than suitable for IBI development, thus protocols involving fish are deemphasized or dismissed altogether in some cases. We believe that the experience of Hughes and Gammon (1987) showed otherwise and that all biological systems have a fundamental organization that is revealed in the structural and functional characteristics and attributes that can be captured by an IBI-type of index regardless of the species richness properties. Key to the success of the Willamette River IBI was the inclusion of the negative influence of non-indigenous species, many of which also function as undesirable generalists and some of which are also members of the tolerant guilds. It is also important to recognize the fundamentals of how cold and cool water systems respond to degradation, particularly in the taxa richness metrics. Unlike warmwater streams and rivers, in which species richness declines in a linear fashion with increasing stress, cold water systems frequently exhibit an overall *increase* in species richness as a response, the result of the addition of non-indigenous mesothermic and eurythermic species that opportunistically invade as conditions become favorable to their life history requirements (Lyons et al. 1996; Mundahl and Simon 1999). More recently, the sampling of fish assemblages of large cold water rivers throughout the western U.S. that was accomplished as part of the U.S. EPA Western Environmental Monitoring and Assessment Program (WEMAP) further demonstrated the utility of this approach including the development of a regional fish IBI for the Pacific northwest (Mebane et al. 2003; Robert Hughes, personal communication).

The recent completion of a virtual statewide coverage of the non-wadeable rivers of Maine first demonstrated the applicability and “doability” of a standardized, single-gear fish assemblage sampling protocol (Yoder et al. 2006 a,b). This project addressed both the methodological issues and also the important data analysis issues including the refinement of structural and functional guilds (Yoder et al. 2008). Taken together this study addressed the needs for a standardized sampling protocol and the development of a project specific QAPP (MBI 2008) for application to other New England rivers via this NELR REMAP project.

Biological Criteria Development

Another important objective of this study is to further contribute to the development and use of biological criteria on a national and regional (New England) basis, specifically in large, non-wadeable rivers. This study fulfills an important prerequisite to the development of biological criteria by testing a standardized collection and analysis of fish assemblage data, including the use of an index of biotic integrity specific to New England rivers. Biological criteria are comprised of numeric expressions that describe the relative biological condition of an aquatic assemblage inhabiting the waters of a given designated use (U.S. EPA 1990, 1995). Benchmarks for designated uses are developed with respect to reference condition, which is derived from fish assemblage data collected at regional reference sites. While the restoration of most U.S. waters to a pristine state is not presently feasible, it is reasonable to base contemporary restoration goals on regional reference conditions that describe the “least and minimally impacted” biological condition and performance (Davis and Simon 1995; Stoddard et al. 2005). Principles for the successful development of numeric biological criteria include developing a reference condition, a regional framework, a characterization of the aquatic assemblage(s), and a habitat evaluation for specifically defined aquatic ecotypes (e.g., large rivers, wadeable streams, headwater streams, wetlands, lakes, etc.).

U.S. EPA (2005) developed a concept termed the Biological Condition Gradient, which is intended to foster the consistent development of biological assessment frameworks and biological criteria development across the U.S. This concept is also intended to enhance communication, understanding, and visualization of biological condition relative to the absolute range of possible biological quality as a gradient ranging between “as naturally occurs” (i.e., pristine) to extremely degraded (Davies and Jackson 2006; Figure 2). A challenge for developing biological criteria for large, non-wadeable rivers is an apparent dearth (or absence) of reference analogs, at least compared to that which is more widely available for wadeable streams. As an alternative, using direct sampling data combined with historical knowledge and a

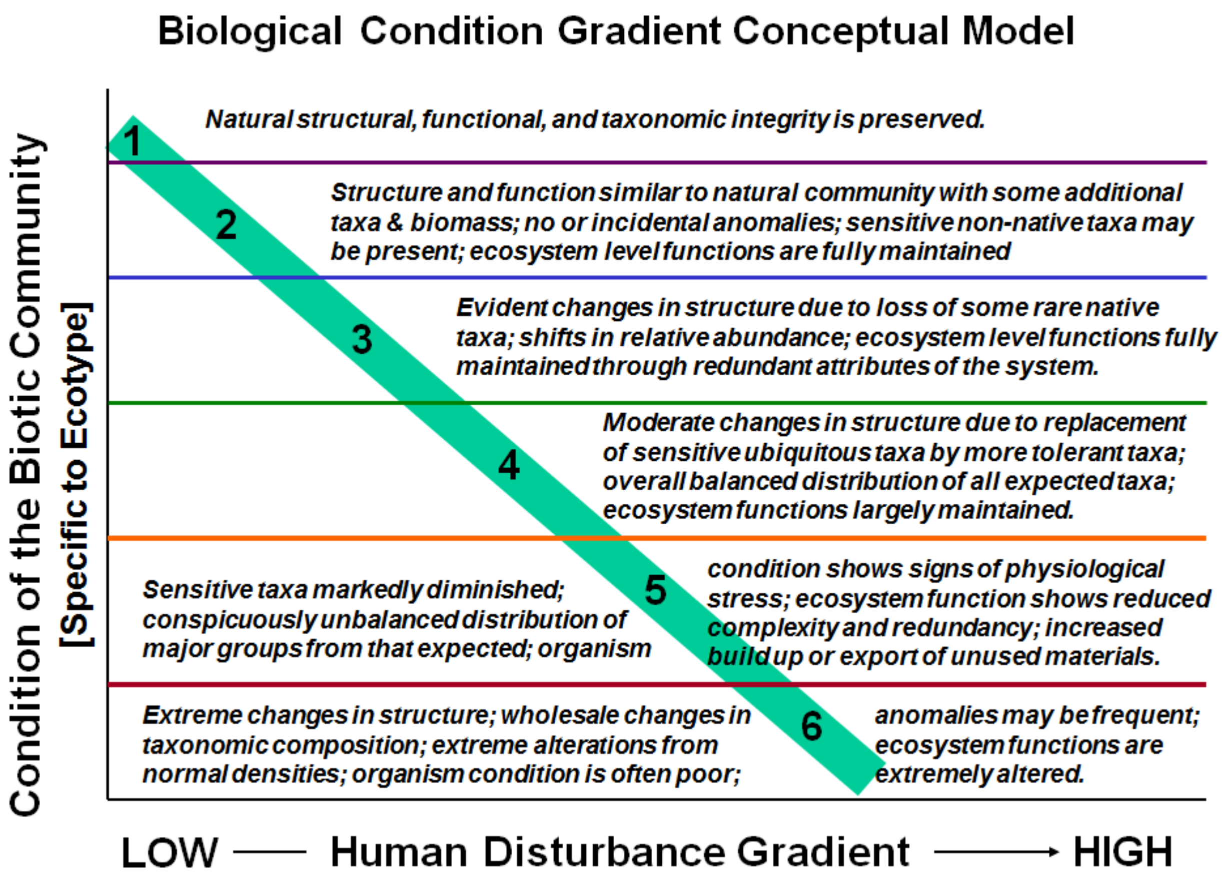


Figure 2. *Tiered aquatic life use conceptual model showing a Biological Condition Gradient (BCG) and descriptive BCG attributes of levels along a gradient of quality and disturbance (modified from U.S. EPA 2011).*

reconstruction of historical assemblages by expert analysis could be used as a partial substitute for directly measured reference condition (Emery et al. 2003; Armitage and Rankin 2009; Rankin and Yoder 2011a). While it is not the purpose of this study to develop a completely formal BCG for New England riverine fish assemblages, it provides much of the essential underlying knowledge that is needed to assign species to the attributes of the BCG and setting up a process to develop and test a regionally relevant suite of multimetric indices. Such a process has already been accomplished for Maine rivers (Yoder et al. 2008) and it employed an initial approximation of a BCG to support the development an IBI (Figure 3). That index likely has application to New England rivers outside of Maine, but its geographic limitations may become apparent in some southern New England rivers, thus undertaking a more formal BCG process is recommended. This would better support a biological criteria development process that New England states could then apply in making biological assessments using fish assemblages in their non-wadeable rivers.

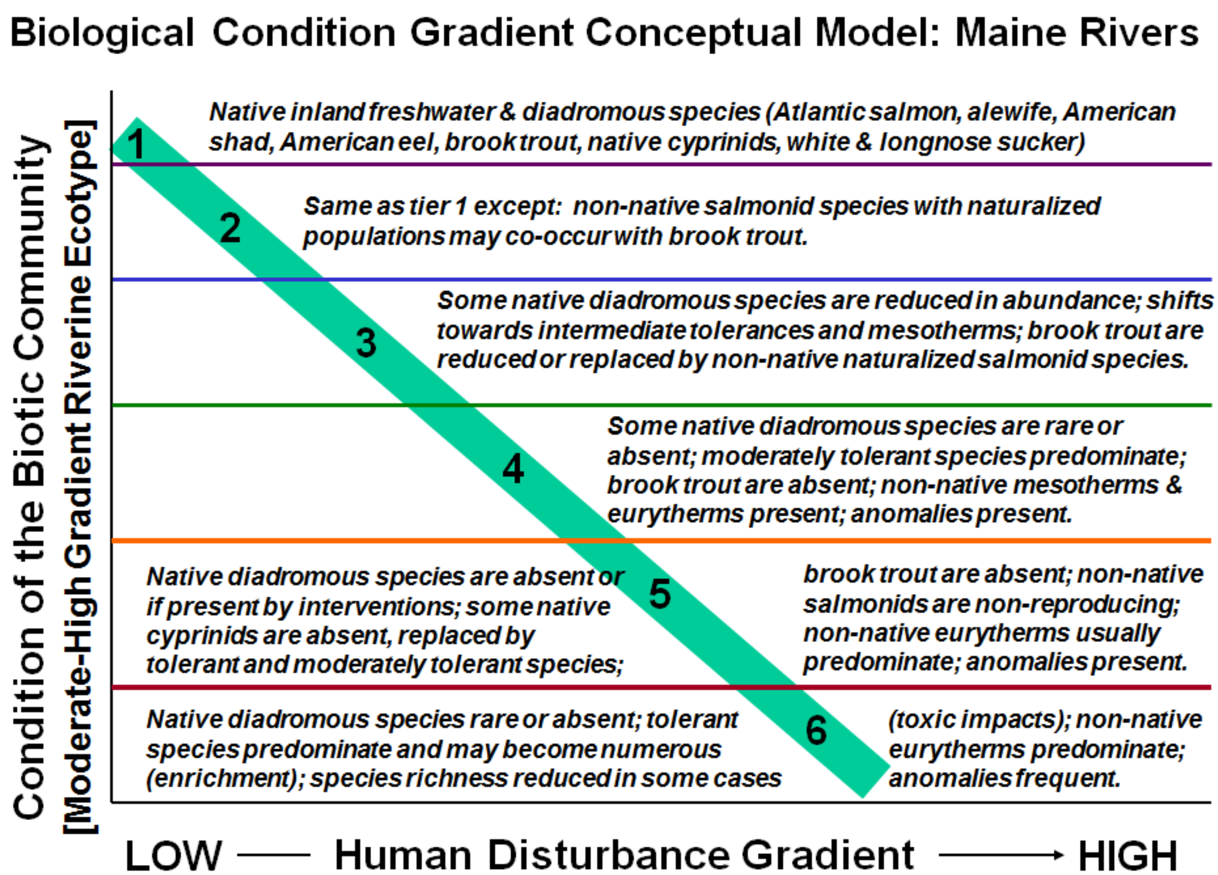


Figure 3. *A Biological Condition Gradient (BCG) model for fish assemblages representative of cool-cold water, moderate-high gradient riverine habitats in Maine (after Yoder et al. 2008).*

Assessment of Riverine Fish Assemblages in New England

As a Regional EMAP project, a primary objective of this study was to produce a statistically valid estimate of the condition of the fish assemblages of New England. We followed what is known as a probabilistic survey design (Olsen et al. 1999) and also following important aspects of the indicators approach developed by the U.S. EPA EMAP program (Hughes et al. 2000). As such this NELR REMAP study includes randomly selected sites from the "population" of non-wadeable rivers throughout New England. In addition we also included targeted sites from intensive surveys of selected rivers that were part of the Maine Rivers project (2002-2007), the Connecticut River survey (2008-9), and selected riverine sites that were sampled as part of this NELR REMAP project in 2008-9. Thus we were able to compare results between probabilistic and targeted sites, particularly as it relates to any differences in the assessment of assemblage condition and the revelation of associated stressors. As such, this study represents the first comprehensive and unified assessment of riverine fish assemblages in New England.

Factors Affecting Riverine Fish Assemblages in New England

A related objective is to determine the key factors that affect riverine fish assemblages in New England. Prior studies on New England wadeable streams showed that the major stressors include urban land use, flow alterations, and thermal modifications (Armstrong et al. 2011; Detenbeck et al. 2010). Hence, while our focus for this aspect of the study includes these categories of stressors, we also focused on other issues such as chemical pollution and the potentially adverse effects of introduced species. Yoder et al. (2008) have already demonstrated the impact of smallmouth bass *(Micropterus dolomieu)* on native fish species in Maine rivers, which corresponds to what other studies have revealed about the adverse impacts of introduced blackbasses (Jackson 2002; Vander Zanden et al. 2004; Whittier et al. 1997; M. Gallagher, Maine IF&W, personal communication).

A key project objective is to demonstrate the response of fish assemblages to stressors that are commonplace in New England. This not only includes developing stress/response relationships, but accessing and using the available stressor databases as an assessment of their relevance and completeness. It also includes an assessment of how each survey design provides for a stressor gradient such that important relationships are either not missed or otherwise masked. Underlying this analysis is the BCG framework and how it relates to setting attainable goals for assemblage quality.

CHAPTER 2: METHODS

Methods employed in this NELR REMAP study were the same as that developed by the Maine rivers project and as described and used by Yoder et al. (2006a,b; 2008). The methodology consists of a single-gear fish sampling method (pulsed D.C. electrofishing) and a qualitative habitat assessment method that were applied at each sampling site. A description of each, including the rationale and development, is described as follows.

NELR REMAP Fish Sampling Methodology

Description and Rationale

A cost-effective, tractable, and reliable sampling method is an essential need for any biological assessment program. The selection of a methodology is a fundamental decision or "cornerstone" in using fish assemblages as an environmental monitoring and assessment tool. While a variety of possible methods and techniques are available, the choice of which one(s) to use is influenced by the objectives of the monitoring program and the conditions that exist in the particular study focus or region (Flotemersch et al. 2010). Another objective of this study was to test and evaluate the Maine rivers and the NRSA fish sampling protocols. Our concept of a cost-effective, tractable, and reliable fish sampling method is one that produces relative abundance data and a sufficiently representative cross-section of the fish assemblage at a site with a "reasonable" effort (i.e., 2-3 hours/site). As such this type of assessment is distinguished from the more intensive efforts that employ longer sampling sites and/or multiple types of sampling gear in an attempt to produce estimates of population(s), standing crop, and/or a virtual inventory of all species present. The methodology used herein was first developed and tested by Yoder et al. (2006a) in Maine during 2001-3. It consists of a single-gear sampling approach that meets the following objectives outlined by Karr et al. (1986):

- Monitor biotic integrity at specific sites, within river reaches, and between different sites, reaches, and rivers.
- Sample and screen large numbers of sites in order to identify those that require attention.
- Assess changes in key fish assemblage parameters and attributes over space and time.
- Interpret large amounts of data from complex fish assemblages where the objective is to assess biotic integrity.

Meeting these objectives requires a methodology that can be used to sample multiple sites during a day, tens of sites within a week, and hundreds of sites over a summer-early fall seasonal index period (e.g., mid-June to early October). The sampling equipment and platform need to be transportable enough to gain access at multiple points along the length of mainstem rivers including comparatively remote reaches. Similar single-gear electrofishing approaches have been successfully tested and used in other parts of the U.S. and Canada to fulfill similar objectives (Yoder and Kulik 2003). Thus it is a primary goal of this project to further test and evaluate the potential for wider application to all New England large rivers. As such the QAPP

(MBI 2008) describes all field, laboratory, and data management procedures in detail along with the rationale and methodological issues associated with the boat/raft electrofishing protocols on which this study is based. The following therefore is a synopsis of the methods we are recommending for routine application to rivers in New England.

An Electrofishing Protocol for Non-wadeable Rivers in New England

This section describes the development process and the essential characteristics of a standardized boat electrofishing protocol for New England non-wadeable rivers. Based on our prior experiences in Maine and other regions where similar approaches are used, this protocol should not only be sufficient for CWA bioassessment purposes, but also for other natural resource management purposes. The approach, equipment, and techniques detailed here are those that were first tested and applied in 2001-2003 in Maine (Yoder et al. 2006a). While this does not preclude nor rule out the use of other designs or equipment to provide supplemental data, the primary goal is to produce comparable results in terms of electrofishing catches (i.e., numbers, biomass, composition, and condition) for the New England states. Thus the equipment choices, specifications, and execution of the sampling protocol has taken meeting these objectives into account.

Crew Composition and Assignments

An electrofishing crew consists of three persons - two netters and a driver - for the boat protocol and two persons - a single netter and driver - for the raft protocol. The primary responsibility of a netter is to capture all fish sighted; the responsibility of a driver is to maneuver the boat or raft so as to provide the netter(s) the best opportunities to capture and land stunned fish (the driver may assist in netting stunned fish that appear near the stern of the boat or raft). The driver also operates the electrofishing unit. Each task requires levels of skill and training, but boat/raft maneuvering requires the most experience to gain adequate proficiency and ensure safe and effective operation. This latter skill is particularly important in the faster flowing sections of riverine sampling sites. In actual practice, the boat/raft driver also functions as the crew leader who should be a skilled professional capable of carrying out and supervising all data collection activities that include fish identification and the accompanying habitat assessment. The netters are usually seasonal technicians with the physical ability to perform all crew member tasks. The netters are clad in chest waders and wear life jackets and rubber gloves; the driver is also clad in chest waders.

A typical sampling day on a continuously navigable river consists of launching the boat at an upstream access point, shuttling the truck and trailer to a downstream retrieval point, and returning to sample sites between the launch and retrieval points by navigating in a downstream direction. Normally, three 1.0 km sites can be sampled each day in river reaches of approximately 10-15 miles in length. If continuous navigation of a river segment is limited or precluded by falls, dams, or other safety concerns, the boat is launched and retrieved from a single access site in close proximity to the sampling site. In New England continuous navigation can be precluded by barriers including dams and other hazardous areas such as falls and heavy rapids. In a few instances, the boat was secured in the river overnight.

In our experience site access is seldom precluded by a lack of launch or retrieval access, although many locations required what is termed as "rough launching" (Figure 4). A few sites necessitated winching the boat and/or trailer, but most can be accessed with the trailer attached to the truck. A four wheel drive truck with the capacity to transport a three-person crew and the electrofishing boat/raft is essential to this type of sampling. Sufficient traction and pulling power is needed to access remote and unimproved access sites.

***Figure 4**. Logistics of boat electrofishing in New England; rough launching is required to gain access at many locations.*

Equipment Specifications

Boat electrofishing was the method of choice based on its successful application as a single gear approach to non-wadeable rivers in other parts of the U.S. and Canada and a successful trial application in the Kennebec River by Kleinschmidt in 2000-1. A 16' john boat was outfitted for initial testing in 2001 (Figure 5). This consisted of a design similar to that originated by Gammon (1973, 1976) and used by Ohio EPA (1989a). Electric current generated by a Smith-Root GPP 5.0 generator/pulsator combination is transmitted via an electrode array positioned 3.6 meters in front of the bow. The anodes (+ electrodes) consist of 3 gangs of 3/16" stainless steel woven cable with a single gang consisting of four to six 2 meter long strands that are doubled over and bundled together with zip ties. Cathodes (- electrodes) consist of four ¾"

***Figure 5**. Electrofishing boat and towing vehicle specifications and configuration used in the Maine rivers fish assemblage assessment, 2002-7.*

diameter flexible stainless steel conduit cut to lengths of 6-10' (longer for deeper sites) that are suspended directly from the bow. Wiring from the pulsator to the electrodes is encased in plastic conduit to protect against electrical shock. A positive pressure foot pedal switch is located on the bow platform and operated by a netter. Emergency cutoff switches are located within easy reach of the boat driver on the rear seat and on the 5.0 GPP pulsator unit. Lights are affixed to the safety railing to enable night sampling, although most sampling is conducted during daylight. The electrofishing boat is propelled by a 25 h.p. outboard mounted on the transom. The electrode and wiring arrangement for the 14' raft is the same except for the distance of the anodes in front of the bow (2.5 meters), cathodes deployed over the side of the raft, and a Smith-Root 2.5 GPP generator/pulsator mounted on a raft frame (Figure 6). A 15 h.p. tiltable outboard is used to propel the raft with a two person crew.

Electrofishing unit settings for the GPP units are typically governed by relative conductivity of the ambient water. At low conductivities (15-40 µS/cm) the GPP unit settings selected includes the high voltage range (500-1000 v) at 120 Hz and ≈100% of the voltage range to produce ≈2-4 A. At sites with higher relative conductivity (>40-100 µS/cm) the same settings at 60-100% of

Figure 6. *Electrofishing raft details showing boom and frame configuration (upper left), "rough" launching (upper right), GPP generator (lower left), and live well (lower right).*

the voltage range produced ≈5-10A. Higher relative conductivities in excess of 200 µS/cm necessitated switching to the low voltage range (maximum = 500 v) and at ≈50-80% of the voltage range at 120 Hz produced 12-18A. The latter situations were rare and occurred in southern New England and in reaches affected by urban runoff and point source effluents. The selection of the 120 Hz pulse frequency was accomplished by trial and error testing in Maine in during testing in 2001 and the initial 2002 river surveys. This was determined to be the most effective pulse setting based on visual observations of the comparative effectiveness in stunning all fish species. Lower settings (30, 60 Hz) were much less effective and are deemed unsuitable for New England rivers. Care is taken to avoid fish injury and all processed fish were examined for visible signs of damage or injury. The selected settings produced very few, if any visible injuries during any of the Maine or New England surveys.

Sampling Site Configuration

Sampling sites were located along the shoreline and in the river channel with the most diverse habitat features in accordance with established methods (Gammon 1973, 1976; Ohio EPA 1989a; Lyons et al. 2001; Yoder et al. 2005) used in the Maine Rivers project (Yoder et al. 2006 a,b). This is generally along the gradual outside bends of a river, but it is not invariable. New

England rivers presented both similarities and dissimilarities compared to the rivers of the Midwestern U.S. where this type of method was originally developed. Dissimilarities included faster current velocities including swift chutes, runs, and rapids and different cover types (e.g., large boulders, log cribs, deep runs, bedrock ledges) that can be positioned away from the shoreline and which required adaptations of the original methods. Boat electrofishing sites were sampled at a fixed distance of 1.0 km as determined for the Maine rivers project (Yoder et al. 2006a). The raft method introduced in 2005 attempted to adhere to the 1.0 km zone length, but some practical limitations of this length were encountered in selected rivers, hence a shorter distance of at least 0.75 km was used at some sites. The rationale is that these smaller rivers require a shorter distance to produce similar relative abundance and species richness results.

An example of a typical site configuration in a moderate to high gradient riverine reach appears in Figure 7 which shows the sampling path of the electrofishing boat produced by a GPS unit in the Kennebec River near Waterville, ME. In such free-flowing riverine reaches, a part of each zone included faster flowing run-riffle habitat in addition to slower flowing pool habitat when the former was available. The fixed sampling distance at each site was determined with a GPS unit or laser range finder. When using the GPS unit each zone is measured by determining the cumulative lineal distance of shoreline. This was done by tracking the cumulative lineal distance of adjacent shoreline as the sampling progressed in a downstream direction. Waypoints were established as necessary to account for the curvature of the shoreline along the sampling path that was followed within each site. Each river was designated with a unique alpha code (e.g., Kennebec River = “KEN”) and each site with a unique numeric descriptor (e.g., “KEN1”). The upstream end, or beginning of each site is designated “A” and subsequent waypoints are designated B, C, D, and so on. The downstream terminus of each zone was designated with a “Z”. This also produces a GPS recorded track of the route that the electrofishing boat/raft actually followed and can be used to determine how thoroughly a particular site was sampled (Figure 7). A detailed description of the sampling track is also recorded on the QHEI data sheet in addition to recording and saving the GPS track. This enables accurate relocation of sites in the event repeat visits are made. If the sampling zone is delineated in disjunct subzones, additional demarcations are necessary. A detailed description of the sampling location should also include proximity to a fixed local landmark such as a bridge, road, discharge outfall, railroad crossing, park, tributary, dam, etc.

Sampling site locations are indexed to UTM coordinates at the beginning, mid-point, and end of each zone. Sites are also delineated by river mile on specially derived maps that depict river mile in 0.1 mile increments. The delineation of river mile proceeds in an upstream direction with mile point zero at the head of tide for coastal rivers or the confluence with a larger river. Sites in the tidal zone of a river are depicted as negative values starting at the head of tide and proceeding towards the Atlantic Ocean.

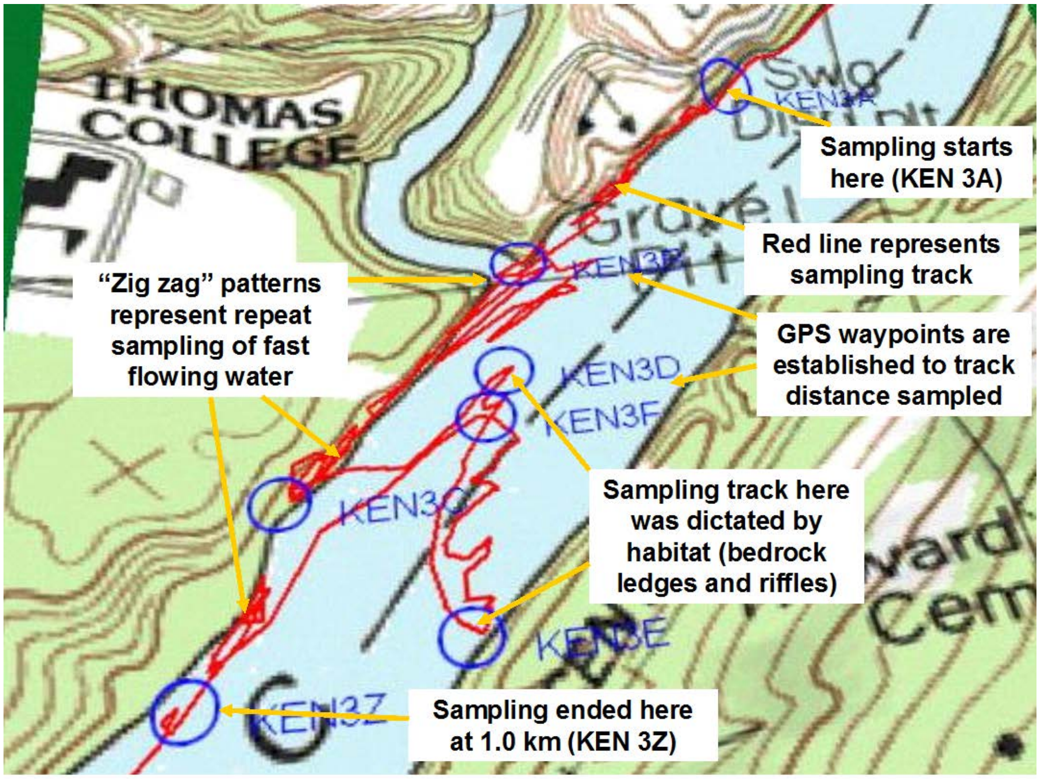


Figure 7. *Track of the electrofishing boat recorded by a GPS unit in the Kennebec River at RM 10.9 on September 9, 2004. The red line is the track followed by the sampling boat and illustrates the technique of thoroughly sampling available habitats and accessing sampleable habitat types within the 1.0 km site. Photo of typical bedrock ledge habitat is shown below.*

General Cautions Concerning Field Conditions

Electrofishing was conducted only during “normal” summer-fall water flow and clarity conditions. What constitutes normal can vary considerably from region to region, but usually include benchmarks such as daily median or 80^{th} percentile low flows as determined by USGS. Generally normal water conditions in New England occurred during below seasonal average river flows. Under these conditions the surface of the water generally had a placid appearance and visibility was > 1-2 meters. Abnormally turbid conditions were avoided as were elevated water levels and abnormal current velocities. Any of these conditions will adversely affect sampling efficiency and will rule out data applicability for bioassessment purposes. Since the ability of the netter to see and capture stunned fish is crucial, sampling took place only during periods of normal water clarity and flow. Floating debris such as twigs, tree limbs, flotsam, leaves, and other trash were usually visible on the surface during prohibitively elevated flow events. Such conditions were avoided and sampling was delayed until the water returned to "normal" flow. Boat and raft mounted methods are particularly susceptible as it becomes more difficult to maneuver the boat into areas of cover and the fish assemblage is locally displaced by most elevated flow events. High flows were also avoided for obvious safety reasons in addition to the sampling efficiency concerns. It usually took several days for the assemblage to return to their normal summer-fall distribution patterns following such flow events. Thus sampling was delayed by a similar time period. Recognizing such conditions requires local knowledge and a familiarity with flow gage readings and conditions. Generally, these conditions coincide with low flow durations of an 80^{th} percentile or less, i.e., flows that are exceeded $\geq$80% of the time for the period of record. These statistics are available for the New England states from the U.S. Geological Survey at: http://waterdata.usgs.gov/[state]/nwis/rt.

Field Sampling and Data Recording

Field data are recorded on water resistant data sheets formatted in the manner that the data is entered into an electronic database (Figures 8a and 8b). Each of the field crew are recorded on the field sheet with crew duties listed (crew leader, boat/raft driver, netters, etc.) along with site information including the alpha-numeric river-site code, a five digit basin-river code, UTM coordinates, river mile, and sampling date. The crew leader will also maintain a field activities log noting all circumstances related to field sampling such as site access, weather, and other relevant observations. All field data sheets are retained indefinitely by MBI.

Upon capture, fish are immediately placed in an aerated live well for later processing. Trout, salmon, and other comparatively fragile species can be placed in separate aerated containers and processed first to minimize their holding time. If necessary, fish are anesthetized to minimize trauma and handling stress, although this practice is rarely used. Adult Atlantic salmon and all sturgeon species that are drawn to the surface during electrofishing were not netted, but were identified and their length estimated by sight. The electric current was temporarily interrupted to minimize their exposure and in accordance with an ESA Section 7 Biological Opinion. Fish weights are derived by length/weight relationship data provided by the Maine DMR. All captured fish are processed by enumerating and recording weights by species or by species age class (Table 1). Individual fish weighing less than 1000 grams are weighed to the nearest gram on a spring dial scale (1000 g x 2g) or a 1000 g hand held spring scale. Fish

***Figure 8a**. Field data sheet for recording electrofishing collection data and for entry into the MBI Maine ECOS database. Front of two sides is shown below.*

Midwest Biodiversity Institute

Fish Data Sheet

Page ____ of ____

Crew Leader Boat Driver Netters

Project Code: ____________

Field Crew: ____________ Time of Day: ____________ Site Code: ____________

River/ Stream: ____________ Location: ____________

Date: ____________ Distance: ____________ Secchi Disk: ____________ Seconds Fished: ____________

River Code: ____________ Sampler Type: ____________ Diss Oxy: ____________ Lat/Long (Beg): ____________

RM: ____________ Depth: ____________ Temp: ____________ Lat/Long (Mid): ____________

Voltage: ____________ Range: ____________ PH: ____________ Lat/Long (End): ____________

% Range: ____________ Amperage: ____________ Conductivity: ____________ Lat/Long (X-Loc): ____________

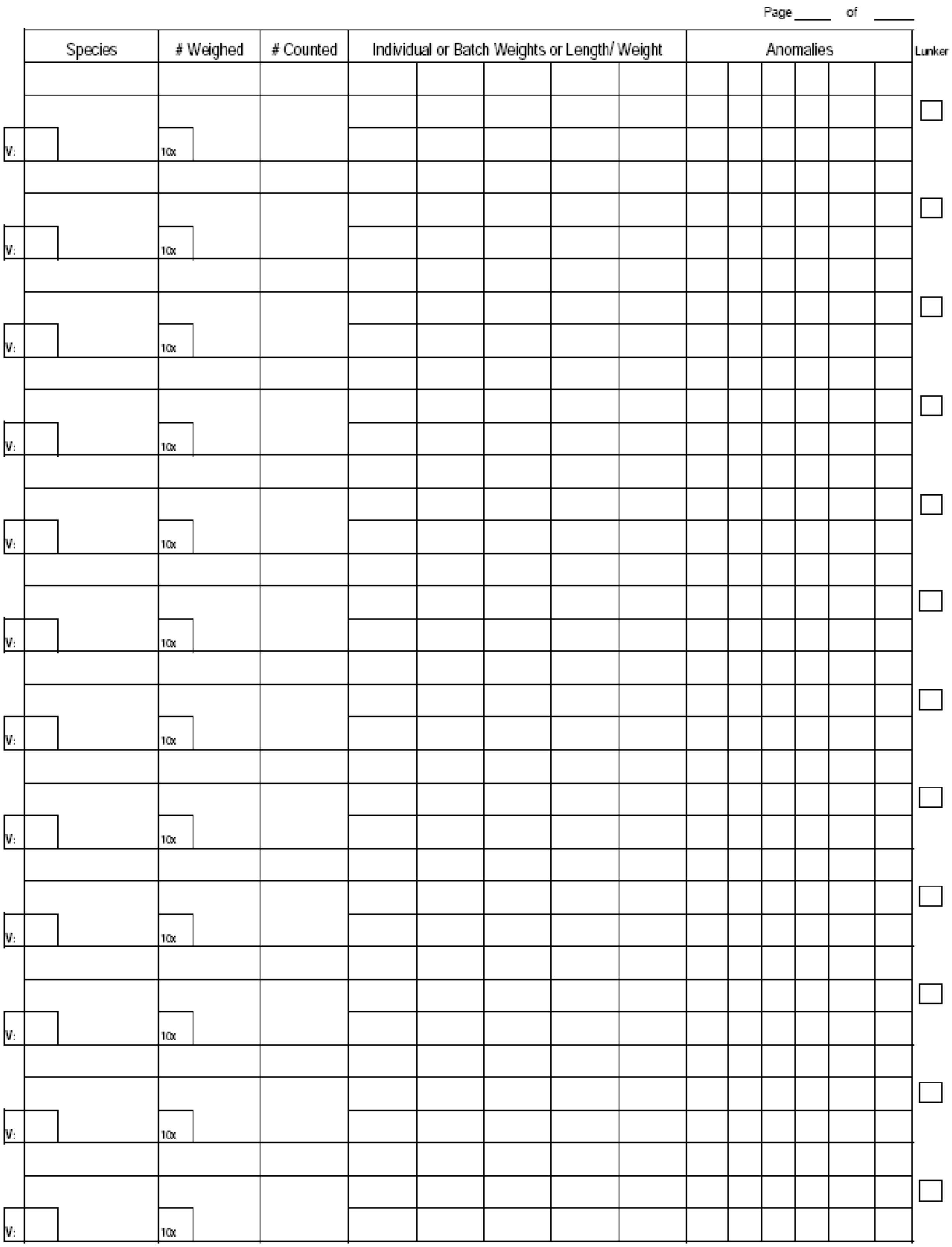
Anomalies: A- anchor worm; B- black spot; C- leeches; D- deformities; E- eroded fins; F- funguns; L- lesions; M- multiple DELT anomalies; N- blind P- parasites; Y- popeye; S- emaciated; W- swirled scales; T- tumors; Z- other. [Heavy (H) or Light (L) code may be combined with above codes.]

Species	# Weighed	# Counted	Individual or Batch Weights or Length/ Weight	Anomalies	Lunker
V:	10x				☐
V:	10x				☐
V:	10x				☐
V:	10x				☐
V:	10x				☐
V:	10x				☐
V:	10x				☐
V:	10x				☐

Mass Weighing Convention: Total Weight → 536 (12) ← Number Weighed Vouchers Collected: ☐

Revised 5/2008

***Figure 8b**. Fish data sheet continued - back of two sides is shown below.*



Page _____ of _____

Species	# Weighed	# Counted	Individual or Batch Weights or Length/ Weight	Anomalies	Lunker
V:	10x				☐
V:	10x				☐
V:	10x				☐
V:	10x				☐
V:	10x				☐
V:	10x				☐
V:	10x				☐
V:	10x				☐
V:	10x				☐
V:	10x				☐
V:	10x				☐
V:	10x				☐

***Table 1**. Criteria (weight, length, or other) used to determine adult (A), 1+ (juvenile; B), and 0+ (young-of-year; Y) designations for New England riverine fish species for the primary purpose of assuring the accuracy of extrapolated total biomass based on subsamples and for IBI guild classification. Not all species are differentiated.*

Species	Adult	1+[1]	0+
Sea lamprey *(Petromyzon marinus)*	fully developed[2]	-[2]	ammocoete
American eel *(Anguilla rostrata)*	>500 g		<10 g
Blueback herring *(Alosa aestivalis)*	>100 g		<10 g
Alewife *(Alosa pseudoharengus)*	>100 g		<10 g
American shad *(Alosa sapidissima)*	>100 g		<10 g
Lake chub *(Couesius plumbeus)*	>10 g		<1 g
Common carp *(Cyprinus carpio)*	>1000 g		<50 g
Common shiner *(Luxilis cornutus)*	>10 g		<1 g
Golden shiner *(Notemigonus crysoleucas)*	>100 g		<10 g
Spottail shiner *(Notropis hudsonius)*	>10 g		<1 g
Blacknose dace *(Rhinichthys atratulus)*		not determined	
Longnose dace *(Rhinichthys cataractae)*		not determined	
Creek chub *(Semotilus atromaculatus)*		not determined	
Fallfish *(Semotilus corporalis)*	>50 g		<3 g
Longnose sucker *(Catostomus catostomus)*	>1000 g		<10 g
White sucker *(Catostomus commersonii)*	>1000 g		<10 g
White catfish *(Ameirus catus)*	>100 g		<10 g
Brown bullhead *(Ameirus nebulosus)*	>100 g		<10 g
Northern pike *(Esox lucius)*	>500 g		<10 g
Chain pickerel *(Esox niger)*	>80 g		<10 g
Rainbow trout *(Oncorhynchus mykiss)*	>100 g		<10 g
Atlantic salmon *(Salmo salar salar)*	>500 mm		<10 g
Landlocked salmon *(Salmo salar sebago)*	>100 g		<10 g
Brown trout *(Salmo trutta)*	>100 g		<10 g
Brook trout *(Salvelinus fontinalis)*	>100 g		<10 g
Burbot *(Lota lota)*	>100 g		<10 g
Banded killifish *(Fundulus diaphanus)*		not determined	
Mummichog *(Fundulus heteroclitus)*		not determined	
Slimy sculpin *(Cottus cognatus)*	>20 g		<2 g
White perch *(Morone americana)*	>100		<10 g
Striped bass *(Morone saxatilis)*	>500 mm		<50 g
Rock bass *(Ambloplites rupestris)*	>80 g		<10 g

[1] Juvenile criteria are <adult, >y-o-y (0+).
[2] Parasitic habits fully developed in adults; buccal funnel is fully developed in juveniles, but is not yet parasitic.

***Table 1**. (continued)*

Species	Adult	1+	0+
Redbreast sunfish *(Lepomis auritus)*	>50 g		<5 g
Pumpkinseed sunfish *(Lepomis gibbosus)*	>50 g		<5 g
Smallmouth bass *(Micropterus dolomieu)*	>150 mm		<10 g
Largemouth bass *(Micropterus salmoides)*	>150 mm		<10 g
Black crappie *(Pomoxis nigromaculatus)*	>100 g		<10 g
Yellow perch *(Perca flavescens)*	>50 g		<5 g

weighing more than 1000 grams are weighed to the nearest 25 grams on a 12 kg spring dial scale (12 kg x 50 g) or 20 kg hand held spring scales. Extremely large fish are weighed using a 50 kg hand held spring scale. Samples that are comprised of two or more distinct size classes of fish (e.g., y-o-y, juveniles, and adults) are processed separately (Table 1). Species that occur in large numbers can be subsampled with a minimum of 15 individuals for large adults and 50 for smaller species and 1+ or 0+ life stages. Fish are distinguished as adults, 1+ (juveniles), or 0+ (young-of-year) in accordance with the criteria in Table 1. These are recorded on the field data sheet by designating an A (adult), B (1+ year), or Y (0+ or young-of-year) to the numeric species code. For example, if both adult and juvenile white suckers occur in the same sample the adult numbers and weights are recorded as family-species code 40-016A with juvenile numbers and weights recorded as 40-016B. Although each is listed separately on the fish data sheet they can be treated in the aggregate as a single sample of the same species in any subsequent data analyses or as distinct size class entities. The principal purpose of this differentiation was to increase the accuracy of extrapolations based on subsampling and for potential IBI guild classification purposes. The data management programs used by MBI are designed to calculate relative numbers and biomass data based on the input of the weighted subsamples. Total lengths can also be recorded for important commercial, recreational, and special interest species on an as needed or as requested basis. Immature and post-larval fish measuring less than 15-20 mm in length are generally not included in the data recording as a matter of practice following the recommendations of Angermeier and Karr (1986) that post-larval fish not be included in IBI calculations. However, specimens may be retained for other purposes.

All fish that are weighed, whether done individually, in the aggregate, or as subsamples, were examined for the presence of gross external anomalies (Figure 9). Light and heavy infestations were noted for certain types of anomalies and follow the guidance in Ohio EPA (1989a) and Sanders et al. (1999). DELT (deformities, erosions, lesions, and tumors) anomalies are a metric in the ME IBI and are also used as a diagnostic indicator.

***Figure 9**. Some of the common external anomalies that occurred in New England rivers and which were recorded as part of the fish assemblage data collection.*

The majority of captured fish are identified to species in the field; however, any uncertainty about the field identification of individual fish requires the retention of voucher specimens for laboratory identification. Vouchered fish are preserved in a solution of borax buffered 10% formalin and labeled by date, river name, and site designation. Identification is made to the species level in all cases and follows the nomenclature of the American Fisheries Society (Nelson et al. 2004; Page et al. 2013). The same is true of new river or regional species distribution records. Fish are preserved for later identification in borax buffered 10% formalin and labeled by date, river or stream, and geographic identifier (e.g., river mile). Large specimens (>50-100 mm) usually require visceral incision (lower right abdominal) to permit proper preservation of internal tissues and organs. After an initial fixation period of at least 3-4 weeks, specimens are washed in plain water and then transferred to increasing dilutions of ethyl alcohol (non-denatured) and water (35%, 50%) and ultimately to a final solution of 70% ethyl alcohol. This process takes approximately 4-5 weeks to complete. Identification is then performed to the species level at a minimum and it may be necessary to the sub-specific level in certain instances. Regional ichthyology keys are used and include the Inland Fishes of Massachusetts (Hartel et al. 2002), Fishes of the Gulf of Maine (Bigelow and Schroeder 1953;

Collette and Klein-McPhee 2000), Freshwater Fishes of Canada (Scott and Crossman 1973), Fishes of Vermont (Langdon et al. 2006), and Inland Fishes of New York (Smith 1985). Assistance with the verification of voucher specimens was provided by Dr. David Halliwell, Maine DEP, Karsten Hartel, Museum of Comparative Zoology, Cambridge, MA, and Marc Kibbey, The Ohio State University Museum of Biodiversity, Columbus, OH. Voucher photographs are also taken to record species occurrences, particularly larger species that are not easily preserved and stored. Photographs are maintained by MBI in an archived electronic file on the MBI data server.

Sampling Procedure and Gear Selection

The selection of sampling gear is determined on a river reach basis and along a continuum of all possible lotic strata from wadeable to non-wadeable (Table 2). We included the closest wadeable strata to illustrate where the transition to non-wadeable equipment and protocols occurs in terms of waterbody size as measured by drainage area and Strahler order. Clearly that choice is not presented until Strahler order IV hence order I-III sites were excluded from the initial REMAP site draw. From that point on the choices are then governed by factors such as gradient and relative conductivity. For the smallest and/or highest gradient non-wadeable sites the 14' raft platform was used transitioning to the 16' boat for the largest non-wadeable sites. The size and power of the accompanying electrofishing unit increases from the 14' raft to the 16' boat platform and corresponds to increases in depth within sampling sites. The choice of which equipment combination to use is made on-site by the crew leader with the directive to take a conservative approach in that the most powerful equipment that a waterbody can safely and effectively accommodate be used.

Habitat and Water Quality Methods

A qualitative habitat assessment using an appropriate modification of the Qualitative Habitat Evaluation Index (QHEI; Ohio EPA 1989a; Rankin 1989, 1995; Ohio EPA 2006; Yoder et al. 2006a) is completed by the crew leader at each electrofishing site. The QHEI is a physical habitat index designed to provide an empirical, qualitative evaluation of the lotic macrohabitat characteristics that are important to fish assemblages. Comprised of seven categories of aquatic habitat (Figures 10a and 10b), the QHEI was developed as a rapid assessment tool and in recognition of the constraints associated with the practicalities of conducting a large-scale monitoring program, i.e., the need for a rapid assessment tool that yields meaningful information and which takes advantage of the knowledge and insights of experienced field biologists who are conducting the biological assessments. This index has been used widely outside of its Ohio origins and parallel habitat evaluation techniques are in widespread existence throughout the U.S. The QHEI incorporates the types and quality substrate, the types and amounts of instream cover, several characteristics of channel morphology, riparian zone extent and quality, bank stability and condition, and pool-run-riffle quality and characteristics. Slope or gradient is also factored into the QHEI score. We followed the guidance and scoring procedures outlined in Ohio EPA (1989a, 2006) and Rankin (1989) with some minor modifications made during 2002 and 2003 in Maine (Yoder et al. 2006a). A QHEI habitat assessment form is completed by the crew leader for each 1.0 km site.

***Table 2**. Key characteristics of electrofishing protocols applicable to New England riverine habitats.*

Category/Attribute	Riverine Wadeable[a] (Low-Mod. Cond.[b])	Riverine High Gradient (Low Cond.)	Riverine Mod. Gradient (Low Cond.)	Riverine Low Gradient (Mod. Cond.)	Impounded (Low Cond.)	Impounded (Mod. Cond.)	Tidal (High Cond.)
1. Drainage Area Strahler Order	<500 mi^2 ≤IV	<500 mi^2 ≥V	>500-1000 mi^2 ≥V	>1000 mi^2 ≥V	NA NA	NA NA	NA NA
2. Platform	Georator[c] (bank set/towboat)	14' raft[d] 16' johnboat	16' johnboat 14-16' raft	16' johnboat	16' johnboat	16' johnboat	16' johnboat
3. Crew Size	3 persons (2 netters)	2 persons (1 netter)	3 persons (2 netters)	3 persons (2 netters)	3 persons (2 netters)	3 persons (2 netters)	3 persons (2 netters)
4. Electrofishing Unit	GPP 2.5, 5.0[e] or equivalent	GPP 2.5, 5.0 or equivalent	GPP 5.0 or equivalent	GPP 5.0 or equivalent	GPP 5.0 or equivalent	GPP 5.0 or equivalent	GPP 5.0 or larger
5. Power Source	2500-5000 Watt Alternator	5000 Watt Alternator	5000 Watt Alternator	5000 Watt Alternator	5000 Watt Alternator	5000 Watt Alternator	≥5000 Watt Alternator
6. Unit Settings[f] (% of Low or High Range)	High 120 Hz	High 120 Hz 2-4 Amperes (100%)	High 120 Hz 2-4 Amperes (100%)	Low or High 120 Hz 4-8 Amperes (60-100%)	High 120 Hz 2-4 Amperes (100%)	Low or High 120 Hz 4-8 Amperes (60-100%)	Low 120 Hz >8-15 Amperes (50-80%)
7. Anodes[g]	Net Ring	2 gangs	3 gangs	3 gangs	3 gangs	3 gangs	2 gangs

[a] Wadeable defined as sites where a raft or boat mounted apparatus cannot be used due to shallowness of depth – accessibility is not a criterion.
[b] Typical relative conductivity ranges: Low (15-40 µS/cm); Moderate (40 – 200 µS/cm); High (>200 µS/cm).
[c] Employs a primary net ring as the anode that is operated by the primary netter backed by an assist netter - the unit is either bank set or towed on a small skiff (towboat).
[d] This platform was more extensively tested in Maine in 2005; it has worked well for other investigators in similar settings.
[e] This does not constitute an endorsement of a particular brand or product name and is for methodological identification only.
[f] Unit settings are selected to produce the highest voltage and amperage output; these are what typically worked in each conductivity range and habitat type.
[g] Anodes consist of gangs or multiple strands of wire as described under Equipment Specifications.

Table 2. Continued.

Category/Attribute	Riverine Wadeable[a] (Low-Mod. Cond.[b])	Riverine High Gradient (Low Cond.)	Riverine Mod. Gradient (Low Cond.)	Riverine Low Gradient (Mod. Cond.)	Impounded (Low Cond.)	Impounded (Mod. Cond.)	Tidal (High Cond.)
8. Cathodes	rat tail	6’	8’	8’	8’	8’	8’
9. Sampling Direction & Distance	Upstream 0.2-0.5 Km	Downstream 0.5-1.0 Km	Downstream 1.0 Km	Downstream 1.0 Km	Downstream 1.0 Km	Downstream 1.0 Km	Downstream 1.0 Km
10. CPUE[h] Basis	Per 0.5 Km	Per Km	Per Km	Per Km	Per Km	Per Km	Per Km
11. Time Sampled[i]	Not tested	3500-4500 s	4000-5500 s	3500-4500 s	3000-4000 s	3000-4000 s	3500-4500 s
12. Time of Day	Day	Day	Day	Day	Day or Night	Day or Night	Day

[h] CPUE = Catch Per Unit Effort; this is the basis for calculating relative abundance estimates.
[i] Typically the *minimum* time required to execute the electrofishing protocol at a 1.0 km site; actual time may be higher in more difficult to sample site

Figure 10a. *Qualitative habitat evaluation index (QHEI) field sheet showing categorical attributes (front side).*

Midwest Biodiversity Institute

Qualitative Habitat Evaluation Index Field Sheet

QHEI Score: ______

River Code: ______ *RM:* ______ *Stream:* ______
Site Code: ______ *Project Code:* ______ *Location:* ______
Date: ______ *Scorer:* ______ *Latitude:* ______ *Longitude:* ______

1.) SUBSTRATE (Check ONLY Two Substrate TYPE BOXES; Estimate % percent

TYPE	POOL	RIFFLE		POOL	RIFFLE
☐ ☐ -BLDR/SLBS [10]	____	____	☐ ☐ -GRAVEL [7]	____	____
☐ ☐ -Lg BOULD [10]	____	____	☐ ☐ -SAND [6]	____	____
☐ ☐ -BOULDER [9]	____	____	☐ ☐ -BEDROCK [5]	____	____
☐ ☐ -COBBLE [8]	____	____	☐ ☐ -DETRITUS [3]	____	____
☐ ☐ -HARDPAN [4]	____	____	☐ ☐ -ARTIFICIAL [0]	____	____
☐ ☐ -MUCK [2]	____	____	☐ ☐ -SILT [2]	____	____

NUMBER OF SUBSTRATE TYPES: ☐ -4 or More [2]
(High Quality Only, Score 5 or >) ☐ -3 or Less [0]

SUBSTRATE ORIGIN
Check ONE (OR 2 & AVERAGE)
☐ -LIMESTONE [1]
☐ -TILLS [1]
☐ -WETLANDS [0]
☐ -HARDPAN [0]
☐ -SANDSTONE [0]
☐ -RIP / RAP [0]
☐ -LACUSTRINE [0]
☐ -SHALE [-1]
☐ -COAL FINES [-2]

SUBSTRATE QUALITY
Check ONE (OR 2 & AVERAGE)
SILT: ☐ -SILT HEAVY [-2]
☐ -SILT MODERATE [-1]
☐ -SILT NORMAL [0]
☐ -SILT FREE [1]
EMBEDDEDNESS: ☐ -EXTENSIVE [-2]
☐ -MODERATE [-1]
☐ -NORMAL [0]
☐ -NONE [1]

Substrate ☐ Max 20

COMMENTS: ______

2.) INSTREAM COVER (Give each cover type a score of 0 to 3; see back for instructions)

(Structure) TYPE: Score All That Occur

____ UNDERCUT BANKS [1]	____ POOLS > 70 cm [2]	____ OXBOWS, BACKWATERS [1]
____ OVERHANGING VEGETATION [1]	____ ROOTWADS [1]	____ AQUATIC MACROPHYTES [1]
____ SHALLOWS (IN SLOW WATER) [1]	____ BOULDERS [1]	____ LOGS OR WOODY DEBRIS [1]
____ ROOTMATS [1]		

AMOUNT: (Check ONLY one or check 2 and AVERAGE)
☐ -EXTENSIVE > 75% [11]
☐ -MODERATE 25 - 75% [7]
☐ -SPARSE 5 - 25% [3]
☐ -NEARLY ABSENT < 5% [1]

Cover ☐ Max 20

COMMENTS: ______

3.) CHANNEL MORPHOLOGY: (Check ONLY one PER Category OR check 2 and AVERAGE)

SINUOSITY	DEVELOPMENT	CHANNELIZATION	STABILTIY	MODIFICATIONS / OTHER	
☐ -HIGH [4]	☐ -EXCELLENT [7]	☐ -NONE [6]	☐ -HIGH [3]	☐ -SNAGGING	☐ -IMPOUNDMENT
☐ -MODERATE [3]	☐ -GOOD [5]	☐ -RECOVERED [4]	☐ -MODERATE [2]	☐ -RELOCATION	☐ -ISLAND
☐ -LOW [2]	☐ -FAIR [3]	☐ -RECOVERING [3]	☐ -LOW [1]	☐ -CANOPY REMOVAL	☐ -LEVEED
☐ -NONE [1]	☐ -POOR [1]	☐ -RECENT OR NO RECOVERY [1]		☐ -DREDGING	☐ -BANK SHAPING
		☐ -IMPOUNDED [-1]		☐ -ONE SIDE CHANNEL MODIFICATIONS	

Channel ☐ Max 20

COMMENTS: ______

4.) RIPARIAN ZONE AND BANK EROSION (check ONE box PER bank or check 2 and AVERAGE per bank) River Right Looking Downstream

RIPARIAN WIDTH	FLOOD PLAIN QUALITY (PAST 100 Meter RIPARIAN)		BANK EROSION
L R (Per Bank)	L R (Most Predominant Per Bank)	L R	L R (Per Bank)
☐ ☐ -VERY WIDE > 100m [5]	☐ ☐ -FOREST, SWAMP [3]	☐ ☐ -CONSERVATION TILLAGE [1]	☐ ☐ -NONE / LITTLE [3]
☐ ☐ -WIDE > 50m [4]	☐ ☐ -SHRUB OR OLD FIELD [2]	☐ ☐ -URBAN OR INDUSTRIAL [0]	☐ ☐ -MODERATE [2]
☐ ☐ -MODERATE 10 - 50m [3]	☐ ☐ -RESIDENTIAL, PARK, NEW FIELD [1]	☐ ☐ -OPEN PASTURE, ROWCROP [0]	☐ ☐ -HEAVY / SEVERE [1]
☐ ☐ -NARROW 5 - 10m [2]	☐ ☐ -FENCED PASTURE [1]	☐ ☐ -MINING / CONSTRUCTION [0]	
☐ ☐ -VERY NARROW < 5m [1]			
☐ ☐ -NONE [0]	COMMENTS: ______		

Riparian ☐ Max 10

5.) POOL / GLIDE AND RIFFLE / RUN QUALITY

MAX. DEPTH (Check 1 ONLY!)	MORPHOLOGY (Check 1 or 2 & AVERAGE)	CURRENT VELOCITY (POOLS & RIFFLES!) (Check All That Apply)	
☐ - 1m [6]	☐ -POOL WIDTH > RIFFLE WIDTH [2]	☐ -EDDIES [1]	☐ -TORRENTIAL [-1]
☐ - 0.7m [4]	☐ -POOL WIDTH = RIFFLE WIDTH [1]	☐ -FAST [1]	☐ -INTERSTITIAL [-1]
☐ - 0.4 to 0.7m [2]	☐ -POOL WIDTH < RIFFLE WIDTH [0]	☐ -MODERATE [1]	☐ -INTERMITTENT [-2]
☐ - 0.2 to 0.4m [1]	☐ -IMPOUNDED [-1]	☐ -SLOW [1]	☐ -VERY FAST [1]
☐ - < 0.2m [POOL = 0]		☐ -NONE [-1]	

Pool / Current ☐ Max 12

COMMENTS: ______

CHECK ONE OR CHECK 2 AND ADVERAGE

RIFFLE DEPTH	RUN DEPTH	RIFFLE / RUN SUBSTRATE	RIFFLE / RUN EMBEDDEDNESS
☐ -*Best Areas > 10cm [2]	☐ - MAX > 50 cm [2]	☐ -STABLE (e.g., Cobble, Boulder) [2]	☐ -NONE [2]
☐ -Best Areas 5 - 10cm [1]	☐ - MAX < 50 cm [1]	☐ -MOD. STABLE (e.g., Large Gravel) [1]	☐ -LOW [1]
☐ -Best Areas < 5cm [0]		☐ -UNSTABLE (Fine Gravel, Sand) [0]	☐ -MODERATE [0]
☐ -NO RIFFLE but RUNS present [0]			☐ -EXTENSIVE [-1]
☐ -NO RIFFLE / NO RUN [Metric = 0]			

Riffle / Run ☐ Max 8

COMMENTS: ______

6.) GRADIENT (ft / mi): ______ DRAINAGE AREA (sq.mi.): ______ % POOL: ☐ % GLIDE: ☐ % RIFFLE: ☐ % RUN: ☐

Gradient Score from Table 2 of Users Manual based on gradient and drainage area.

Gradient ☐ Max 10

**Best areas must be large enough to support a population of riffle-obligate species*

***Figure 10b**. Qualitative habitat evaluation index (QHEI) field sheet (back side).*

Is Sampling Reach Representative of the Stream? (Y/ N) ____ If Not, Explain: ____

Lat / Long (Beg): ____

Lat / Long (Mid): ____

Lat / Long (End): ____

Lat / Long (X-Loc): ____

Subjective Rating (1-10)	Aesthetic Rating (1-10)

Gradient:
☐ -Low ☐ -Moderate ☐ -High

	Gear:	Distance:	Water Clarity:	Water Stage:	Canopy- % open:
First Sampling Pass					

Yes	No	
☐	☐	Is Stream Ephemeral (no pools, totally dry of only damp spots)?
☐	☐	Is there water upstream? How far:
☐	☐	Is there water close downstream? How far:
☐	☐	Is Dry Channel mostly natural?

Major Suspected Sources of Impacts (Check All That Apply):
- None ☐
- Industrial ☐
- WWTP ☐
- Agriculture ☐
- Livestock ☐
- Silviculture ☐
- Construction ☐
- Urban Runoff ☐
- CSOs ☐
- Suburban Impacts ☐
- Mining ☐
- Channelization ☐
- Riparian Removal ☐
- Landfills ☐
- Natural ☐
- Dams ☐
- Other Flow Alteration ☐

Other: ____

Stream Drawing:

Instructions for scoring the alternate cover metric: Each cover type should receive a score of between 0 and 3, where: 0 = Cover type absent; 1 = cover type in very small amounts or if more common of marginal quality; 2 = cover type present in moderate amounts, but not of highest quality or in small amounts of highest quality; 3 = cover type of highest quality in moderate of greater amounts. Examples of highest quality include, very large boulders in deep or fast water, large diameter logs that are stable, well developed rootwads in deep / fast water, or deep, well-defined, functional pools.

Field chemical/physical measurements are taken in the field during fish sampling and include temperature (°C), dissolved oxygen (D.O., mg/l and % saturation), relative conductivity (µS/cm), pH (S.U.) with YSI 556 meters. The meters were maintained and calibrated in accordance with the project QAPP (MBI 2008).

Data Management

MBI used the Maine ECOS data management system developed to store, retrieve, and analyze biological and habitat assessment data and information. Fish assemblage and qualitative habitat data were entered via an electronic data entry routine from the field sheets (see Figures 6 and 8). All data entry codes followed those developed specifically for New England in conformance to the Maine ECOS data management system. Each entered data sheet contains the basin-river code, date of entry, river mile, and date of sampling. Each entry was checked, initialed, and dated by a data entry analyst; any subsequent changes that were made to the fish data sheets were also initialed and dated. After all data was entered into Maine ECOS the entries were proofread by the data entry analyst and a crew leader for accuracy. All corrections or updates were then made in the database. The initialed data sheets also served as the chain-of-custody for the data collection and management process. The data sheets were then assembled in a notebook along with a data sheet log, any site description sheets, maps of the sampling sites, and the site characterization forms and retained on permanent file at MBI.

NELR Sampling Designs

The sampling design for the primary objective of providing a statistically valid assessment of the status of the fish assemblages of non-wadeable rivers was based on a spatially probabilistic design. The target population included non-wadeable freshwater rivers (as previously defined) throughout New England. This included coastal rivers to the head-of-tide thus excluding any part of a river that was influenced by tides (these areas were sampled by the Maine and Connecticut river surveys). A targeted site design used in selected NELR mainstem rivers and the 2002-7 Maine rivers assessment were included primarily to compare the assessment outcomes between the baseline probabilistic assessment and that from the targeted sites.

NRSA Survey Design (2008-9)

The basis for the selection of the probabilistic sites for NELR REMAP project was the nationally stratified and unequal probability survey design of the 2008-9 National River and Stream Assessment (NRSA). Nationally the sample size was set at 900 sites for Strahler 5th order and larger rivers (Olsen et al. 2007). Approximately 200 sites were allocated within each of nine aggregated level III ecoregions (Omernik 1987) as follow-up sites from the 2004-5 Wadeable Streams Assessment (WSA) and also allocating a minimum number of sites per state. The primary goal of the NRSA was to provide national and regional (i.e., at the aggregated level III ecoregion scale) estimates of the status of non-wadeable rivers.

Unequal selection probabilities were defined for 5th, 6th, 7th, and 8th Strahler order rivers such that the expected number of sites for each panel would be 350, 275, 175, and 100 sites, respectively. These unequal selection probabilities were then adjusted by the nine aggregated level III ecoregion strata so that an equal number of sites would occur in each region. The design also included a minimum number of sites by state. This comprised the non-wadeable stratum of the 2008-9 NRSA (Olsen 2007).

The NELR REMAP study area occurs within the Northern Appalachians region of the NRSA (Figure 11). The effect of having a minimum number of sites in each state resulted in a higher concentration of sites in southern New England and accounts for the differential distribution of probabilistic sites by latitude (Table 3).

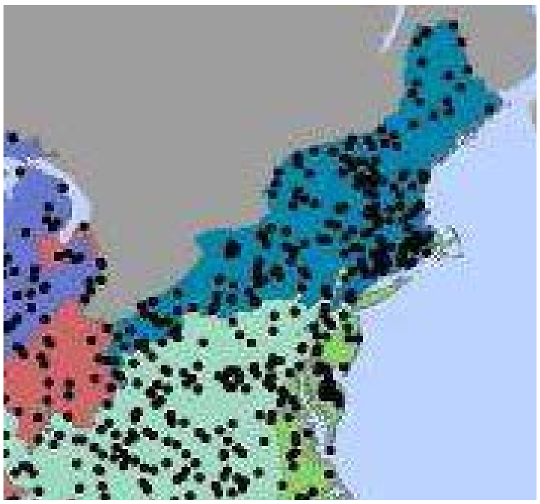


Figure 11. *Map of the Northeastern portion of the U.S. showing the base sites of the 2008-9 NRSA (Tarquinio 2011). The Northern Appalachians region incorporates all of New England (blue shaded).*

NELR REMAP Probabilistic Design

The probabilistic aspect of this project was based on the NRSA draw of sites that were made available for each of the six New England states by U.S. EPA. The NRSA base draw of sites was intensified by selecting an approximate equal number of overdraw sites to comprise the NELR REMAP sample.

All Strahler order sites 1-3 were eliminated from NELR REMAP consideration as these were assumed to be wadeable. The remaining sites of 4th order and larger were retained to provide a first estimate of potentially non-wadeable sites as the target population for the NELR REMAP project. It seemed plausible that a portion of the 4th and perhaps some 5th order sites would not be sampleable with either the boat or the raft method and, if so, they were rejected as being non-target for this project. Probabilistic sites were accepted or rejected via an initial on-site reconnaissance and/or during the initial sampling visits and generally in accordance with the site acceptance procedures of the NRSA. Our determination of the transition between wadeability and non-wadeability was "conservative", i.e., we attempted to gain access for using non-wadeable methods by unusual means if necessary and as previously described in the non-wadeable methods protocol discussion. For the NRSA base sites, we also verified the decisions made by the other state and/or contractor NRSA crews since some of the NRSA base sites were visited by these crews ahead of the NELR REMAP project crew. It was a major objective of this project to compare the NELR REMAP fish sampling method with the NRSA fish sampling method hence the crew efforts for each project were coordinated.

The procedure was to visit the NRSA base sites first to determine sampleability with the boat/raft methods and to also determine accessibility. Sites that were either not accessible nor sampleable with the non-wadeable equipment were rejected and replaced with the next overdraw site from within the same state following the order in the 2008-9 NRSA state sites lists. Inaccessible and otherwise non-target sites were also replaced in a like manner. The total of base and overdraw sites in Table 3 reflect the target number of sites within each state (see Appendix A for the full list of sites by state).

Following the process just outlined, the NELR REMAP sample included 75 NRSA base and 66 overdraw sites of Strahler order 4 and larger (Table 3). Nearly 90% of the sites were in Strahler orders 5 and 6 and only 4 total sites in Strahler order 4. River locations were sampled once within a July 1 – October 15 (September 30 for Maine, New Hampshire, and Vermont north of ≈43-44 N latitude) seasonal index period as river flow, water clarity, and weather conditions permitted. An approximate 10% resample of the NELR REMAP sites was accomplished (see Appendix A).

***Table 3**. NRSA base and overdraw sites that were sampled in 2008-9 listed by state and Strahler order. These comprise the NELR REMAP probabilistic design.*

State - Type	4th Order	5th Order	6th Order	7th Order
Connecticut – Base [16]	-	9	7	-
Connecticut – Overdraw [10]	-	7	3	-
Massachusetts – Base [15]	1	6	4	4
Massachusetts – Overdraw [15]	-	7	4	4
Maine – Base [12]	1	3	8	-
Maine – Overdraw [12]	-	7	4	1
New Hampshire – Base [10]	-	4	5	1
New Hampshire – Overdraw [10]	-	4	3	3
Rhode Island – Base [12]	1	11	-	-
Rhode Island – Overdraw [10]	-	10	-	-
Vermont – Base [10]	1	8	1	-
Vermont – Overdraw [9]	-	9	-	-
TOTAL – Base [75]	4	41	25	5
TOTAL – Overdraw [66]	-	44	14	8

Intensive Pollution Survey Design

A longitudinally stratified, intensive pollution survey design was used to select sampling sites in selected mainstem rivers. This design emanates from the pollution continuum concept of Bartsch (1948) and Bartsch and Ingram (1967) that has been employed previously by Gammon (1976), Hughes and Gammon (1987), Ohio EPA (Yoder and Smith 1999; Yoder et al. 2005), Kovacs et al. (2002), and Lyons et al. (2001) to accomplish an understanding of pollution effects on riverine fish assemblages. This consisted of locating sites in proximity to major sources of potential stress (major point sources, hydroelectric peaking facilities, dams, tributary confluences), including major habitat types (free-flowing, impounded, tidal estuary), and being spatially arrayed so that a longitudinal profile of fish assemblage attributes and indices could be analyzed and interpreted. Such a design represents a systematic and stratified census of the mainstem river fish assemblage that also includes the coverage of diverse pollution gradients that exist in longitudinal space. Following an initial allocation of sites the exact sampling locations were determined in the field and also included a representative proportion of reaches along each mainstem with respect to the transition from cold water to cool and warmwater habitats, modified (i.e., usually impounded sections behind dams and reaches affected by water level fluctuations below hydroelectric facilities), relatively unmodified and free-flowing reaches, reaches affected by point source discharges, and segments affected by dam removals. Specific sites were selected during the initial sampling runs to include representative environmental conditions and habitats available along each mainstem river and to capture the pollution impact continuum. Outside of Maine, the most complete of this design was the Connecticut River mainstem survey that included all raftable and boatable sites from northern New Hampshire to the salt wedge in southern Connecticut. In order to take advantage of the NRSA base and overdraw sites that were already available, these also functioned as intensive pollution survey sites with targeted sites added to fill in for gaps left by the probabilistic sites along the longitudinal continuum. These and other targeted sites from the 2002-7 Maine Rivers survey and in other selected New England rivers were included in the regional analyses to provide a basis for comparing probabilistic and targeted assessment outcomes. An accounting of the targeted sites that were sampled in 2008-9 appears in Appendix A that is organized by each New England state and the Connecticut River survey.

Spatial Distribution of NELR Sampling Sites

The spatial distribution of rivers across New England is depicted in Table 4 and Figure 12 and is based on the NHDPlus coverage for New England (excluding the Lake Champlain drainage in Vermont). This was done to determine the occurrence of non-wadeable river reaches by state and across New England and to determine if there was a spatial bias in the probabilistic or targeted sites data sets. Strahler order 5 and larger were used to represent the non-wadeable target population for this study. Only four order 4 sites were included in the NELR REMAP probabilistic data set as most were rejected as being wadeable.

Nearly one-half (49.3%) of the order 5 and larger rivers are in Maine, followed by New Hampshire (21.1%), Massachusetts (13.2%), and Connecticut (12.1%) with Vermont and Rhode Island combined being <5%. The Connecticut River mainstem was assigned to New Hampshire which shares their state boundary with Vermont in the upper one-half of the mainstem. The Vermont totals do not include the Winooski R. as the NHDPlus coverage we used excluded the Lake Champlain drainage. Also, the Champlain drainage fish fauna has more in common with the New York and the Midwest than the rest of New England because the eastward post-glacial reinvasion by western species ceased at elevations between 400 and 800 in Vermont and Quebec (Langdon 2006).

We also examined the occurrence of the NELR REMAP probabilistic and targeted sites used in the analyses by increments of latitude (Table 5). This aspect is thought to be important in that it potentially represents both a natural climatic and an anthropogenic impact gradient as the latter increases in a southerly direction in New England. The influence of the natural gradient is less certain and difficult to show with our empirical datasets as the confounding aspect of the anthropogenic gradient is difficult to distinguish.

***Table 4**. Total length of non-wadeable rivers in New England by Strahler stream order (≥5) and by state. Data are from NHDPlus.*

State and Stream Order	Reaches	Total Length (km)	Percent
Maine – Order 5	1083	1299	32.8
Maine – Order 6	440	540	13.6
Maine – Order 7	96	111	2.8
Maine Totals	1619	1950	49.2
Connecticut – Order 5	207	240	6.1
Connecticut – Order 6	197	239	6.0
Connecticut – Order 7	0	0	0
Connecticut Totals	404	479	12.1
Massachusetts – Order 5	297	324	8.2
Massachusetts – Order 6	88	114	2.9
Massachusetts – Order 7	50	84	2.1
Massachusetts Totals	435	522	13.2
New Hampshire – Order 5	480	509	12.8
New Hampshire – Order 6	225	249	6.3
New Hampshire – Order 7	65	81	2.0
New Hampshire Totals	770	839	21.1
Rhode Island – Order 5	46	84	2.1
Rhode Island – Order 6	4	5	0.1
Rhode Island – Order 7	0	0	0
Rhode Island Totals	50	89	2.2
Vermont[1] – Order 5	88	82	2.1
Vermont[1] – Order 6	2	1	<0.1
Vermont[1] – Order 7	0	0	0
Vermont Totals	90	83	2.1
TOTAL - Order 5	2201	2538	64.1
TOTAL - Order 6	956	1148	29.0
TOTAL - Order 7	211	276	6.9
GRAND TOTAL	3368	3962	100.0

[1]Data from Lake Champlain watershed was excluded from NHDPlus coverage.

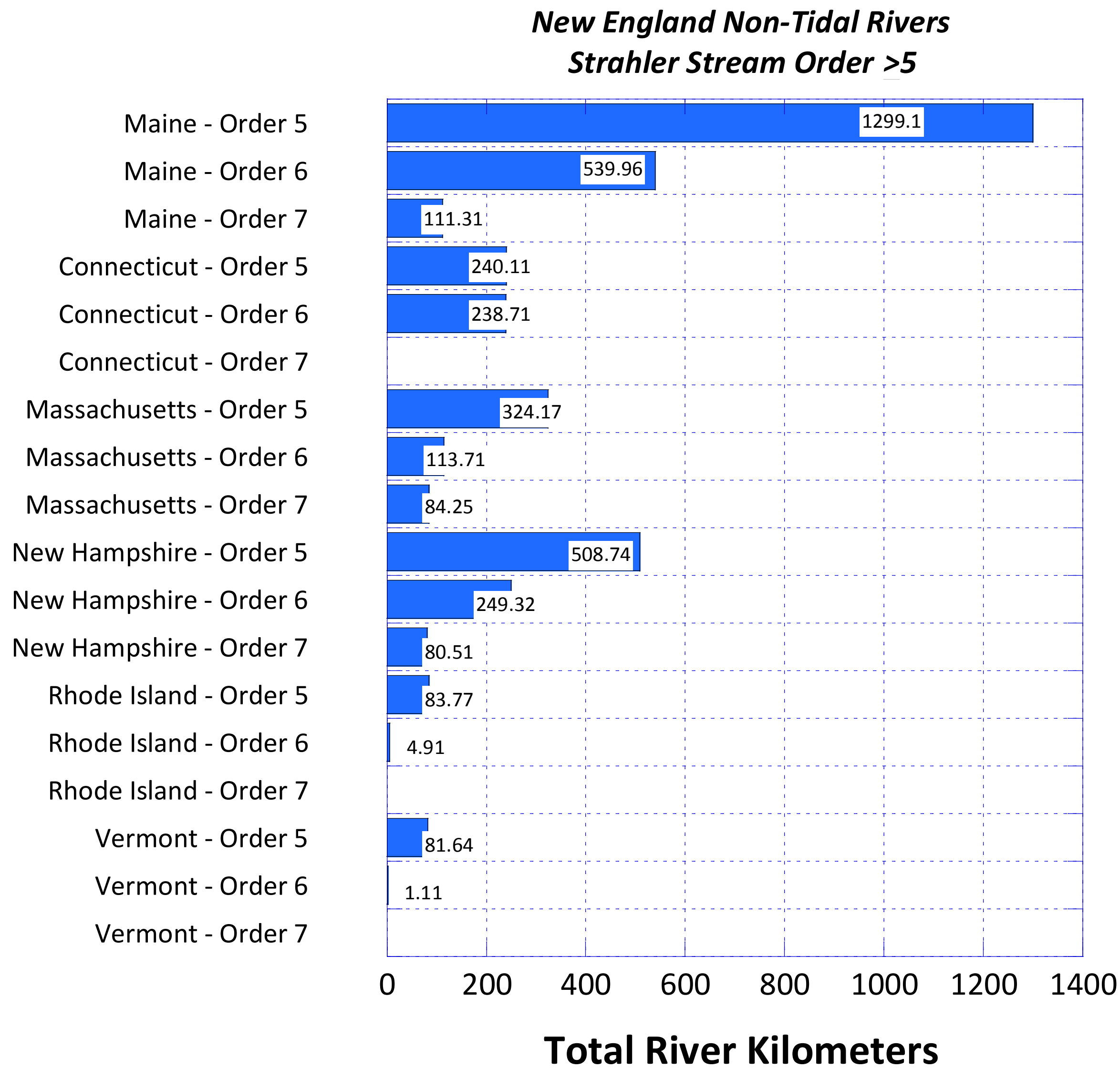


Figure 12. *Distribution of river reaches and distance by Strahler order ≥5 and by each New England state covered by the New England REMAP project. Data are from NHDPlus.*

The distribution of sites between the NELR REMAP probabilistic and targeted designs shows distinct differences by latitude in terms of the proportions for each dataset. We would expect a design to have a proportional representation similar to the proportion of the available reaches within each panel of latitude to be considered as being proportionately represented. On this basis, the NELR REMAP probabilistic data set is somewhat skewed by having a higher proportion of sites south of latitude 45°N where nearly 90% of the sites were located, but where 73% of the river miles were located. This is a partial reflection of the allocation of sites to each state by the NRSA design. Only 10.1% of the probabilistic sites (15 sites) were located in the 46-47°N panels where 21.8% of the rivers occurred. Nearly 80% of the targeted sites were located in the latitude 43-45°N panels where more than 64% of the river length occurred. Still, the latitudes >46°N were well represented in this data set having 11.6% of the sites (43) where 11% of the rivers occurred. In terms of the latitudinal availability of rivers of >5th order, the targeted design appeared to provide the most equitable coverage.

***Table 5**. The distribution of NELR REMAP probabilistic and targeted sites ≥5th order by latitude and the availability of all order ≥5 reaches in NHD Plus.*

	REMAP Probabilistic		Targeted		Strahler Order ≥5	
N Latitude	**Samples**	**Percent**	**Samples**	**Percent**	**Reaches**	**Km (%)**
>41 to ≤ 42	47	31.5	9	2.4	447	634 (15.3)
>42 to ≤ 43	42	28.2	26	7.0	521	630 (15.2)
>43 to ≤ 44	22	14.8	83	22.4	782	916 (22.2)
>44 to ≤ 45	23	15.4	142	38.3	541	845 (20.4)
>45 to ≤ 46	8	5.4	68	18.3	500	654 (15.8)
>46 to ≤ 47	0	0.0	24	6.5	187	227 (5.5)
>47	7	4.7	19	5.1	193	226 (5.5)
Totals	149	100.0	371	100.0	3,171	4,133

CHAPTER 3: ASSESSMENT OF THE CONDITION AND STRESSORS AFFECTING FISH ASSEMBLAGES IN NEW ENGLAND LARGE RIVERS

The condition of fish assemblages in the non-wadeable rivers in New England is less well known than that of the smaller, wadeable streams. Much of this is attributable to the difficulties in effectively sampling these waters and the lack of standardized approaches and assessment tools (e.g., multimetric indices) for this region of the U.S. The objectives of this chapter include:

1. Describing gradients of environmental variables and stressors in New England rivers.
2. Providing estimates of the biological condition in New England rivers as expressed by fish assemblages.
3. Exploring associations between fish assemblage measures and stressors including habitat, water quality, land use, barriers, thermal gradients, and measures of hydrologic alteration to identify the key limiting stressors to New England river fish assemblages.

We addressed each objective by conducting a number of different analyses on the NELR REMAP probabilistic and targeted databases both separately and combined.

Background

There is a very limited amount of systematic monitoring data on the extant riverine fish faunas in New England beyond the commonly managed sport (e.g., trout and salmon) and diadromous species. To more effectively assess and manage these rivers there is a need to document key stressor associations and their respective influences on fish assemblages. The riverine fish fauna of New England is naturally depauperate in terms of species richness because of the position in the landscape and the fact that most are cold or cool water, coastal drainages. It is therefore important to explore whether the native fish assemblages respond differently compared to other regions of North America and to a cadre of stressors that include introduced species, barriers to movement, hydrologic alterations, and thermal alterations.

We used the ME IBI (Table 6a; Yoder et al. 2008) as an overall measure of fish assemblage condition in New England large rivers. Although the ME IBI (ME IBI) was calibrated with the Maine rivers database it should be applicable *at a minimum* to rivers in mid to northern New England (i.e., >43-44 N latitude). We also explored and commented on its applicability in southern New England how it may eventually need to be refined for those latitudes. The guild definitions developed by Yoder et al. (2006b, 2008) were used and updated for the additional fish species that were encountered in the 2008-9 NELR REMAP study area (Table 7). A number of multivariate approaches were used to explore the variation in the fish assemblage data and to aid in identifying stressors that were most likely to be influencing the assemblages in New England large rivers.

***Table 6a**. The Maine non-wadeable rivers IBI metrics with calibrated scoring equations and manual scoring adjustment criteria. Proportional (%) metrics are based on numbers unless indicated otherwise (after Yoder et al. 2008).*

Metric	**Scoring Equation**	**Scoring Adjustments**	
		Score = 0	**Score = 10**
Native Species Richness	10 * (-0.2462 + (0.0828*numspec2)))	<3 sp.	≥15 sp.
Native Cyprinid Species (excluding fallfish)	(10 * (0.4457 + (0.0109*allcyp_ff) - (0.00005629 * (allcyp_ff^{2}))))	Eq[1]	Eq
Adult white & longnose sucker abundance (biomass)	(10 * (0.3667 + (0.008*ws_lns_pb) - (0.000023592 * (ws_lns_pb^{2}))))	0	≥128 kg/km
%Native Salmonids	(10 * (0.9537 + (0.00000000039*nat_salm) - (0.000078892 * (nat_salm^{2}))))	0	≥20%
%Benthic Insectivores	10 * (0.010966*benth_pc_n)	0	≥91.2%
%Black bass	10 - (10 * (-0.09684 + (0.5638*log10(black bass))))	Eq	0
%Fluvial Specialist/ Dependent	(10 * (0.2775 + (0.0073*fluv_pc_n)))	0%	Eq
%Macrohabitat Generalists	10 - (10 * (0.1017 + (0.0096*macro_gen)))	>90%	Eq
Temperate Stenothermic Species	(10 * (0.7154 + (0.4047*(log10(steno)))))	0 sp.	>5 sp.
Non-guarding Lithophilic Species	(10 * (0.2979 + (0.8975*log10(lith_ng))))	<1	>10
Non-indigenous Species	10 - (10 * (0.1063 + (0.3271*Non-indigenous_sp) - (0.029*(Non-indigenous_sp^{2}))))	≥5	0
%DELT Anomalies	10 - (10 * (0.8965 + (0.1074*log10(delta))))	Eq	0

[1] No scoring adjustments are necessary; scoring determined by equation (Eq) across entire metric scoring range of 0-10.

A set of four diadromous metrics (Table 6b) were developed in 2011 in order to better highlight and assess the role of diadromous fishes in rivers where they have historically had access. This is also in keeping with the attributes of the BCG (Figure 3) which includes diadromous species. The diadromous IBI (DIBI) is reported as a standalone value and is additive to the ME IBI in keeping with the BCG and to maintain the distinction between the "core" inland freshwater fish assemblage and the temporal influx of diadromous species that can vary seasonally. As such analyses and displays of the results are done as the ME IBI and the ME IBI + DIBI (see Figure 32a).

***Table 6b**. Diadromous IBI metrics intended to represent the diadromous component of a riverine fish assemblage in Maine and New England expressed as the Diadromous IBI (DIBI). These are additive to the ME IBI in the NELR REMAP analyses.*

Metric	Scoring Equation	Scoring Adjustments	
		Score = 0	Score = 10
Diadromous Species Richness	Score = 0.0318 + 0.227*(Diadromous Species Richness)	0	≥5 sp.
Number of American Eel	Score = 0.0689 + 0.2*(Log Eel Rel. No.) + 0.0616*(Log Eel Rel. No.)	0	≥389/Km
Number of Clupeidae	Score = 0.832*Log10(Rel. No. Clupeids)^ (0.269)	0	≥96/Km
Number of Diadromous Fish (all diadromous species)	Score = 0.0522 + 0.168*(Log(Diad Rel. No.) + 0.0644^(Log(Diad Rel. No.))	0	≥560/Km

Programming to calculate ME IBI and DIBI scores for individual electrofishing samples was accomplished based on the derivation and calibration of the ME IBI metrics by Yoder et al. (2008) and the later addition of the DIBI. The routines are stored in Maine ECOS where the entry, storage, retrieval, and calculation of metrics and indices are accomplished. The programming currently exists in FoxPro and outputs are exported in a variety of formats including Excel and Adobe Acrobat. Equations for each of the 12 ME IBI and 4 DIBI metrics were developed from continuous calibration plots and include adjustments (if necessary) at the upper and lower terminus of each plot to normalize each to metric scoring ranges from 0 to 10.

***Table 7**. Native, tolerance, habitat, foraging, and reproductive guild designations and other notes on the distribution and occurrence of 87 fish species documented or suspected to occur in New England non-wadeable rivers (including freshwater tidal). Sources for guild and metric assignments appear in the footnotes (scientific nomenclature adheres to Page et al. 2013).*

Species	Native Status[1]	Environmental Tolerance[2]	Target Fish Classification[3]	Common Habitat(s)[4]	Spatial Occurrence[5]	Thermal Guild[6]	Foraging Guild[7]	Reproductive Guild[8]	Habitat Guild[9]	Notes
Petromyzondidae										
Sea lamprey *(Petromyzon marinus)*	N	M	A	T1,R1	C	M	D	LN	B	Occurs primarily as ammocoetes.
Amer. brook lamprey (*Lethenteron appendix*)	N	I	[FS]	R1	S	M	D	LN	B,E	Blackstone R. (MA).
Lepisosteidae										
Longnose gar (*Lepisosteus osseus*)	-	P	[MG]	All	LC	E	P	P	W	Winooski R. only (native)
Amiidae										
Bowfin (*Amia calva*)	IC	P	[MG]	R2	S	E	O	VN	W	Winooski R. (native), Taunton R. (introduced)
Acipenseridae										
Shortnose sturgeon *(Acipenser brevirostrum)*	N	I	A	T1	C	M	I	NGL	W	Presumpscot R. only
Atlantic sturgeon *(Acipenser oxyrinchus)*	N	I	A	R1	C	M	I	NGL	W	Kennebec R., Presumpscot R.
Anguillidae										
American eel *(Anguilla rostrata)*	N	T	FD,C	All	C	M	C	na	W,B	Common in lower coastal rivers.
Clupeidae										
Blueback herring *(Alosa aestivalis)*	N	M	A	T1,T2	C	M	P	NGL	W	All y-o-y, no adults collected.

[1] After Halliwell (2005) for Maine and Hartel et al. (2002) for New England proper: N – native; E – exotic of inter-continental origin; IC – introduced of intracontinental origin; IS – introduced of interstate origin;; U – undetermined origin.
[2] I – highly intolerant; S – sensitive (moderately intolerant); M – intermediate; P – moderately tolerant; T – highly tolerant.; sources used include Ohio EPA (1987), Whittier and Hughes (1998), Halliwell et al. (1999), Langdon (2001)
[3] After Bain and Meixler (2000) : FS – fluvial specialist; FD – fluvial dependent; MG – macrohabitat generalist; A – anadromous; C – catadromous; [ ] - designations in brackets were not classified by Bain and Meixler (2000).
[4] R1 – high gradient riverine; R2 – low gradient riverine; I1 – impounded riverine; T1 – tidal riverine freshwater; T2 – tidal embayment brackish
[5] Spatial distribution within New England: C – primarily coastal rivers; S – primarily south of 46.000° latitude; N – primarily north of 45.500° latitude; W – west of longitude 71°W; LC – Lake Champlain drainage only; U – ubiquitous regional occurrence.
[6] After Hokanson (1977); S – temperate stenotherm; M – temperate mesotherm; E – temperate eurytherm.
[7] After Goldstein and Simon (1999); H – herbivore, D – detritivore, I – invertivore, BI – benthic insectivore, C – top carnivore, P – piscivore, G – generalist, O – omnivore, P – planktivore.
[8] After Ohio EPA (1987) and Hughes et al. (1998),; NGL – non-guarding lithophil [simple lithophil], LN – lithophilic nester, L – lithophil, V – vegetation, P – psammophil [sand-fine gravel], CN – cavity nester, VN – vegetation nester, PN – psammophil nester.
[9] After Hughes et al. (1998); W – water column, B – benthic, E – edge, H – hider, G – generalist.

***Table 7**. continued.*

Species	Native Status[1]	Environmental Tolerance[2]	Target Fish Classification[3]	Common Habitat(s)[4]	Spatial Occurrence[5]	Thermal Guild[6]	Foraging Guild[7]	Reproductive Guild[8]	Habitat Guild[9]	Notes
Alewife *(Alosa pseudoharengus)*	N	M	A	T1-R2	C	M	P	PS	W	Mostly y-o-y, few adults collected.
American shad *(Alosa sapidissima)*	N	M	A	R1,T1-2	C	M	P	PS	W	Mostly y-o-y, few adults collected.
Gizzard shad *(Dorosoma cepedianum)*	IC	T	[MG]	-	-	E	D	L	W	Collected in Kennebec R. in 2000.
Cyprinidae										
Lake chub *(Couesius plumbeus)*	N	I	[FD]	R1	N	S	BI	NGL	B	Cold-cool rivers
Common carp *(Cyprinus carpio)*	E	T	MG	T1-2	C	E	O	V	W	Lower Kennebec R., S. of 43ºN
Eastern silvery minnow *(Hybognathus regius)*	N	M	[FD]	R2	N	E	O	NGL	E	Winooski R., Kennebec R.
Common shiner *(Luxilis cornutus)*	N	M	FD	R1-T1	U	E	I	NGL	W	Ubiquitous across New England
Golden shiner *(Notemigonus crysoleucas)*	N,IS	T	MG	R2,I1	U	E	G	L	W	Common in low gradient rivers
Bridle shiner *(Notropis bifrenatus)*	N	I	MG	R2	S	E	I	L	W	Rare – 4 disjunct locations
Blacknose shiner *(Notropis heterolepis)*	N	M	MG	R1-2	N	E	I	L	W	Uncommon N. of 44ºN.
Spottail shiner *(Notropis hudsonius)*	N,IS	M	MG	T1,I1	S	E	I	L	W	Native to Connecticut R.; elsewhere introduced
Mimic shiner *(Notropis volucellus)*	IC	S	[FS]	R1	S	E	I	NGL	W	Winooski R.; upper Connecticut
Emerald shiner *(Notropis atherinoides)*	N	I	[MG]	R1	LC	E	I	NGL	W	Winooski R. only
Rosyface shiner *(Notropis rubellus)*	IC	I	[FS]	R1	W	M	I	NGL	W	Winooski R., White R., & West R.
Spotfin shiner *(Cyprinella spiloptera)*	-	M	[FD]	R1	LC	E	I	NGL	W	Winooski R. only
N. Redbelly dace *(Chrosomus eos)*	N	M	MG	R1-2	N	M	G	NGL	W	Uncommon – N. of 45ºN
Northern Pearl dace *(Margariscus nachtriebi)*	N	S	[FD]	R1-2	N	S	G	NGL	W	Uncommon – N. of 44ºN
Bluntnose minnow *(Pimephales promelas)*	IC	T	MG	R1-2,I1	S	E	G	CN	G	Winooski R. (native), Housatonic R. (introduced)

[1] After Halliwell (2005) for Maine and Hartel et al. (2002) for New England proper: N – native; E – exotic of inter-continental origin; IC – introduced of intracontinental origin; IS – introduced of interstate origin; U – undetermined origin.
[2] I – highly intolerant; S – sensitive (moderately intolerant); M – intermediate; P – moderately tolerant; T – highly tolerant.; sources used include Ohio EPA (1987), Whittier and Hughes (1998), Halliwell et al. (1999), Langdon (2001)
[3] After Bain and Meixler (2000) : FS – fluvial specialist; FD – fluvial dependent; MG – macrohabitat generalist; A – anadromous; [ ] - designations in brackets were not classified by Bain and Meixler (2000).
[4] R1 – high gradient riverine; R2 – low gradient riverine; I1 – impounded riverine; T1 – tidal riverine freshwater; T2 – tidal embayment brackish
[5] Spatial distribution within New England: C – primarily coastal rivers; S – primarily south of 46.000° latitude; N – primarily north of 45.500° latitude; W – west of longitude 71°W; LC – Lake Champlain drainage only; U – ubiquitous regional occurrence.
[6] After Hokanson (1977); S – temperate stenotherm; M – temperate mesotherm; E – temperate eurytherm.
[7] After Goldstein and Simon (1999); H – herbivore, D – detritivore, I – invertivore, BI – benthic insectivore, C – top carnivore, P – piscivore, G – generalist, O – omnivore, P – planktivore.
[8] After Ohio EPA (1987) and Hughes et al. (1998); NGL – non-guarding lithophil [simple lithophil], LN – lithophilic nester, L – lithophil, V – vegetation, P – psammophil [sand-fine gravel], CN – cavity nester, VN – vegetation nester, PN – psammophil nester.
[9] After Hughes et al. (1998); W – water column, B – benthic, E – edge, H – hider, G – generalist.

***Table 7**. continued.*

Species	Native Status[1]	Environmental Tolerance[2]	Target Fish Classification[3]	Common Habitat(s)[4]	Spatial Occurrence[5]	Thermal Guild[6]	Foraging Guild[7]	Reproductive Guild[8]	Habitat Guild[9]	Notes
Fathead minnow *(Pimephales promelas)*	N,IS	T	MG	R1	N	E	G	CN	W	Uncommon – N. of 44°N
Blacknose dace *(Rhinichthys atratulus)*	N	S	FS	R1	N	M	BI	NGL	B	Moderate to high gradient rivers
Longnose dace *(Rhinichthys cataractae)*	N	M	FS	R1	W	M	BI	NGL	B	Common W. of 70°W.
Rudd *(Scardinius erythrophthalmus)*	E	T	[MG]	-	-	E	G	L	W	Did not occur in NELR samples
Creek chub *(Semotilus atromaculatus)*	N	M	MG	R1	N	E	G	LN	W	Wide occurrence, but low numbers
Fallfish *(Semotilus corporalis)*	N	M	FS	R1-I1	U	E	G	LN	W	Ubiquitous across New England
Cutlip minnow *(Exoglossum maxillingua)*	N	M	[FS]	R1	S	M	I	LN	W	Housatonic R. only
Catostomidae										
Longnose sucker *(Catostomus catostomus)*	N	I	[FD]	R1	N	S	BI	NGL	B	Common N. of 46°N.
White sucker *(Catostomus commersonii)*	N	P	FD	R1-T2	U	M	I,D	NGL	W	Ubiquitous across New England
Eastern Creek chubsucker *(Erimyzon oblongus)*	N	I	[FD]	R2	S	W	G	NGL	W	Saco R., Wood R., Taunton R.
Silver redhorse *(Moxostoma anisurum)*	N	I	[FD]	R1-2	LC	S	I	NGL	B	Winooski R. only
Shorthead redhorse *(M. macrolepidotum)*	N	I	[FS]	R1	LC	S	BI	NGL	B	Winooski R. only
Ictaluridae										
White catfish *(Ameiurus catus)*	IC	T	MG	R1,T1-2	C	E	I,C	P	W	Common in selected coastal rivers
Yellow bullhead *(Ameiurus natalis)*	IC	T	MG	R1-2, I1	S	E	G	P,CN	W	Common S. of 43°N
Brown bullhead *(Ameirus nebulosus)*	N	T	MG	R2,I1	U	E	G	P,CN	W	Common across New England
Black bullhead *(Ameiurus melas)*	IC	T	MG	R2	S	E	G	P,CN	W	Rare – Lower Connecticut R., Taunton R.

[1] [1] After Halliwell (2005) for Maine and Hartel et al. (2002) for New England proper: N – native; E – exotic of inter-continental origin; IC – introduced of intracontinental origin; IS – introduced of interstate origin; U – undetermined origin.
[2] I – highly intolerant; S – sensitive (moderately intolerant); M – intermediate; P – moderately tolerant; T – highly tolerant.; sources used include Ohio EPA (1987), Whittier and Hughes (1998), Halliwell et al. (1999), Langdon (2001)
[3] After Bain and Meixler (2000) : FS – fluvial specialist; FD – fluvial dependent; MG – macrohabitat generalist; A – anadromous; [ ] - designations in brackets were not classified by Bain and Meixler (2000).
[4] R1 – high gradient riverine; R2 – low gradient riverine; I1 – impounded riverine; T1 – tidal riverine freshwater; T2 – tidal embayment brackish
[5] Spatial distribution within New England: C – primarily coastal rivers; S – primarily south of 46.000° latitude; N – primarily north of 45.500° latitude; W – west of longitude 71°W; LC – Lake Champlain drainage only; U – ubiquitous regional occurrence.
[6] After Hokanson (1977); S – temperate stenotherm; M – temperate mesotherm; E – temperate eurytherm.
[7] After Goldstein and Simon (1999); H – herbivore, D – detritivore, I – invertivore, BI – benthic insectivore, C – top carnivore, P – piscivore, G – generalist, O – omnivore, P – planktivore.
[8] After Ohio EPA (1987) and Hughes et al. (1998); NGL – non-guarding lithophil [simple lithophil], LN – lithophilic nester, L – lithophil, V – vegetation, P – psammophil [sand-fine gravel], CN – cavity nester, VN – vegetation nester, PN – psammophil nester.
[9] After Hughes et al. (1998); W – water column, B – benthic, E – edge, H – hider, G – generalist.

Table 7. continued.

Species	Native Status[1]	Environmental Tolerance[2]	Target Fish Classification[3]	Common Habitat(s)[4]	Spatial Occurrence[5]	Thermal Guild[6]	Foraging Guild[7]	Reproductive Guild[8]	Habitat Guild[9]	Notes
Tadpole madtom *(Noturus gyrinus)*	IC	M	[MG]	R2-I1	S	E	I,O	CN	W	Merrimack R. only
Margined madtom *(Noturus insignis)*	IC	M	[FS]	R1	S	M	I	NGL	B	Merrimack R. only
Esocidae										
Muskellunge *(Esox masquinongy)*	IC	I	MG	R1-2	N	M	P	L	H	St. John R., Housatonic R.
Northern pike *(Esox lucius)*	IC	M	MG	I1	S	M	P	L	H	Common in Connecticut R. basin
Chain pickerel *(Esox niger)*	N,IS	P	MG	I1,R2	S	M	P	L	H	Low gradient & impounded S. of 46°N
Redfin pickerel *(Esox americanus)*	N	N	[MG[	R2	S	M	P	P	W,H	Common S. of 42°N
Percopsidae										
Trout-perch *(Percopsis omiscomaycus)*	N	M	[MG]	R1-2	LC	M	I	P	B	Winooski R. only
Umbridae										
Central mudminnow *(Umbra limi)*	N	T	[MG]	R1	S	E	I	VN	H	Lower Connecticut R., St. John R.
Osmeridae										
Rainbow smelt *(Osmerus mordax)*	N	M	A	T2	C	M	I,C	L	W	Isolated locations in Maine
Salmonidae										
Rainbow trout *(Oncorhynchus mykiss)*	IC	I	FD	R1	W	S	C	LN	W	S. of 45°N; widely stocked

[1] [1] After Halliwell (2005) for Maine and Hartel et al. (2002) for New England proper: N – native; E – exotic of inter-continental origin; IC – introduced of intracontinental origin; IS – introduced of interstate origin; U – undetermined origin.
[2] I – highly intolerant; S – sensitive (moderately intolerant); M – intermediate; P – moderately tolerant; T – highly tolerant.; sources used include Ohio EPA (1987), Whittier and Hughes (1998), Halliwell et al. (1999), Langdon (2001)
[3] After Bain and Meixler (2000) : FS – fluvial specialist; FD – fluvial dependent; MG – macrohabitat generalist; A – anadromous; [ ] - designations in brackets were not classified by Bain and Meixler (2000).
[4] R1 – high gradient riverine; R2 – low gradient riverine; I1 – impounded riverine; T1 – tidal riverine freshwater; T2 – tidal embayment brackish
[5] Spatial distribution within New England: C – primarily coastal rivers; S – primarily south of 46.000° latitude; N – primarily north of 45.500° latitude; W – west of longitude 71°W; LC – Lake Champlain drainage only; U – ubiquitous regional occurrence.
[6] After Hokanson (1977); S – temperate stenotherm; M – temperate mesotherm; E – temperate eurytherm.
[7] After Goldstein and Simon (1999); H – herbivore, D – detritivore, I – invertivore, BI – benthic insectivore, C – top carnivore, P – piscivore, G – generalist, O – omnivore, P – planktivore.
[8] After Ohio EPA (1987) and Hughes et al. (1998); NGL – non-guarding lithophil [simple lithophil], LN – lithophilic nester, L – lithophil, V – vegetation, P – psammophil [sand-fine gravel], CN – cavity nester, VN – vegetation nester, PN – psammophil nester.
[9] After Hughes et al. (1998); W – water column, B – benthic, E – edge, H – hider, G – generalist.

Table 7. *continued.*

Species	Native Status[1]	Environmental Tolerance[2]	Target Fish Classification[3]	Common Habitat(s)[4]	Spatial Occurrence[5]	Thermal Guild[6]	Foraging Guild[7]	Reproductive Guild[8]	Habitat Guild[9]	Notes
Atlantic salmon *(Salmo salar)*	N	I	A	R1	C	S	C	LN	W	Sea run - limited occurrence
Atlantic salmon *(Salmo salar)*	N,IS	I	[FD]	R1	N	S	C	LN	W	Hatchery origin – stocked in interior rivers
Brown trout *(Salmo trutta)*	E	I	FD	R1	W	S	C	LN	W	Present where stocked
Brook trout *(Salvelinus fontinalis)*	N	I	FS	FS	N	S	C	LN	W	Cold rivers lacking blackbasses
Round whitefish *(Prosopium cylindraceum)*	N	I	[FD]	R1	N	S	C	L	W	Cold rivers N. of 45°N
Gadidae										
Burbot *(Lota lota)*	N	S	[FD]	R1	N	S	C	NGL	B	Cold-cool rivers N. of 44°N
Atlantic tomcod *(Microgadus tomcod)*	N	P	[TS]	T1-2	C	E	G	P	G	Presumpscot R., Pawcatuk R.
Fundulidae										
Banded killifish *(Fundulus diaphanus)*	N	M	MG	R1-T2	U	E	I	VN	E	Common in Kennebec R.
Mummichog *(Fundulus heteroclitus)*	N	T	[TS]	T1-2	C	E	D	VN	E	Tidal habitats only
Atherinopsidae										
Inland silverside *(Menidia beryllina)*	N	M	[TS]	T2	C	E	P	V	E	Rare – tidal coastal rivers
Gasterosteidae										
Brook stickleback *(Culea inconstans)*	IC	I	[MG]	I1	N	M	P,I	VCN	H	Sporadic occurrence
Fourspine stickleback *(Apeltes quadracus)*	N	M	[TS]	T1-2	C	M	P	VCN	E	Common in Lower Kennebec R.

[1] [1] After Halliwell (2005) for Maine and Hartel et al. (2002) for New England proper: N – native; E – exotic of inter-continental origin; IC – introduced of intracontinental origin; IS – introduced of interstate origin; U – undetermined origin.
[2] I – highly intolerant; S – sensitive (moderately intolerant); M – intermediate; P – moderately tolerant; T – highly tolerant.; sources used include Ohio EPA (1987), Whittier and Hughes (1998), Halliwell et al. (1999), Langdon (2001)
[3] After Bain and Meixler (2000) : FS – fluvial specialist; FD – fluvial dependent; MG – macrohabitat generalist; A – anadromous; [ ] - designations in brackets were not classified by Bain and Meixler (2000).
[4] R1 – high gradient riverine; R2 – low gradient riverine; I1 – impounded riverine; T1 – tidal riverine freshwater; T2 – tidal embayment brackish
[5] Spatial distribution within New England: C – primarily coastal rivers; S – primarily south of 46.000° latitude; N – primarily north of 45.500° latitude; W – west of longitude 71°W; LC – Lake Champlain drainage only; U – ubiquitous regional occurrence.
[6] After Hokanson (1977); S – temperate stenotherm; M – temperate mesotherm; E – temperate eurytherm.
[7] After Goldstein and Simon (1999); H – herbivore, D – detritivore, I – invertivore, BI – benthic insectivore, C – top carnivore, P – piscivore, G – generalist, O – omnivore, P – planktivore.
[8] After Ohio EPA (1987) and Hughes et al. (1998); NGL – non-guarding lithophil [simple lithophil], LN – lithophilic nester, L – lithophil, V – vegetation, P – psammophil [sand-fine gravel], CN – cavity nester, VN – vegetation nester, PN – psammophil nester.
[9] After Hughes et al. (1998); W – water column, B – benthic, E – edge, H – hider, G – generalist.

***Table 7**. continued.*

Species	Native Status[1]	Environmental Tolerance[2]	Target Fish Classification[3]	Common Habitat(s)[4]	Spatial Occurrence[5]	Thermal Guild[6]	Foraging Guild[7]	Reproductive Guild[8]	Habitat Guild[9]	Notes
3-spine stickleback *(Gasterosteus aculeatus)*	N	M	[MG]	R1	N	M	I	PN	E	N. of 46°N; absent Blackbasses
Ninespine stickleback *(Pungitius pungitius)*	N	M	[MG]	R1,T2	N,C	M	P	VCN	E	N. of 46°N; absent Blackbasses
Cottidae										
Slimy sculpin *(Cottus cognatus)*	N	I	FS	R1	N	S	BI	NGL	B	Cold-cool rivers; N of 44°N
Moronidae										
White perch *(Morone americana)*	N,IS	M	MG	I1,T1-2	C	M	C	L	W	Lower coastal rivers, impoundments
Striped bass *(Morone saxatilis)*	N	I	A	R1,T1-2	C	M	P	L	W	Lower coastal rivers
Centrarchidae										
Rock bass *(Ambloplites rupestris)*	IC	M	MG	I1	W	E	C	LN	W	Common W. of 71°W
Banded sunfish *(Enneacanthus obesus)*	N	M	MG	R2	W	E	I	VN	H	Millers R. only
Redbreast sunfish *(Lepomis auritus)*	N	M	MG	R1-T1	U	E	I	PN	W	Common in lower coastal rivers
Green sunfish *(Lepomis cyanellus)*	IC	T	MG	na	S	E	I	PN	W	Scattered locations CT, MA, ME
Pumpkinseed *(Lepomis gibbosus)*	N	T	MG	R2-T1	U	E	I	VN	W	Common S. of 46°N, low gradient
Bluegill *(Lepomis macrochirus)*	IC	T	MG	R1-2,I1,T1	W	E	I	VN	W	Common S. of 45°N, W of 70°W
Smallmouth bass *(Micropterus dolomieu)*	IC	M	MG	R1-T1	U	E	C	LN	W	Common within sphere of introduction
Largemouth bass *(Micropterus salmoides)*	IC	P	MG	R2-T1	U	E	C	PN	W	Low gradient and impounded rivers
White crappie *(Pomoxis annularis)*	IC	P	MG	I1	W	E	I	VN	W	Isolated occurrence

[1] [1] After Halliwell (2005) for Maine and Hartel et al. (2002) for New England proper: N – native; E – exotic of inter-continental origin; IC – introduced of intracontinental origin; IS – introduced of interstate origin; U – undetermined origin.
[2] I – highly intolerant; S – sensitive (moderately intolerant); M – intermediate; P – moderately tolerant; T – highly tolerant.; sources used include Ohio EPA (1987), Whittier and Hughes (1998), Halliwell et al. (1999), Langdon (2001)
[3] After Bain and Meixler (2000) : FS – fluvial specialist; FD – fluvial dependent; MG – macrohabitat generalist; A – anadromous; [ ] - designations in brackets were not classified by Bain and Meixler (2000).
[4] R1 – high gradient riverine; R2 – low gradient riverine; I1 – impounded riverine; T1 – tidal riverine freshwater; T2 – tidal embayment brackish
[5] Spatial distribution within New England: C – primarily coastal rivers; S – primarily south of 46.000° latitude; N – primarily north of 45.500° latitude; W – west of longitude 71°W; LC – Lake Champlain drainage only; U – ubiquitous regional occurrence.
[6] After Hokanson (1977); S – temperate stenotherm; M – temperate mesotherm; E – temperate eurytherm.
[7] After Goldstein and Simon (1999); H – herbivore, D – detritivore, I – invertivore, BI – benthic insectivore, C – top carnivore, P – piscivore, G – generalist, O – omnivore, P – planktivore.
[8] After Ohio EPA (1987) and Hughes et al. (1998); NGL – non-guarding lithophil [simple lithophil], LN – lithophilic nester, L – lithophil, V – vegetation, P – psammophil [sand-fine gravel], CN – cavity nester, VN – vegetation nester, PN – psammophil nester.
[9] After Hughes et al. (1998); W – water column, B – benthic, E – edge, H – hider, G – generalist.

***Table 7**. continued.*

Species	Native Status[1]	Environmental Tolerance[2]	Target Fish Classification[3]	Common Habitat(s)[4]	Spatial Occurrence[5]	Thermal Guild[6]	Foraging Guild[7]	Reproductive Guild[8]	Habitat Guild[9]	Notes
Black crappie *(Pomoxis nigromaculatus)*	IC	P	MG	R1,I1	S	E	I	VN	W	Sporadic occurrence
Percidae										
Walleye *(Sander vitreum)*	IC	S	MG	I1	W	M	P	P	W	Quinebaug River only
Yellow perch *(Perca flavescens)*	N,IS	P	MG	I1,T1-2	U	M	C	V	W	Commonly occurring in New England
Logperch *(Percina caprodes)*	N	P	[FD]	R1	LC	M	I	NGL	B	Winsooski R. only
Tessellated darter *(Etheostoma olmstedi)*	N	I	[FS]	R1	N	M	I	NGL	B	Connecticut R. basin
Eastern sand darter *(Ammocrypta pellucida)*	N	I	[FS]	R1	LC	M	I	NGL	B	Winsooski R. only
Fantail darter *(Etheostoma flabellare)*	N	I	[FS]	R1	LC	S	I	NGL	B	Missisquoi R. only
Pomatomidae										
Bluefish *(Pomatomus saltatrix)*	N	M	[TS]	T1-2	C	E	P	P	W	Tidal Connecticut R. only
Gobidae										
Naked goby *(Gobiosoma bosc)*	N	M	[TS]	T1-2	C	E	I	P	B	Taunton R. only
Achiridae										
Hogchoker *(Trinectes maculatus)*	N	M	[TS]	T1-2	C	M	I	P	B	Taunton R., tidal Connecticut R.

[1] [1] After Halliwell (2005) for Maine and Hartel et al. (2002) for New England proper: N – native; E – exotic of inter-continental origin; IC – introduced of intracontinental origin; IS – introduced of interstate origin; U – undetermined origin.
[2] I – highly intolerant; S – sensitive (moderately intolerant); M – intermediate; P – moderately tolerant; T – highly tolerant.; sources used include Ohio EPA (1987), Whittier and Hughes (1998), Halliwell et al. (1999), Langdon (2001)
[3] After Bain and Meixler (2000) : FS – fluvial specialist; FD – fluvial dependent; MG – macrohabitat generalist; A – anadromous; [ ] - designations in brackets were not classified by Bain and Meixler (2000).
[4] R1 – high gradient riverine; R2 – low gradient riverine; I1 – impounded riverine; T1 – tidal riverine freshwater; T2 – tidal embayment brackish
[5] Spatial distribution within New England: C – primarily coastal rivers; S – primarily south of 46.000° latitude; N – primarily north of 45.500° latitude; W – west of longitude 71°W; LC – Lake Champlain drainage only; U – ubiquitous regional occurrence.
[6] After Hokanson (1977); S – temperate stenotherm; M – temperate mesotherm; E – temperate eurytherm.
[7] After Goldstein and Simon (1999); H – herbivore, D – detritivore, I – invertivore, BI – benthic insectivore, C – top carnivore, P – piscivore, G – generalist, O – omnivore, P – planktivore.
[8] After Ohio EPA (1987) and Hughes et al. (1998); NGL – non-guarding lithophil [simple lithophil], LN – lithophilic nester, L – lithophil, V – vegetation, P – psammophil [sand-fine gravel], CN – cavity nester, VN – vegetation nester, PN – psammophil nester.
[9] After Hughes et al. (1998); W – water column, B – benthic, E – edge, H – hider, G – generalist.

The analyses also included making comparisons between the probabilistic and targeted designs for determining fish assemblage condition estimates and in identifying various stressors.

Analytical Methods

Data Sets

Fish assemblage data produced by this project included the 2008-9 NELR REMAP probability sites, targeted sites sampled by NELR REMAP in 2008-9, the Maine rivers assessment of 2002-2007, and the Connecticut River mainstem assessment of 2008-9 (Table 8). These data are directly comparable since all were collected using the same methods as described in Chapter 2.

***Table 8**. List of sites and samples by state and survey design that were used in the assessment of fish assemblage condition and the stressor analyses.*

State	NRSA Base	NRSA Overdraw[1]	Targeted Samples
Connecticut	13	8	4
Maine	18	13	288
Massachusetts	21	15	23
New Hampshire	13	12	46
Rhode Island	14	10	0
Vermont	13	9	0
Totals	92	57	371

[1] NRSA base + overdraw samples comprised the NELR REMAP probabilistic sample.

Stressor Analyses

Our approach to establishing linkages between various stressors and fish assemblages was to first examine the range and distribution of key environmental variables across New England large rivers. A number of different (and readily available) data sets were accessed to estimate stressor levels or sources of potential stressor types. We used the Qualitative Habitat Evaluation Index (QHEI) originally developed for Midwestern streams and rivers and as modified for application to Maine rivers (Yoder et al. 2006a) as our primary measure of habitat quality at the site-specific level. The QHEI consists of multiple attributes of riverine habitat and we used these attributes to estimate the number of habitat "niches" available at a given site. In addition to the typical attributes of structural habitat, the QHEI also includes attributes that are flow dependent and these were extracted as a measure of hydrological habitat related to current or depth (i.e., Hydro-QHEI) as previously described and used by Rankin et al. (2011b) to describe the ecological flow requirements of fish. The series of field parameters collected during fish sampling including dissolved oxygen and relative conductivity was also used. Temperature grab samples were not used as field measurements are sensitive to locational bias and pH was inconsistently available due to probe reliability issues. We attempted to obtain other chemical water quality data through EPA's WQX, but there were too few matches with our sampling sites and within the same seasonal index period to attempt using the data in the analyses.

GIS Data Sets

We accessed the following GIS coverages as part of the stressor data sets:

1. Ecological Connectivity – Northeast Aquatic Connectivity (NAC) report which provided information and metrics about dams, anadromous fish habitat, and other parameters (Table 9).
2. Temperature Classification - the Northeast Aquatic Habitat Classification (NEAHCS) system results were used (Olivero and Anderson 2008).
3. Land Cover - consisted of land cover types in the total catchment upstream of a site and locally nearer a site.
4. Nutrient Enrichment - Sparrow predicted nutrient concentrations for total phosphorus and nitrogen.

Table 9*. Eight metrics from the NAC dataset (Martin and Apse 2011) accessed for use in the stressor analyses.*

Metric	Description
ConnStatus	Overall connectivity status metric
Ust_Barr Dst_Barr	Number of upstream and downstream barriers
Dst_Impass	Downstream impassable dam count
Ust_Dens	Upstream dam density
Dst_Dens	Downstream dam density
RMS_Mouth	Distance (mi) to River Mouth
PresAnad AnadHist	Presence of anadromous fish (current & historical)
AnadCount	Number of anadromous species downstream
Dst_Hydro	Number of Hydro Dams on Downstream Flowpath

Ecological Connectivity

We accessed the Northeast Aquatic Connectivity (NAC) databases that contain metrics related to the presence and type of dams and barriers in rivers in the Northeast (Martin and Apse 2011). This effort derived more than 70 metrics related to ecological connectivity and we selected eight that we thought were potentially relevant to fish assemblages (Table 10) which included ones weighted most heavily

***Table 10**. Northeast Aquatic Habitat Classification System (NEAHCS) categories and definitions.*

STREAM/RIVER SIZE		
NEAHCS Size Class	**Description**	**Units (miles^2)**
1a	Headwaters	< 3.861
1b	Creeks	≥3.861 – <38.61
2	Small Rivers	≥38.61 - <200
3a	Medium Tributary Rivers	≥200 - <1000
3b	Medium Mainstem Rivers	≥1000 - <3861
4	Large Rivers	≥3861 - <9653
5	Great Rivers	≥9653
GRADIENT		
NEAHCS Gradient Class	**Description**	**Slope**
1	Very Low Gradient	< 0.02%
2	Low Gradient	≥0.02 - < 0.1%
3	Moderate – Low Gradient	≥0.1 - < 0.5%
4	Moderate – High Gradient	≥0.5 - < 2%
5	High Gradient	≥0.2 - < 5%
6	Very High Gradient	>5%
GEOLOGICAL BUFFERING		
NEAHCS Buffering Class	**Description**	**Norton Index**
1	Acidic, Low Buffered	100-174
2	Neutral, Moderately Buffered	175-324
3	Calcareous-Neutral, Highly Buffered	325-400
0	Size 3, 4, 5 rivers, Assume Neutral	Any
WATER TEMPERATURE CLASSIFICATION		
NEAHCS Temperature Class	**Description**	**Species Characteristics**
1	Cold	Coldwater species >50%
2	Transitional Cool	Increased %cool & warmwater species relative to cold
3	Transitional Warm	Increased dominance warmwater species relative to cool & cold
4	Warm	Warmwater species >75%

to anadromous fish. We linked these metrics to NHDPlus reaches. The dam statistics were assigned to each reach where a fish and habitat sampling site was located and we likewise assigned all variables (e.g., impassable dam count) to upstream sites until another dam intervened. We also classified whether a NELR REMAP site was free-flowing or impounded based on the QHEI.

NEAHCS Aquatic Habitat Classification
The Northeast Aquatic Habitat Classification System (NEAHCS; Olivero and Anderson 2008) categories were used for river reaches using the NHDPlus reaches and the NEAHCS classification dataset[65]. The NEAHCS focuses on four key and readily obtainable habitat categories including stream and river size, gradient, geological buffering, and temperature classification (Table 10). We used drainage area (mi.^{2}) values generated for the QHEI instead of the NEAHCS size classes because it was a continuous variable and we did not use the buffering classes because large rivers were assumed to be neutral in their buffering capacity.

Land Cover
The modified SPARROW (Spatially Referenced Regressions on Watershed Attributes) National Hydrography Dataset (NHD) based on a 1:100,000 scale provided land use data. It is comprised of 42,000 reaches (Moore et al. 2004) to associate National Land Cover Data (NLCD) from 1992 that were matched to the NELR REMAP fish sampling sites. This data also included mean annual flow estimates, temperature, and rainfall data. Land cover data was estimated for each reach for the immediate catchment ("local") within which a sampling site was located and for the entire watershed upstream of that catchment ("cumulative"). Other variables extracted from the NHDPlus included mean slope (%), mean precipitation and air temperature from PRISM, mean flow (annual), and a base flow index. The mean slope is based on the minimum and maximum elevation for a given flow line segment and is unitless (meter/meter; USGS 2007). Mean annual precipitation and air temperature were for a catchment within which a reach is located (USGS 2007) and was calculated as described by Daly and Taylor (1988).

Nutrient Enrichment
Outputs of the SPARROW model were used to obtain estimates of mean annual total phosphorus and total nitrogen at each REMAP site. This served as a surrogate for not having water quality data for N and P.

Statistical Analyses
We used cumulative frequency distribution graphs, histograms, and maps to explore the distribution and variation of stressors and riverine fish assemblage measures across New

[65] http://rcngrants.org/content/northeastern-aquatic-habitat-classification-project

England. We calculated selected quantiles with 95% confidence intervals (normal based, XLSTAT Version 2012.5.01, Addinsoft Software) for key stressors and the ME IBI. Non-metric multidimensional scaling (NMS), cluster analysis, and indicator species analysis (ISA) was used to explore fish species composition in response to natural and anthropogenic variables. Bray–Curtis dissimilarity was used as the distance measure for all analyses, a commonly accepted approach for ecological data with consistent relative abundance properties. For NMS ordinations and cluster analyses we removed species that occurred at <5% of the sampling sites to reduce environmental noise McCune and Grace (2002). All fish abundance data was log transformed prior to running this analysis. River samples that were near one another in the ordination plots had more similar species composition and abundance than sample points that were more distant. NMS is considered well suited for ecological assemblage data because it has fewer data assumptions and analytical problems compared to other ordination methods (McCune and Grace 2002). Ordination sample points were coded by key environmental variables and differences in site groupings were interpreted visually to identify important environmental variables for New England large rivers.

A stressor variable reduction process was conducted, using principal component analysis and correlation analysis to select a reduced set of environmental and stressor variables. It was included in a Canonical Correspondence Analyses (CCA) to identify the strength and direction of significant stressor relationships and to create an overall Human Disturbance Index (HDI). We generally followed the methods of Wang et al. (2008) with some necessary simplifications. We plotted selected stressors vs. the number of intolerant fish species to identify threshold values, by eye, in each stressor-response relationship. We then standardized each stressor on a 1-10 scale and then weighed each stressor score by the average of the coefficients from the CCA analyses (1st and 2nd axes). The metric scores, now weighted by CCA coefficient, were then summed and standardized on a 0-100 scale.

Stressor-specific species sensitivity and tolerance metrics (richness and proportion metrics) were derived by:

- calculating weighted stressor values (WSVs) for each stressor and environmental variable;
- ranking weighted stressor values by species for each stressor;
- selecting the upper and lower 15 percent of species as sensitive and tolerant species, respectively;
- calculating positive and negative species richness and proportional metrics for each stressor; and,
- plotting these metrics against the original stressor variables and the ME IBI.

The usefulness and strength of each variable was assessed using linear regression coefficients and by visually examining plots for threshold responses and classifying them as strong, moderate, or weak. These metrics can now be used in the stressor identification process and later to explore making changes to the ME IBI that might make it more responsive to key stressors in New England large rivers.

We also examined the weighted mean stressor value (WSVs) for each stressor and species by plotting the median stressor value vs. the WSV, coded by the applicable metric (e.g., fluvial specialist or tolerance assignment) to illustrate the location of each species in relation to each stressor. These plots can be used to examine, using ambient data, the consequences of including or excluding species from these metrics. For example, did fish species designated as fluvial specialists respond accordingly to independent, ambient measures of flow habitat (e.g., Hydro QHEI, QHEI riffle metrics, etc.)?

Results

Environmental Variables in New England Large Rivers

Environmental variables in New England large rivers were compared to results from the NELR REMAP probabilistic and targeted sites. The targeted coverage represents a spatially more dense distribution of sites within selected rivers than do the probabilistic sites and the potential biases and consequences of were highlighted as they arose. The results for each of 11 stressor variables that comprised a combination of measured values and modeled outputs are depicted in Table 11.

Physical Habitat

The QHEI score represents a type of physical habitat indicator that has not been widely used in New England (Figure 13). It is responsive to anthropogenic changes to

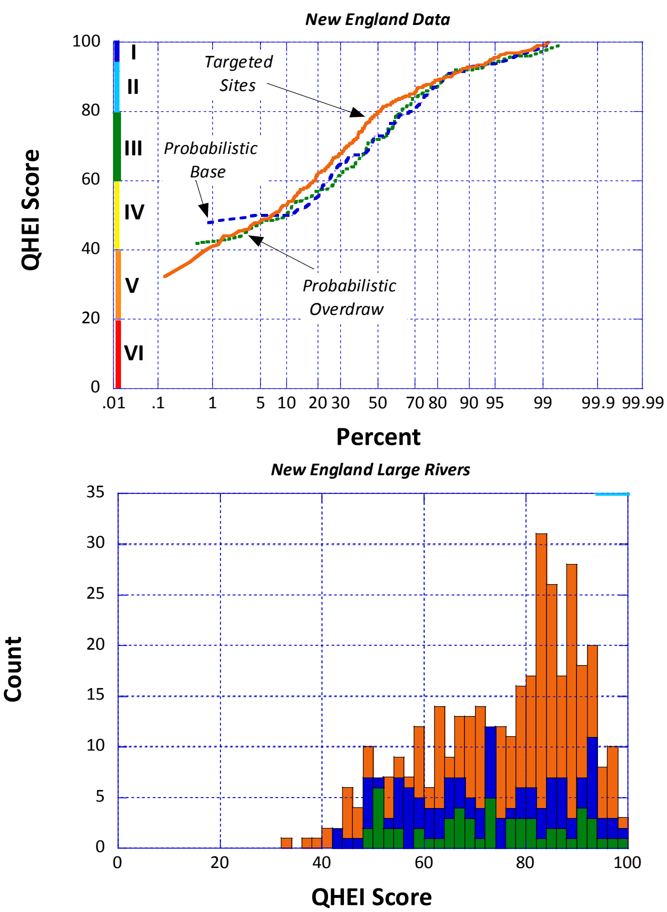


***Figure 13**. Cumulative frequency distribution plot (top) of targeted sites (orange), base probabilistic sites (green) and overdraw probabilistic sites (blue) and frequency histogram (bottom) of QHEI score, separately for targeted sites (orange) and probabilistic sites (stacked and colored coded by base vs. overdraw).*

***Table 11**. Frequency statistics for 11 measured and modeled variables from probabilistic sample groups (NRSA base and overdraw) and targeted samples for non-wadeable New England rivers (95% confidence intervals for each statistic appear in parentheses).*

Variable/Sample Type	5th	25th	Median	75th	95th
QHEI					
Probabilistic	48.5 (43.0-50.0)	59.5 (55.5-64.0)	72.0 (68.0-75.0)	85.0 (80.8-88.0)	94.0 (92.5-96.0)
Targeted	48.0 (45.5-50.3)	64.8 (62.0-67.5)	80.0 (77.0-82.0)	88.0 (86.0-89.0)	95.5 (93.5-97.0)
QHEI Riffle/Run Score					
Probabilistic	0.00 (0.0-0.0)	0.00 (0.0-0.0)	4.0 (2.0-5.0)	7.0 (6.0-8.0)	8.0 (8.0-8.0)
Targeted	0.00 (0.0-0.0)	0.00 (0.0-0.0)	7.0 (7.0-8.0)	8.0 8.0-8.0)	8.0 (8.0-8.0)
Dissolved Oxygen (mg/l)					
Probabilistic	7.40 (6.1-7.6)	8.30 (8.11-8.53)	8.88 (8.70-9.10)	9.65 (9.43-9.88)	11.08 (10.76-11.97)
Targeted	7.10 (6.65-7.20)	8.0 (7.83-8.13)	8.60 (8.50-8.70-)	9.20 (9.00-9.35)	10.70 (10.3-11.16)
Conductivity (µS/cm)					
Probabilistic	28.0 (24.0-37.0)	68.0 (56.0-91.0)	119.0 (102.0-142.0)	202.0 (169.0-231.0)	339.0 (314.0-509.0)
Targeted	28.0 (24.0-29.0)	42.7 (39.7-46.0)	63.0 (58.5-68.0)	113.0 (103.0-130.0)	563.5 (321.0-602.0)
Mean Annual Air Temperature					
Probabilistic	3.2 (3.0-4.1)	6.7 (5.9-7.0)	8.5 (7.6-8.9)	9.5 (9.3-9.8)	10.0 (10.0-10.1)
Targeted	3.2 (3.0-4.1)	6.7 (5.9-7.0)	8.5 (7.6-8.9)	9.5 (9.3-9.8)	10.0 (10.0-10.1)
SPARROW Mean Annual TP Concentration (mg/l)					
Probabilistic	0.016 (0.0150.020)	0.027 (0.024-0.037)	0.046 (0.042-0.047)	0.065 (0.059-0.086)	0.296 (0.153-0.766)
Targeted	0.011 (0.0080.013)	0.020 (0.019-0.022)	0.028 (0.026-0.030)	0.044 (0.041-0.045)	0.067 (0.063-0.113)
SPARROW Mean Annual TN Concentration (mg/l)					
Probabilistic	0.016 (0.0150.020)	0.027 (0.023-0.034)	0.045 (0.040-0.047)	0.065 (0.059-0.085)	0.295 (0.153-0.766)
Targeted	0.011 (0.0080.013)	0.020 (0.019-0.022)	0.028 (0.026-0.030)	0.044 (0.041-0.045)	0.067 (0.063-0.113)

***Table 11**. Frequency statistics for 11 measured and modeled variables from probabilistic sample groups (NRSA base and overdraw) and targeted samples for non-wadeable New England rivers (95% confidence intervals for each statistic appear in parentheses).*

Variable/Sample Type	5th	25th	Median	75th	95th
Number of Impassable Barriers					
Probabilistic	0.00 (0.0-0.0)	0.00 (0.0-0.0)	0.0 (0.0-0.0)	2.0 (1.0-3.0)	6.0 (5.0-7.0)
Targeted	0.00 (0.0-0.0)	0.00 (0.0-0.0)	0.0 (0.0-0.0)	2.0 (1.0-3.0)	12.0 (6.0-17.0)
Number of Barriers					
Probabilistic	0.00 (0.0-0.0)	0.00 (0.0-0.0)	2.0 (2.0-3.0)	3.0 (3.0-5.0)	10.0 (7.0-16.0)
Targeted	0.00 (0.0-0.0)	1.00 (0.0-2.0)	3.0 (3.0-3.0)	6.0 5.0-7.0)	17.0 (14.0-21.0)
Cumulative Natural Land Cover					
Probabilistic	71.8 (59.1-73.6)	81.6 (79.3-83.3)	88.2 (86.6-88.7)	92.6 (90.7-93.2)	96.8 (94.3-97.9)
Targeted	74.3 (58.8-83.3)	90.0 (89.3-90.4)	93.0 (92.7-93.6)	95.7 95.3-96.3)	98.4 (98.3-98.7)
Local Natural Land Cover					
Probabilistic	23.4 (13.4-25.5)	49.7 (38.8-55.5)	66.3 (61.7-72.1)	84.4 (82.5-88.4)	97.8 (94.8-99.6)
Targeted	26.8 (24.4-35.2)	60.6 (54.5-63.7)	77.9 (75.8-81.0)	92.6 90.5-95.0)	99.8 (99.7-100)

physical habitat caused by channelization, impoundment, hydrological alterations, or watershed scale modifications to habitat and sediment dynamics in rivers. The median QHEI score at the probabilistic sites was slightly lower than at the targeted sites (Table 11; Figure 13, upper panel), which is likely related to a higher number of impounded sites and other habitat alterations in southern New England compared to Maine. The skew in the targeted data is most evident in the frequency histogram in Figure 13, lower panel. Note that on this and subsequent histograms the targeted sites are plotted separately and are not stacked as are the NRSA base and overdraw probabilistic sites, thus each targeted sites bar starts at zero.

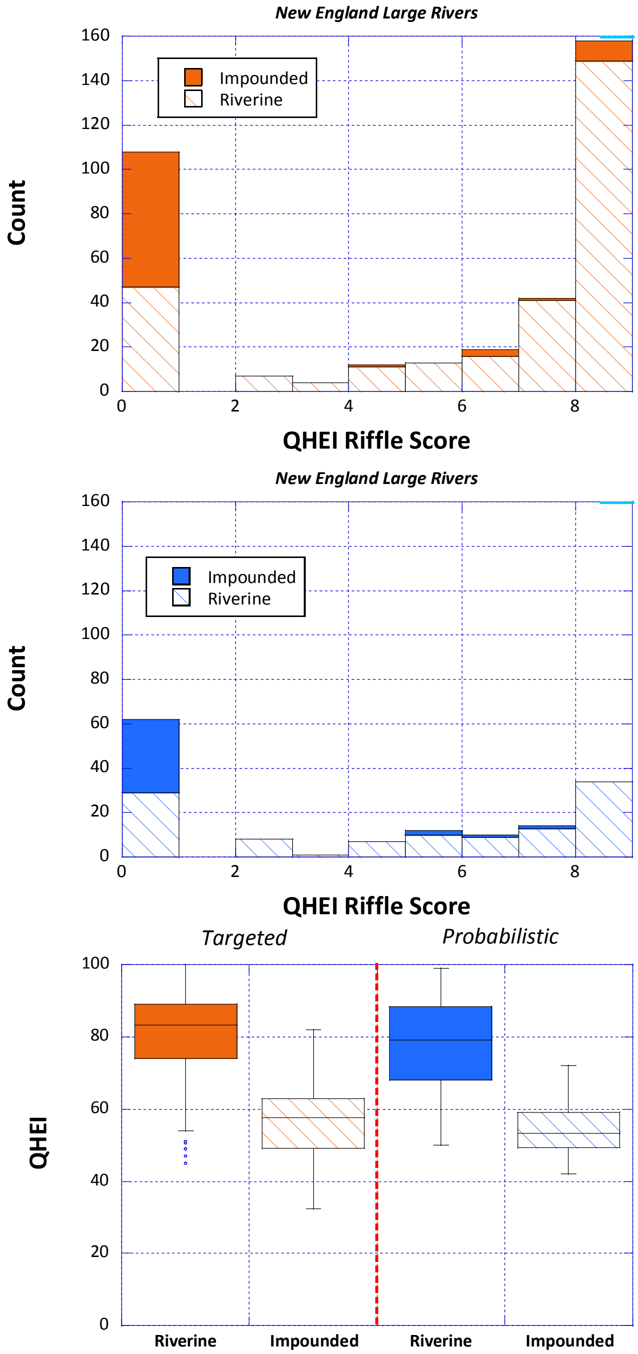


Figure 14. Frequency histogram of QHEI riffle score, separately for targeted sites (upper panel -orange) and probabilistic sites (middle panel - blue, stacked and colored coded by base vs. overdraw). QHEI scores between riverine and impounded appear in the lower panel for targeted (orange) and all probabilistic sites (blue).

In the plot of the QHEI riffle/run metric score, which rates the quality of riffle/run habitats, the targeted sites (orange) had proportionately fewer sites with poor riffle scores (zero) which indicates either impounded or low gradient sites (e.g., low gradient, pool habitat dominated sites), whereas probabilistic sites had more poor riffle/run scores (zero) than high riffle/run scores (Table 11; Figure 14). Habitat quality was generally good (>60) to excellent (>80) in the absence of impoundment, which was the leading source for lower QHEI scores. Mapped QHEI scores show that many high quality sites do exist throughout southern New England (Figure 15).

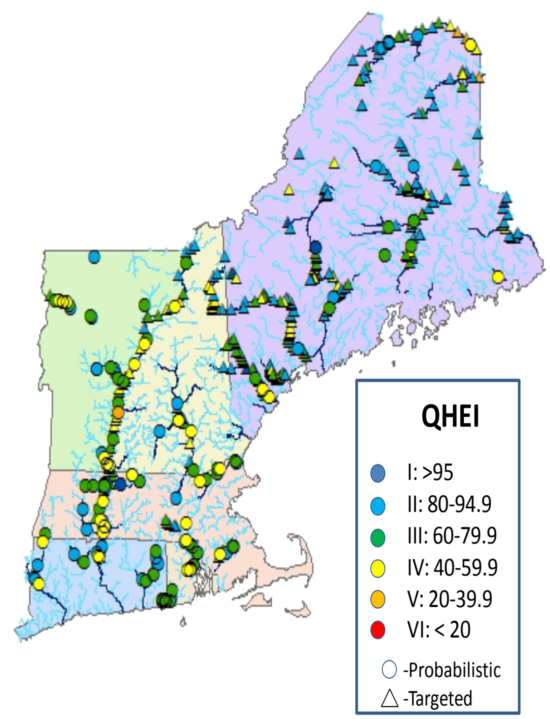


Figure 15. *Map of QHEI scores in New England rivers with sites colored coded by QHEI narrative ranges and BCG level. Probabilistic sites are circles, target sites are triangles.*

Dissolved Oxygen and Conductivity

We used data collected at each site consisting of instantaneous values for dissolved oxygen and conductivity that were collected during each fish sampling event. Plots of these parameters are found in Figures 16 and 17 and frequency statistics in Table 11. Dissolved oxygen (D.O.) at the

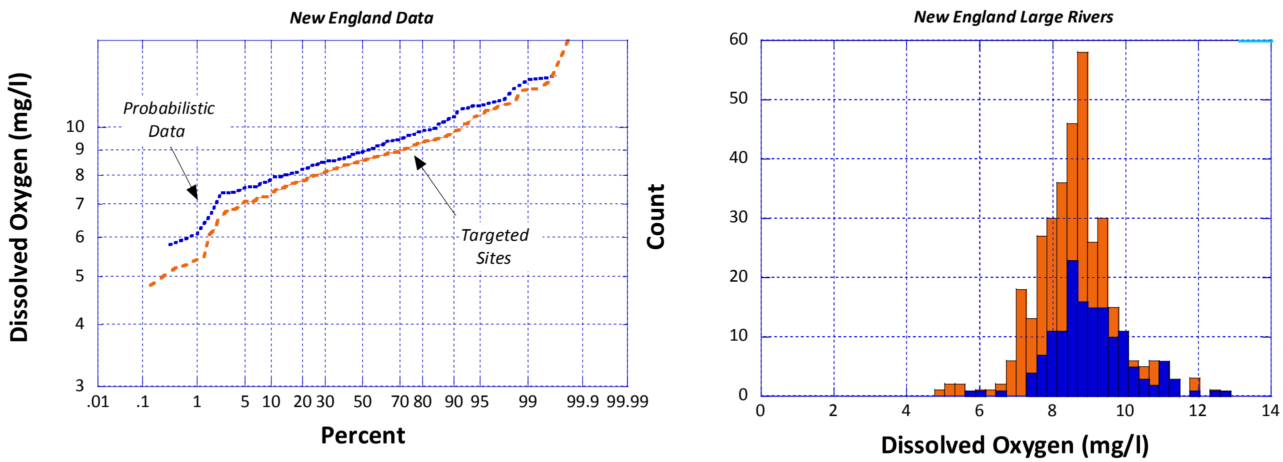


***Figure 16**. Cumulative frequency distribution (left) plot of dissolved oxygen (D.O.) at REMAP probabilistic sites (blue) vs. targeted sites (orange) and as a stacked histogram (right).*

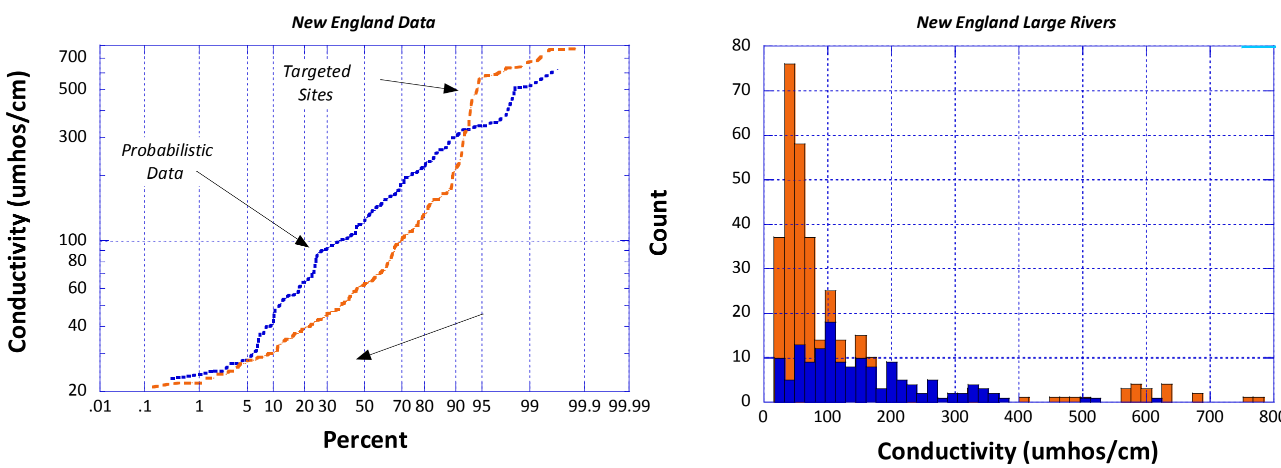


***Figure 17**. Cumulative frequency distribution (left) plot of conductivity at NELR REMAP probabilistic sites (blue) vs. targeted sites (orange) and as a stacked histogram (right).*

targeted sites was consistently, but only slightly lower along most of the New England rivers (Table 11; Figure 16). Daytime D.O. values ranged mostly between 8-10 mg/l with only a few sites showing extremely high or low values. This seems typical of daytime D.O. values which do not always depict the most critical aspects of the D.O. regime that can vary between day and night within a diel cycle. These results are likely to be of limited value in a stressor analysis. Conductivity varied widely across New England with differences in geochemical variation (e.g., parent geology), natural runoff over these formations (USGS 2004), and differences in land use and anthropogenic influences (e.g., discharges from effluents, urbanization) being apparent in the results. Median conductivity was significantly lower at the targeted than at probabilistic sites (Table 11; Figure 17) reflecting the more equitable distribution of these sites by latitude. The targeted sites also uniquely identified a cluster of sites at the very upper end of the observed conductivity gradient (Table 11; Figure 17). Conductivity ranged widely from very low values of <75 µS/cm to >500 µS/cm generally along a north to south gradient (Figure 18, left panel). Values from an interpolated map developed by USGS (2004; Figure 16, right panel) also reflected this gradient that is related to both natural phenomena and anthropogenic sources, particularly in revealing urban areas in southern New England. Conductivity values were also elevated downstream of large volume point sources in selected rivers (Androscoggin, Connecticut, Kennebec, St. John, Blackstone) that were sampled with an intensive pollution survey design showing its value as an indicator of potential effluent impacts and urbanization.

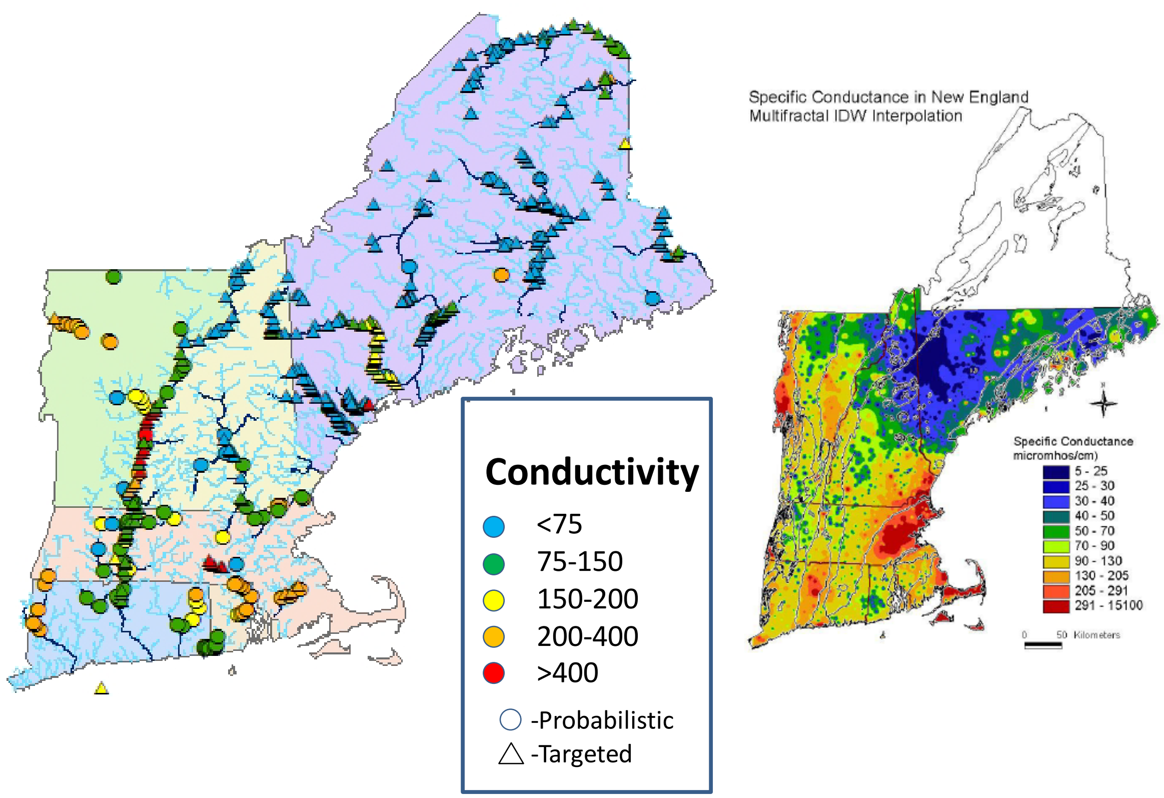


***Figure 18**. Map of conductivity (µS/cm) in New England rivers (left) with sites colored coded by conductivity ranges. A USGS (2004) interpolated map of specific conductance is inset to the right (analysis not available for northern one-half of Maine).*

The measured values at REMAP and targeted sampling sites seem to confirm the USGS (2004) interpolated map, the latter revealing where conductivity values are naturally elevated compared to the northern latitudes where it is comparatively low.

SPARROW Predicted Total Phosphorus and Total Nitrogen

We used the outputs of the SPARROW model to provide a simulated water quality result in lieu of having sufficient and readily available water chemistry data at NELR REMAP probabilistic and targeted sampling sites. SPARROW model outputs for mean annual total phosphorus (TP) and total nitrogen (TN) concentrations were used as a surrogate indicator of potential nutrient enrichment. Targeted sites had lower modeled concentrations of TP and TN than did the probability sites except for the upper tail of the CFD distribution where the extreme highest values were revealed at targeted sites (Table 11; Figure 19).

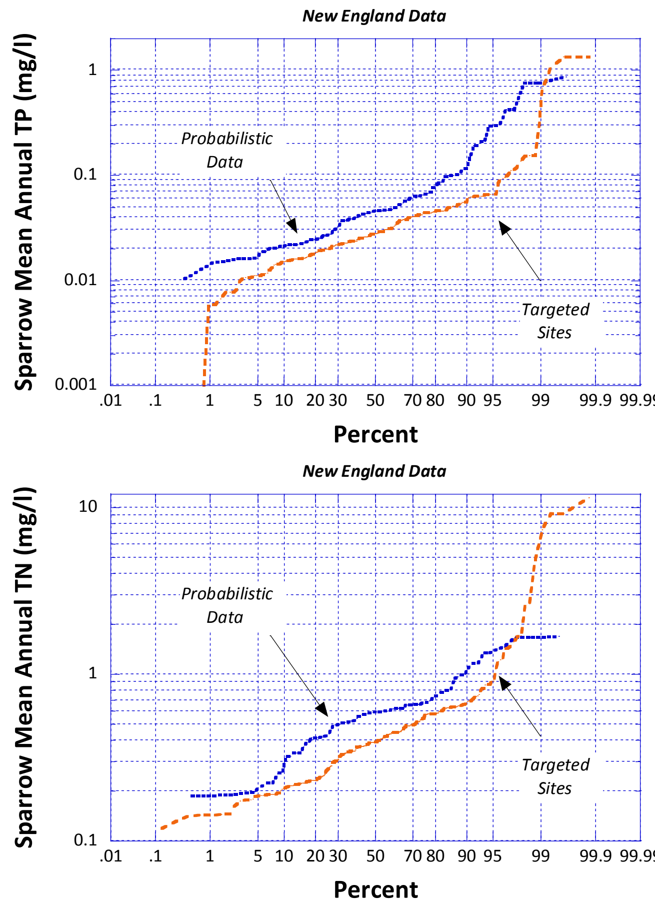


***Figure 19**. Cumulative frequency distribution plot of targeted sites (orange) and probabilistic sites (blue) for Sparrow TP (top) and Sparrow TN (bottom).*

The SPARROW Model considers landscape variables (e.g., land cover, land use, temperature, and soil permeability) that each exhibit spatial gradients throughout New England. As illustrated in the mapped values (Figure 20) modeled TP and TN increased in a general southerly direction and roughly corresponds to increases in non-natural land cover and increased population density. Pollution gradients are especially apparent in the TP predictions in the same selected mainstem rivers that showed observed increases in conductivity, thus reflecting the influence of large point source effluents and urbanization. It also illustrates the distinctiveness in the results between the probabilistic and targeted intensive pollution survey sites.

Temperature

Two temperature measures were used to depict thermal gradients in New England rivers. Mean annual air temperature and the thermal classification results from NEAHCS which

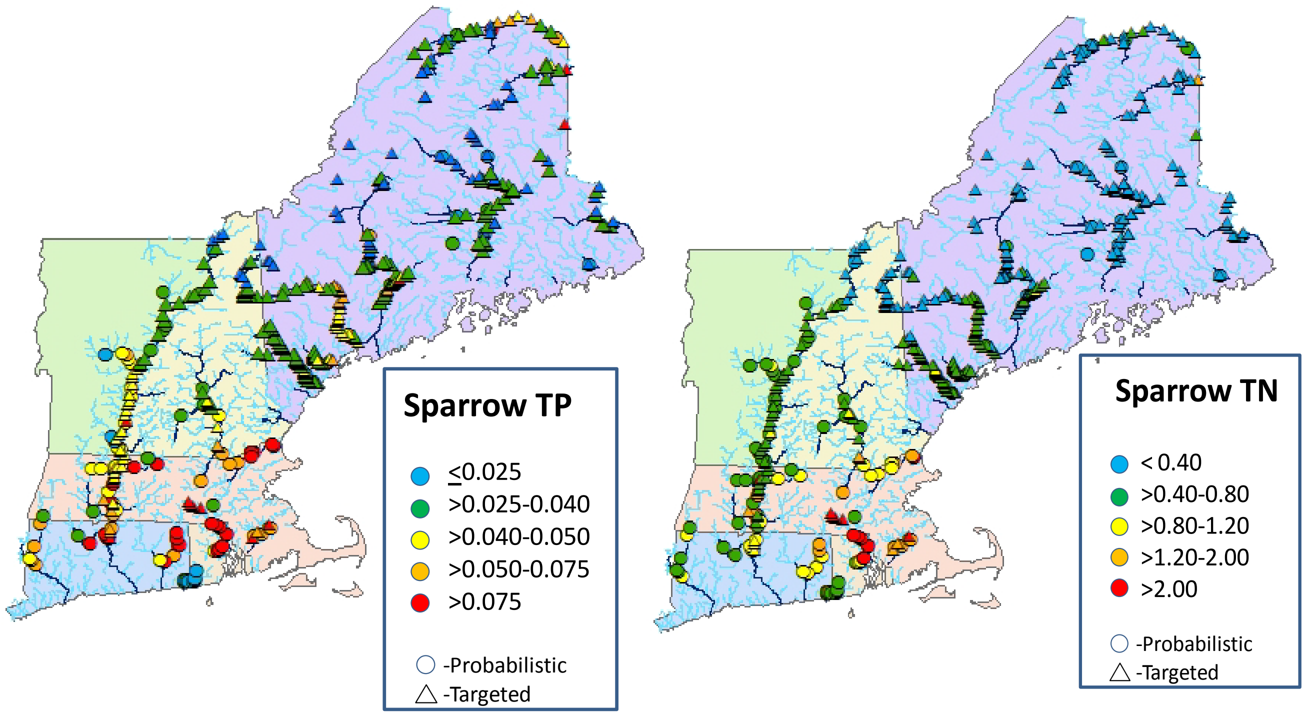


***Figure 20**. Maps of SPARROW modeled mean annual total phosphorus (left panel, mg/l) and total nitrogen (right panel, mg/) in New England rivers with sites colored coded by TP and TN ranges.*

classified each site as cold water, cold transitional, warm transitional, or warmwater based on the expected fish species composition (see Table 10). While we did measure ambient temperature at all fish sampling sites, we found that it was too variable within a site to be sufficiently representative of the prevailing thermal regime to explain biological variation. Mean annual air temperature within the watershed within which a sampling site was located was significantly lower for targeted than for probabilistic sites (Table 11; Figure 21) again likely due to the fact that nearly one-half of New England large rivers occur in the more northern latitudes.

A similar pattern was revealed by the NEACHS thermal classifications (Figure 22) with cold water and cold transitional sites shifting to warm transitional sites in southern New England. However, with the exception of the Merrimac R. in New Hampshire and Massachusetts, cool transitional sites occurred as far south as northwestern Connecticut and southwestern Rhode Island. While there is undoubtedly a natural latitudinal gradient in water temperature, changes to the southern New England landscape over time have likely resulted in the artificial warming of rivers with increased land development and hydrological alterations thus confounding the determination of true potential.

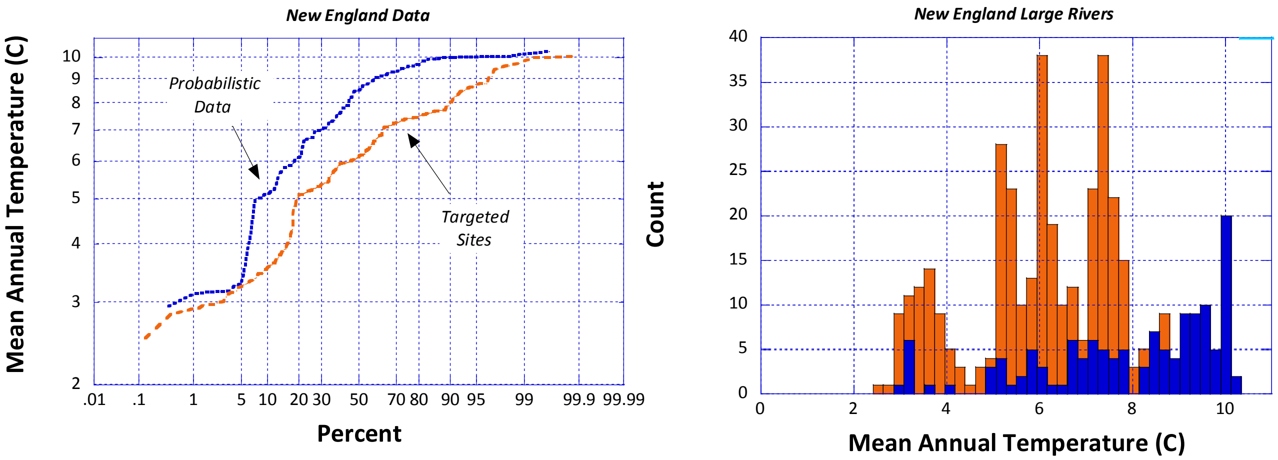


***Figure 21**. Cumulative frequency distribution plot (left panel) of mean annual air temperature (℃) at targeted sites (orange) and probabilistic sites (blue) and a frequency histogram of mean annual air temperature (right panel) for targeted sites (orange) and probabilistic sites (blue).*

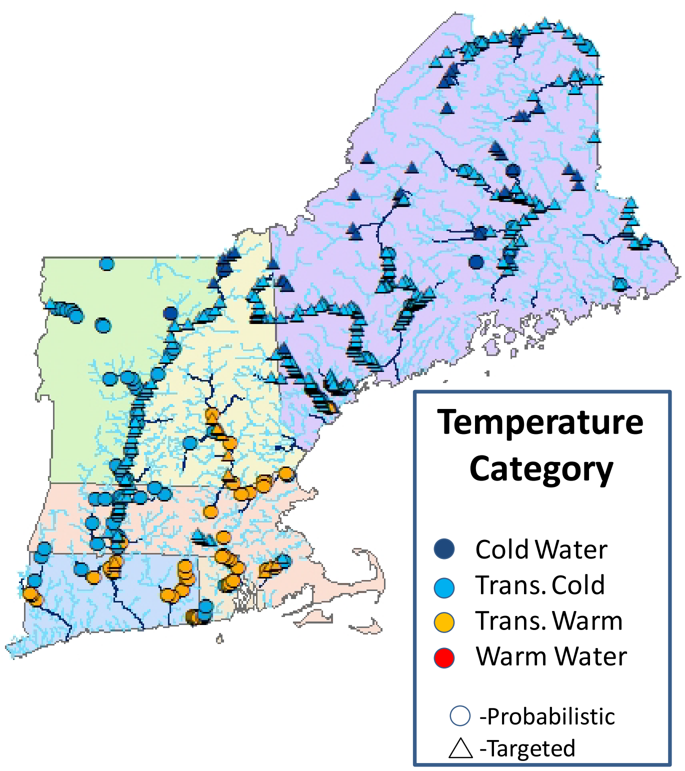


***Figure 22**. Map of thermal classification of New England river sites color coded by thermal classification from the NEAHCS project.*

Land Cover

Changes in land cover from natural (e.g., forest, wetland) to agricultural and developed land cover can result in altered hydrology, increases in polluted runoff, and a decreased export of natural organic matter that forms the baseline trophic structure of rivers. Natural hydrologic processes in rivers also include periodic flooding of the adjacent floodplains and periodic connections with adjacent wetlands. Frequently, floodplains are either eliminated or restricted and wetlands are filled as land use changes to a more developed state.

Two forms of natural land cover data were used in this analysis; cumulative and local (Figure 23). Cumulative natural land cover represents that of the entire watershed upstream of the downstream most point of the NHDPlus river segment within which a site was located. Local

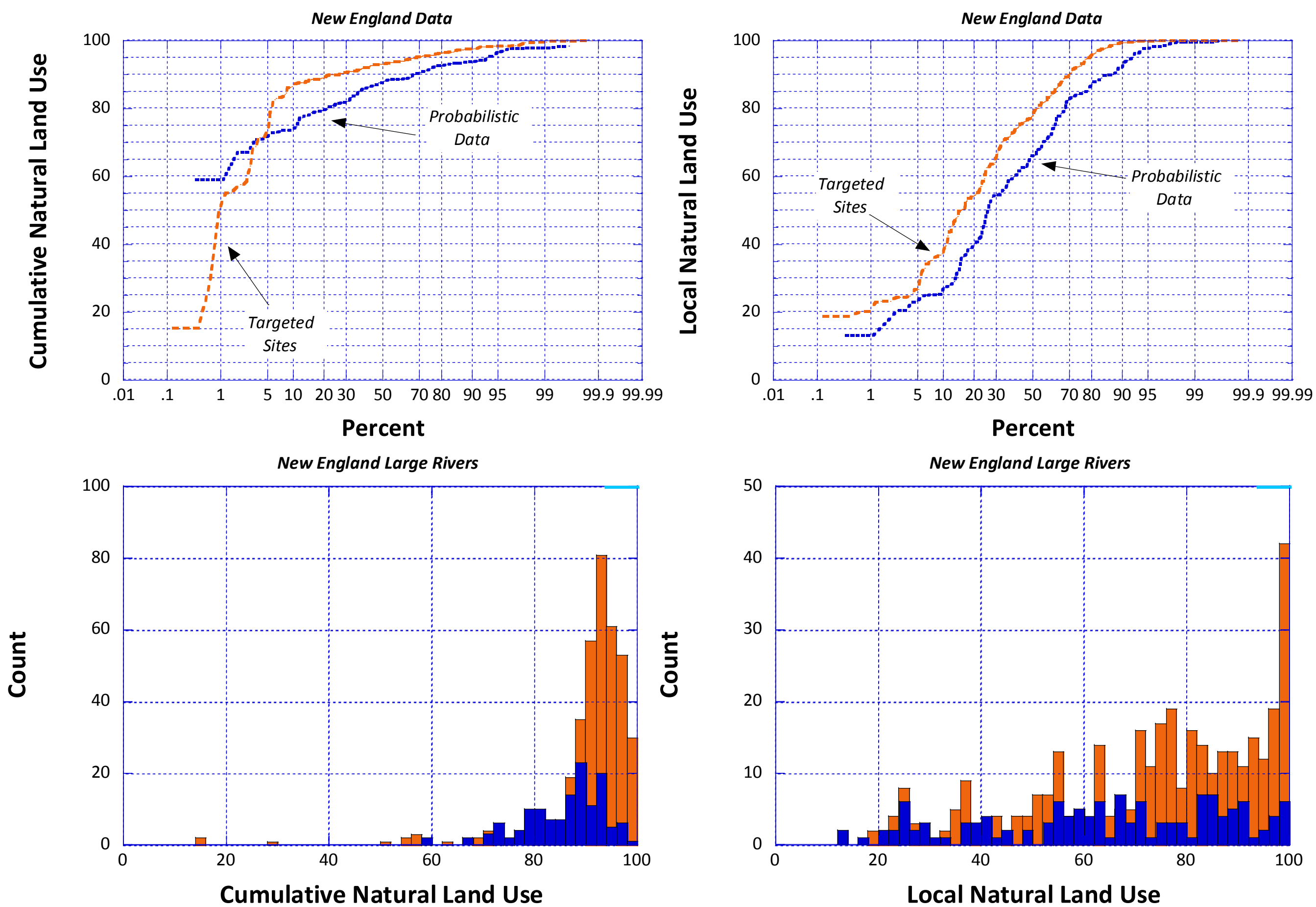


Figure 23*. Cumulative frequency distribution plots (upper left and right) and frequency histograms (lower left and right) of targeted (orange) and probabilistic sites (blue) for cumulative natural land cover (left) and local natural land cover (right).*

land cover includes the land use in the immediate watershed in a single NHDPlus reach within which a site was located. Local natural land cover percentages had a wider range than cumulative land cover percentages and were potentially more informative about the potential for local scale impacts. A higher proportion of targeted sites had higher natural cover values (Table 11; Figures 23) because of the greater number of sites in the less developed and sparsely populated northern latitudes, although this population of sites had a longer tail of local natural land cover <80% down to <20% (Figure 23, upper left panel).

The map of cumulative natural land cover revealed the same overall north to south gradient that was observed with the other stressor variables (Figure 24). Local land cover revealed a

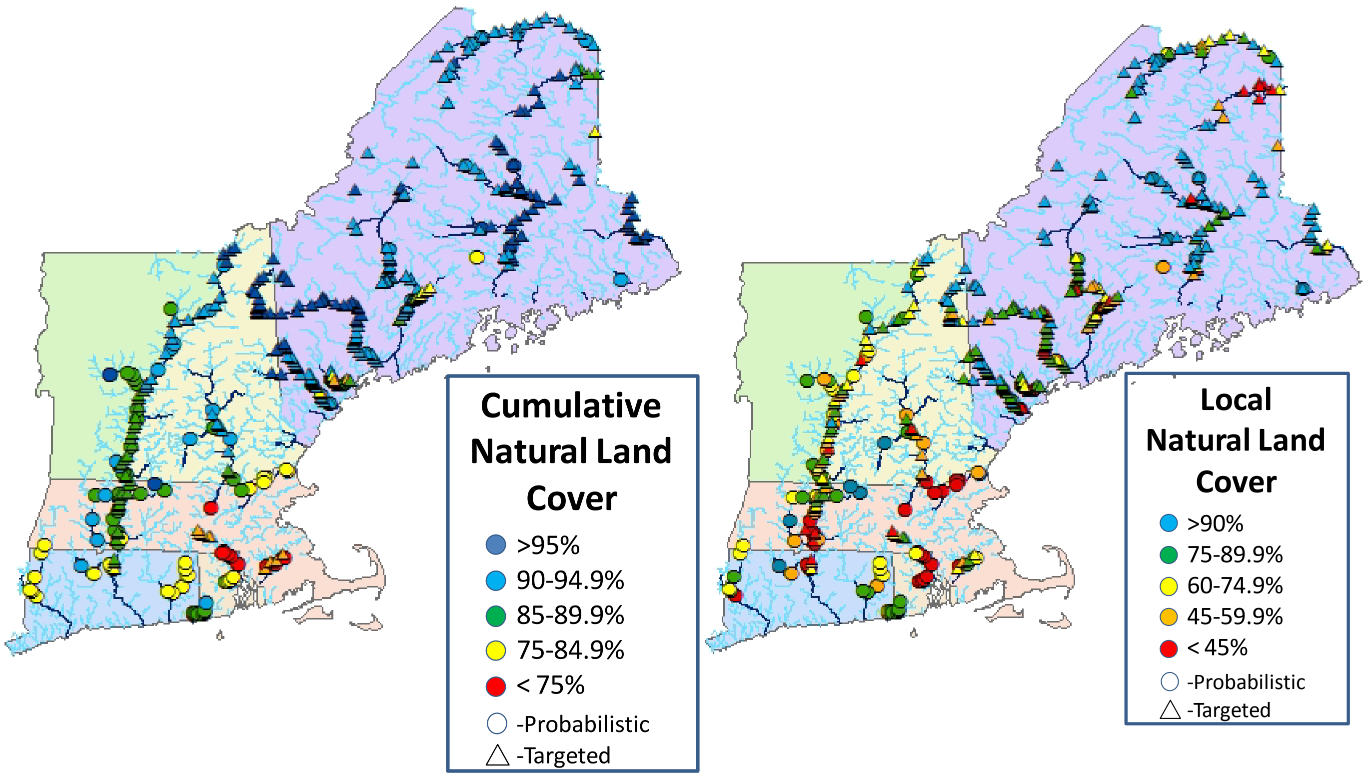


***Figure 24**. Map of cumulative (top) and local (bottom) land cover data at sites sampled in New England rivers (left) with sites colored coded by percent of cover type.*

more detailed pattern and reflected some of the more localized and river reach stressor gradients that were also revealed by conductivity and the SPARROW TP results.

Connectivity

Dams and other barriers (e.g., waterfalls) have an important influence on the fish assemblages of large rivers by impeding or blocking movements and migrations of fish and particularly of diadromous species that rely on open connections between their riverine and ocean habitats. We used the Northeast Aquatic Connectivity (NAC) database that contains metrics related to the presence and types of dams and barriers on rivers in the Northeast U.S. (Martin and Apse 2011). We then linked these metrics to the NHDPlus reaches. More than 50% of the probabilistic and targeted sites had no impassable barriers and approximately 25% had no barriers of any kind. The upper tail of the distribution of targeted sites generally had more barriers and impassable barriers than did the probabilistic sites (Figure 25). All of the coastal rivers have multiple barriers at some point along their respective lengths that impedes connectivity between reaches (Figure 26). These accumulate in an upstream direction making access to historical spawning grounds for anadromous species and freshwater habitat for adult and juvenile American eel more difficult to access. Of note is that some of the rivers labeled in Figure 26 as having multiple barriers were historically inaccessible to anadromous fish species. This is true of the St. John River drainage in Maine that was isolated by a large falls in New Brunswick just over the border with Maine. However, the portion of that river in New Brunswick was historically accessible and is now impeded by at least two dams.

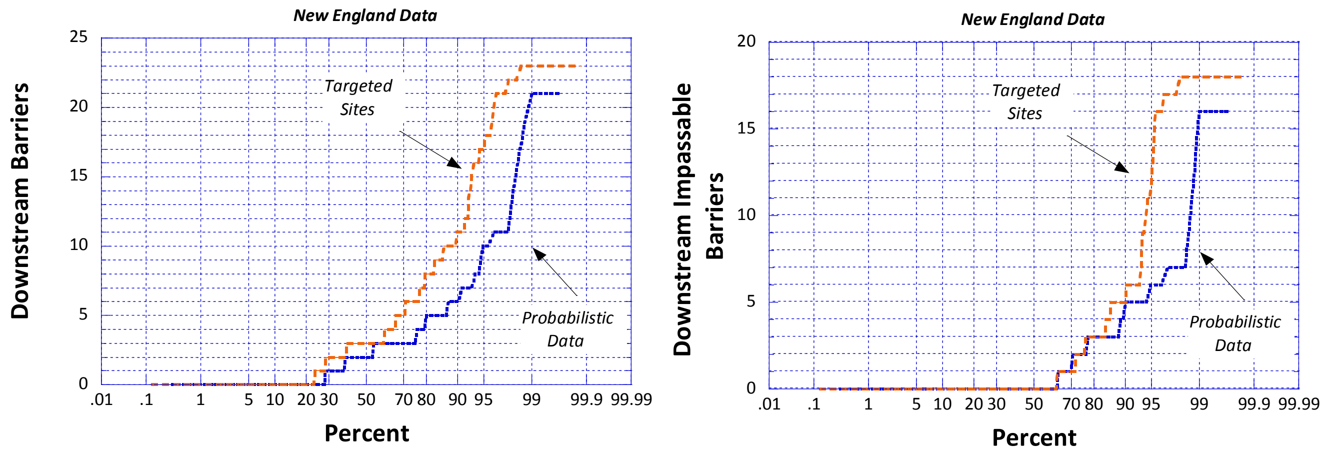


***Figure 25**. Cumulative frequency distribution plot of targeted sites (orange) and probabilistic sites (blue) for Downstream Impassable Barriers (top) and Downstream Barriers (bottom).*

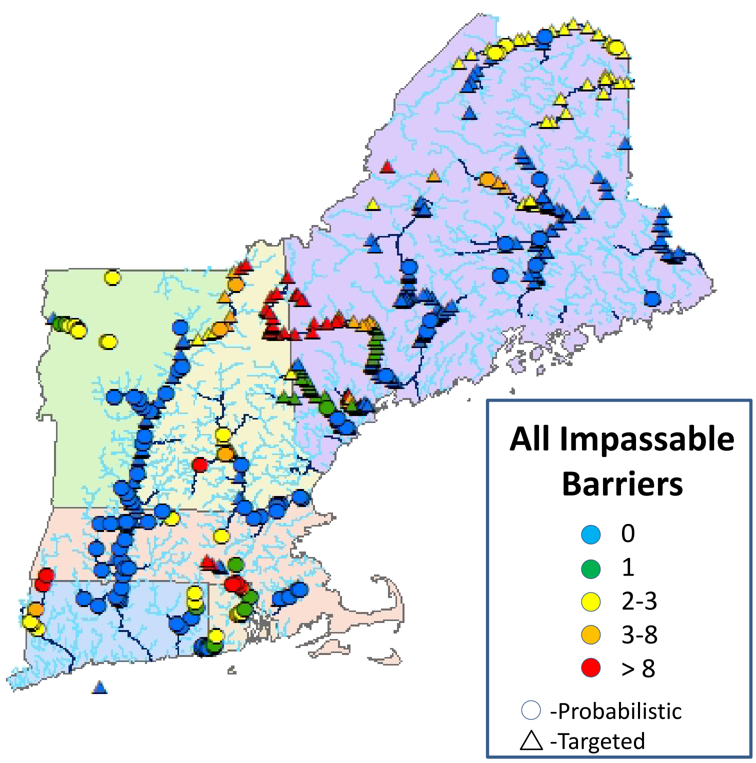


***Figure 26**. Map of all impassible downstream barriers with respect to each REMAP and New England targeted sampling locations.*

Fish Assemblage Condition Assessment

Assessing the condition of the fish assemblage in New England large rivers is a primary objective of the study. Determining the effects of associated stressors is also a part of this project objective. The NELR REMAP probabilistic study design was intended to provide a statistically valid assessment of condition hence various analyses were performed to yield the assessment statistics. In addition, we evaluated the results from targeted sites that were sampled in New England prior to and alongside the 2008-9 NELR REMAP study.

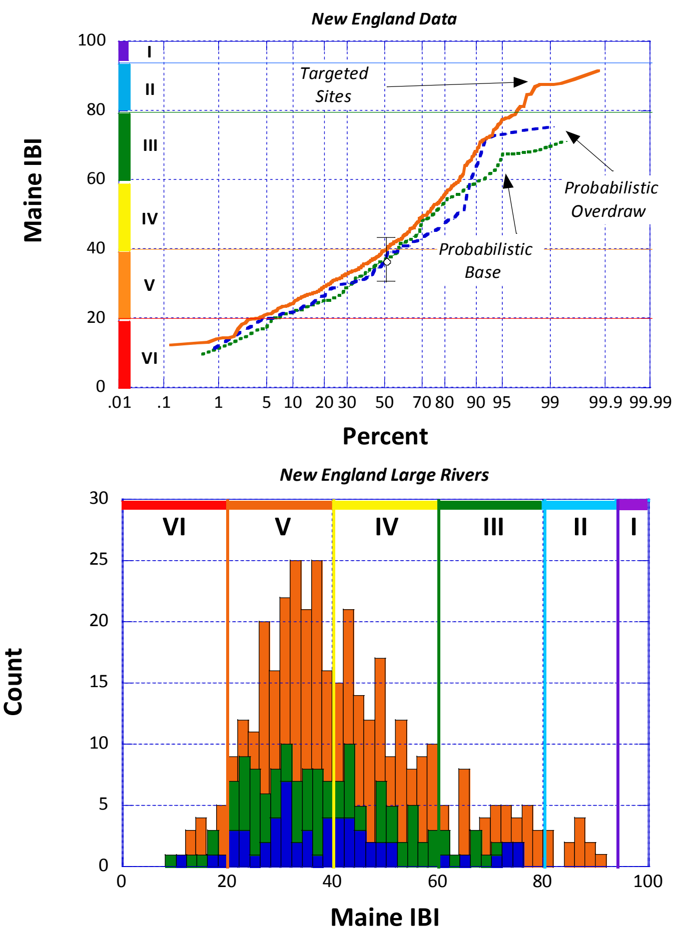


__Figure 27__. Cumulative frequency distribution plot of the Maine IBI (upper panel) for targeted sites (orange) and NELR REMAP (NRSA base [green] and overdraw [blue]) probabilistic sites and a frequency histogram (lower panel) of the Maine IBI for targeted sites (orange) and stacked for NELR REMAP probabilistic sites (color coded by base vs. overdraw).

Regional Assessment of Condition

The ME IBI (Yoder et al. 2008) was used as the primary measure of fish assemblage condition for New England large rivers. This index was initially calibrated for Maine large rivers and for a cold to cool water ecotype and evaluating its applicability across New England was a sub-objective. It was the only readily available IBI type of index that was specifically derived and calibrated for non-wadeable rivers in New England. The ME IBI also has a supplemental set of diadromous metrics to calculate a Diadromous IBI that adds to the ME IBI. While still experimental in its testing and application, it should be useful in assessing the condition of coastal rivers that have historically harbored diadromous fish species.

Maine IBI (ME IBI)

Figure 27 (upper panel) illustrates a cumulative frequency distribution of the ME IBI separately for targeted sites, NRSA base probabilistic sites, and overdraw probabilistic sites with the biological condition gradient (BCG) tiers superimposed on the y-axis. Figure 27 (lower panel) uses the same data depicted as frequency histograms for the targeted sites (orange bars) and as a stacked histogram for the NELR REMAP probability sites, color coded as NRSA base sites (green) or overdraw sites (blue).

Maine IBI with Diadromous Metrics (DIBI)
The frequency distribution of the ME IBI with diadromous metrics (DIBI) was similar to that of the ME IBI alone (Figure 28, upper panel). The distribution of higher DIBI scores relative to ME IBI results corresponded to rivers where diadromous species have at least partial access to upstream reaches. The DIBI also resulted in a few more sites in BCG Level 2 (Figure 28, lower panel). This included two tributaries to the Penobscot R. in Maine and the Farmington R. in Connecticut. The histogram of the DIBI was less skewed than the ME IBI alone suggesting that it is indeed capturing these important ecological attributes of coastal river fish assemblages that should be apparent where they are expected components of the fish assemblage.

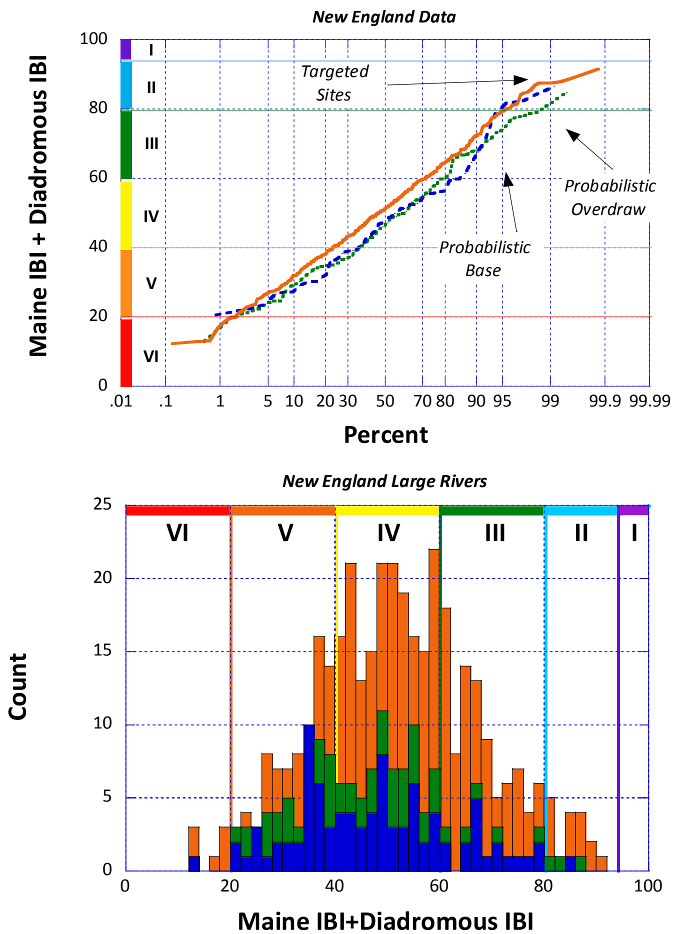
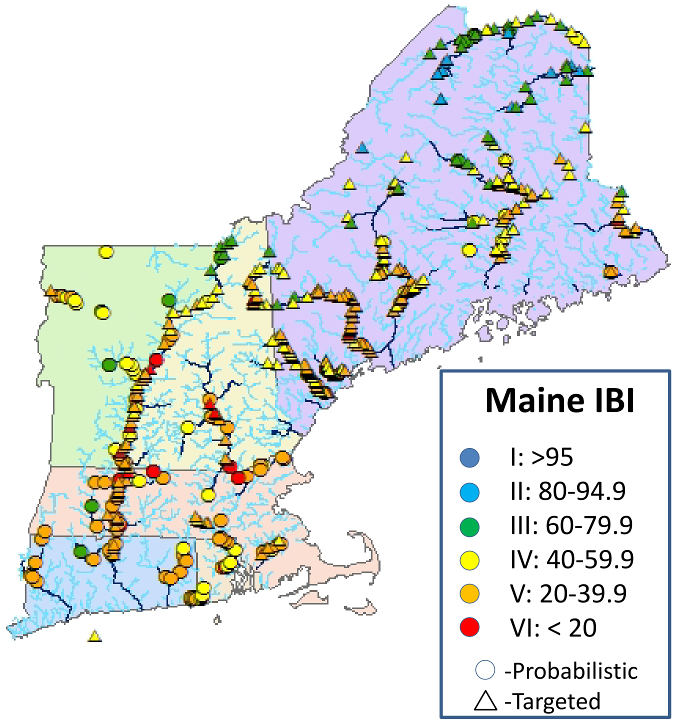


***Figure 28**. Cumulative frequency distribution plot (left) of targeted sites (orange), base probabilistic sites (green) and overdraw probabilistic sites (blue) and frequency histogram (right) of Maine IBI + Diadromous IBI, separately for targeted sites (orange) and probabilistic sites (stacked and colored coded by base vs. overdraw).*

Prototype Condition Assessment
At the spatial density represented by the REMAP probabilistic design the upper tail of the distribution of ME IBI scores missed the highest quality sites seen in the targeted data set and which are mostly concentrated in northern Maine and New Hampshire (Figure 29). We used the ranges of the ME IBI that correspond to the six levels of the BCG as defined by Yoder et al. (2008) and as depicted in Figures 28 and 29. The results of the ME IBI and the DIBI with diadromous metrics added by BCG level are presented in Table 12. As with all of our analyses, the results at the REMAP probabilistic and targeted sites are presented separately and for comparison purposes. The proportion of samples that reflected BCG Levels 3 through 6 were not substantially different between the REMAP probabilistic and New England



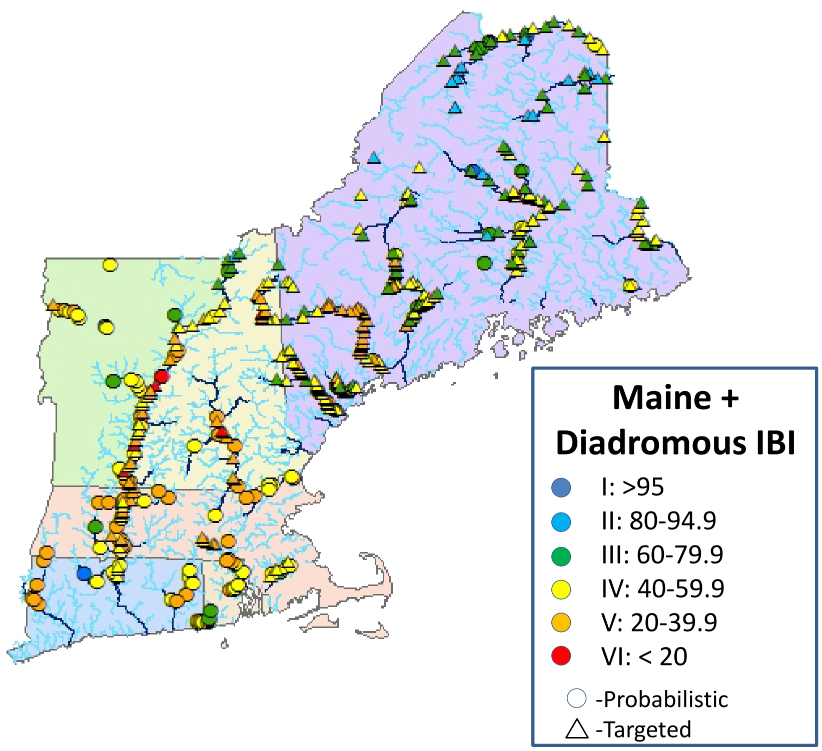


Figure 29. *Map of non-wadeable fish sampling sites in New England with Maine IBI and Maine plus Diadromous IBI (DIBI) values colored coded by BCG Levels 2-6 (II-VI).*

***Table 12**. The number and percentage of NELR REMAP probabilistic and targeted samples arranged by corresponding BCG level for the Maine IBI (top) and the Diadromous IBI (bottom).*

	NELR REMAP Probabilistic		**NELR REMAP Targeted**	
BCG Level	**Samples**	**Percent**	**Samples**	**Percent**
	Maine IBI			
Level 1: IBI >95	0	*0*	0	*0*
Level 2: IBI >80 and IBI <95	0	*0*	12	*3.2*
Level 3: IBI >60 and IBI <80	15	*10.1*	42	*11.3*
Level 4: IBI >40 and IBI <60	48	*32.2*	127	*34.2*
Level 5: IBI >20 and IBI <40	78	*52.3*	177	*47.7*
Level 6: IBI <20	8	*5.4*	13	*3.5*
Totals	149	*100*	371	*100*
	Diadromous IBI			
Level 1: IBI >95	0	*0*	0	*0*
Level 2: IBI >80 and IBI <95	4	*2.7*	16	*4.3*
Level 3: IBI >60 and IBI <80	22	*14.8*	90	*24.3*
Level 4: IBI >40 and IBI <60	70	*47.0*	179	*48.2*
Level 5: IBI >20 and IBI <40	52	*34.9*	79	*21.3*
Level 6: IBI <20	1	*0.7*	7	*1.9*
Totals	149	*100*	371	*100*

targeted results being <5% at most. No sites in either dataset were in BCG Level 1. The most revealing difference was the absence of any BCG Level 2 samples in the NELR REMAP data set for the ME IBI. The DIBI had 4 NELR REMAP samples at BCG Level 2, but the corresponding increase in targeted BCG Level 2 sites made the difference essentially the same.

Table 13 lists the highest scoring NELR REMAP probabilistic and targeted sites that correspond to BCG Level 2 and the upper portion of Level 3. A total of 19 targeted sites in Table 13 had higher IBI scores than the highest scoring REMAP probability site and only 4 of the 27 highest

***Table 13**. Fish sample locations of top 27 sampling locations ordered by IBI score. Sample type: T=NELR REMAP Targeted; P-O=NELR REMAP probabilistic. Sites in blue shading are BCG Level 2 and green represents BCG Level 3.*

Basin	River	River Name	River Mile	Date Sampled	Maine IBI	Sample Design
30	800	Aroostook River	44.00	07/20/2005	91.6	T
70	660	N. Br. Penobscot R.	11.80	07/22/2004	89.1	T
30	800	Aroostook River	15.40	08/22/2005	88.0	T
30	500	Allagash River	25.00	08/26/2005	87.6	T
30	500	Allagash River	32.20	08/26/2005	87.6	T
30	500	Allagash River	36.80	08/26/2005	87.6	T
30	500	Allagash River	36.80	09/14/2006	87.0	T
30	800	Aroostook River	9.80	07/18/2005	84.9	T
30	500	Allagash River	63.20	09/16/2006	84.6	T
30	500	Allagash River	21.10	09/15/2006	81.1	T
30	600	Fish River	8.10	09/13/2006	81.1	T
30	800	Aroostook River	94.00	07/19/2005	80.2	T
30	800	Aroostook River	65.30	07/19/2005	78.8	T
80	001	Connecticut River	322.00	08/28/2008	78.6	T
30	800	Aroostook River	19.70	08/22/2005	78.3	T
70	700	E. Br. Penobscot R.	41.57	08/08/2007	77.9	T
70	600	W. Br. Penobscot R.	37.60	07/21/2004	77.8	T
30	800	Aroostook River	76.60	07/19/2005	77.6	T
60	700	Magalloway River	15.82	09/13/2007	77.6	T
30	800	Aroostook River	91.80	07/13/2009	76.3	P-O
30	500	Allagash River	6.00	08/25/2005	75.9	T
70	600	W. Br. Penobscot R.	47.20	07/22/2009	75.4	P-O
30	001	St. John River	303.80	07/21/2005	75.3	T
30	001	St. John River	290.60	07/22/2005	74.7	T
80	001	Connecticut River	291.00	08/30/2008	74.5	T
30	600	Fish River	2.40	07/11/2009	74.2	P-O
30	001	St. John River	286.80	07/12/2009	73.3	P-O

scoring sites were NELR REMAP probabilistic sites. Only two of the 27 highest scoring sites occurred outside of Maine and both were in the upper Connecticut River in northern New Hampshire. These would typify sites that are of the highest quality in the region and as a result would merit more than baseline protections under the CWA. Based on the order of remaining overdraw sites in the NRSA sites draw, it would have required an additional 23 site replacements to have a site in a river (Allagash) that would have had a high likelihood of a Level 2 ME IBI score. Essentially there is little to no chance that a Level 2 ME IBI would have been yielded by any probabilistic site under the NRSA site draw. The estimate of the median for the NELR REMAP targeted sites is within the confidence interval of the probabilistic sites. Despite the non-random selection of the targeted sites the median is a relatively close approximation of the median NELR REMAP probabilistic sample.

We also recalculated statistics by restricting targeted and probabilistic sites to those above latitude 45°N and the difference in median scores was even less (Table 14). The NELR REMAP probabilistic data set was essential to provide a randomly generated estimate of a median IBI

***Table 14**. Median and percentile statistics for the Maine IBI from probabilistic sample groups (base and overdraw) and targeted samples from non-wadeable New England Rivers. Data in parentheses are 95% confidence intervals for each statistic.*

Data	5^{th}	25^{th}	Median	75^{th}	95th
Maine DIBI (w Diadromous Component)					
Probabilistic	23.7 (20.3-27.2)	36.0 (34.0-38.6)	47.2 (42.8-49.9)	56.2 (54.1-59.8)	78.0 (70.4-82.4)
Targeted	27.3 (23.0-28.8)	40.6 (38.4-42.8)	51.2 (49.4-53.2)	61.5 (59.8-64.3)	78.8 (75.7-84.1)
Maine IBI (without Diadromous Component)					
Probabilistic	19.8 (11.3-21.4)	27.1 (24.6-30.3)	37.1 (34.2-40.9)	48.6 (44.4-53.3)	68.4 (61.8-74.2)
Targeted	21.2 (18.6-23.0)	31.2 (29.3-32.6)	39.6 (37.3-41.6)	51.7 (49.6-54.8)	77.6 (72.3-81.1)
Maine IBI – Northern Sites Only Above Latitude 45					
Probabilistic	42.8 (42.0-43.0)	45.2 (42.0-45.0)	57.9 (45.2-64.7)	71.2 (58.9-75.4)	76.3 (73.3-76.6)
Targeted	31.8 (18.3 – 35.3)	44.1 (39.6 – 48.0)	54.0 (51.4-60.9)	71.0 (65.8-75.9)	87.6 (81.1-89.1)

score, without which we would have been uncertain about how representative of regional conditions the targeted data set actually was. Such a comparison provides much needed confirmation that a high density targeted sampling effort within a regional scope can provide reliable estimates of condition and it certainly seems needed to reveal the highest quality sites.

While we do not purport to be recommending an impairment threshold for CWA purposes as an objective of this study, the results in Table 12 do offer a template for an initial assessment of the condition of New England riverine fish assemblages. If BCG Level 4 for the ME IBI is used as the minimum CWA goal impairment threshold, then 42.3% of the non-wadeable rivers in New England perform at or above this level. A higher proportion of targeted sites (48.2%) performed above this level, but 3.2% were at BCG Level 2 which was not represented in the NELR REMAP probabilistic data set. The DIBI results showed a higher proportion of sites at BCG Level 4 or higher with 35.5% of REMAP probabilistic sites and 23.2% of targeted sites below BCG Level 4. This leaves little doubt that including the supplemental diadromous metrics not only alters these statistics, it boosts the BCG level of many sites. However, this seems to occur mostly within BCG Levels 3 and higher. Many of the highest scoring sites were in parts of northern Maine where diadromous species are not an expected part of the fish assemblage, thus these sites are reflecting the core cold and cool water attributes around which the ME IBI is constructed.

Mainstem River Assessment Case Example

Another NELR REMAP project objective was to compare the assessment outcomes produced by different spatial sampling designs in a large mainstem river. To meet this objective we located additional sites on selected mainstem rivers in addition to the NRSA probabilistic and overdraw sites that comprise the REMAP sampling design. For this analysis we focused on the Connecticut River mainstem based on sampling conducted in 2008-9.

Connecticut River Assessment

The Connecticut River mainstem was sampled in 2008 and 2009 from the Third Connecticut Lake in New Hampshire downstream to the “salt wedge” just upstream from I-95 in Connecticut. The upper portion of this study area was sampled between the Third Connecticut Lake and the Turners Falls dam in 2008 and between Turners Falls and I-95 in 2009. Probabilistic sites were selected from the 2008-9 NRSA draw of sites for two levels of coverage with targeted sites added to fill in “gaps” for a longitudinal pollution survey design. For the latter, all of the probabilistic sites served as part of a more spatially intensive design. The NRSA sites encompassed a 4.0 km distance sampled by a different electrofishing protocol, thus two 1.0 km NELR REMAP sites were included within each of the 4 km long NRSA sampling reaches. One of those 1.0 km sites was randomly selected to function as a NELR REMAP probabilistic site for this analysis. The objective was to compare the estimates of condition between the two probabilistic sample draws (NRSA base and NELR REMAP) and from the intensive pollution survey design. The ME IBI and DIBI were used to illustrate these comparisons. Figure 28 (left panels) is a cumulative frequency diagram of the NRSA (NRSA Base Only), NELR REMAP (NRSA base + overdraw sites), and the intensive pollution survey results. All of the ME IBI and DIBI percentiles were statistically similar between the 3 designs except for the 95th percentiles which were highest for the intensive pollution survey design (Table 15). This observation is similar to that seen in the regional NELR REMAP results where the upper tail of the ME IBI distribution was similarly skewed upward for the targeted sampling sites.

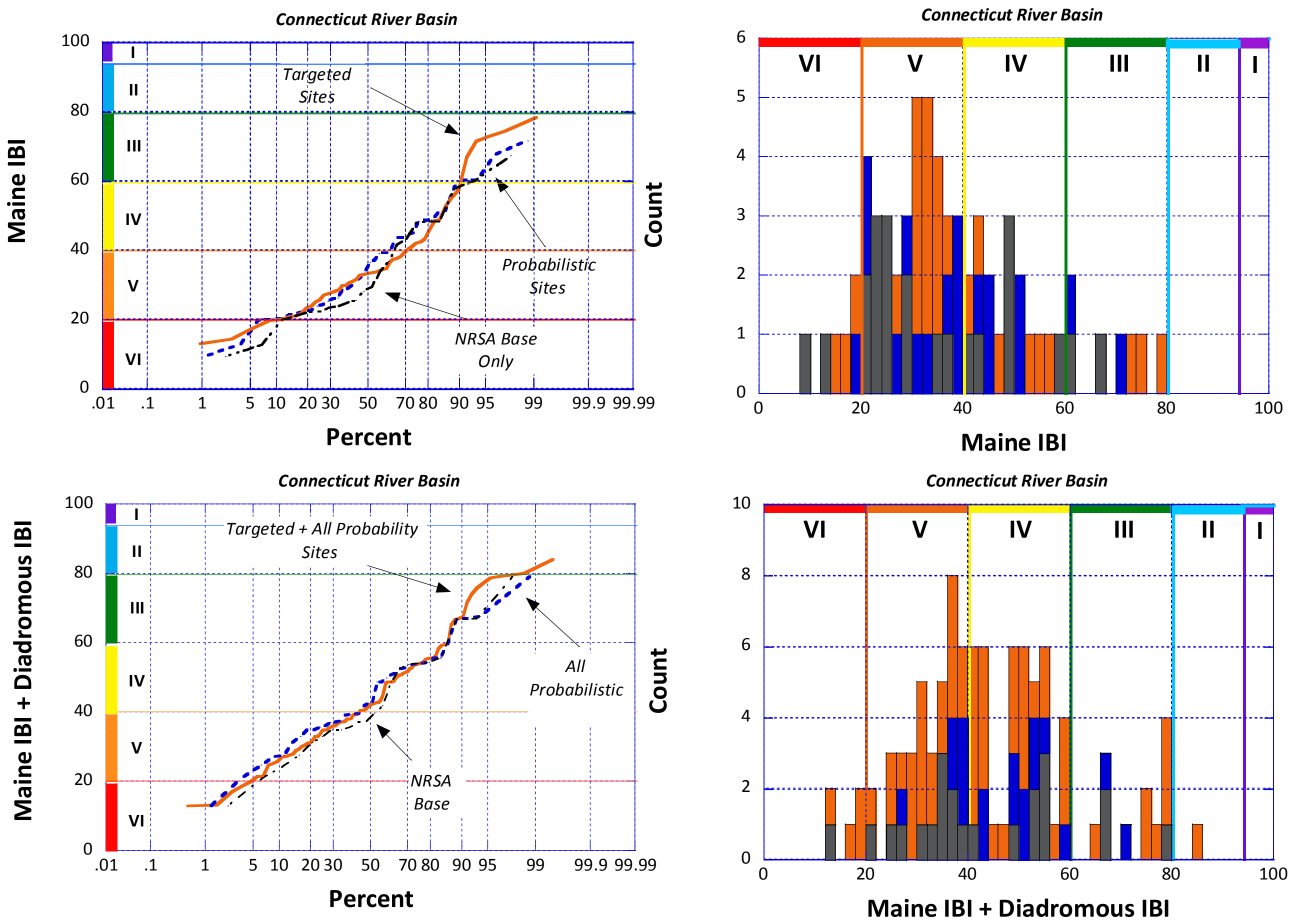


***Figure 30**. Cumulative frequency distribution plots (left) of targeted sites (orange), base probabilistic sites (green) and overdraw probabilistic sites (blue) and frequency histograms (right) separately for targeted sites (orange) and probabilistic sites (stacked and colored coded by base vs. overdraw) of the Maine IBI (upper panels) and the Maine IBI + Diadromous IBI (lower panels) in the Connecticut River mainstem 2008-9.*

The targeted design produced slightly higher IBI statistics than the NELR REMAP and even higher ones than the NRSA base sites alone. It also produced a disproportionately higher 95th percentile revealing sites with IBIs that were more than 12 points higher than either the NELR REMAP or NRSA base sites. This is consistent with the regional NELR REMAP project data that tends to show the same results. The highest scoring Connecticut River sites were in BCG Level 3 with most in BCG Level 4 and 5, the lowest scoring sites in BCG Level 6. The Maine DIBI resulted in an upward shift of all values (Table 15a), as expected, but this was not uniform at all Connecticut River sites. Figure 31 shows a longitudinal plot of the two IBIs along the mainstem from upstream to downstream (including the tidal reach downstream from Hartford, CT). The

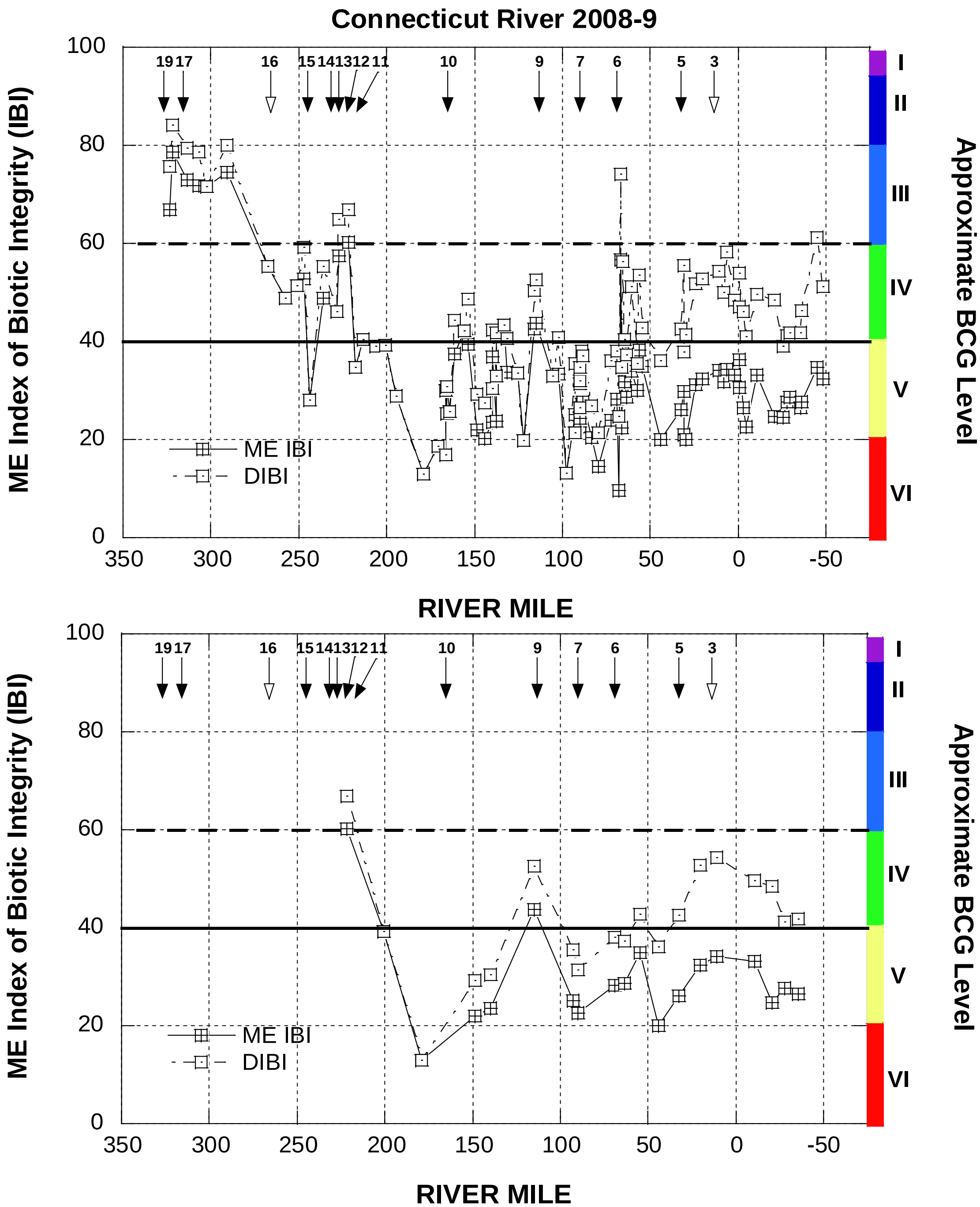


***Figure 31**. Connecticut River mainstem results for the Maine IBI (solid line) and Maine IBI + Diadromous IBI (DIBI) for all sites sampled from below Third Connecticut Lake (New Hampshire) to upstream I-95 in Connecticut (upper panel) and probabilistic sites only (lower panel). The corresponding BCG level is labeled on the y2 axis. River mile 0 is at the head of tide with negative values in the tidal affected segments. Arrows and numbers indicate dams and hydropower projects (see Table 15b). Dashed and solid lines indicate BCG Levels 3 and 4 respectively.*

***Table 15a**. Median and percentile statistics for Maine IBI for NRSA base samples, all probabilistic sample groups (base and overdraw) and targeted samples from non-wadeable Connecticut River data. Data in parentheses are 95% confidence intervals for each statistic.*

Data	5th	25th	Median	75th	95th
Maine IBI					
REMAP (All Probabilistic)	13.0 (9.6-20.2)	23.8 (20.2-28.6)	34.2 (26.1-41.8)	45.1 (39.3-51.3)	60.3 (51.3-71.6)
NRSA Base Sites	13.0 (9.6-20.2)	20.2 (9.6-20.2)	28.6 (22.6-43.7)	48.2 (34.2-58.2)	60.3 (48.6-67.6)
Targeted Sites	16.9 (13.1-14.5)	25.8 (20.4-30.0)	33.4 (30.0-36.8)	42.1 (36.8-52.8)	72.9 (55.3-78.6)
Maine DIBI					
REMAP (All Probabilistic)	21.4 (13.0-27.4)	35.5 (27.4-39.1)	42.6 (37.3-51.3)	53.9 (50.6-59.6)	67.5 (59.6-79.0)
NRSA Base Sites	21.4 (13.0-24.7)	33.0 (21.4-35.5)	37.3 (33.0-53.7)	54.1 (41.8-66.8)	67.0 (55.6-79.0)
Targeted Sites	18.6 (13.1-25.3)	30.9 (25.8-36.0)	40.9 (36.0-46.0)	51.8 (46.0-59.2)	79.4 (59.3-84.1)

differences between the DIBI and ME IBI scores were most apparent in the tidal segment and generally becoming less upstream. However, differences continued to occur well upstream into New Hampshire and enough so that a single site in the upper mainstem moved into BCG Level 2. Given the importance of diadromous species management in the Connecticut River the DIBI concept provides a way to better utilize the fish assemblage as an indicator of connectivity.

The plot of REMAP probabilistic sites only (Fig. 31, lower panel) shows how this design missed the highest quality part of the upper mainstem and also the most impacted site immediately below the Turners Falls dam. The general trend is similar to the intensive survey design, but it missed both the highest and lowest ME IBI and DIBI scores in the mainstem.

Lake Champlain Drainage

Eleven (11) of the NELR REMAP sites include the Winooski and Missisquoi Rivers in Vermont. Each is tributary to Lake Champlain which lies in a distinctly different ichthyofaunal region than

Table 15b. *Location and description of major dams and hydroelectric projects on the mainstem Connecticut River corresponding to numbers on Figure 31.*

Dam Number in Fig. 31	Dam Name	Hydroelectric Project Status	Dam/Impound-ment Size	Fish Passage
19	Murphy Dam	TransCanada Power	117 feet/2000 Acres	None
17	Lower Canaan	PSNH	30 feet/≈1.5 mi.	None
16	Groveton Dam	Breached	N/A	N/A
15	Moore Reservoir	TransCanada Power	178 feet/11 mi.	None
14	Gillman Dam	Ampersand Gillman Hydro LLC	24 feet/2.9 mi.	None
13	Comeford Station	TransCanada Power	170 feet/8.0 mi.	None
12	McIndoe Station	TransCanada Power	25 feet/5.0 mi.	None
11	Dodge Falls Dam	DFHC	15.5 feet/?? mi.	None
10	Wilder Dam	TransCanada Power	59 feet/45 mi.	Yes – ust. (ladder) & dst.
9	Bellow Falls Dam	TransCanada Power	30 feet/26 mi.	Yes – ust. (ladder) & dst.
7	Vernon Dam	TransCanada Power	20 feet/26 mi.	Yes – ust. (ladder); No – dst.
6	Turners Falls Dam	First Light	≈50 feet/20 mi.	Yes – ust. (3 ladders) & dst.
5	Holyoke Dam	HG&E	30 feet/2290 Acres	Yes – ust. (ladder) & dst.
3	Enfield Dam	Breached	N/A	N/A

the remainder of the NELR REMAP study area. At least 9 fish species in Table 7 are unique to the Lake Chaplain drainage and were not included in the original calibration of the ME IBI. Because of the potential inapplicability of the ME IBI to these rivers and the lack of a riverine IBI for Vermont (Langdon [2001] developed *wadeable* fish IBIs for cold and warm water streams in

Vermont), two other IBIs that are based on similar ichthyofaunas were also calculated. This included the Atlantic slope IBI (Daniels et al. 2005) and the Ohio EPA boatable sites IBI (Ohio EPA 1987, 1989b). While neither IBI was specifically derived or calibrated for the Lake Champlain tributary rivers in Vermont there is some value in using each to initially portray the mainstem fish assemblages. The Atlantic slope IBI was developed for the neighboring Hudson and Delaware River basins and the authors (Daniels et al. 2003) surmised that it would be relevant in adjacent drainages. The Ohio EPA boatable IBI is developed for large rivers and includes many of the same fish species as occur in the Lake Champlain drainage.

All three IBIs were plotted by river mile for the Winooski River which had 10 sampling locations (Figure 32). The IBIs for the Missisquoi R. at the single sampling site were also included on the same plot. The thresholds for CWA aquatic life use attainment for the Ohio EPA boatable IBI

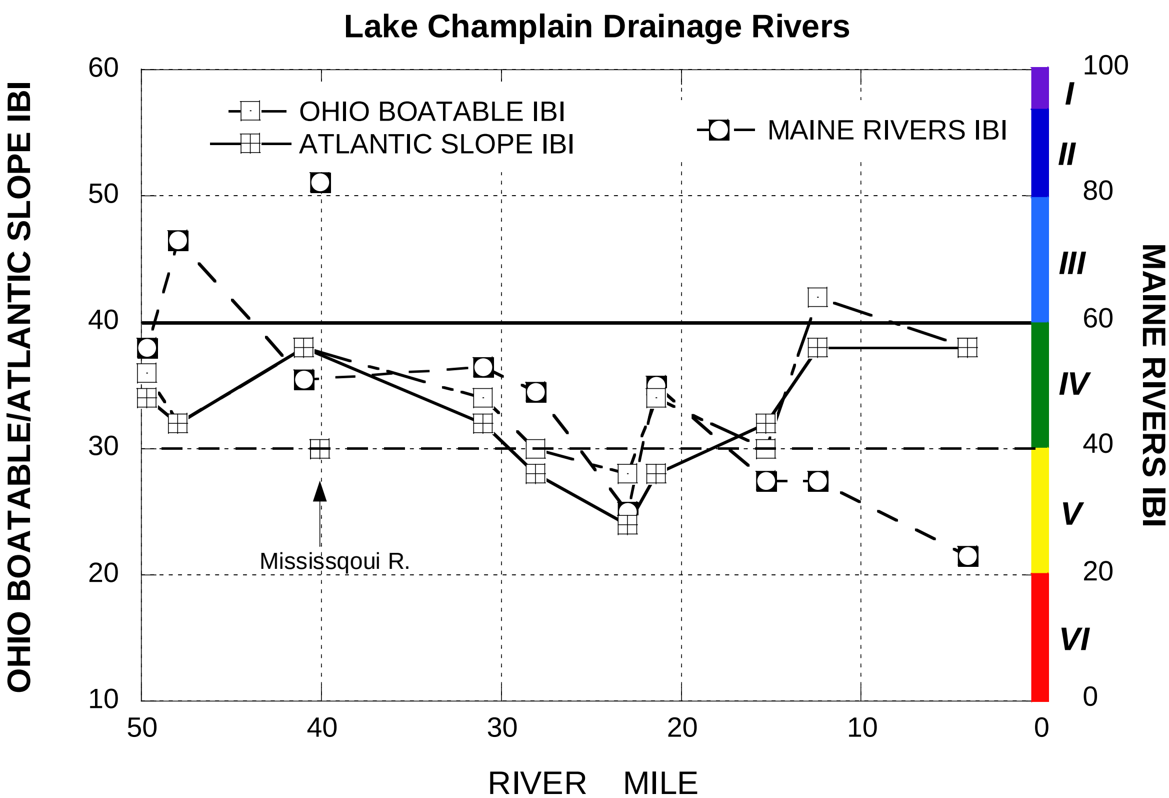


Figure 32. *Plots of the Maine IBI, Atlantic slope IBI, and Ohio EPA boatable IBI by river mile for 10 sites in the Winooski R. and a single site in the Missisquoi R. The biocriteria for the Ohio EPA boatable IBI is indicated by the solid line. BCG level 4 for the Maine IBI is represented by the dashed line and is the threshold for CWA aquatic life use attainment used for the NELR REMAP assessment. The corresponding BCG levels of the Maine IBI are depicted along the y-2 axis.*

and the ME IBI are shown as an initial assessment of these rivers, with the latter used for the NELR REMAP assessment. The Atlantic slope and Ohio IBIs showed a very similar trend in terms of longitudinal direction from upstream to downstream and IBI values being identical or within 2-4 units at most sites. Only one value was above the Ohio threshold of 40, but at least 4 were within 4 IBI units which is the nonsignificant departure used by Ohio EPA for assessment purposes. By contrast the ME IBI showed a consistent decline from upstream to downstream being above or within 2 IBI units of the NELR REMAP minimum CWA threshold at all except the downstream most site. The single site on the Missisquoi R. had the highest ME IBI value which was at BCG Level 2, an indication of high quality in terms of other NELR REMAP sites. From a regional development standpoint the Atlantic slope IBI is perhaps the most relevant, but it was developed primarily for wadeable streams. Deciding which IBI is most relevant would require further analysis and consideration. To be conservative these sites were restricted from the New England-wide assessment, stressor analyses, and mapping of ME IBI metrics.

Major New England Mainstem Rivers and Reaches

Sufficient data was available to make a comparative assessment of 36 individual rivers or river reaches using the ME IBI and DIBI (Figure 32a) and the Index of Well-Being (Iwb; Figure 32b). Box-and-whisker plots were used to display and rank each river by the 75th percentile value of the ME IBI + DIBI and Iwb and shading the boxes with the corresponding BCG level in accordance with the median (50th percentile) value. Ranking rivers by the 75th percentile value reflects the "best sites" and either the protection level or restoration potential of each. The ME IBI + DIBI rankings were done according to the sum of both indices which best represents the BCG attributes, the current quality of each river, and where the core freshwater and/or diadromous components of the assemblage show differing results. It also allows for a fair comparison with river reaches that naturally lack diadromous species due to natural barriers. The Iwb rankings were done to determine if there were different river rankings in terms of an index that only considers abundance and biomass without regard to assemblage guilds such as diadromy, tolerance, native status, organism condition, or function. It should reveal instances where rivers may have a comparatively high abundance, diversity, and biomass of fish, yet lack the other attributes that are embodied in the BCG and not measured by abundance and diversity.

Maine IBI and DIBI

The most intact rivers in terms of the ME IBI occurred in northern Maine that are generally north of latitude 44-45N and included the Allagash, Aroostook, upper Connecticut R., and upper branches of the Penobscot R. These all included sites where the ME IBI corresponded to BCG Level 2. The Allagash R. has no diadromous potential being removed from the natural barrier

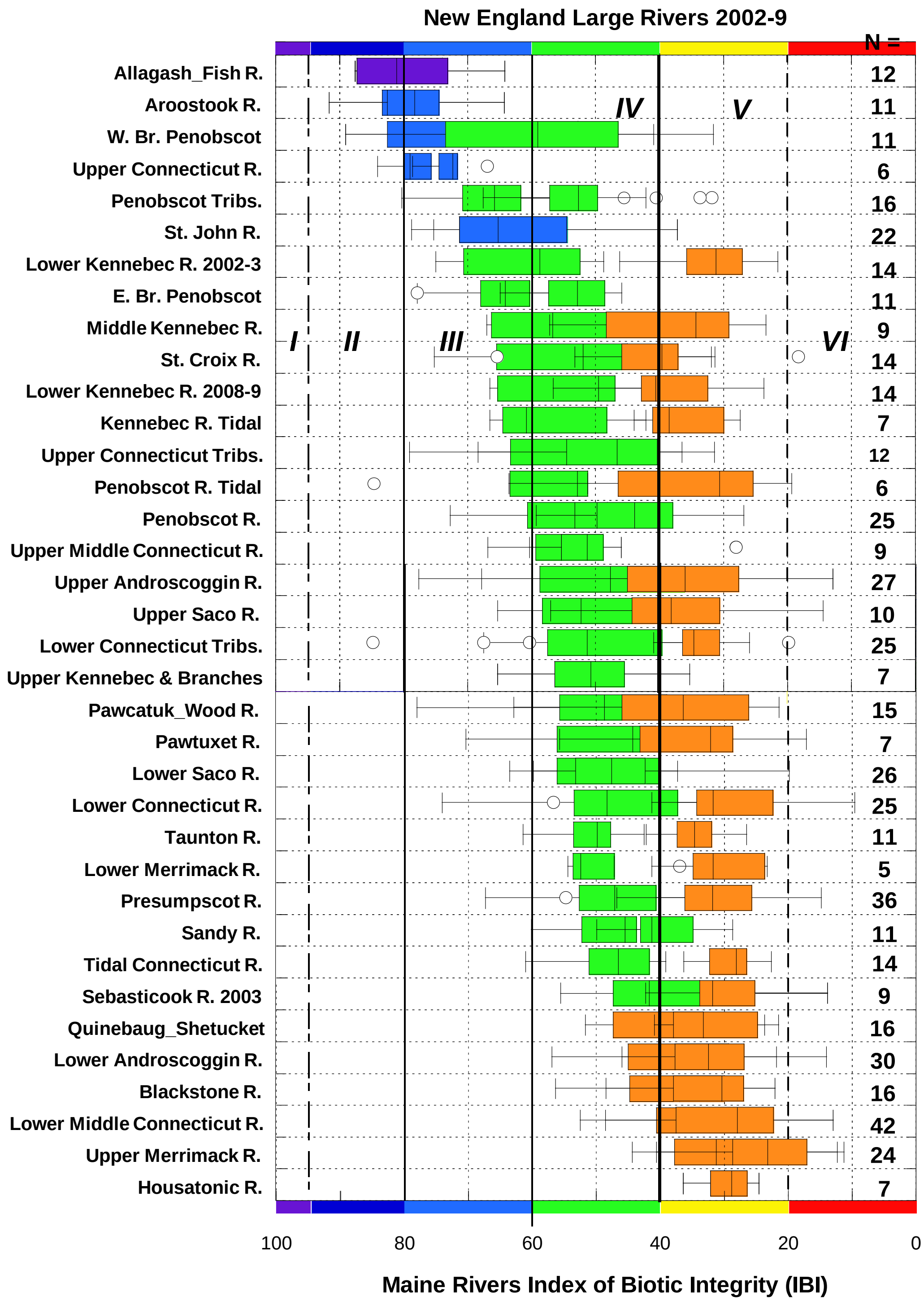


***Figure 32a**. Maine Rivers IBI and plus Diadromous IBI box-and-whisker plots for all sites sampled during 2002-9 in 36 major riverine segments in New England in ordered by the 75th percentile ME IBI+DIBI. Fill color corresponds to the BCG range using the 50th percentile value for each river and reach.*

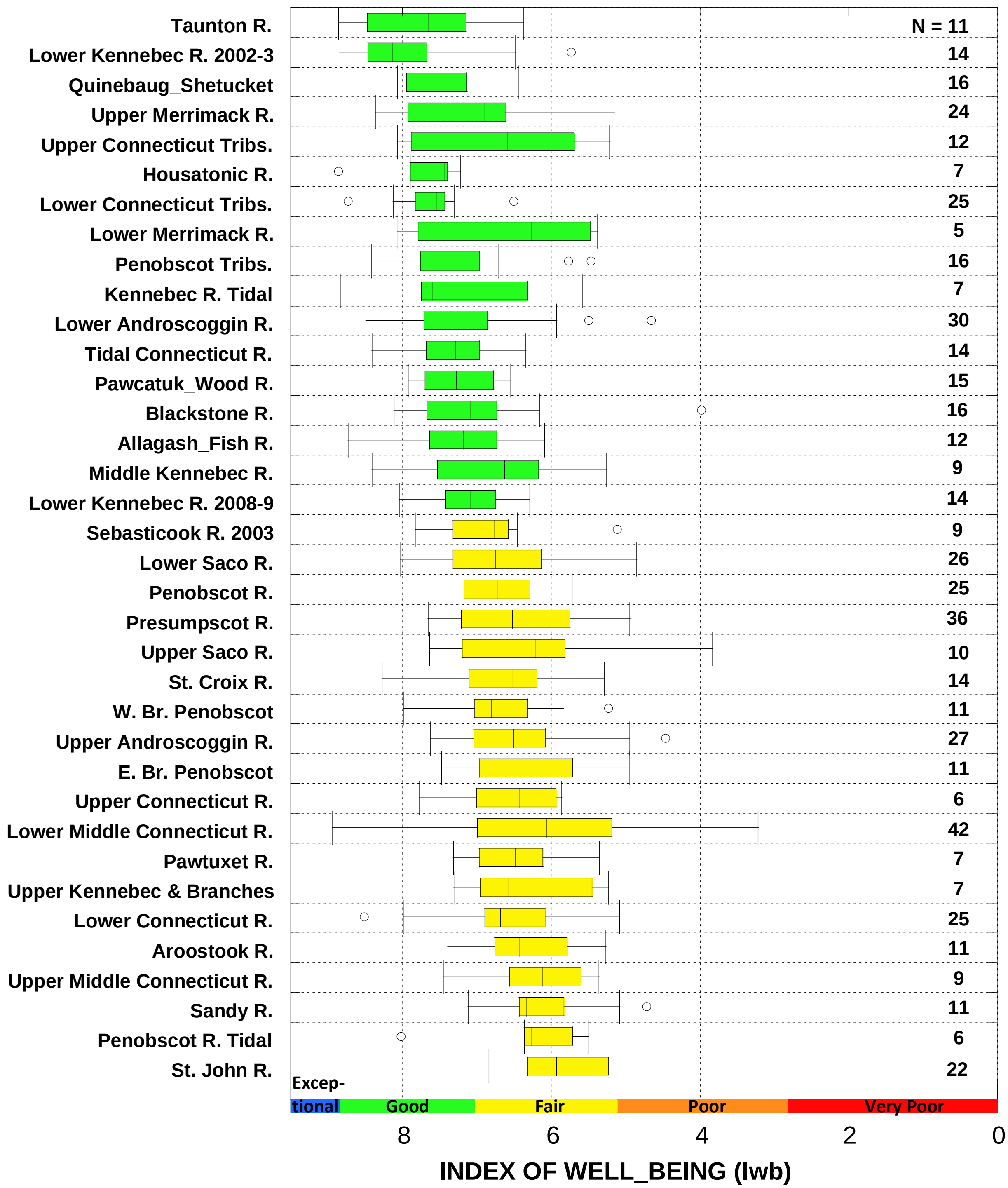


***Figure 32b**. Index of Well-Being (Iwb) box-and-whisker plots for all sites sampled during 2002-9 in 36 major riverine segments in New England ordered by the 75th percentile Iwb. Fill color corresponds to a narrative quality range using the 50th percentile value for each river and reach.*

on the St. John R. while the other rivers have historical or current day access for American eel only. The fall off to ME IBI values reflective of BCG Levels 4 and 5 is the predominant condition for the remaining rivers which includes those that lie south of the 44-45 N latitude. The DIBI "effect" was the most pronounced for the Lower Kennebec in 2002-3 and 2008-9 where diadromous fish restoration has been active and has included most of the species that are included in the DIBI metrics. The addition of the DIBI boosted the BCG from Level 5 to Level 4 in other coastal rivers throughout the study area including the Presumpscot, lower Connecticut, lower Merrimack, Taunton Rivers.

The ordering of rivers by the Iwb was in contrast to the ME IBI + DIBI with the possible exception of the Lower Kennebec R. in 2002-3, where a remarkable increase in fish abundance and biomass was observed in the immediate years following the Edwards Dam removal (Yoder et al. 2006b). The contrast in rankings is attributable to the Iwb weighting all species the same including attributes that are negative to the ME IBI including the abundance of blackbasses, macrohabitat generalists, eurytherms, and non-native species. The Iwb does not incorporate many of the attributes of the BCG and in some cases counteracts its intent, thus exhibiting some of the problematic outcomes of judging fish assemblage quality on abundance and diversity alone.

CHAPTER 4: KEY LIMITING STRESSORS IN NEW ENGLAND LARGE RIVERS

The preceding analyses illustrated the distribution of stressors across New England and provide an estimate of biological condition using the ME IBI, with and without its diadromous component (DIBI). While some inferences can be made simply by examining the plots and distributions of stressors and assemblage indices and metrics, it is also important to explore relationships using the raw fish assemblage data linked to stressor parameters to provide estimates of the strength of any apparent relationships. This process can also reveal patterns that were not evident from examining the univariate plots. The results of multivariate analyses however, require a well-founded biological interpretation of the results to ensure that they reflect ecological reality and are not simply artifacts that can be inherent to complex data sets. The stressor databases lack sufficient chemical information hence we instead relied on modeled outputs. Also lacking are geomorphic data that might provide additional insights into habitat patterns and daily flow data that could have provided more detailed insights into the influence of altered hydrology. Even so, our analyses do provide an initial assessment of key limiting factors at the categorical level.

Principal Component Analysis (PCA) Results

Principal Component Analysis (PCA) by variable category was conducted because sample sizes were not equal across all categories (e.g., fewer water chemistry data were available than for other categories). This analysis was employed to choose variables for further examination with NMS ordination coding variables and for selecting variables for developing weighted stressor values (WSV) for individual fish species.

National Land Cover Data (NCLD)

We obtained NLCD from the NHDPlus data set which included:

- Cumulative land cover at the downstream end of each NHD flow line reach for which each fish assemblage site was associated. This essentially includes the all of the aggregate land cover upstream of a sampling site.
- Local land cover includes only the land cover within the immediate catchment where a flow line was located. As such it includes only the land cover in close proximity to a sampling site.

Cumulative Land Cover Data

The first and second vectors for the PCA analysis contained a majority of the variation in land cover variables (Table 16). Important variables included developed land, natural land cover types, forest-shrub, water, and agricultural land cover types. The natural land cover category is a combination of multiple land cover types including water, wetland and forest-shrub land cover types. For further stressor analyses we selected natural, agricultural, and developed land cover types which contributed the highest loading to the first two factors of the PCA.

Local Land Cover Data
Local land cover data, which represents land cover within the immediate catchment within a NHDPlus reach, showed a similar pattern to cumulative land cover except that agricultural land cover types showed a stronger relation to the second vector wetland land cover types loaded more heavily (Table 17). Developed, agricultural, and natural land cover types (which include water and wetland as well as forest shrub types) were selected for further analysis.

***Table 16**. PCA results for cumulative NLCD from all sites with available fish assemblage and NLCD data in this study. The upper table contains eigenvalues and variability of each factor and lower table the percent of the variability attributed to each variable.*

	F1	F2	F3	F4	F5	F6	F7
Eigenvalue	3.423	1.328	1.098	0.698	0.453	0.000	0.000
Variability	48.903	18.977	15.679	9.972	6.467	0.001	0.000
Cumulative	48.903	67.881	83.560	93.532	99.999	100.000	100.000

	F1	F2	F3	F4	F5	F6	F7
c_waterpc	0.251	52.403	11.277	21.522	4.698	1.758	8.091
c_devpc	20.923	4.721	12.193	0.098	19.114	16.559	26.392
c_distpc	7.966	9.577	29.990	18.234	31.736	0.990	1.507
c_agpc	13.559	21.635	1.176	7.256	40.838	6.011	9.524
c_forshrpc	24.723	10.650	0.001	0.000	2.691	11.437	50.499
c_grasspc	0.000	0.000	0.000	0.000	0.000	0.000	0.000
c_wetlpc	7.206	0.997	33.651	52.833	0.433	0.894	3.987
c_natural	25.372	0.016	11.713	0.056	0.491	62.351	0.000

Habitat Data
Habitat data consisted of the QHEI and its component metrics, good and poor quality attributes, and the Hydro-QHEI which is a recombination of select QHEI attributes that are most directly related to the flow aspects of habitat. The first factor explained the greatest variability

***Table 17**. PCA results for local NLCD from all sites with available fish and NLCD data in this study. Upper table contains eigenvalues and variability of each factor and lower table the percent of the variability attributed to each variable.*

	F1	F2	F3	F4	F5	F6
n_waterpc	0.005	9.035	63.470	5.600	8.868	3.235
n_devpc	26.929	6.264	6.033	10.095	3.779	15.053
n_distpc	0.002	15.395	6.319	40.890	36.923	0.087
n_agpc	4.144	44.588	5.161	21.881	4.258	6.315
n_forshrpc	32.827	1.909	5.397	0.304	3.983	14.020
n_grasspc	0.000	0.000	0.000	0.000	0.000	0.000
n_wetlpc	0.037	20.968	13.168	21.057	41.081	0.921
n_natural	36.056	1.842	0.453	0.172	1.108	60.369

	F1	F2	F3	F4	F5	F6
Eigenvalue	2.659	1.300	1.180	0.965	0.895	0.001
Variability	37.983	18.568	16.857	13.788	12.792	0.011
Cumulative	37.983	56.551	73.409	87.197	99.989	100.000

in the habitat data (Table 18). The variables loading most strongly on the first factor included aggregate measures such as total QHEI score and attribute counts and the channel and riffle metrics (Table 18). The Hydro-QHEI loaded most strongly on the second factor with all other variables contributing much less suggesting this factor indeed represents flow related habitat influences. The total QHEI, good and poor habitat attribute counts, channel and riffle metrics, and the Hydro-QHEI and its current and depth metrics were selected for further analyses.

Water Quality

Two types of water quality data were linked with the NELR REMAP sites:

- Measured data collected during the fish sampling event (dissolved oxygen and conductivity); and,
- Total phosphorus and total nitrogen extracted from the New England SPARROW model.

***Table 18**. PCA results for habitat data from all sites with available fish and QHEI data in this study. Upper table contains eigenvalues and variability of each factor and lower table the percent of the variability attributed to each variable.*

	F1	F2	F3	F4	F5	F6	F7	F8	F9	F10	F11	F12
qhei	10.975	1.127	0.167	0.001	2.387	0.676	2.056	0.065	2.804	1.508	0.235	0.141
substrate	6.448	3.200	2.001	0.211	1.160	5.671	9.122	13.561	37.779	13.814	0.000	0.466
cover	0.774	4.636	48.238	3.912	0.000	0.706	0.236	28.559	7.812	1.763	0.201	0.005
channel	8.698	0.267	0.206	3.791	0.288	4.376	6.587	0.605	2.016	0.000	27.918	22.656
riparian	1.170	2.030	1.619	48.986	1.307	13.221	18.341	8.136	0.045	0.092	1.799	1.511
pool	7.870	0.000	0.450	11.629	0.403	5.232	1.680	0.034	0.729	0.991	49.817	8.186
riffle	9.008	0.096	1.093	6.916	0.124	0.422	1.517	0.470	0.787	0.631	0.010	56.260
gradient_v	2.082	0.874	0.093	0.096	20.221	49.929	25.059	0.414	0.003	0.060	0.429	0.550
gradient_s	0.879	1.272	3.161	5.624	64.105	11.386	3.807	6.405	1.301	0.147	0.260	0.110
hydroqhei	7.815	17.353	0.657	0.157	0.591	0.003	0.224	0.015	0.044	0.002	1.091	0.146
current	6.629	20.816	0.925	0.650	0.900	0.008	0.408	0.000	0.011	0.158	2.213	0.906
depth	2.925	32.295	0.884	6.449	1.553	0.370	1.982	0.072	0.003	0.148	5.460	0.097
wwh_attrib	10.753	1.218	0.100	1.858	0.000	0.364	0.013	0.009	0.473	0.026	5.236	0.256
mwh_attrib	10.154	1.237	0.327	0.754	0.207	0.809	0.181	1.682	0.681	2.195	3.116	3.052
mwh_h_att	2.625	3.808	30.563	0.338	2.755	5.971	2.931	28.948	12.923	7.263	0.086	0.983
silt	5.454	4.968	3.474	4.008	2.632	0.853	14.561	6.168	29.758	24.274	0.321	0.127
embedded	5.741	4.803	6.043	4.619	1.366	0.003	11.294	4.858	2.830	46.927	1.808	4.548

	F1	F2	F3	F4	F5	F6	F7	F8	F9	F10	F11	F12
Eigenvalue	8.294	1.867	1.308	1.097	1.000	0.829	0.750	0.463	0.371	0.326	0.251	0.208
Variability	48.791	10.983	7.696	6.454	5.883	4.878	4.413	2.721	2.185	1.916	1.477	1.222
Cumulative	48.791	59.774	67.470	73.923	79.806	84.684	89.097	91.818	94.003	95.919	97.396	98.618

Chemical water quality data collected by New England state programs were considered, but there were too few matches with NELR REMAP fish sites in WQX. The SPARROW results are modeled concentrations for the mean annual flow and for large rivers it can provide a reasonable estimate of comparative nutrient enrichment (Moore et al. 2004). The PCA analyses (Table 19) showed that the modeled TP and TN data loaded most strongly on the first factor, temperature and dissolved oxygen on the second factor and conductivity on the third factor. We decided to use all of these parameters in the later analyses, particularly because of the sparseness of water chemistry data in general.

Natural Environmental Gradients

This group of variables consists of factors that are mostly invariant to anthropogenic influences (e.g., latitude) or short-term anthropogenic influences (e.g., mean flow). The purpose of these analyses was to detect important natural gradients in New England large rivers. Latitude, river size, and slope variables loaded most strongly on the first three factors of the PCA (Table 20). Latitude, drainage area, and the NAHC temperature classification were selected as key environmental variables to explore in additional analyses. The NAHC temperature classification was selected over the mean catchment temperature because it is a more direct reflection of the potential fish assemblage because it is a reach based variable and mean catchment temperature was highly correlated with latitude (r=-0.923).

Table 19*. PCA results for water chemistry data from all sites with available fish and chemistry data in this study. Upper table contains eigenvalues and variability of each factor and lower table the percent of the variability attributed to each variable.*

	F1	F2	F3	F4	F5
Eigenvalue	1.625	1.135	0.882	0.866	0.492
Variability (	32.500	22.708	17.641	17.316	9.836
Cumulative	32.500	55.208	72.849	90.164	100.000

	F1	F2	F3	F4	F5
p10	0.406	48.527	3.129	47.882	0.056
p94	16.274	1.362	68.155	13.113	1.095
p299	0.227	48.243	14.933	36.596	0.001
sparrow_tp	42.958	0.970	3.519	0.385	52.169
sparrow_tn	40.135	0.898	10.264	2.024	46.680

Connectivity and Other Variables

Key connectivity variables included the number of downstream barriers, the number of impassable barriers, and counts of anadromous species. Other variables included percent base flow, population density (1990 census), and a soil permeability rating. Impassable barriers, anadromous species counts, a base flow index, population density, and soil permeability were selected for further analysis. We excluded downstream barriers because it was correlated with impassable barriers (r = 0.867) and the number of anadromous species (r = -0.606; Table 20).

Nonmetric Dimensional Scaling (NMS)

NMS ordination was used to further explore associations between fish assemblage data and the key environmental and stressor variables that were selected based on the PCA analyses. It

***Table 20**. PCA results of environmental variables from all sites with available fish and environmental data in this study. Upper table contains eigenvalues and variability of each factor and lower table the percent of the variability attributed to each variable.*

	F1	F2	F3	F4	F5	F6	F7	F8	F9	F10	F11
Eigenvalue	3.481	2.586	1.608	1.026	0.857	0.570	0.316	0.295	0.149	0.092	0.021
Variability	31.649	23.506	14.614	9.324	7.788	5.184	2.872	2.685	1.357	0.834	0.187
Cumulative	31.649	55.154	69.768	79.093	86.880	92.065	94.937	97.621	98.979	99.813	100.000

	F1	F2	F3	F4	F5	F6	F7	F8	F9	F10	F11
lat	24.238	3.697	0.002	0.369	3.830	0.756	0.466	0.023	3.392	0.083	63.143
long	14.443	1.056	0.782	1.268	46.098	0.847	3.230	0.777	19.888	0.007	11.605
nahc_grad	0.467	9.030	34.035	4.348	0.659	0.464	1.358	49.213	0.399	0.019	0.008
nahc_temp	16.386	0.001	1.574	2.973	6.756	37.592	29.404	0.437	4.864	0.009	0.003
slope	0.189	4.233	44.634	2.398	0.037	0.050	0.010	47.412	0.524	0.456	0.057
meanflow_	1.942	29.370	4.177	0.007	0.011	7.319	3.909	1.008	1.987	49.562	0.707
meanflow_	0.016	0.009	4.915	81.390	4.079	8.481	0.342	0.117	0.176	0.473	0.002
strm_order	4.861	17.616	1.868	5.757	6.719	11.997	50.107	0.446	0.056	0.552	0.019
meanprecip	9.636	8.145	0.458	0.406	31.304	22.993	2.153	0.098	23.876	0.253	0.678
meantempe	24.962	1.515	0.003	0.001	0.439	2.282	0.064	0.467	44.831	1.744	23.692
drnarea	2.858	25.330	7.552	1.081	0.069	7.218	8.958	0.000	0.006	46.841	0.086

seemed reasonable to assume that a relationship between fish assemblage composition and latitude would exist given the wide geographic distribution of the study. The association of fish assemblage similarity with latitude appears to be a relatively strong one based on the NMS ordination, but we already know that the natural latitudinal gradient is confounded by other environmental and anthropogenic influences. The data points in Figure 33 are coded by the integer value of the latitude of each data point. There appears to be a strong gradient for latitude with the northern-most sites above latitude 45°N (Figure 34) and especially so for sites north of 46°N latitude. Sites at latitudes below 45°N tended to exhibit a wider degree of overlap.

An examination of other variables that are correlated with latitude indicate that the NEAHC temperature classification (1=cold . . . 4=warm) and soil permeability were also highly correlated with latitude (Table 22) as were the various land cover variables, especially cumulative natural and agricultural land cover. The vectors on the NMS ordination represented the three strongest continuous environmental variables of the variables we selected based on the PCA analyses, of which latitude was the strongest. The other two variables included QHEI riffle score and counts of anadromous species from the NAC (Martin and Apse 2011). These three variable categories seem to make ecological sense in explaining fish species composition

***Table 21**. PCA results of connectivity and miscellaneous variables from all sites with available fish and connectivity data in this study. Upper table contains eigenvalues and variability of each factor and lower table the percent of the variability attributed to each variable.*

	F1	F2	F3	F4	F5	F6	F7
ust_barr	10.154	38.407	0.004	0.692	8.514	41.243	0.986
dst_barr	33.177	4.526	0.183	0.247	3.935	1.436	56.495
dst_impass	29.203	3.726	0.034	3.203	25.411	0.081	38.342
anadcount	24.407	0.295	5.708	0.005	34.668	33.898	1.019
baseflow_i	0.444	26.127	43.623	0.163	21.927	5.616	2.100
pop90	2.607	6.331	8.565	77.309	3.385	1.651	0.153
soilperm	0.008	20.589	41.882	18.381	2.159	16.075	0.905
	F1	F2	F3	F4	F5	F6	F7
Eigenvalue	2.557	1.388	1.119	0.939	0.515	0.378	0.104
Variability	36.535	19.826	15.986	13.412	7.359	5.400	1.482
Cumulative	36.535	56.361	72.347	85.759	93.118	98.518	100.000

changes which would be expected as the proportion of cold water vs. warmwater species varies along the latitude/temperature gradient. The proportion of fluvial specialist and dependent species changed with differences in riffle/run habitat and especially with the lack of such features. Anadromous species changed in relation to the presence of impassable barriers that limit or inhibit movements of these fish species (Figure 35).

Riffle/Run Features

The QHEI riffle score was weakly correlated with one NMS axis (r^2 >0.2; Figure 36). The QHEI riffle score is composed of both riffle and run depth and quality which includes the predominant riffle/run substrates and embeddedness attributes. The sites that are most distinct on the ordination graph tended to have a maximum riffle score ≥8 which indicates clean substrates and deep, fast riffle/run attributes. Sites with riffle scores = 0 in large rivers are most likely low gradient sites predominated by pool habitat or impounded riverine where riffle/run features have been lost due to the artificial ponding effects. The riffle score was most correlated with the Hydro-QHEI which is a compilation of the current and depth-related attributes of the QHEI, the number of “good” habitat attributes (which include all of the flow-related attributes), and gradient, all of which are negatively correlated with silt cover which is associated with slow or non-flowing depositional habitat types (e.g., impoundments).

***Table 22**. Pearson correlation coefficients for selected stressors and sensitive and tolerant fish species richness measures and with the Maine IBI. Yellow shaded cells have r^2 values > 0.30.*

Parameter	Metric Form	Sensitive Species			Tolerant Species		
		Pearson r^2	Threshold Relationship	Variable vs. Maine IBI (r^2)	Pearson r^2	Threshold Relationship	Variable vs. Maine IBI (r^2)
Total QHEI	#	0.16	Strong	0.36	0.09	Weak	0.04
	%	0.08	Mod. Strong	0.15	0.07	Moderate	0.01
QHEI Riffle	#	0.12	Strong	0.53	0.04	Weak	0.03
	%	0.08	Mod. Strong	0.25	0.06	Weak	0.03
QHEI Substrate	#	0.10	Strong	0.50	0.13	Weak	0.14
	%	0.08	Mod. Strong	0.28	0.09	Weak	0.09
Hydro-QHEI Total	#	0.16	Mod. Strong	0.26	0.13	Mod. Weak	0.07
	%	0.09	Moderate	0.09	0.05	Weak	0.07
Hydro-QHEI Current	#	0.18	Mod. Strong	0.34	0.05	Weak	0.01
	%	0.11	Moderate	0.13	0.02	Mos. Weak	0.02
Hydro-QHEI Depth	#	0.07	Weak	0.58	0.12	Weak	0.09
	%	0.09	Mod. Weak	0.29	0.06	Weak	0.07
Cum. Nat. Land Cover	#	0.09	Moderate	0.62	0.22	Mod. Weak	0.10
	%	0.06	Strong	0.30	0.22	Weak	0.07
Sparrow TP	#	0.08	Mod. Strong	0.50	0.13	Weak	0.09
	%	0.07	Mod. Strong	0.28	0.04	Weak	0.05
Sparrow TN	#	0.13	Mod. Strong	0.60	0.18	Mod. Strong	0.06
	%	0.08	Weak	0.32	0.08	Weak	0.04
Dst. Impassable	#	0.15	Mod. Strong	0.02	0.09	Weak	0.09
	%	0.07	Mod. Strong	0.04	0.03	Weak	0.03
Mean Annual Air Temperature	#	0.42	Strong	0.55	0.42	Strong	0.11
	%	0.29	Strong	0.28	0.20	Mod. Strong	0.08

[1] Relationship shows a unimodal threshold response.

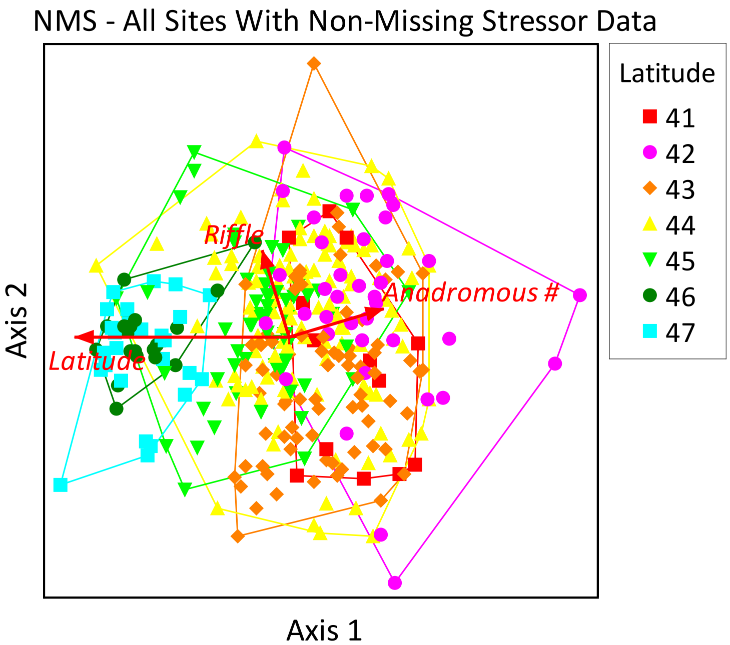


Figure 33. *Plot of latitude vs. cumulative percent land cover (natural – top; agriculture – bottom) for river sites in New England with fish data.*

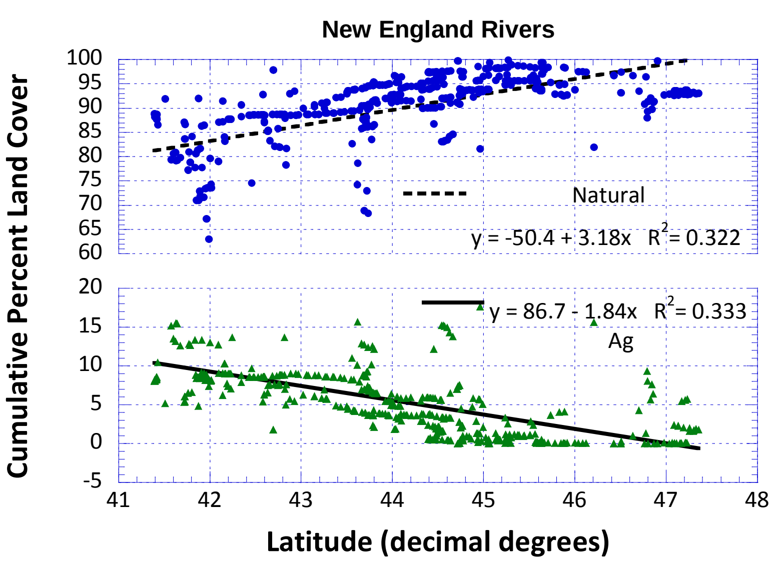


Figure 34. *NMS ordination of fish assemblage data (targeted and probability) grouped by the integer value of latitude.*

Connectivity

Connectivity is another potential influence on fish assemblage composition in New England large rivers as measured by the number of anadromous species identified in an NHDPlus reach based on the NAC study (Martin and Apse 2011). This assemblage association was somewhat more variable with latitude. However, in northern New England large rivers, where few or no anadromous species are expected, were relatively distinct from the sites with higher numbers of expected anadromous species (5-7). Differences in the distribution of anadromous species limited by impassable dams or where migratory routes are hindered by the frequency of smaller barriers is an obvious source of fish composition differences that are reflected in the NMS ordination. Other variables exhibited correlations with these key variables, but latitude, QHEI riffle score, and the number of anadromous species best captured these patterns in the fish assemblage data.

Figure 35 illustrates the NMS ordination of the fish data coded from the results of all of the cluster analyses. The cluster analysis groups represent the maximum variation that can be extracted from the species composition

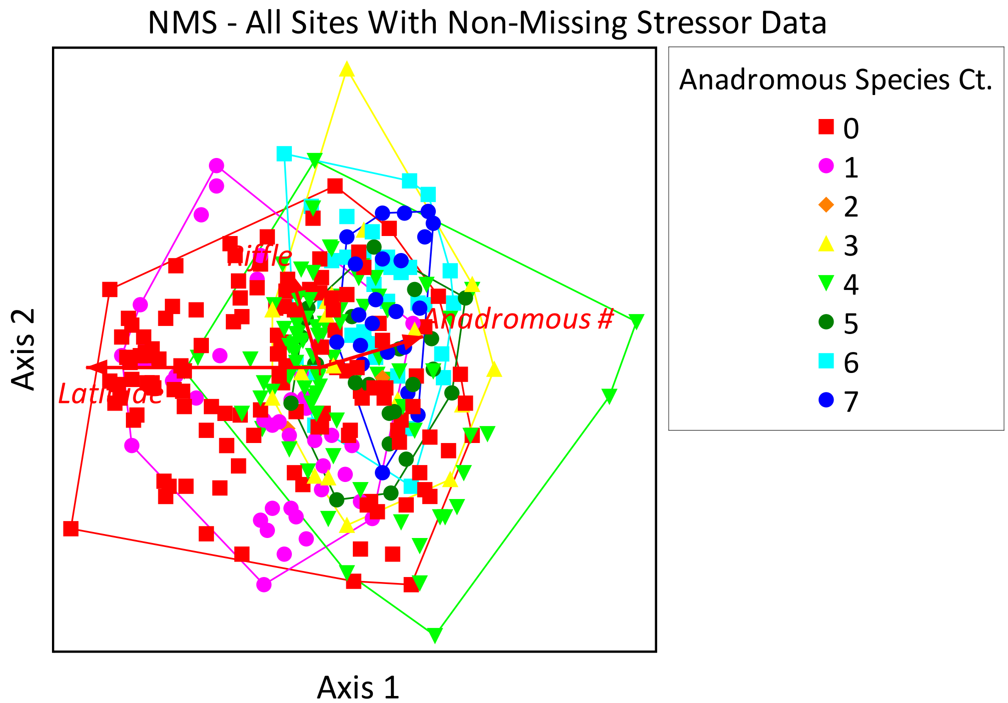


Figure 35. NMS ordination of fish assemblage data (targeted and probability) grouped by the count of anadromous fish species from the NAC study (Martin and Apse 2011).

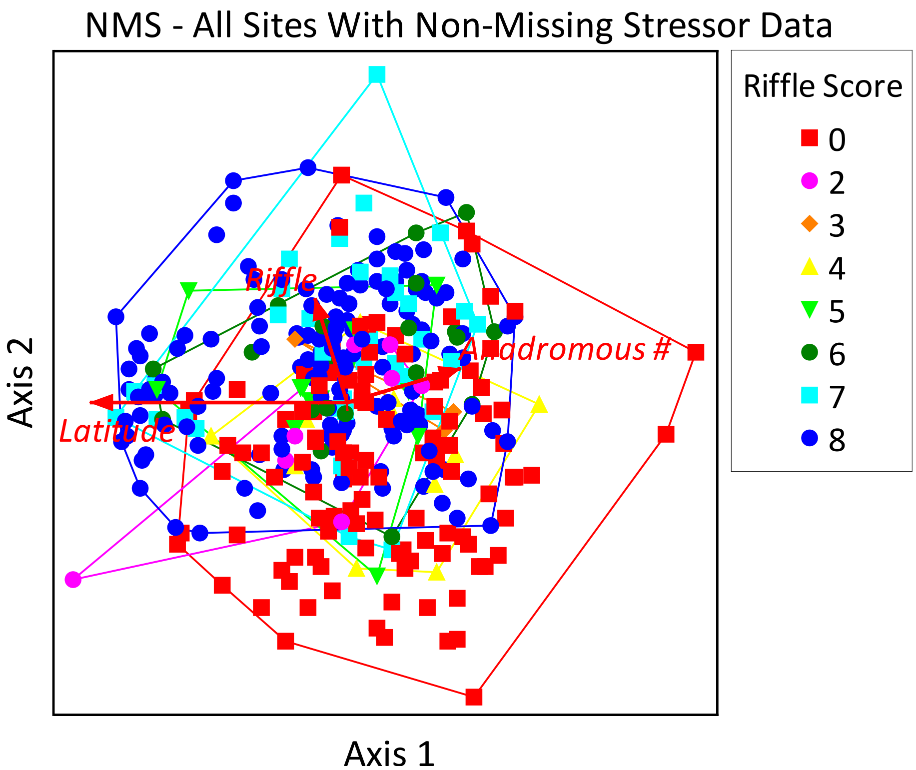


Figure 36. NMS ordination of fish assemblage data (targeted and probability) grouped by the QHEI riffle score from each site.

differences among sites for a cluster of six groupings. Although they represented maximum differences among groups the pattern and degree of overlap is not substantially different from the composite differences that emerged out of the NMS ordinations in relation to groups of sites distinguished by latitude, riffle score, and anadromous species. Figure 37 includes labeling based on an interpretation of the ecological relevance of each cluster.

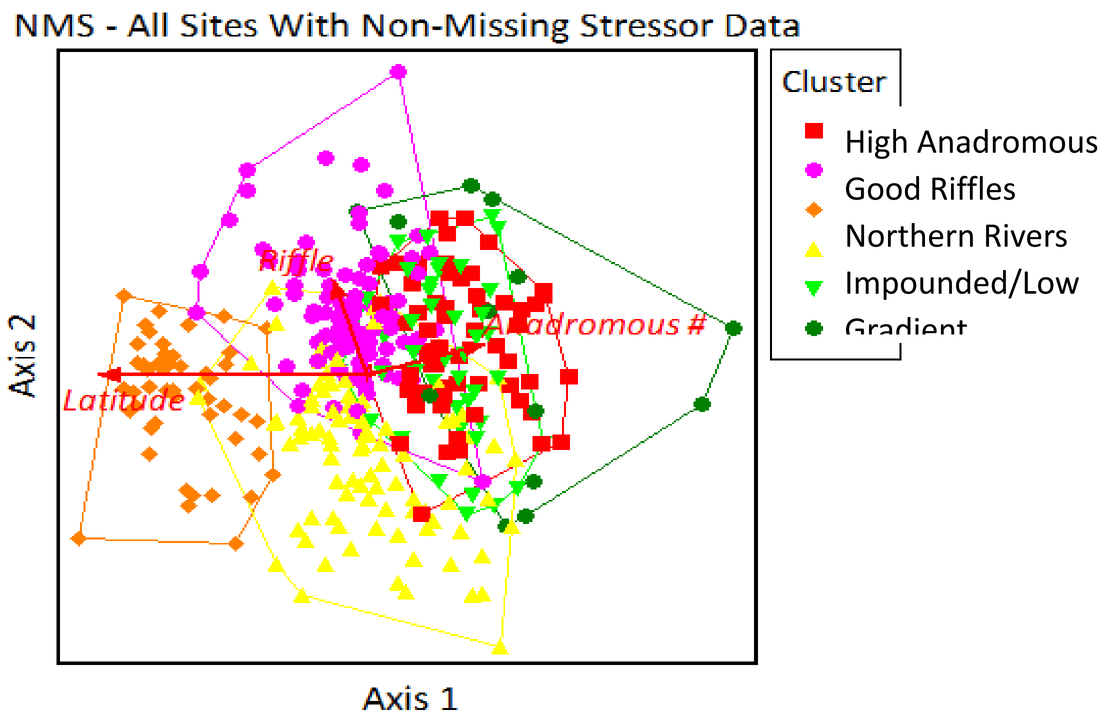


Figure 37. *NMS ordination of fish assemblage data (targeted and probability) grouped by the 6-variable cluster analysis group membership for each site. Legend reflects interpretation of the meaning of each cluster.*

Weighted Stressor Values (WSVs)

Weighted stressor values (WSVs) were derived for each species in the NELR REMAP dataset that occurred at 10 or more sites (Appendix B). This process helped to enhance the understanding about how common stressors are related to fish assemblage composition and condition. This also included correlations between the stressor-specific metrics and the current ME IBI metrics to identify how they responded to the gradient of stressors that we examined across New England.

Individual Stressor-Based Biological Metrics

From a regional perspective based on NMS ordination it appears that dams, impoundments, and flow-related habitat alterations are the predominant influences on riverine fish assemblages in New England. These stressors coincide with the density of development and human population and likewise occur along a latitudinal gradient that reflects both zoogeographical and thermal regimes. Local scale stressors are also limiting, but these also comprise a dimension that contributes to observed variability in the regional scale results. We were not able to more directly examine the influence of chemical variables that are known to have influenced New England rivers from diverse point sources, particularly prior to full implementation of controls under the CWA prior to the late 1990s. Heavy metals and ammonia, for example, were commonly listed as causes of impairment in state 305[b] reports in the 1980s and early 1990s. While the contributions of these pollutants to aquatic life impairment have undoubtedly decreased, chemical pollution from urban sources still contribute significant loadings of these and other chemical parameters to selected New England rivers. The conductivity data collected during the project suggests continuing influences from point sources in segments of rivers that have historically had degraded biological assemblages. Hence, we also examined local and river-reach scale patterns in key biological response indicators to enhance the overall stressor identification process. Such site- and reach-specific stressor identification requires detailed knowledge about how metrics, species, and/or species-traits respond to stressors at this scale. Stressor-specific species richness and proportional metrics were developed using WSVs to identify stressor-specific metrics including tolerant and sensitive groupings. These were then used to conduct site-specific stressor identification.

Site-specific Habitat Variables (QHEI)

The QHEI results were used as the principal indicator of local habitat quality. The QHEI WSV vs. median QHEI score of each species with at least 5 occurrences were used to develop a response gradient to site-specific habitat (Figure 38). Each data point was also coded by the tolerance assignments used in the ME IBI (see Table 7). The sequence of relative sensitivity in this classification is intolerant > sensitive > intermediate > moderately tolerant > tolerant.

Intolerant species were strongly associated with high QHEI scores and most, but not all tolerant species with lower QHEI scores; however, sensitive species showed a wider degree of variability in relation to habitat quality. The breadth and degree of habitat degradation in New England is generally less than what has been observed in more severely modified landscapes such as the Midwestern U.S. (Figure 36) and it seems likely that the distinction between sensitive and tolerant species would "widen" along a wider gradient in New England. In addition some New England species that are classified as tolerant do occur in high numbers in natural riverine habitats (e.g., adult white sucker), but they can also be equally abundant in modified habitats (e.g., juvenile and y-o-y white suckers). This was evident in some of the tolerant species WSVs for QHEI.

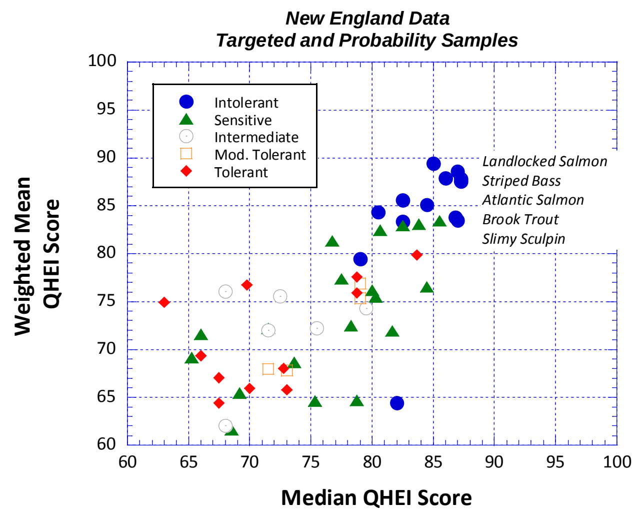


***Figure 38**. Plot of median QHEI score vs. QHEI weighted mean score (WSV) for fish species in New England rivers. Each pointed coded by Maine IBI intolerance designation. Species with the highest median QHEI labeled.*

The derivation of QHEI related metrics included both species richness and proportional metrics and as sensitive and tolerant formulations. Pearson correlation coefficients were derived through mean and best fit threshold regressions and also "by eye". The sensitive species richness metric showed a strong positive association with high QHEI scores (Figure 39, top) and with the ME IBI (Figure 39, bottom) exhibiting a strong threshold response. The QHEI tolerant species richness metric was only weakly correlated with either the QHEI or ME IBI. In addition to the reasons stated above, the presence of some tolerant species can be expected at good and exceptional quality sites. The QHEI sensitive proportional metric was positively correlated with the QHEI and ME IBI, but was weaker than the species richness metric. This may be related to the inherent variability in relative abundance estimates, especially relative to other species abundances (Figure 39). At QHEI scores <70 the percentage of QHEI sensitive individuals was usually lower. A predominance of QHEI habitat sensitive individuals was more common at QHEI >80. Higher proportions of QHEI tolerant individuals were generally found where QHEI scores were <60 (Figure 40, middle). Thus, QHEI seems to be a useful tool for understanding the influence of habitat disturbance on New England riverine fishes. Riverine habitat conditions in New England exhibited variation among sites, but the frequency and severity of degraded habitat was less when compared to rivers with more intensive agricultural land use such as in Ohio (Figure 39).

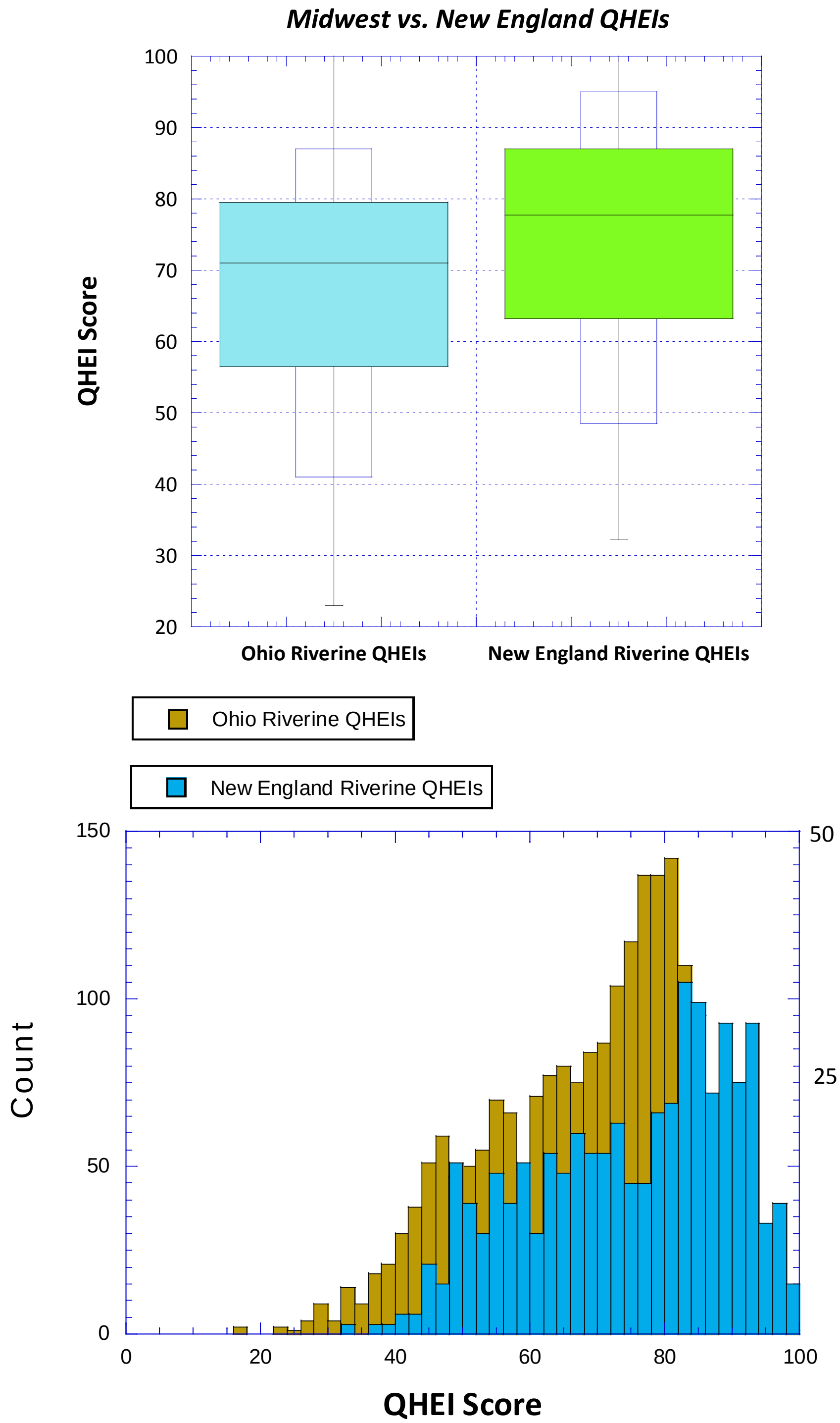


Figure 39. *Illustration of the distribution of QHEI score at riverine sites in Ohio vs. New England using a frequency histogram (top) and box and whisker plot (bottom).*

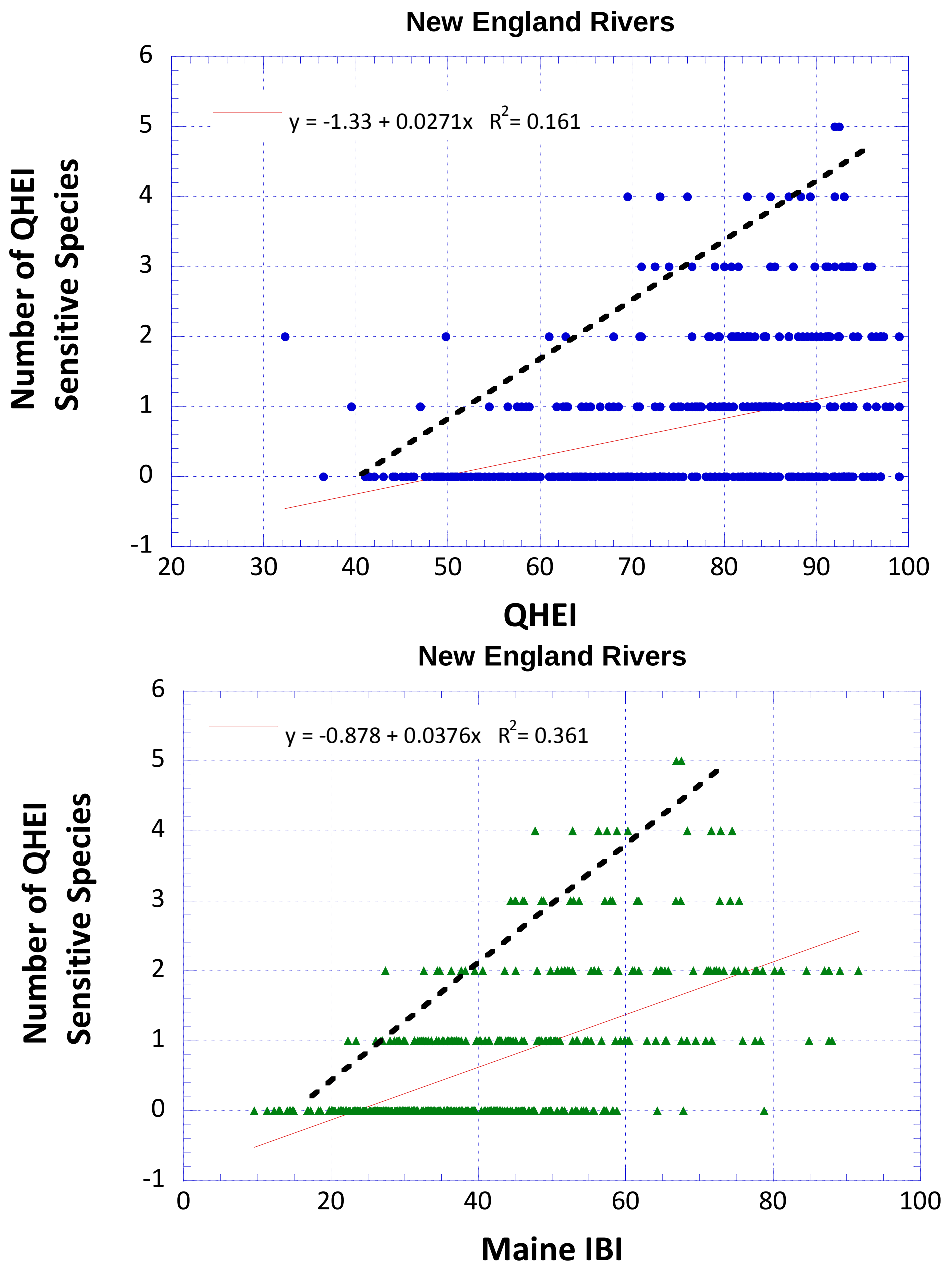


***Figure 40**. Plots of QHEI (top) and Maine IBI (bottom) and the number of QHEI sensitive species. Linear correlation is depicted by red lines and dotted lines were drawn by eye to represent thresholds. All targeted and probabilistic sites from New England rivers with matching QHEI data were included.*

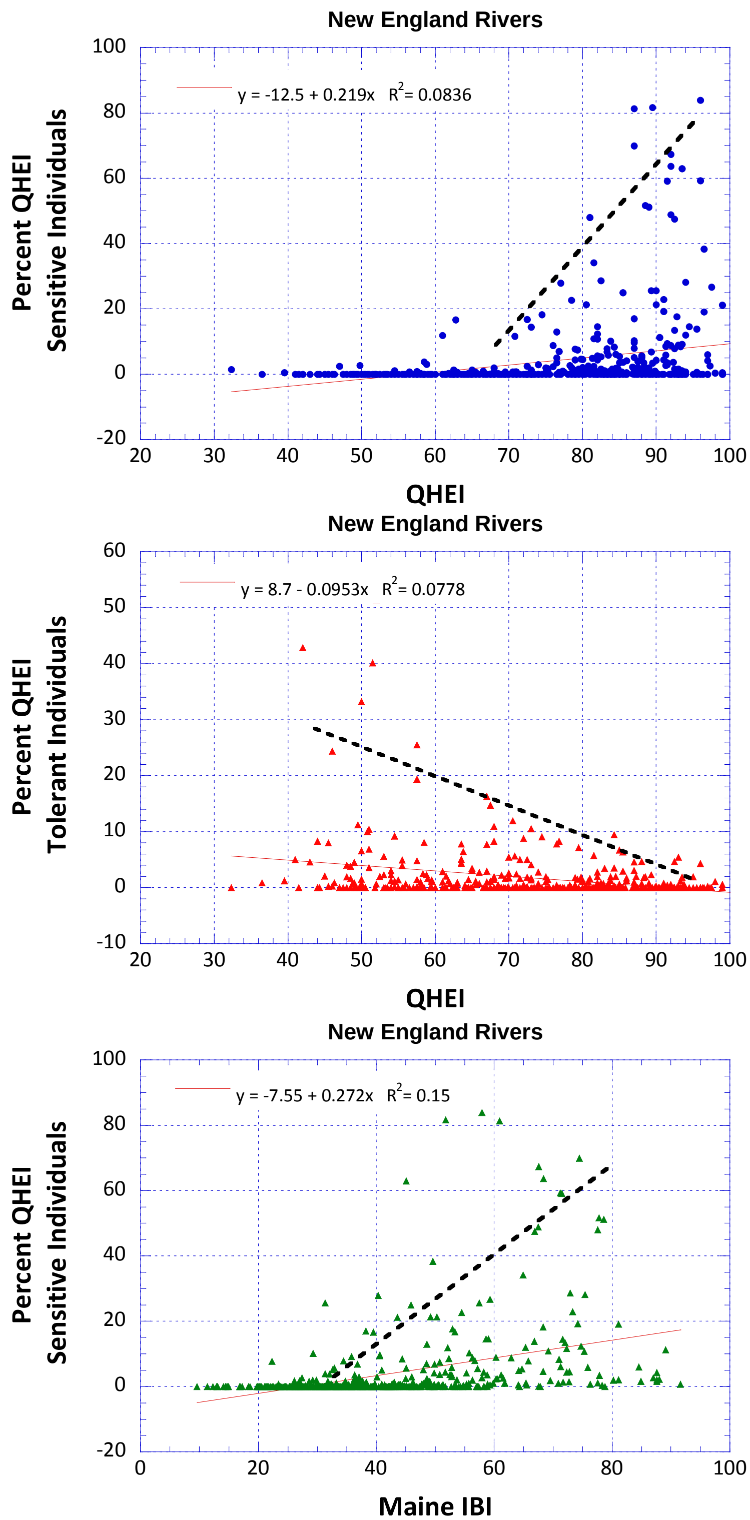


***Figure 41**. Plots of QHEI (top, middle) and Maine IBI (bottom) vs. the percent of QHEI sensitive (top, bottom) or tolerant (middle) individuals. Linear correlation is depicted by red lines and dotted lines were drawn by eye to represent thresholds. All targeted and probabilistic sites from New England rivers with matching QHEI data were included.*

QHEI Metrics

The QHEI riffle metric was correlated most strongly with the NMS vectors in the fish species ordination (see Figure 34). It also exhibited a strong association with fluvial specialist and fluvial dependent species as illustrated in the plot of the median riffle score for each species vs. riffle WSVs (Figure 42). All of the fluvial specialist and most of the fluvial dependent species are "pegged" to the maximum value of the QHEI riffle score (which is 8; Figure 38). Two fluvial dependent species that had lower riffle scores, tessellated darter and creek chubsucker, both prefer slower flows and were restricted to specific rivers in the NELR REMAP dataset. In addition, some of the species classified as diadromous (e.g., Atlantic salmon) were also correlated with high riffle scores and perhaps could be redefined as a fluvial specialists. As expected, most species classified as macrohabitat generalists by being less dependent on riffle features tended to have lower riffle score WSVs.

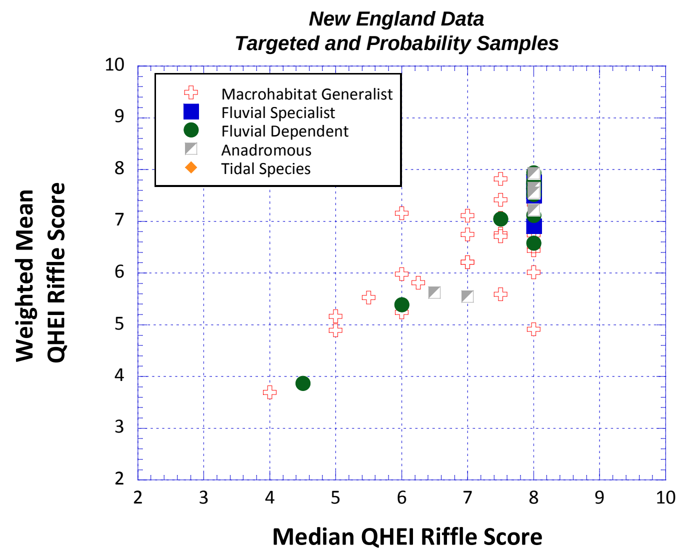


***Figure 42**. Plot of median QHEI riffle score vs. QHEI riffle weighted mean score (WSV) for fish species in New England rivers. Each pointed coded by Maine IBI fluvial designation.*

Riffle sensitive species richness showed a strong association with the riffle metric score, especially as a threshold response and also with the ME IBI (Figure 43) because it tracked the occurrence of fluvial specialist and dependent species. The correlation with the ME IBI ($r^2 = 0.53$) was among the highest for any of the habitat metrics. The proportional metric association with the ME IBI was weaker, but still comparatively strong ($r^2=0.25$) indicating that riffle sensitive species are an important signal for the ME IBI, again likely related to the fluvial guilds represented in the IBI.

Flow-Related Habitat QHEI Variables

The Hydro-QHEI is a recalculation of the QHEI created by selecting metrics most associated with current or depth aspects of habitat that are generally associated with greater relative flow. A plot of median Hydro-QHEI scores vs. Hydro-QHEI WSV scores for each species, coded by fluvial designation results in a similar separation of fluvial specialist and dependent species from macrohabitat generalists as did the QHEI riffle metric (Figure 43). As with the riffle metric, the Hydro-QHEI sensitive species richness metric showed the strongest relation with the ME IBI (Figure 44, bottom) and showed a strong threshold relationship with the Hydro-QHEI itself

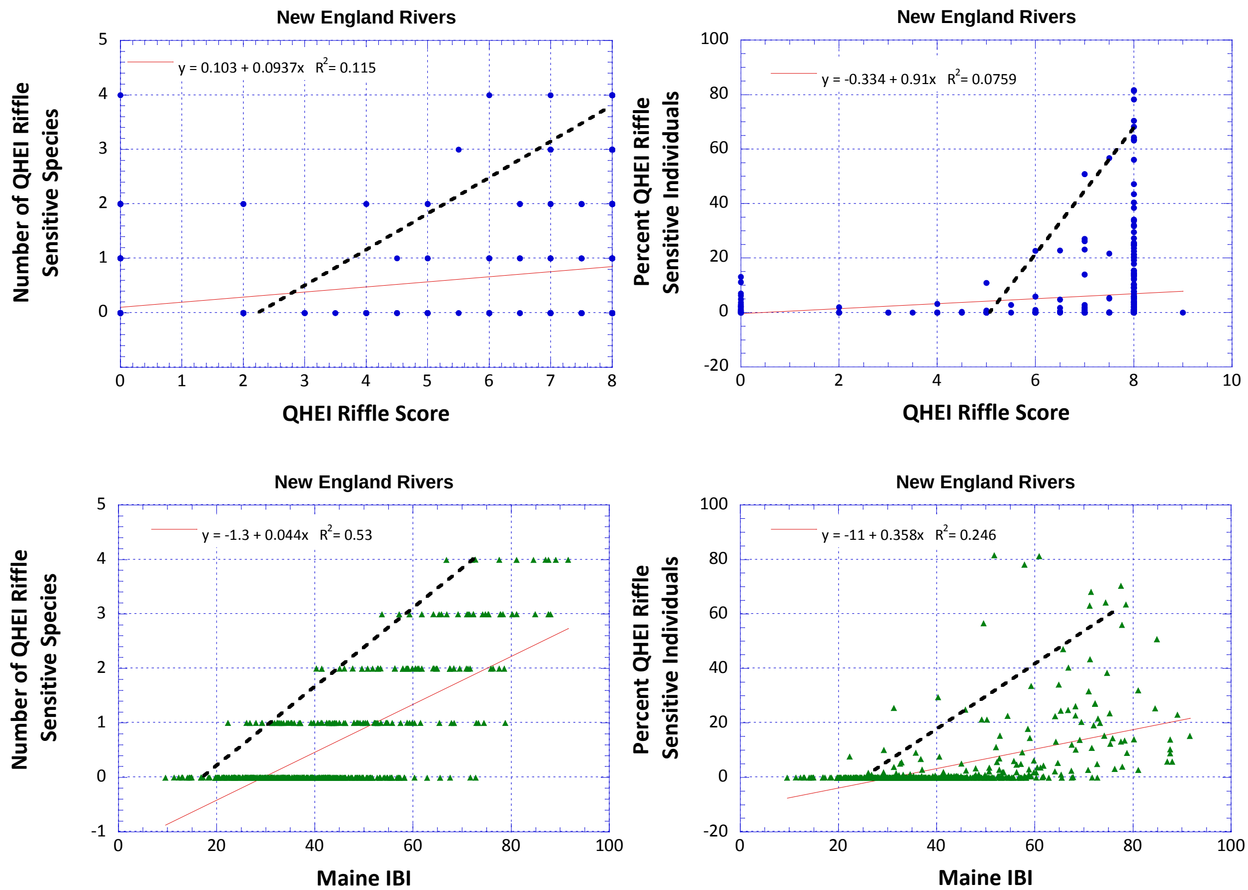


***Figure 43**. Plots of QHEI riffle score (top) and Maine IBI (bottom) vs. the percent of QHEI sensitive species richness (left) or percent QHEI sensitive individuals (right). Linear correlation is depicted by red lines and dotted lines were drawn by eye to represent thresholds. All targeted and probabilistic sites from New England rivers with matching QHEI data were included.*

(Figure 45, top). The Hydro-QHEI provides an easily collected correlate for more complex hydraulic variables such as the Froude number that has been suggested as a useful indicator for fish assemblage response to hydraulic complexity in rivers and streams (Lamouroux et al. 2002).

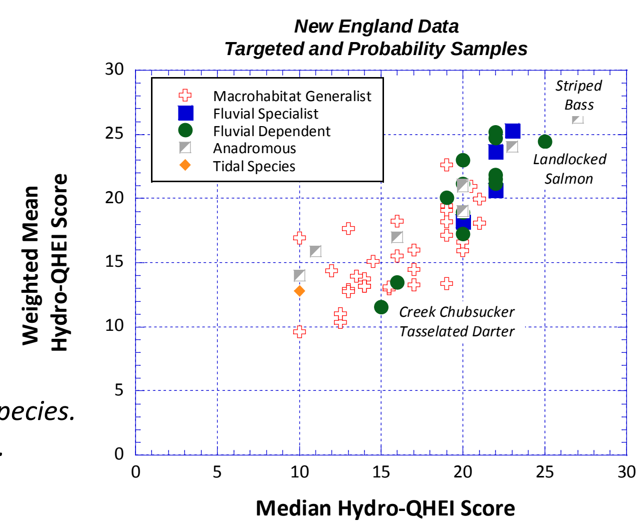
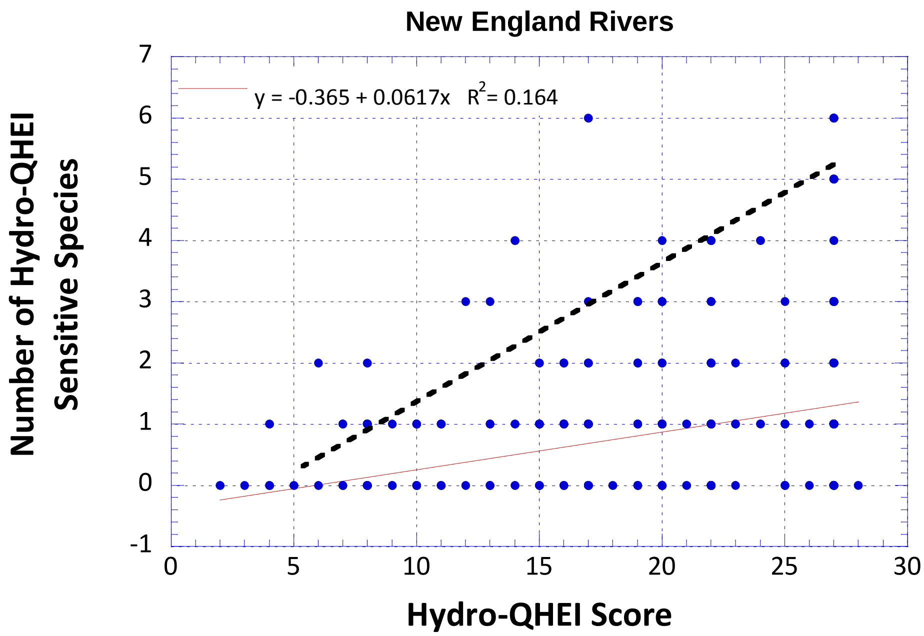


***Figure 44**. Median Hydro-QHEI score vs. Hydro-QHEI weighted mean score (WSV) for New England fish species. Each point is coded by Maine IBI fluvial designation.*



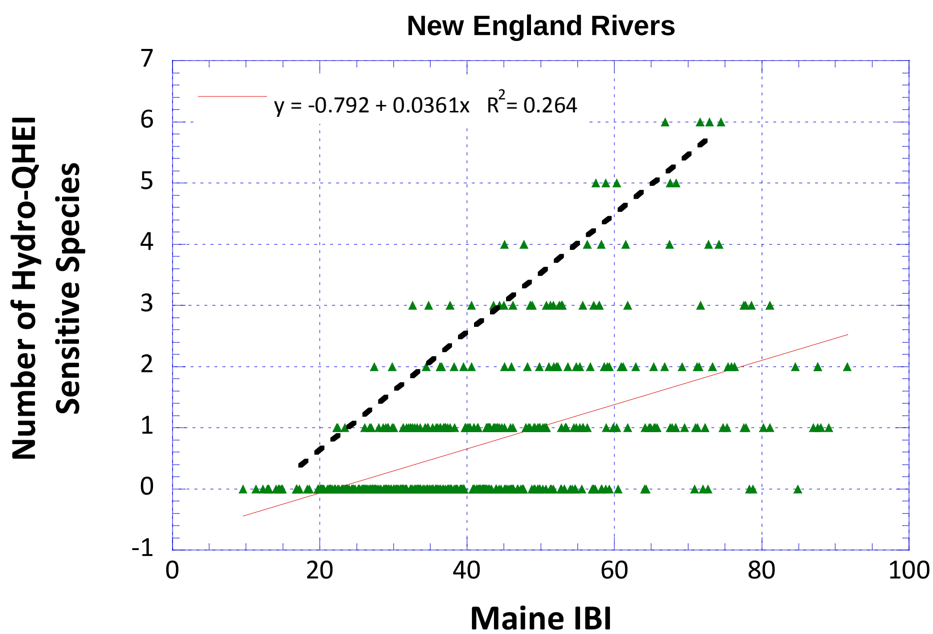


***Figure 45**. Plots of Hydro-QHEI (top) and Maine IBI (bottom) vs. the number of Hydro-QHEI sensitive species. Linear correlation is depicted by red lines and dotted lines were drawn by eye to represent thresholds. All targeted and probabilistic sites from New England rivers with matching QHEI data were included.*

Land Cover Metrics

These are widely used as a surrogate measure for anthropogenic influences. WSVs were derived for two land cover variables generated using the NHDPlus database. Intolerant fish species were most strongly associated with cumulative natural land cover >90% (Figure 46). Cumulative natural land cover represents the cumulative total percent of natural land cover types (e.g., forest, shrub, water, wetlands) at the downstream end of the NHDPlus reach within which a sampling site was located. Sensitive species showed a broader association with natural land cover and tolerant species were more variable across a range of natural land cover percentages (Figure 46, upper). Local natural land cover is within the same catchment in which the site is located. Intolerant and tolerant fish species showed the same relationship as with aggregate land cover, but sensitive fish species varied more widely (Figure 46, lower).

The cumulative natural land cover type sensitive species metric showed a strong threshold response to the original cumulative land cover variables with a sharp decline in the number of sensitive species and percent of sensitive individuals <90% natural land cover types (Fig 47, upper). These metrics also have a particularly strong association with the ME IBI.

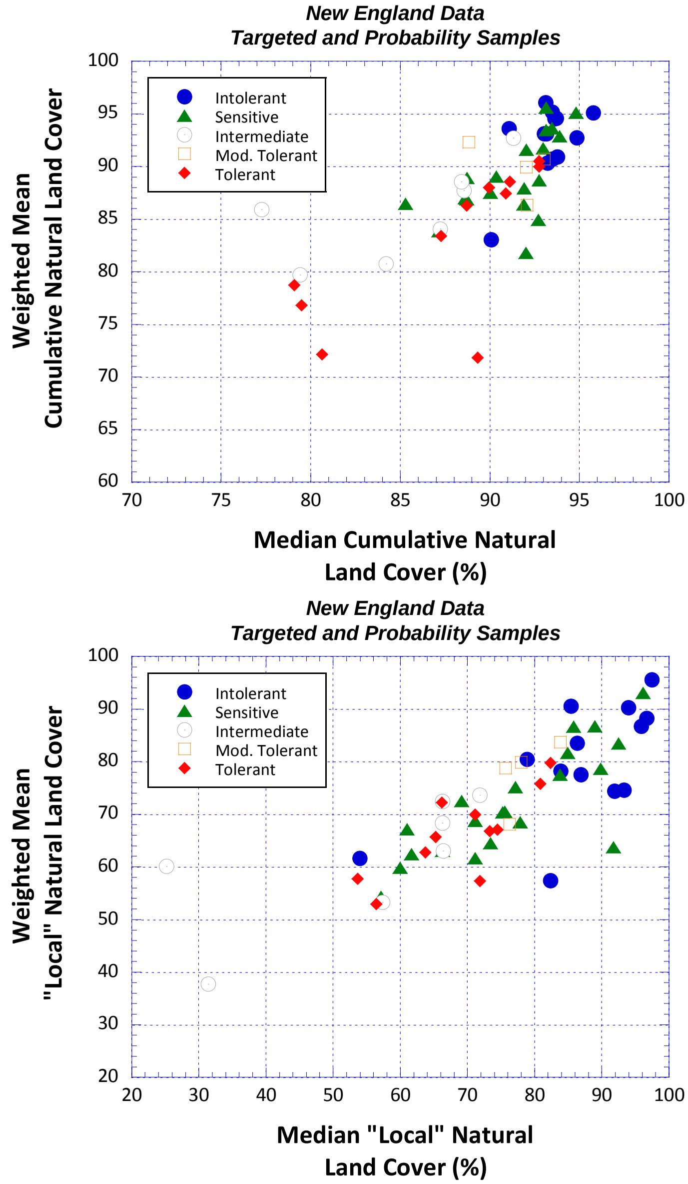


***Figure 46**. Plots of median cumulative (upper) and local (lower) natural land cover vs. weighted mean cumulative and local natural land cover (WSV) for fish species in New England rivers. Each pointed coded by Maine IBI tolerance designations.*

Generalized Human Disturbance Index (HDI)

We used the sum of individual stressor-response relationship results to calculate an overall HDI

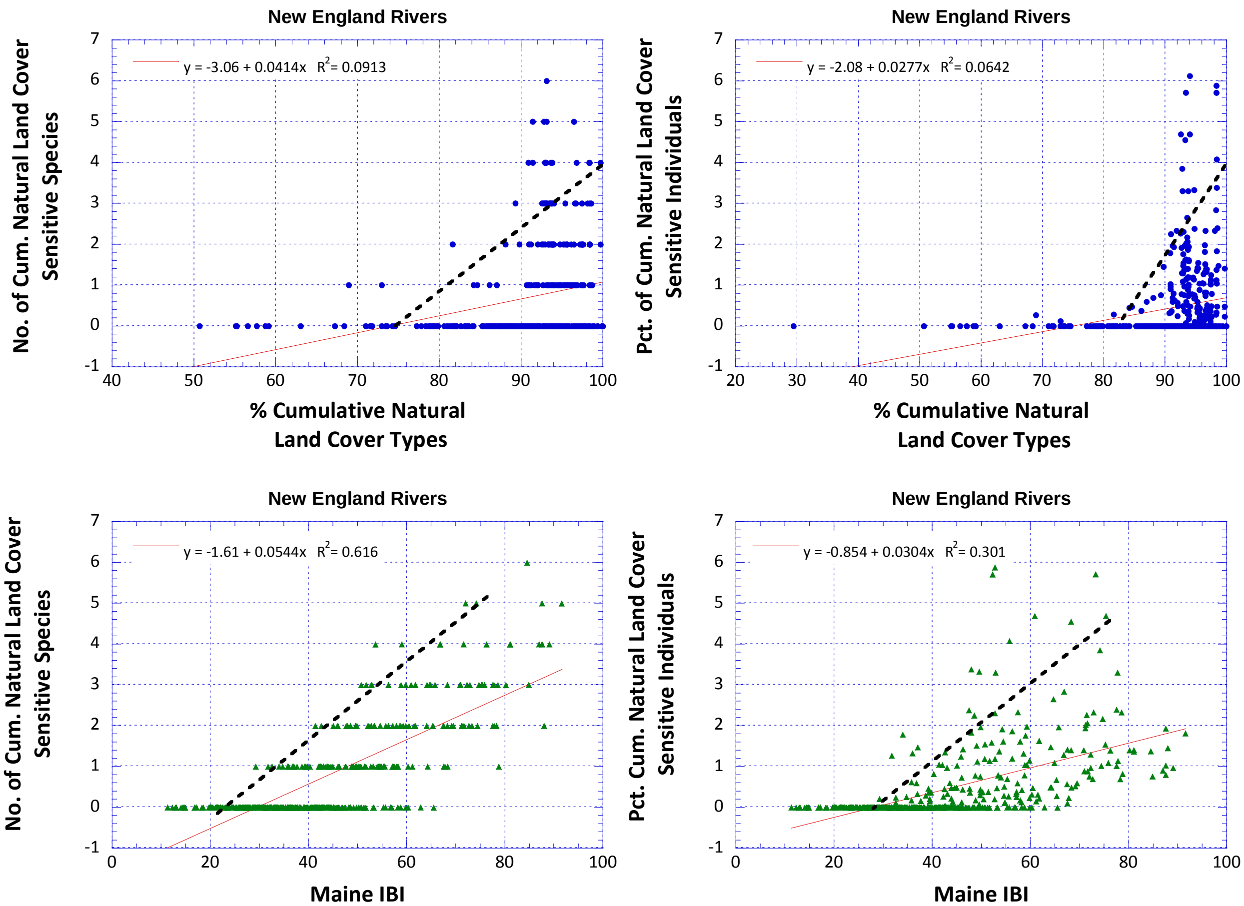


***Figure 47**. Plots of Percent cumulative natural land cover (top) and Maine IBI (bottom) vs. the number of natural land cover (cumulative) sensitive species richness (left) or percent of natural land cover (cumulative) sensitive individuals (right). Linear correlation is depicted by red lines and dotted lines were drawn by eye to represent thresholds. All targeted and probabilistic sites from New England rivers with matching QHEI data were included.*

based on a selection of human influenced stressor variables. We excluded environmental variables that do not reflect human disturbance (e.g., latitude). To calculate the index we identified, by eye, the portion of the curve where the stressor began to clearly influence fish assemblage condition as measured by the number of intolerant species. Curves and thresholds are illustrated in Figures 47 through 49. As discussed in the methods each metric was standardized on a scale of 1-10, weighted by the average of CCA coefficients from the 1st and 2nd axis and the final score standardized to a score of 0-100. We created a total HDI score and then a second where ecological connectivity stressors were eliminated (*i.e.,* downstream impassable barriers, all downstream barriers, and the NAC count of anadromous species in a reach). Both the ME IBI and number of intolerant species showed a clear threshold response to both forms of the HDI (Figure 50). The HDI, however, did not capture all of the variability in the ME IBI or intolerant species as there was substantial variation in the IBI at low HDI levels. As was shown earlier there is a strong latitudinal gradient in the fish assemblages of New England large rivers. In addition, we lacked information about some stressors such as local water quality variables (e.g., metals, toxicants).

The inclusion of the connectivity metrics introduced some variability where high performing sites as measured by the ME IBI occur where the HDI is also elevated (see circled areas on Figure 50). The arrows on these plots (Figure 50, left) identify sites where the HDI is elevated due to a loss of connectivity in northern latitude rivers (e.g., St. John R.) and where it excluded very low HDI scores (Figure 50, right). The loss of anadromous species is obviously a loss in overall biological condition, but other aspects of the assemblage can still perform well enough to score high IBIs or have high numbers of intolerant species where other human disturbances (e.g., habitat, fluvial, land use, and water chemistry stressors) are low. In addition, the connectivity values did not always recognize rivers that historically never had diadromous species, one such example being the St. John River in Maine which is above an impassable natural barrier (Grand Falls) just across the border in New Brunswick, Canada.

Maine IBI Stressor Relationships

This section focuses on exploring associations between the previously described stressors and the component metrics of the ME IBI and DIBI. The goal was to determine which metrics of the existing ME IBI and DIBI vary with the suite of stressor variables in our dataset. Some IBI metrics may not be strongly associated with these stressors because they were selected with other stressors in mind or the metric (e.g., DELT anomalies) responds strongly to only a portion of the stressor gradient (e.g., severely degraded conditions) that were not commonly observed or to a stressor category (e.g., toxic chemicals) that was not as well represented in the stressor variables. Pearson coefficients of determination were derived for sites where all of the variables co-occurred, which includes most of the probabilistic and targeted sites across New England. An alternate analysis included a visual examination of the spatial response of each metric to determine if there were any subregional, reach level, or site-specific patterns that might reveal stressors not included in the HDI. The Lake Champlain drainage was excluded since it falls outside the domain and applicability of several ME IBI metrics.

Relationship of Environmental Variables to the Maine IBI and Metrics

The most strongly associated environmental variables with the ME IBI were latitude (r^2=0.41) and mean annual air temperature (r^2=-0.48; Table 23). The NAHC thermal classification was also significant for this analysis (r^2=0.11). Habitat variables were significantly associated with the ME IBI and included the QHEI score (r^2=0.10), good QHEI attributes (r^2=0.11), QHEI riffle score (r^2=0.10), and channel score (r^2=0.10). Of the land use variables, locally developed land cover was the only significant variable (r^2=-0.11). All of these variables are related to both natural and human disturbance gradients, the latter of which generally increases from north to

***Table 23**. Pearson coefficients of determination (r^2) values between the Maine IBI and its individual metrics and environmental variables in New England large rivers. Data used only from sites where all environmental variables were available (i.e., no missing data). Yellow shaded shells are considered significant correlation coefficients with an r^2 >0.10 (p<0.05).*

Environmental Variable	Maine IBI	Native Species	%Native Cyprinids	Sucker Biomass	Native Salmonid Species	Benthic Insect.	%Black-basses	Fluv. Spec. Depend Species	Macrohab. Generalists	Steno-thermic Species	Non-guarding Lithophils	Non-indig. Species	DELT Anom.	Amer. Eel	Diad. Abund.	Clupeid Abun.	Diad. Species
Latitude	0.41	0.16	0.15	0.00	0.02	0.21	0.02	0.25	0.15	0.31	0.21	0.47	0.00	0.04	0.08	0.04	0.14
NAHC Thermal Class.	0.11	0.02	0.00	0.00	0.05	0.01	0.00	0.06	0.03	0.08	0.17	0.21	0.00	0.01	0.02	0.03	0.05
Mean Air Temp.	0.48	0.10	0.17	0.00	0.05	0.24	0.01	0.33	0.13	0.42	0.49	0.39	0.00	0.17	0.25	0.10	0.29
Dst. Barriers	0.04	0.03	0.00	0.00	0.04	0.00	0.02	0.03	0.00	0.10	0.05	0.01	0.01	0.18	0.25	0.08	0.26
Dst. Impassable	0.04	0.01	0.01	0.00	0.03	0.01	0.01	0.03	0.00	0.10	0.03	0.00	0.00	0.10	0.14	0.03	0.15
QHEI Total Score	0.10	0.06	0.01	0.00	0.01	0.02	0.01	0.05	0.11	0.12	0.11	0.05	0.02	0.04	0.03	0.00	0.01
QHEI Substrate	0.09	0.07	0.00	0.00	0.01	0.03	0.00	0.03	0.08	0.08	0.10	0.10	0.00	0.06	0.02	0.00	0.00
QHEI Cover	0.00	0.00	0.02	0.00	0.00	0.03	0.01	0.02	0.02	0.00	0.01	0.00	0.01	0.01	0.01	0.00	0.00
QHEI Channel	0.10	0.06	0.01	0.00	0.01	0.02	0.01	0.07	0.15	0.14	0.11	0.02	0.02	0.02	0.02	0.00	0.01
QHEI Riffle	0.10	0.07	0.01	0.00	0.01	0.04	0.03	0.06	0.09	0.16	0.13	0.04	0.01	0.01	0.01	0.00	0.00
Hydro-QHEI	0.08	0.05	0.02	0.01	0.01	0.02	0.02	0.04	0.09	0.11	0.08	0.04	0.02	0.02	0.01	0.00	0.00
QHEI "Good" Attributes	0.11	0.07	0.01	0.00	0.01	0.02	0.03	0.05	0.09	0.15	0.13	0.06	0.02	0.03	0.01	0.00	0.00
QHEI "Modified" Attributes	0.09	0.04	0.01	0.00	0.01	0.02	0.01	0.03	0.07	0.10	0.11	0.06	0.02	0.03	0.01	0.00	0.00
Conductivity	0.04	0.00	0.01	0.00	0.00	0.00	0.01	0.01	0.01	0.03	0.06	0.05	0.02	0.01	0.00	0.00	0.03
Dissolved Oxygen	0.00	0.03	0.00	0.10	0.01	0.00	0.01	0.00	0.03	0.00	0.01	0.00	0.00	0.02	0.00	0.01	0.04
Sparrow TP	0.03	0.01	0.00	0.00	0.00	0.00	0.00	0.02	0.00	0.02	0.03	0.02	0.00	0.00	0.02	0.04	0.02
Sparrow TN	0.01	0.00	0.00	0.00	0.00	0.01	0.00	0.01	0.00	0.01	0.04	0.03	0.00	0.00	0.02	0.01	0.03
Local Developed Land Cover	0.11	0.01	0.02	0.00	0.01	0.02	0.01	0.10	0.03	0.07	0.10	0.08	0.00	0.10	0.10	0.02	0.08
Local Natural Land Cover	0.08	0.01	0.02	0.00	0.01	0.01	0.01	0.06	0.01	0.07	0.12	0.08	0.00	0.08	0.09	0.02	0.06
Cum. Developed Land Cover	0.05	0.00	0.02	0.00	0.00	0.02	0.01	0.04	0.02	0.06	0.07	0.08	0.00	0.02	0.04	0.04	0.11
Cum. Natural Land Cover	0.05	0.00	0.02	0.00	0.00	0.01	0.01	0.04	0.00	0.06	0.09	0.09	0.00	0.04	0.07	0.09	0.12

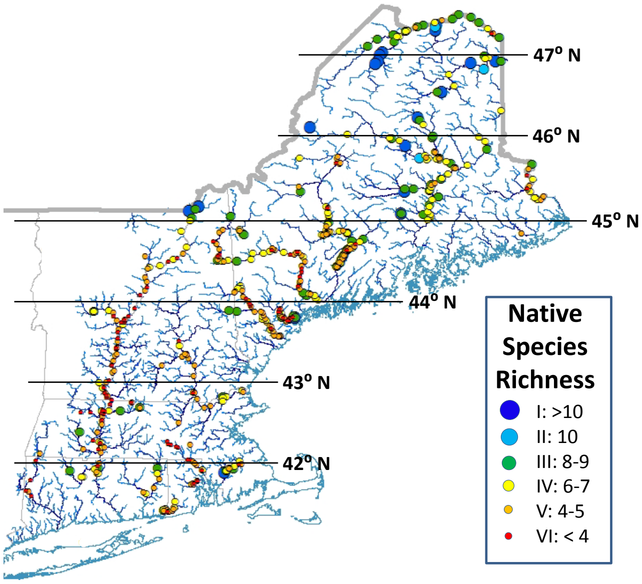


Figure 48. *Native species richness at NELR REMAP sites with symbols coded by the Maine IBI metric value that contributes to BCG Levels I-VI (Table 24).*

south across New England. A map of each metric was also included and color coded to its equivalent contribution to the BCG level depicted by the ME IBI (Table 24). This allowed for the visualization of general patterns across New England and also to highlight any site or river reach specific issues that can otherwise escape regionally focused analyses and which could warrant more detailed follow up investigations. Lake Champlain tributary sites in northwestern Vermont were not included as these contain several species indigenous to that drainage that are not considered native to the majority of New England.

Native Species Richness

The native species richness metric was positively associated with latitude (r^2=0.41) and negatively associated with air temperature (r^2=-0.48) the same as a number of other ME IBI metrics (Table 23). The strongest associations with native species richness were cumulative natural land cover (r^2=0.36) and habitat (e.g., QHEI - r^2=0.25; and substrate score, riffle score, good habitat attributes - all r^2=0.26). Other land cover variables were associated at less than r^2=0.20. For the most part this metric exhibited a north to south gradient with BCG Levels 1 (>10 native species) and 2 (<9 native species) and were restricted to selected sites in the St. John (Allagash R., Fish R.), Aroostook, upper Penobscot, and upper Connecticut R. drainages (Figure 48). No BCG 1 or 2 values were evident south of 45°N where BCG 5 (4-5 native species) and 6 (<4 native species) values predominated. These correspond to the previously described anthropogenic disturbance gradient and latitude.

Percent Native Cyprinid Species Richness

This metric showed significant results only for latitude (r^2= 0.15) and air temperature (r^2= 0.17)– all other values were $\leq$0.02 (Table 23). The mapped BCG "equivalents" for this metric show BCG 1 and 2 (>47%) concentrated north of 46°N, but this was not invariable (Figure 49). Isolated sites and river reaches had BCG 1 and 2 results at all latitudes throughout New England. BCG 5 and 6 sites were evident as far north as 46°N and this corresponds to the presence and abundance of black basses. Yoder et al. (2008) showed a negative relationship between native Cyprinids and Blackbass relative abundance which is likely the operative influence across New England. However, pollutional influences were also apparent, especially in the St. John R.

Table 24. *Maine IBI and Diadromous IBI metrics and BCG level equivalents with the method of estimation (e.g., regression equation, by eye). These cutoffs were used in the mapping of the Maine IBI metrics.*

IBI Metric	Biological Condition Tier						Equation/Method for BCG Cutoff Estimation
	BCG 1	BCG 2	BCG 3	BCG 4	BCG 5	BCG 6	
No. of Native Species	>10	10	8-9	6-7	4-5	<4	Numspec=1.83+0.018*IBI
No. Temperate Stenothermic Species	≥5	4	3	1-2	0	-	Stenotherms=-1.94+0.0713*IBI
No. of Non-Guarding Lithophilic Species	≥7	6	4-5	3	1-2	0	Lithophil NG=-1.36+0.0866*IBI
%. of Cyprinid Species*	≥58.2	>47.3-58.2	>32.8-47.3	>18.3-32.8	>3.8-18.3	≤ 3.8	% Cyprinids=-10.6+0.724*IBI
% Native Salmonids	≥4.20	>3.22-4.20	>1.91-3.22	>0.59-1.91	0	-	% Nat. Salm.=-2.03+0.0656*IBI
% Benthic Insectivores	≥39.2	>30-39.2	>17.7-30.0	>5.3-17.7	≤5.3	-	% Benth. Ins.=-19.3+0.616*IBI
% Black Bass	-	0	>0-9.2	>9.2-19.3	>19.3-29.4	≥29.4	% Blackbass=39.5-0.505*IBI
% Fluvial Specialists and Dependents	≥96.8	>86.3-96.8	>68.7-86.3	>43.9-68.7	>1.4-43.9	≤1.4	% Fluvial Specialists= -182+141*log(IBI)
% Macrohabitat Generalists	≤0.6	>0.6-9.4	>9.4-24.2	>24.2-45.0	>45.0-80.5	>80.5	%Macrohab. Gen.=234-118*log(IBI)
Adult White, Longnose Sucker Biomass	≥63.4	>52.8-63.5	>38.7-63.5	>24.6-38.7	>10.5-24.6	≤10.5	White, LN Sucker=-3.62+0.705*IBI
Non-Indigenous Species	0	1	2	3	4	≥5	By Eye
% DELT Anomalies	0	>0-0.30	>0.30-0.50	>0.50-1.0	>1.0-2.0	>2.0	Threshold by eye
Log American Eel Number/Km	≥2.5	>2.0-2.5	>1.5-2.0	>1.0-1.5	>0.5-1.0	≤0.50	By Eye
Log Diadromous Number/Km	≥2.5	>2.0-2.5	>1.5-2.0	>1.0-1.5	>0.5-1.0	≤0.50	By Eye
Log Clupeid Number/Km	≥2.5	>2.0-2.5	>1.5-2.0	>1.0-1.5	>0.5-1.0	≤0.50	By Eye
Diadromous Species Richness	5	4	3	2	1	0	By Eye

*excludes fallfish.

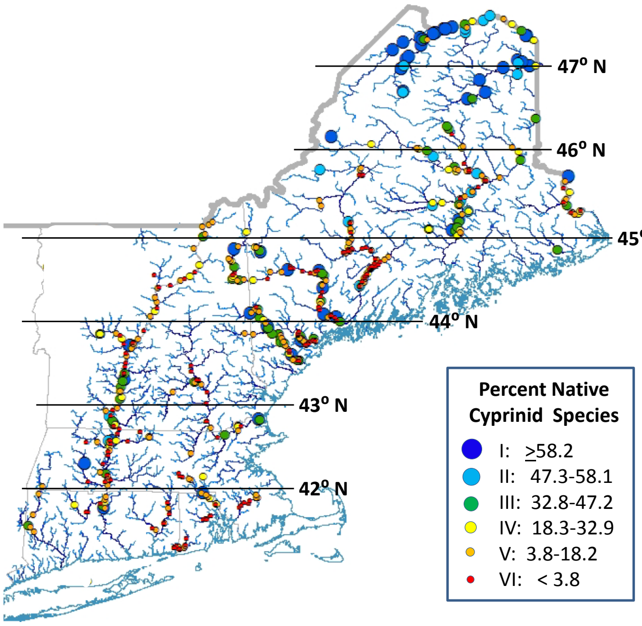


Figure 49*. Native Cyprinid species richness at NELR REMAP sites with symbols coded by the Maine IBI metric value that contributes to BCG Levels I-VI (Table 24).*

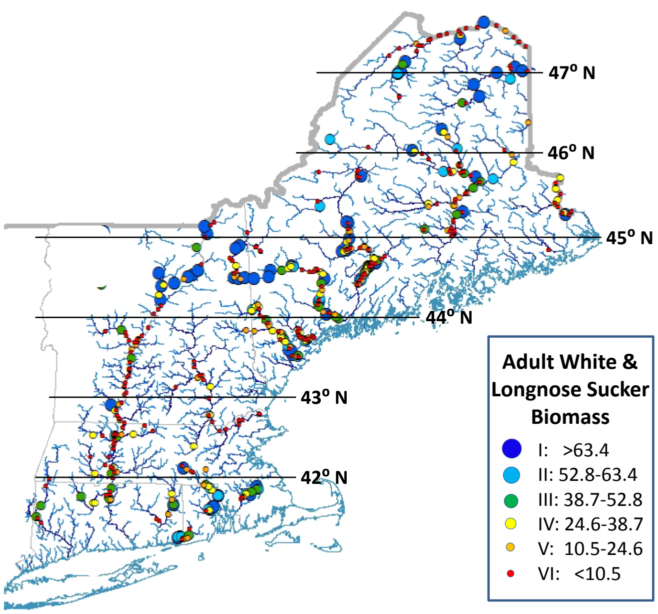


Figure 50a*. Adult white and longnose sucker biomass results at NELR REMAP sites with symbols coded by the Maine IBI metric value that contributes to BCG Levels I-VI (Table 24).*

downstream from the Madawaska, ME – Edmundston, NB area that has several large point sources. Similar patterns in the W. Branch Penobscot, Penobscot R., and St. Croix R., all of physical habitat caused by channelization, impoundment, hydrological alterations, or watershed scale modifications to habitat and sediment dynamics in rivers which have point sources were also evident.

Adult White and Longnose Sucker Biomass

This metric showed a significant relationship with D.O. (r^2=-0.10) only – all others were 0. The intent of this metric is to include the commonly occurring Catostomidae species in New England and the observed occurrence of adult white and longnose suckers in deep run habitats. As such it was included by Yoder et al. (2008) in the ME IBI on a representative ecological basis, not an observed statistical relationship with an *a priori* stressor gradient. It is the only biomass based metric in the ME IBI. While the statistical relationships with the selection of stressors used in the regional analyses were not revealing, the pattern of distribution in the BCG equivalents (Figure 50a) showed some interesting results. Generally, the north to south pattern evident for other metrics and the ME IBI was apparent. Most BCG 1 and 2 results occurred north of 44°N, but some significant instances occurred in selected rivers throughout New England and as far south as southwestern Rhode Island. The observed tendency of high biomass seemed to parallel sites that had high ME IBI scores where the cold water fish assemblage was the most intact and where non-native species were absent (Allagash R., Aroostook R.). However, this was not invariable as some isolated northern sites had low biomass for this metric (Moose R.). These instances tended to be low gradient reaches that lacked a prominence of the deep run habitat that this metric seems to best represent. The reduced biomass to a BCG Level 6 at all except two sites in the St. John R. may well be due to the presence of muskellunge *(Esox masquinongy)* which is an invasive predator in New England. Support for this includes the observation of BCG 1 and 2 values above the barriers that prevent the ingress of muskellunge in the Allagash and Fish Rivers and the numerous BCG 1 and 2 results in the Aroostook R., which is likewise restricted. This metric also exhibited reduced BCG level equivalents in rivers with a higher frequency of impoundments. White and longnose sucker biomass showed reduced values at impounded sites compared to riverine sites (Figure 50b), which reinforces the need to look at more than common stressor gradients on a landscape basis.

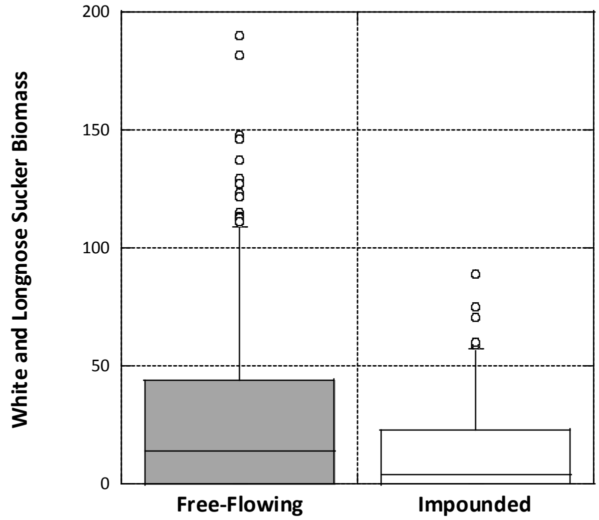


***Figure 50b**. Relative biomass (kg/km) of white and longnose sucker at riverine and impounded sites in NELR REMAP rivers.*

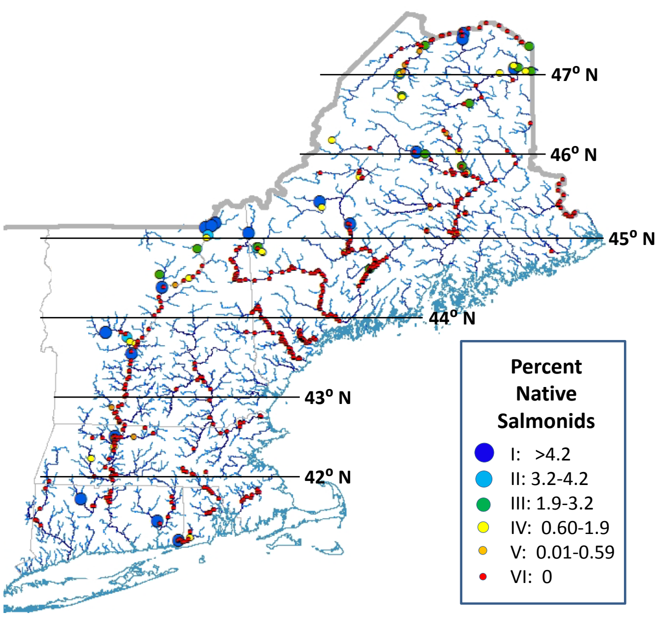


***Figure 51**. Percent of native salmonids results at NELR REMAP sites with symbols coded by the Maine IBI metric value that contributes to BCG Levels I-VI (Table 24).*

Native Salmonid Species

This metric was not significantly correlated with any of the stressor variables (Table 23). In keeping with the BCG basis of the ME IBI this metric includes only native Salmonids of which there are only two indigenous to New England rivers, brook trout *(Salvelinus fontinalis)* and Atlantic salmon *(Salmo salar)*. Each species has been historically reduced in range and abundance in large rivers throughout New England and as such their current distribution is likely not consistently related to the stressors we examined on a regional basis. However, there were sufficient results where these species did occur to at least reveal the general conditions under which they occurred at the higher BCG level equivalents (Figure 51). There were fewer than 20 sites with BCG Level 1 and 2 results throughout New England and these were surprisingly not restricted to the northern latitudes. The results at two sites in Connecticut were inflated by stocking of hatchery origin adult Atlantic salmon and these should be dismissed. Selected sites in Maine were the result of naturalized populations of landlocked Atlantic salmon and technically while these are not native, they do occur naturally at this latitude thus they were considered a legitimate part of this metric. The occurrence of native brook trout were limited to isolated sites in the Allagash R., Fish R., Aroostook R., Moose R., Magalloway R., and Rapid R. in Maine and the upper Connecticut R. in New Hampshire and in segments where smallmouth bass have not yet become established. In turn, this metric was either 0 (BCG Level 6) or in very low abundance (<1%) throughout the now established range of blackbasses throughout New England.

Benthic Insectivores

This metric showed significant results only for latitude (r^2= 0.21) and air temperature (r^2= 0.24) – all other values were $\leq$0.02-0.04 (Table 23). The mapped BCG "equivalents" for this metric show BCG 1 and 2 (>30%) concentrated north of 45°N (Figure 52). Only three isolated sites south of 45°N had BCG 2 results. BCG 5 and 6 sites were predominant throughout the remainder of New England.

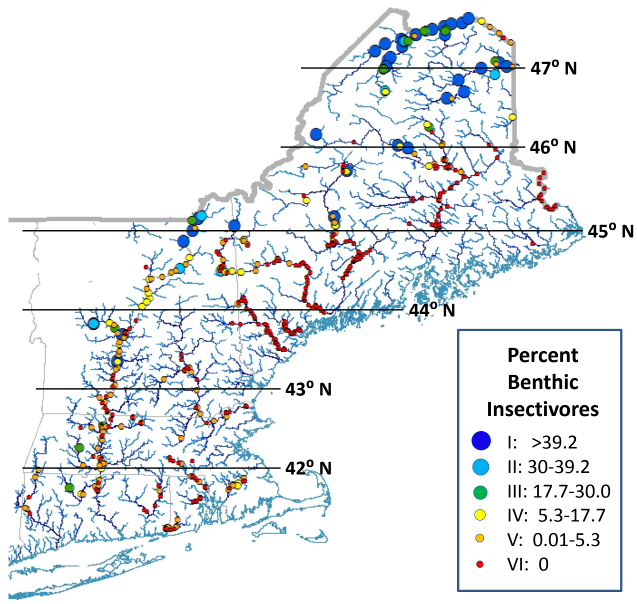


Figure 52. *Percent benthic insectivores results at NELR REMAP sites with symbols coded by the Maine IBI metric value that contributes to BCG Levels I-VI (Table 24).*

Blackbasses

This metric reflects the non-native black bass species, mostly Smallmouth Bass, that were introduced into New England lakes, rivers, and streams in the late 19th century. They were not significantly associated with any environmental variable (Table 23). However, they are absent from several northern New England river basins which may explain the absence of any region-wide correlations. As described earlier, their apparent effect on native fish species was documented by Yoder et al. (2008) in Maine and this corresponds to observations elsewhere in Maine lakes (Whittier et al. 1997), Ontario (Vander Zanden et al. 2004), and elsewhere in North America (Jackson 2002). BCG Level 1 and 2 equivalents occurred primarily north of 45-46°N and in rivers that they have not reached either by ingress or illegal stocking (Figure 53). BCG 6 values were evident north of 45-46°N and included mainstem rivers influenced by sources of point source pollution and organic enrichment. These included the St. Croix R., Mattawamkeag R., Penobscot R., and Kennebec R. Their abundance was highest in the Androscoggin R. downstream from the series of paper mill discharges in Berlin, NH, Rumford, ME, and Jay, ME. Blackbasses were less abundant in southern New England, except where they were abundant in the Connecticut R. in southern Vermont and northern Massachusetts.

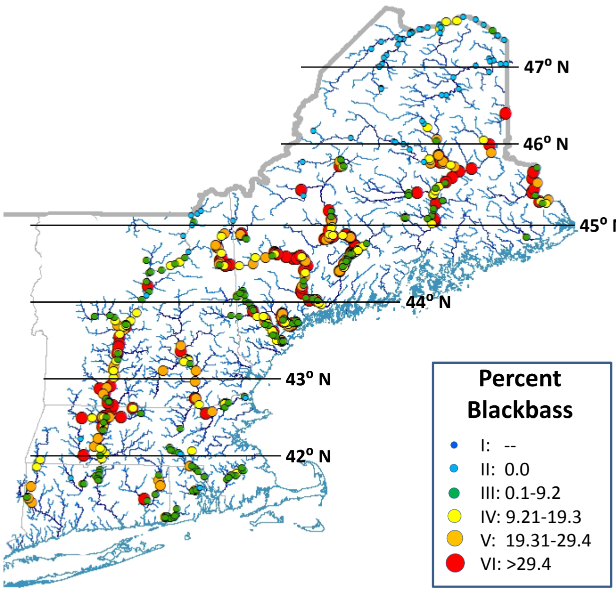


***Figure 53**. Percent blackbass results at NELR REMAP sites with symbols coded by the Maine IBI metric value that contributes to BCG Levels I-VI (Table 24).*

Fluvial Specialist/Dependent Species

This metric was positively associated with latitude (r^2=0.25), air temperature (r^2=0.33), and local developed land cover (r^2=-0.10). As with several other of the ME IBI metrics, BCG Level 1 and 2 equivalent values occurred mostly north of 46°N (Figure 54). However, some BCG 1 and 2 values occurred south of 45°N mostly in the upper Connecticut R. and as far south as 42°N with a single site in the Farmington R. BCG 5 and 6 values occurred in reaches of New England rivers that either had the highest density of impoundments and/or deliberate flow fluctuations and several of these occurred north of 45°N. It is probably why this metric responded poorly to the region-wide gradients.

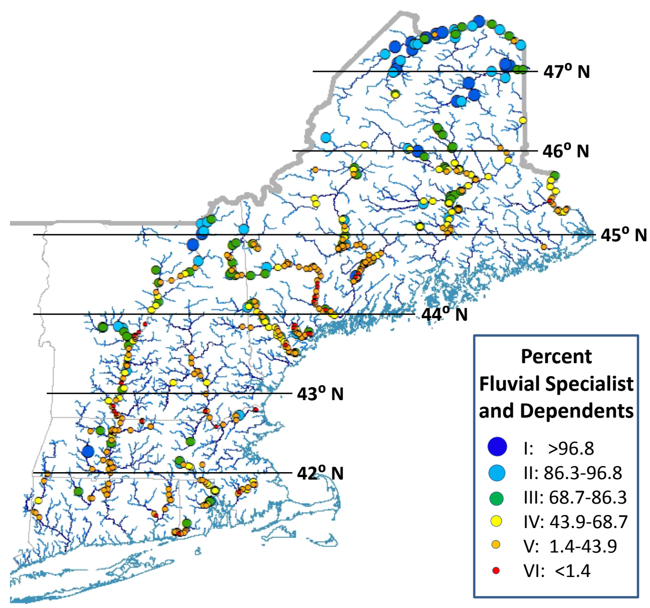


***Figure 54**. Percent fluvial specialist and dependent species results at NELR REMAP sites with symbols coded by the Maine IBI metric value that contributes to BCG Levels I-VI (Table 24).*

Macrohabitat Generalists

This metric was significantly correlated with latitude (r^2=0.15), air temperature (r^2=0.13), QHEI channel score (r^2=-0.15), and the QHEI score (r^2=-0.11). Several metrics had r^2= -0.07-0.09 (just below P<0.05) and included QHEI substrate score, QHEI riffle score, the Hydro-QHEI, and QHEI modified attributes. This metric

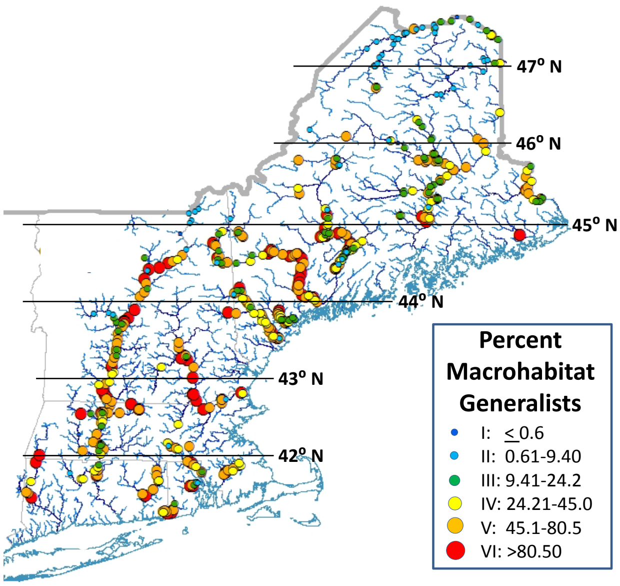


***Figure 55**. Percent macrohabitat generalist results at NELR REMAP sites with symbols coded by the Maine IBI metric value that contributes to BCG Levels I-VI (Table 24).*

(r^2=0.42), downstream barriers (r^2=0.10), the QHEI score (r^2=0.12), QHEI channel score (r^2=0.14), QHEI riffle score (r^2=0.16), Hydro-QHEI (r^2=0.11), QHEI good attributes (r^2=0.15), and QHEI modified attributes (r^2=-0.11). BCG Levels 1 and 2 occurred in the upper and colder reaches of most large rivers including those above 46°N, the upper Androscoggin R., and the upper Connecticut R. BCG 2 values occurred as far south as Connecticut (Figure 56). BCG 5 and 6 scores were prominent in the lower parts of the mainstem rivers and again, corresponded to the onset of a higher density of dams, impoundments, and deliberate flow modifications all of which affect the thermal regime.

Non-guarding Lithophils
This metric reflects spawning habits and it was significantly correlated with 11 variables,

reflects a degree of riverine habitat modification being comprised of species that are tolerant of impounded and otherwise habitat modified conditions. This is reflected in the distribution of BCG equivalents with Level 1 and 2 values being largely restricted to northern Maine and the upper Connecticut R. (Figure 55). One particular river with BCG 2 values is the lower Kennebec R. below Waterville which is now openly riverine since the removal of the Edwards Dam in 2001. BCG 5 and 6 values were evident elsewhere and 1 rivers with the highest density of dams and impounded conditions.

Temperate Stenothermic Species
Better known as cold water species, temperate stenotherms were significantly correlated with a number of variables including latitude (r^2=0.31), air temperature

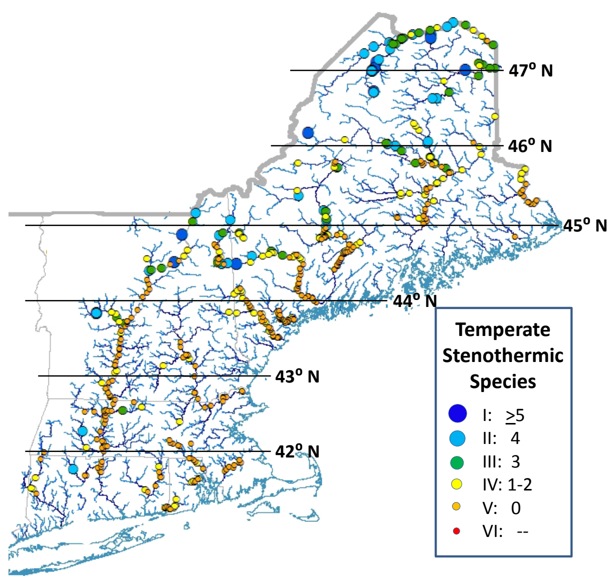


***Figure 56**. Number of temperate stenothermic species results at NELR REMAP sites with symbols coded by the Maine IBI metric value that contributes to BCG Levels I-VI (Table 24).*

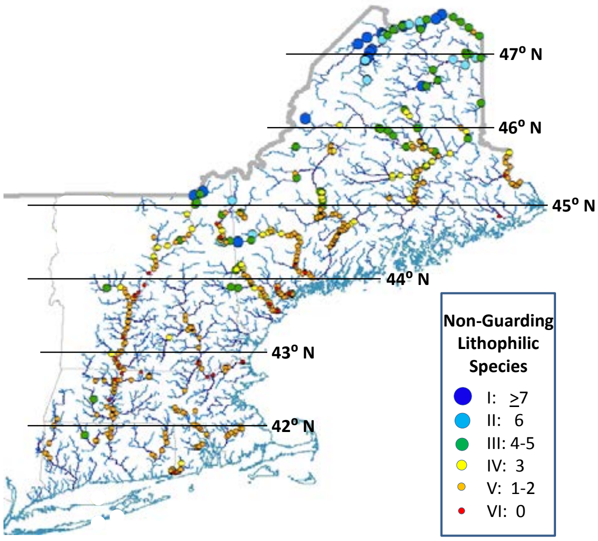


***Figure 57a**. Number of non-guarding lithophilic species results at NELR REMAP sites with symbols coded by the Maine IBI metric value that contributes to BCG Levels I-VI (Table 24).*

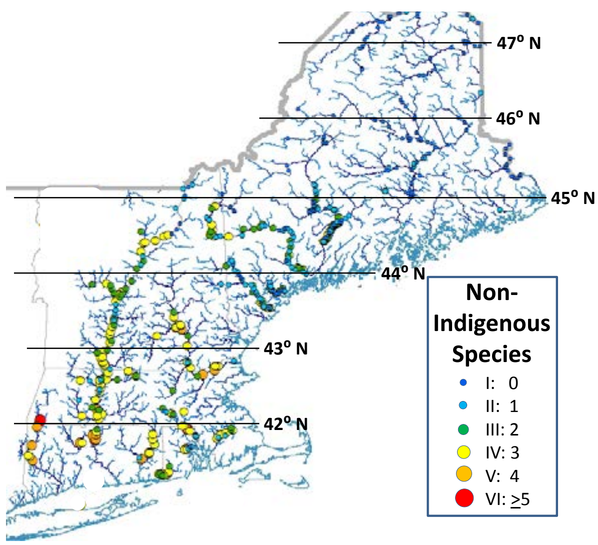


***Figure 57b**. Number of non-indigenous species results at NELR REMAP sites with symbols coded by the Maine IBI metric value that contributes to BCG Levels I-VI (Table 24).*

the most of any ME IBI metric. This included latitude (r^2=0.21), NAHC thermal class (r^2=0.17), (air temperature (r^2=0.49), the QHEI score (r^2=0.11), QHEI substrate score (r^2=0.10), QHEI channel score (r^2=0.11), QHEI riffle score (r^2=0.13), QHEI good attributes (r^2=0.13), QHEI modified attributes (r^2=-0.11), local developed land cover (r^2=0.10), and local natural land cover (r^2=0.11). The BCG Level 1 and 2 equivalent values occurred in northern Maine above 46^{o}N and the upper Androscoggin and Connecticut Rivers (Figure 57a). BCG Level 5 and 6 scores predominated the lower reaches of all mainstem rivers, particularly southern New England and to the onset of a higher density of dams, impoundments, and deliberate flow modifications, all of which affect coarse substrates over which species that this metric represents utilize for spawning.

Non-indigenous Species

This metric was significantly related to latitude (r^2=0.47), NAHC thermal class (r^2=0.21), air temperature (r^2=0.39), and QHEI substrate score (r^2=0.10). Four metrics had r^2= -0.08-0.09 (just below P<0.05) and included local developed land cover, local natural land cover, cumulative developed cover, and cumulative natural land cover. BCG level 1 and 2 values occurred above 45N and below that in Maine (Figure 57b). BCG Level 4 and 5 values were common in southern New England where introduced species comprise more of the fish fauna.

DELT Anomalies

DELT anomalies did not exhibit an association with any variable with an r^2-value of greater than 0.02. This, however, is not unexpected given the relatively low influence of acutely toxic stressors in New England rivers and the patchy occurrence of DELTs. DELTs can reflect the more severe impacts of industrial, CSO, and sewage pollution in rivers which was more prominent during the 1970s and 1980s (Yoder et al. 2005). The spatial footprint of these types of impacts has declined substantially in the past 20+ years as toxic discharges have been reduced. This metric is still important conceptually, as a site-specific indicator of stress where localized toxic impacts may well still occur (e.g., urban areas), and to detect novel compounds that may cause an increase in DELTs in fish assemblages. We lacked direct toxic stressor variables (e.g., metal concentrations) to include in the analyses and source-level variables (e.g., developed land cover) generally are poor predictors of site-specific "hot-spots" of toxic accumulations and stress. The regional pattern of BCG level equivalents illustrates the more reach- and site-specific response of this metric (Figure 58). BCG Level 1 and 2 values occurred throughout New England and independent of the latitudinal patterns evident in the other ME IBI metrics. BCG 5 and 6 values were isolated to specific sites or reaches and appeared to coincide with the location of active point sources and urban areas. As such it exhibited a response pattern that did not correlate with any region-wide stressor variables.

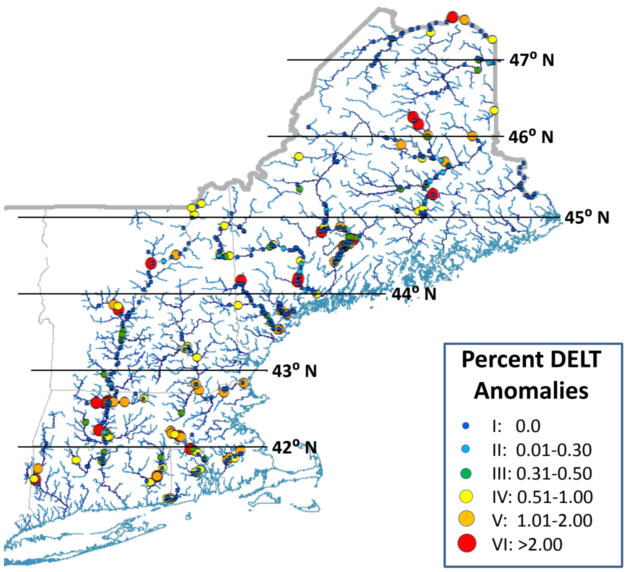


***Figure 58**. Percent DELT anomalies results at NELR REMAP sites with symbols coded by the Maine IBI metric value that contributes to BCG Levels I-VI (Table 24).*

Relationship of Environmental Variables to the Maine DIBI Metrics

The four metrics that comprise the diadromous metrics of the Maine DIBI were included in the metric specific analyses. These metrics were added to the ME IBI after its initial development and testing by Yoder et al. (2008) to better include any responses by the fish assemblage where diadromous species occur now and where they occurred historically. Diadromous species were historically restricted by natural barriers in selected rivers such as the upper St. John, Aroostook, upper Kennebec, upper Androscoggin, and upper Connecticut River basins. Otherwise, diadromous species have had historical access to most of New England. Presently that access is restricted by dams many of which are impassable to some, but not all diadromous species in New England. Of note in the Pearson coefficients were the significant correlations with the downstream barriers and impassable barriers variables for three of the four DIBI metrics.

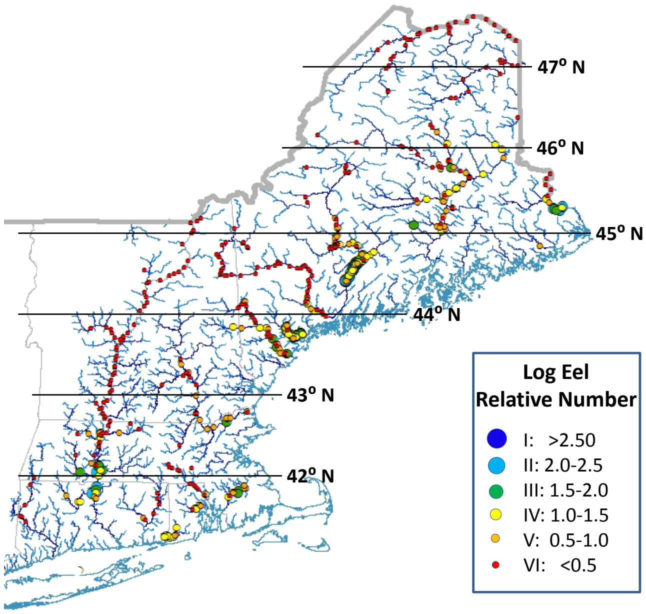


***Figure 59**. Relative numbers of American Eel at NELR REMAP sites with symbols coded by the Maine IBI metric value that contributes to BCG Levels I-VI (Table 24).*

American Eel Abundance

This metric includes the relative numbers of American Eels and it was significantly correlated with mean air temperature (r^2=0.17), downstream barriers (r^2=0.18), downstream impassable barriers (r^2=0.10), and local developed land cover (r^2=0.10). The BCG Level equivalents mapping demonstrates the comparative restricted distribution of American Eel throughout New England, with most sites at BCG 6 (Figure 59). No BCG Level 1 results were observed and BCG 2 was restricted to the lower Kennebec River in the open reach downstream from Waterville. American Eel were distributed throughout the Penobscot R. and tributaries and other coastal rivers throughout New England albeit at reduced numbers.

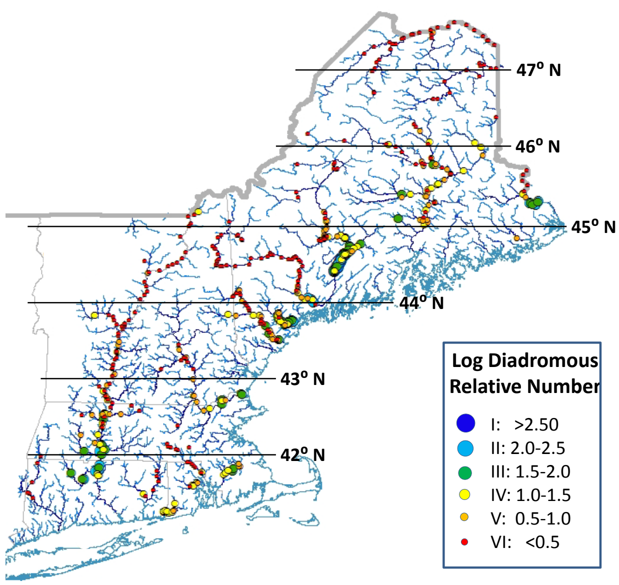


***Figure 60**. Relative abundance of diadromous species at NELR REMAP sites with symbols coded by the Maine IBI metric value that contributes to BCG Levels I-VI (Table 24).*

Diadromous Species Abundance

The relative numbers of all diadromous species was significantly correlated with mean air temperature (r^2=0.25), downstream barriers (r^2=0.25), downstream impassable barriers (r^2=0.14), and local developed land cover (r^2=0.10). The BCG Level equivalents mapping demonstrates the comparative restricted distribution of diadromous species throughout New England being comparable to that for American Eel, with most sites at BCG 6 (Figure 60). No BCG Level 1 results were observed and BCG 2 was restricted to the lower Kennebec River in the open reach downstream from Waterville and the lower Connecticut R. below the Holyoke dam. Diadromous species were distributed throughout the Penobscot R. and tributaries and other coastal rivers

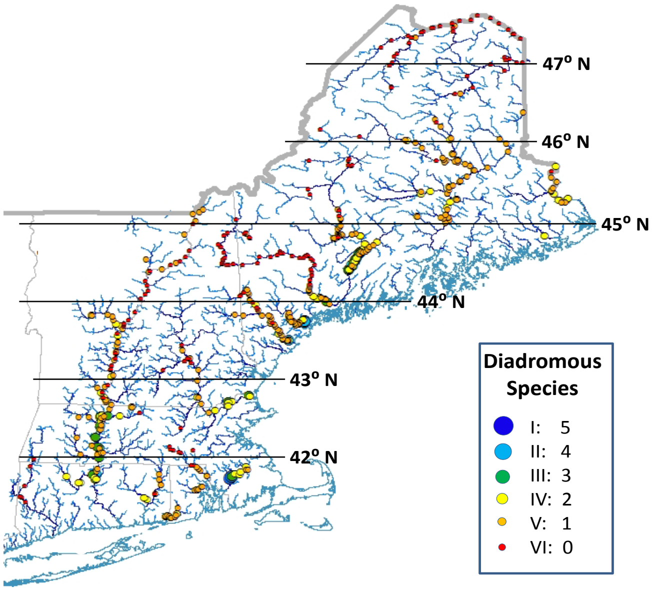


***Figure 61**. Diadromous species richness at NELR REMAP sites with symbols coded by the Maine IBI metric value that contributes to BCG Levels I-VI (Table 24).*

Clupeidae Abundance

This metric includes the relative number of Clupeidae and it was significantly correlated only with mean air temperature (r^2=0.10) and just less than p<0.05 for downstream barriers (r^2=0.08) and cumulative natural land cover (r^2=0.09). The BCG Level equivalents mapping demonstrates the very restricted distribution of Clupeidae throughout New England occurring only in isolated reaches with the highest abundance in the open reach of the lower Kennebec River below Waterville (Figure 62) which has been the concerted focus of restoration efforts. These results are comprised almost exclusively of young-of-year individuals that are out migrating from their parent streams and lakes during the summer and fall months (July – October).

throughout New England. As expected relative numbers were higher in closest proximity to the lower reaches of the major coastal rivers.

Diadromous Species Richness

The number of diadromous species was significantly correlated with latitude (r^2=0.14), mean air temperature (r^2=0.29), downstream barriers (r^2=0.26), downstream impassable barriers (r^2=0.15), cumulative developed land cover (r^2=0.11), and cumulative natural land cover (r^2=0.11). The BCG level equivalents mapping overlaps with the results for American Eel since this was the sole diadromous species at many locations (Figure 61). The Taunton R. had a BCG Level 1 result with 5 diadromous species. The lower Connecticut and lower Kennebec Rivers had the next highest number of diadromous species.

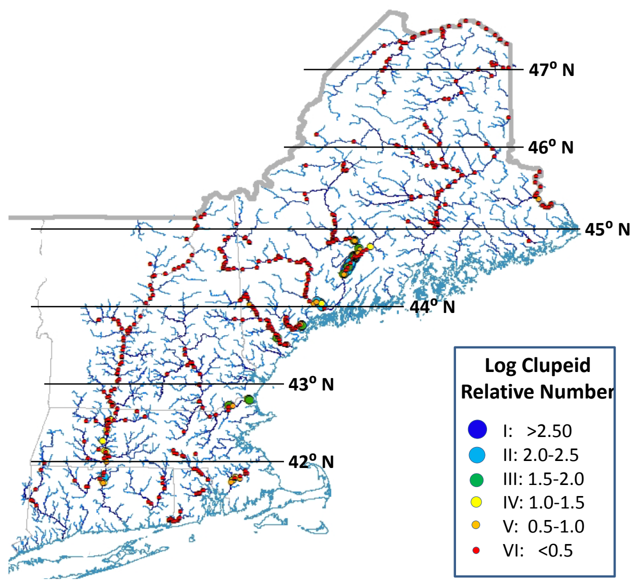


***Figure 62**. Relative numbers of Clupeidae at NELR REMAP sites with symbols coded by the Maine IBI metric value that contributes to BCG Levels I-VI (Table 24).*

Discussion of Stressor Identification Results

The predominant influences on New England riverine fish assemblages included both fluvial and structural aspects of habitat (includes dam impoundments), the presence of dams, non-native species, and land uses. This was demonstrated by both multivariate analyses against the array of stressors that were analyzed and by examining the distribution and values of the ME IBI and metrics. The typical result is a continuous gradient of indicator response related to the magnitude and extent of the *effect* of one or more stressors and as expressed by correlative or multivariate analyses. However, not all of the indicators emulate such responses to all stressors. For example, impassable dams act as a discrete impact thus the typical pollution tolerance, functional, or species richness metrics may not appear to be responsive to such an impact. This was reflected in the response of the ME IBI and intolerant fish species measures to the human disturbance gradient calculated with and without connectivity metrics included (see Figure 24). Fluvial dependent and specialist species metrics tended to be more strongly associated with specific habitat niches (e.g., riffle/run metric of the QHEI) and higher scores of the Hydro-QHEI as illustrated by WSVs being skewed towards the maximum scores in most instances.

It also needs to be understood that any exploratory analysis of stressors and their apparent responses as measured by biological indicators is only as meaningful as are the actual mechanisms that are at work. In this or any other analysis a stressor is the measure of the *presence* of an impact or alteration while the biological response is an indication of the *effect* of that stressor or aggregation of stressors. The mere presence of a stressor does not equate to an effect by that stress on the biota. Simply compiling an array of stressors and then subjecting them to correlative and multivariate analyses seldom explains even the majority of the variation in biological responses. Such was the case in this study as most of the relationships explained less than one-third of the variation on a regional basis. This is a tacit admission that the analyses herein either did not capture all of the stressors that are important to riverine fish assemblages in New England or all of their responses. Analyses that are keyed on “reading” the biological responses first and then diagnosing the stressors based on details about the setting in which the responses were observed are also needed to ensure a more complete approach.

It is seems likely that the variation in some key fish assemblage indicators (e.g., DELT anomalies) that were only evident at the local reach and site-specific scales could be better explained if important chemical stressor data that characterized the input sources and ambient conditions was more widely available and at a meaningful spatial scale. Coupled with the regional approach taken by this study it would provide the template for developing a more monitoring dependent and data driven stressor identification process for New England rivers. A targeted design at least as intensive as that employed in this study would be needed to capture effects at this scale which operate along pollution continuums in a downstream direction. Such a non-random, but spatially adequate and equitable design can provide substantial benefits particularly where there is a need to detect, characterize, and resolve reach and site-specific issues in rivers. It also needs to include sufficient site density to assure that the full range of resource quality is captured as was demonstrated in this study with the level 2 BCG ME IBI scores being uniquely revealed by the targeted sampling design on both a regional and river specific basis.

The results of the stressor analyses revealed a strong latitudinal effect that includes both natural phenomena and anthropogenic stressors which corresponds to a general decline in the ME IBI and metric values rated against the BCG. Separating the influence of natural and anthropogenic gradients in these observations is challenging because these gradients are also spatially correlated. The ME IBI was used in the NELR REMAP project with the understanding that it may not apply equally well to all rivers throughout New England. However, other potentially applicable IBIs simply do not exist and those that are either available or under development for the Northeastern U.S. are restricted to wadeable streams. The latitudinal pattern in ME IBI scores is illustrated in Figure 63 in seven increments of latitude. While the general pattern of decreasing median IBI scores in a southerly direction is supported by this analysis, it is not invariable. High outlier IBIs equivalent to BCG Level 3 occur at all of the southerly latitudes. The pattern is almost the opposite in Vermont where the highest quality streams occur at the higher elevations in the south (R. Langdon, personal communication). Based on the analysis of regional stressors and the examination of metric responses, it is concluded that this is the result of an increasing stressor gradient related to habitat alterations and land use changes that increase from north to south. What part natural factors play in this pattern is less clear, but one of the regional variables that was used in the stressor analyses suggest that it is not completely due to natural factors. The thermal classification scheme developed by the Northeast Aquatic Habitat Classification System (NEAHCS; Martin and Apse 2011) shows the occurrence of transitional cold water fish faunas as far south as mid-Connecticut (see Figure 22), which would support the contention that the *potential* for a cold-cool fish assemblage is farther south than is demonstrated by the 2008-9 NELR REMAP results. Whether a cold-cool water and native fish assemblage is widely attainable in southern New England is a question that needs to be addressed as part of the implementation of a yet to be developed framework consisting of natural fish assemblage classification, index derivation and calibration, and numeric biocriteria in support of tiered aquatic life uses that is discussed in more detail Chapter 6.

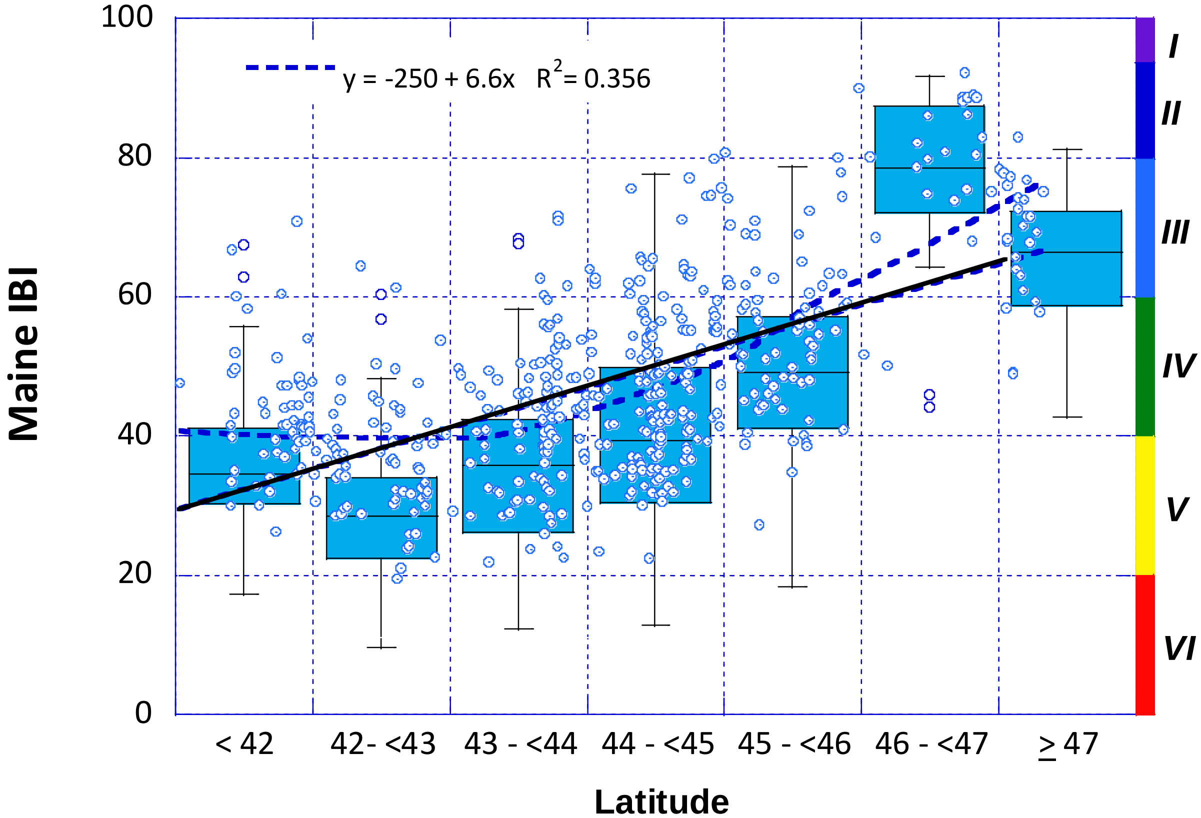


***Figure 63**. Box-and-whisker plot of Maine IBI results at all NELR REMAP sites stratified in increments of latitude. Individual Maine IBI site data points are also shown by decimal degree with linear regression (solid line) and weighted smoothing curve (dashed curve). Maine IBI ranges corresponding to Levels I-VI of the Biological Condition Gradient are shown on the y-2 axis.*

CHAPTER 5: INITIAL COMPARISON OF NELR REMAP AND NRSA FISH SAMPLING METHODS

One of the major objectives of the NELR REMAP project was to compare the outputs of the methods employed in this study with those produced by the 2008-9 NRSA methods. Because of the overlapping goals of the NRSA and this project it is important to understand their comparability in terms of data characteristics, logistics, and assessment outputs. Each project purports to provide the states (and others) with a set of standardized protocols for conducting biological assessments for CWA purposes. This comparison was limited by the scope of the NELR REMAP project to the fish assemblage data that each produced.

Background

Biological methods and assessment comparability is an issue of importance to U.S. EPA because of the variation in techniques that are employed by states throughout the U.S. These differences have raised questions about how comparable are biological assessments for purposes such as assessing attainment of the CWA 101[a][2] goals. How differences in methods potentially affect how an impairment of aquatic life uses is expressed in state reporting for 305[b] and 303[d] purposes is a major issue of concern. Different states with technically different sampling and data analysis frameworks should be able produce similar determinations of aquatic life use impairment provided that the technical rigor of their respective approaches is sufficiently robust (U.S. EPA 2013). While the concepts of comparability apply to other bioassessment program aspects such as being able to incrementally measure condition along the BCG and conducting stressor characterization and identification, the NELR REMAP project is focused here on the comparability of overall aquatic life condition assessment outcomes.

Study Design

Paired fish sampling was conducted as part of the NELR REMAP project at 64 NRSA sites during 2008-9. Because the NRSA base and oversample draw of sites were used for the probabilistic aspect of the NELR REMAP assessment, data was available for each method. Independent MBI crews conducted the fish sampling for each project which made the required coordination easier to accomplish. An exception to this was with 10 sites located in New Hampshire that were sampled by a U.S. EPA, Region I crew for the NRSA. All paired sampling events were accomplished within the same field year (2008 or 2009) and with sufficient time between sampling events (>3-4 weeks) so as to minimize any electrofishing induced effects on the resulting data. In most cases the NRSA crew reconnoitered and sampled each site to conduct the NRSA methodology first being followed by the NELR REMAP crew employing that methodology in an overlapping fashion. Overlapping means that the NRSA site was located following the probabilistic draw of the site coordinates that served as the “center” or x-point for delineating the NRSA site. The corresponding NELR REMAP site was setup to contain at least a significant portion of the NRSA site without violating the principles of site location and configuration. There was a degree of flexibility in adapting an NELR REMAP site within an NRSA site because the latter was usually much longer. When the NRSA site measured the maximum of 4.0 km, two 1.0 km NELR REMAP sites were located within the 4.0 km NRSA sampling reach.

NRSA and NELR REMAP Method Characteristics

The NRSA sites varied in distance by river size with the cumulative distance being determined by multiples of river width. Forty (40) times the mean river width defined an NRSA sampling site with a set minimum of 0.15 km and a maximum of 4.0 km. As a result NRSA site distances predictably varied by river size as is demonstrated by box-and whisker plots by Strahler order and a scatterplot by drainage area (Figure 64). New England NRSA site distances varied from ≈0.5 km to the maximum of 4.0 km which is the upper limit set by the NRSA protocol (U.S. EPA 2009). Within each site either a raft or boat-mounted electrofishing apparatus was used to conduct the NRSA fish sampling. The same equipment specifications were employed in the NRSA and NELR REMAP projects thus standardizing that aspect of the comparability study. From here the two methods varied considerably in their setup and execution (Table 25).

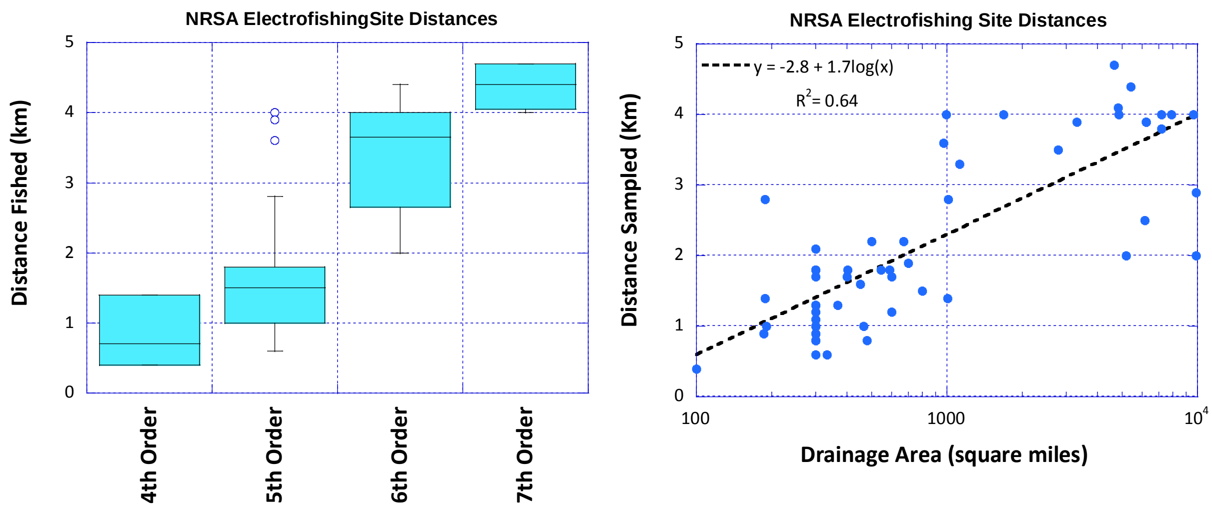


***Figure 64**. Distance electrofished by Strahler order (left) and drainage area (right) for NRSA sites sampled during 2008-9 and used in the NELR REMAP electrofishing methods comparison.*

The NELR REMAP method was described extensively in Chapter 2 and key aspects are summarized in Table 25 for comparison to the NRSA methodology. The key differences that are summarized include both well-defined and perhaps more subjective characteristics of each method. Characteristics such as distance sampled, time sampled, number of netters, and data attributes are discrete and quantitative. Some aspects of executing each method are inherently qualitative, but are nonetheless critical characteristics that can affect the data outputs of each. These aspects were considered by Tewes et al. (2007) in their electrofishing comparability study in major upper Ohio and upper Mississippi River tributary rivers and it explained many of the differences they found between different programs.

Site Configuration

To analyze the differences between the NELR REMAP and NRSA methods it is important to understand how each method configures a riverine sampling site. The NELR REMAP site configuration is shown in Figure 65 (upper panel) with two types of river reaches – slow current

***Table 25**. Selected characteristics of the NELR REMAP and NRSA fish sampling methods including aspects of sampling effort, method execution, and data recording.*

Method Characteristic	NELR REMAP	NRSA
1. Electrofishing distance	1.0 km fixed continuous distance	40X mean width; 4 km maximum
2. Time electrofished	>2500 seconds min.; no max.	3500 seconds 1st 5 transects; 700 seconds max./transect
3. Site configuration	All -“best” habitats - continuous	Random shoreline; 5-10 transects
4. Method execution	“Attack” all habitats	“Passive”
5. “Late” fish	Go after all fish sighted	Leave late fish “behind”
6. Netters	1 (raft) or 2 (boat); all crew members have a net	Single netter
7. Data – age/length	3 age classes (A,B,Y); selective lengths for some species	Max./min. length each species
8. Data - biomass	Individual or subsampled weights	None
9. Data - numbers	All fish counted in 1.0 km	Cease sampling if >500 ct. after 5 of 10 transects, otherwise continue until >500 is reached
10. Data – external anomalies	DELTs and all other anomalies	DELTs

with abundant woody debris and moderate to fast current with swift runs and in-channel cover such as boulders. The latter is the more commonly occurring situation in New England, but impoundment by run-of-river dams is exemplified by the former. The sampling track retained from a NELR REMAP electrofishing site in the Kennebec River is also shown in Figure 65 (lower panel). The track is from a GPS recording of the path taken by the electrofishing boat and superimposed on a topographic representation of the river channel. The 1.0 km distance criterion is a lineal measurement along the shoreline that is sampled and includes any changes from bank to bank and mid-channel sampling around structure such as bed rock ledges and deep run habitats. Waypoints are set along the general path followed by the boat (Figure 65) and are used to calculate the cumulative site distance. As such, the NELR REMAP sampling distance is not an accumulated distance from the GPS track and the cumulative distance traveled by the boat is almost always greater than 1.0 km. The choice to index sampling effort by lineal site distance was made because different sites require different levels of effort to sample effectively. In addition, fish are generally distributed by their preferred habitats which may require varying efforts and time to sample each habitat effectively. Effort is standardized by “thoroughness” with time serving as a *minimum* proxy for that criterion, but also recognizing that time electrofished alone is an insufficient measure of the overall effectiveness of sampling effort. While some of the protocol attributes might appear to be subjective, crew leader

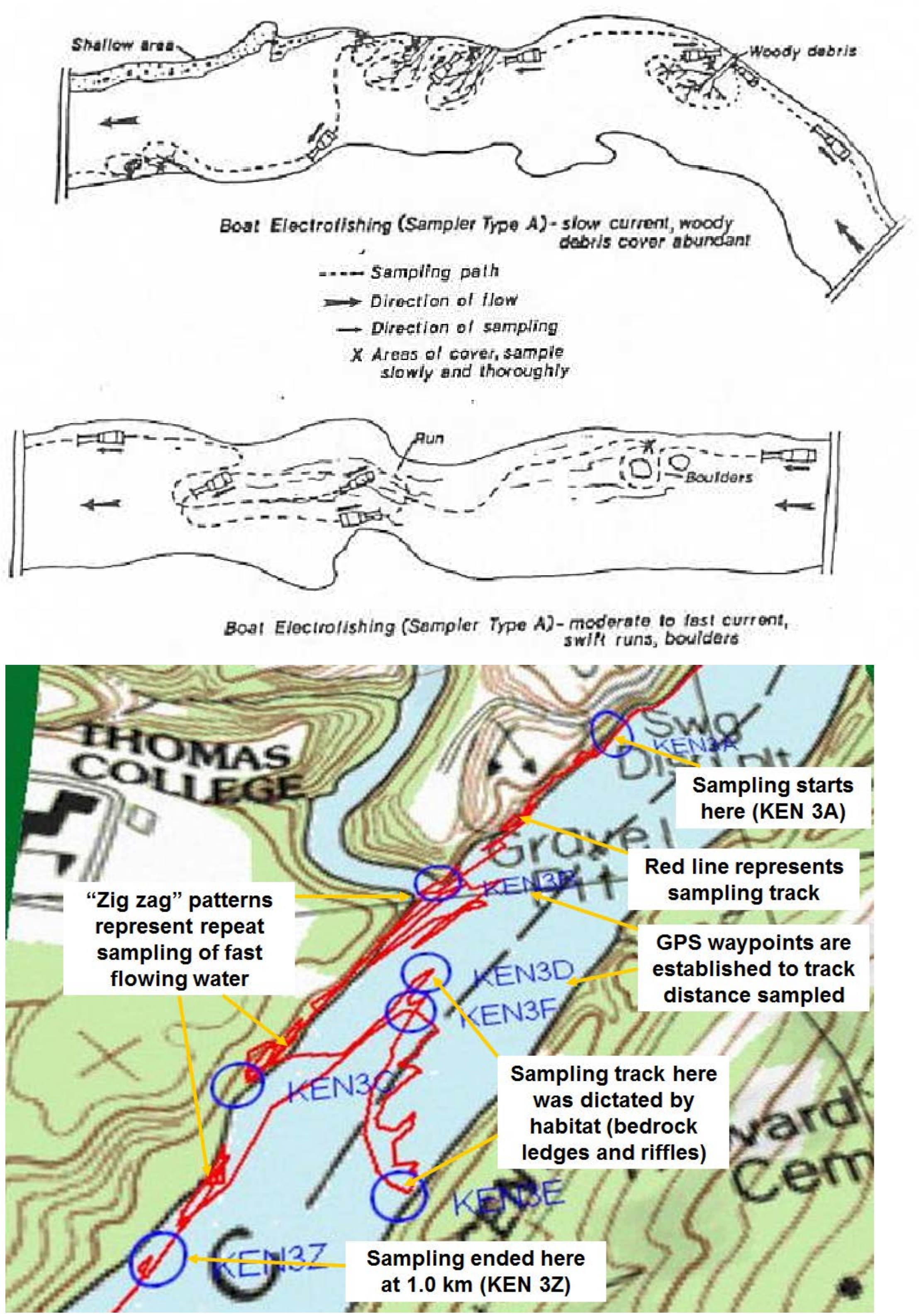


***Figure 65**. Configuration of a NELR REMAP non-wadeable electrofishing site showing the site layout for a predominantly pooled and slow flowing site and a moderate to high gradient site with deep runs with in-channel structure (upper panel). The GPS sampling track from a site in the Kennebec River near Waterville, ME reveals how a site is to be sampled.*

training and apprenticeship has been proven effective in making this protocol reproducible between crews (Yoder and Smith 1999).

The NRSA non-wadeable electrofishing site configuration follows the overall transect design of an NRSA site within which samples and measurements are collected for water quality, physical habitat, periphyton, fish tissue, and macroinvertebrates in addition to the fish assemblage (U.S. EPA 2009). When a site is located and verified as being a "target" site, an overall sampling reach is established based on 40 times the mean wetted width. The minimum reach length is 0.15 km and the maximum is 4.0 km regardless of the mean width. Ten (10) transects are spaced at equal intervals of 4 times the mean wetted width (Figure 66, upper panel). The river bank (i.e., river left or river right) along which sampling will take place within each transect is determined randomly and without regard to habitat quality. The fish sampling procedure was conducted with the same boat and raft mounted electrofishing apparatus that was used for the NELR REMAP project. Continuous shoreline electrofishing begins upstream at transect A and proceeds in a downstream direction through transects B, C, etc. Transects A-C are sampled along the same bank then alternated to the opposite river bank for transects D and E (Figure 65, lower panel). The entire length of each transect is sampled and the fish sample is processed at the end of each transect thus producing 5-10 subsamples. Sampling is continued in transects F-J, alternating the river bank every two transects, only if fewer than 500 individuals are collected in transects A-E. As such, electrofishing distance can vary between 20 times and 40 times the mean width of an NRSA site (see Figure 65). Each transect is sampled by moving the electrofishing platform at a velocity slightly more than the current speed and within "close proximity" to the shoreline. The NRSA sampling approach is labeled here as "passive" (Table 25) as a comparison to how the NELR REMAP method is described as "attacking" each habitat. Simply stated the NELR REMAP method thoroughly samples all habitat types by repeated stopping, turning, reversing, and resampling swift flowing areas multiple times. By comparison the NRSA sampling platform is kept moving in a continuous manner with limited turning into and stopping in cover and with no return attempts in swift runs and chutes. The NRSA protocol states ". . . Crews may 'nose in' to habitat to effectively sample, but should not remain in that habitat for too long" which reinforces our characterization of the NRSA method as "passive".

The different elements of the NRSA method that relate to site configuration and method execution include the random selection of alternating banks, the continuous sampling in a downstream direction, limitations for sampling around habitat structure, the >500 fish count subsampling procedure, and the variable distance that is eventually sampled. As such, the respective configurations of the NELR REMAP and NRSA methodologies have a direct effect on how electrofishing is conducted at a site, the habitats that are sampled, and as a result the resultant data. The track followed by the electrofishing craft for two overlapping sites is shown in Figure 67 which demonstrates the site configuration differences of each protocol.

Data Elements

While many of the baseline data elements included in each protocol are the same, there are some important differences. The NELR REMAP method included the collection of biomass data by species, whereas the NRSA method included counts of individuals only. The ME IBI that

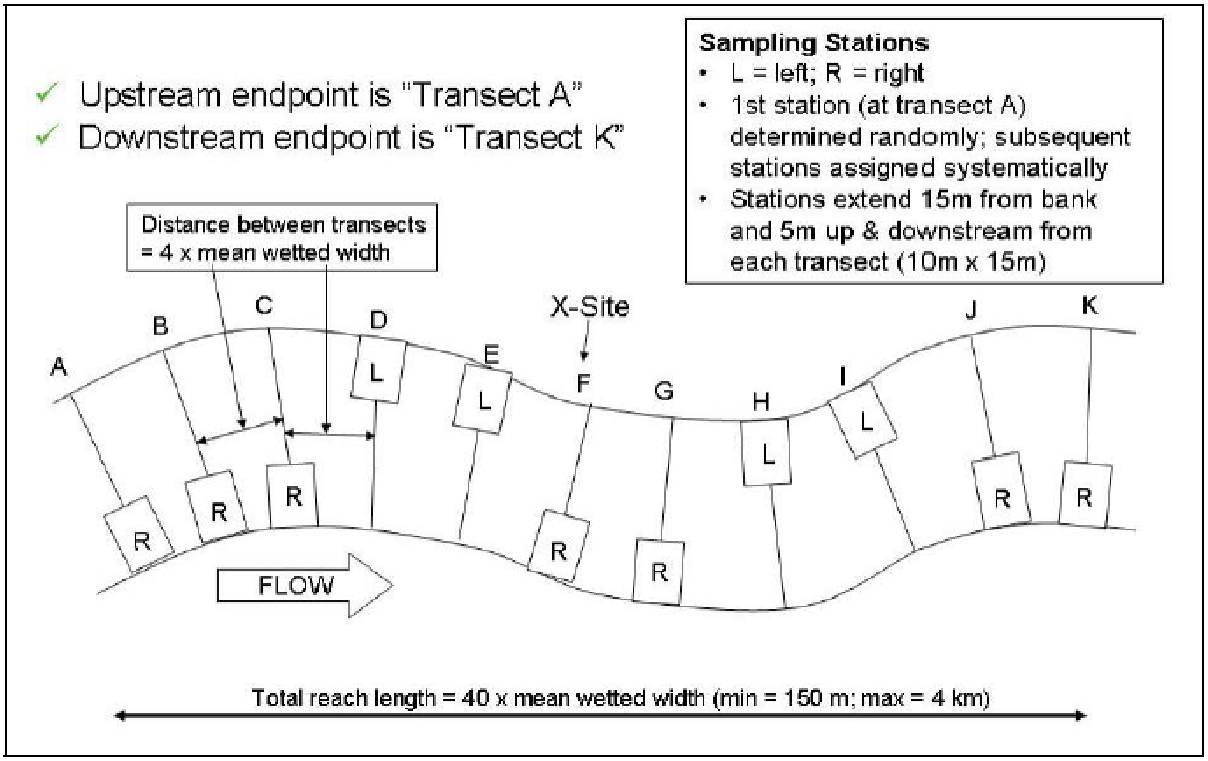


Figure 4-5. Sampling reach features for a non-wadeable site.

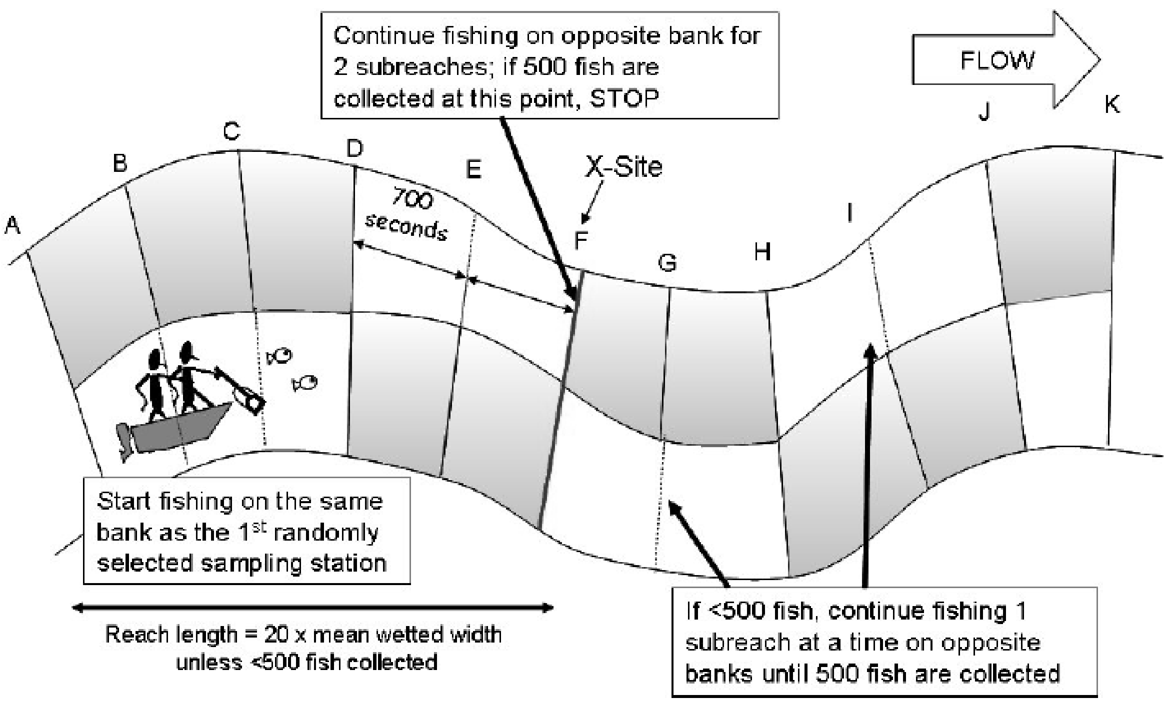


Figure 5.5-2. Transect sampling design for fish sampling at non-wadeable sites.

***Figure 66**. NRSA non-wadeable site configuration (Fig. 4-5; upper panel) and boat electrofishing site configuration (Fig. 5.5-2; lower panel). Operational descriptions are indicated on each (after U.S. EPA 2009).*

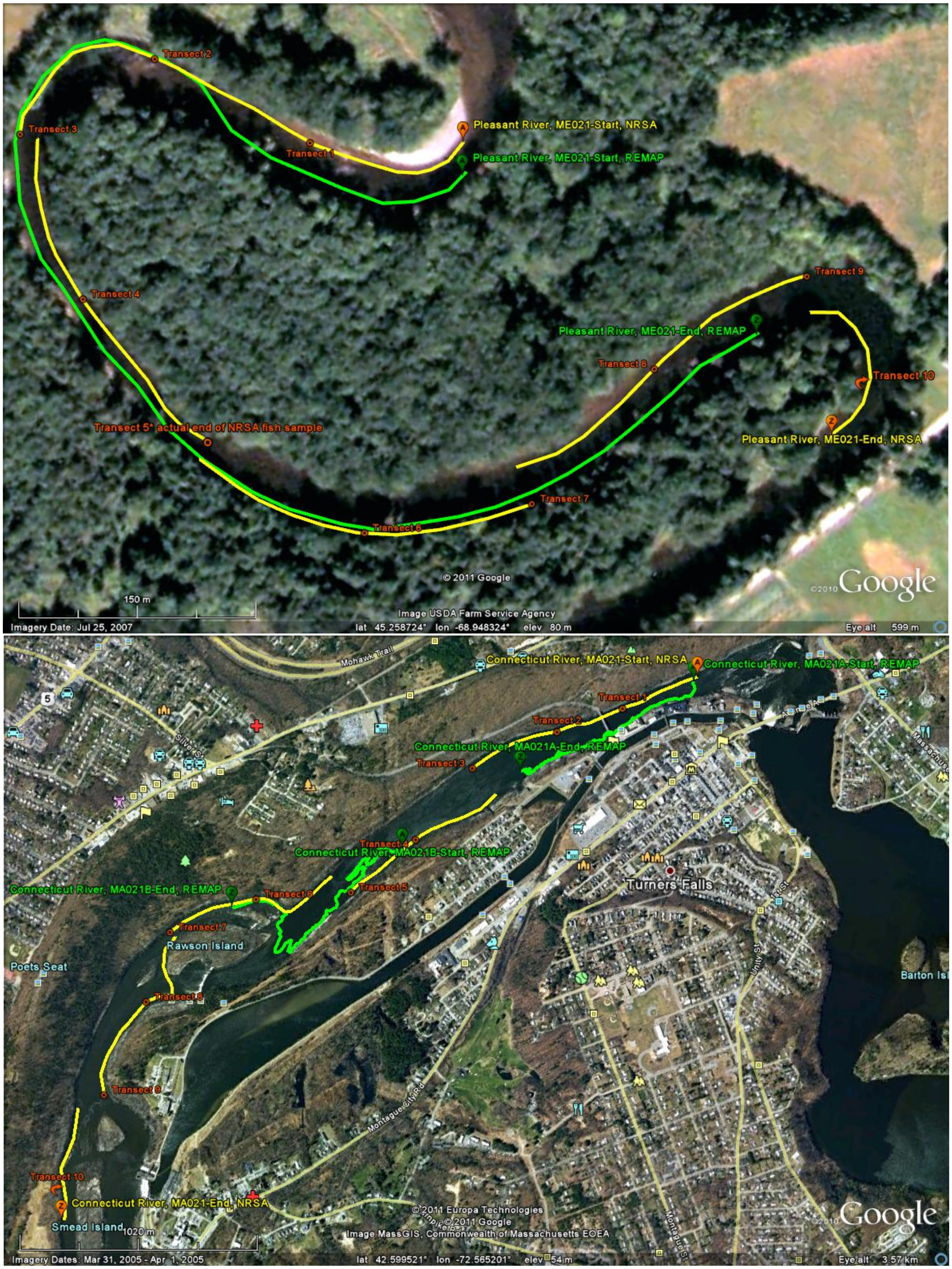


***Figure 67**. GPS derived sampling tracks of NELR REMAP fixed distance (green trace) and NRSA 40X mean width sites (yellow trace) for two comparability sites; the Pleasant R. (Maine; upper) and the Connecticut R. (Massachusetts; lower). Two NELR REMAP sites were sampled within a 4.0 km NRSA site in the Connecticut River.*

was used to assess fish assemblage condition for the NELR REMAP study includes a biomass based metric, something that would be precluded for an NRSA-based index. While the remaining data is categorically similar, some detailed differences exist. These include how fish size classes are determined, how a site sample is determined, and how external anomalies were identified (Table 25). For the latter, the NELR REMAP method follows the Ohio EPA (1996) and Sanders et al. (1999) procedure and training for how external anomalies are recorded. While the NRSA method indicates the collection of "DELT" anomalies, there are no apparent references to a particular procedure or training. The NELR REMAP method also included the more refined developments of Sanders et al. (1999) which includes the recording of "light" and "heavy" infestations and a separate category for multiple DELT anomalies. It was not clear if this additional detail was required by the NRSA method, but there is no mention of these refined procedures in the NRSA field methods manual (U.S. EPA 2009).

Results

The comparability of the NELR REMAP and NRSA non-wadeable fish assemblage data was assessed by comparing selected data outputs such as the ME IBI, relative numbers, commonly occurring fish species, and ME IBI metrics. Other types of data and comparisons are possible, but this initial comparison of the results yielded by each method was focused first on assessment outcomes in terms of reporting aquatic life goal attainment since that would be the primary concern for New England states and EPA Region I for determining the utility of either method. As such the ME IBI was calculated for both NRSA and NELR REMAP data and the results compared using the same thresholds as reported in Chapter 3.

Maine IBI Adjustments and Comparisons

The comparability of the NELR REMAP and NRSA non-wadeable fish assemblage data was assessed by comparing selected data outputs such as the ME IBI, relative numbers, commonly occurring fish species, and ME IBI metrics. Other types of data and comparisons are possible, but this initial comparison of the results yielded by each method was focused first on assessment outcomes in terms of reporting aquatic life goal attainment since that would be the primary concern for New England states and EPA Region I for determining the utility of either method. As such the ME IBI was calculated for both NRSA and NELR REMAP data and the results compared using the same thresholds as reported in Chapter 3. To determine if this modification had an effect on the utility of the NRSA data for supporting aquatic life status assessments the ME IBI based on NELR REMAP data was calculated with and without adult white and longnose sucker biomass. This allowed for a comparison of the NRSA based ME IBI, the NELR REMAP based ME IBI, and the same with the adult white and longnose sucker metric removed (Figure 68). While there were mostly similarities, some differences existed in the distribution of the ME IBIs among the 64 NRSA samples included in the comparison. The non-adjusted NELR REMAP and NRSA based IBIs produced the same median and 75th percentile values, but the former had lower 25th percentile and minimum values. The NRSA based IBI had one value >80 which equates to a BCG Level 2 result. Recall that none of the NRSA base or overdraw probabilistic sites in the NELR REMAP dataset had any BCG Level 2 values (see Chapter 3). However, the results shown here are an adjusted ME IBI to compensate for the lack

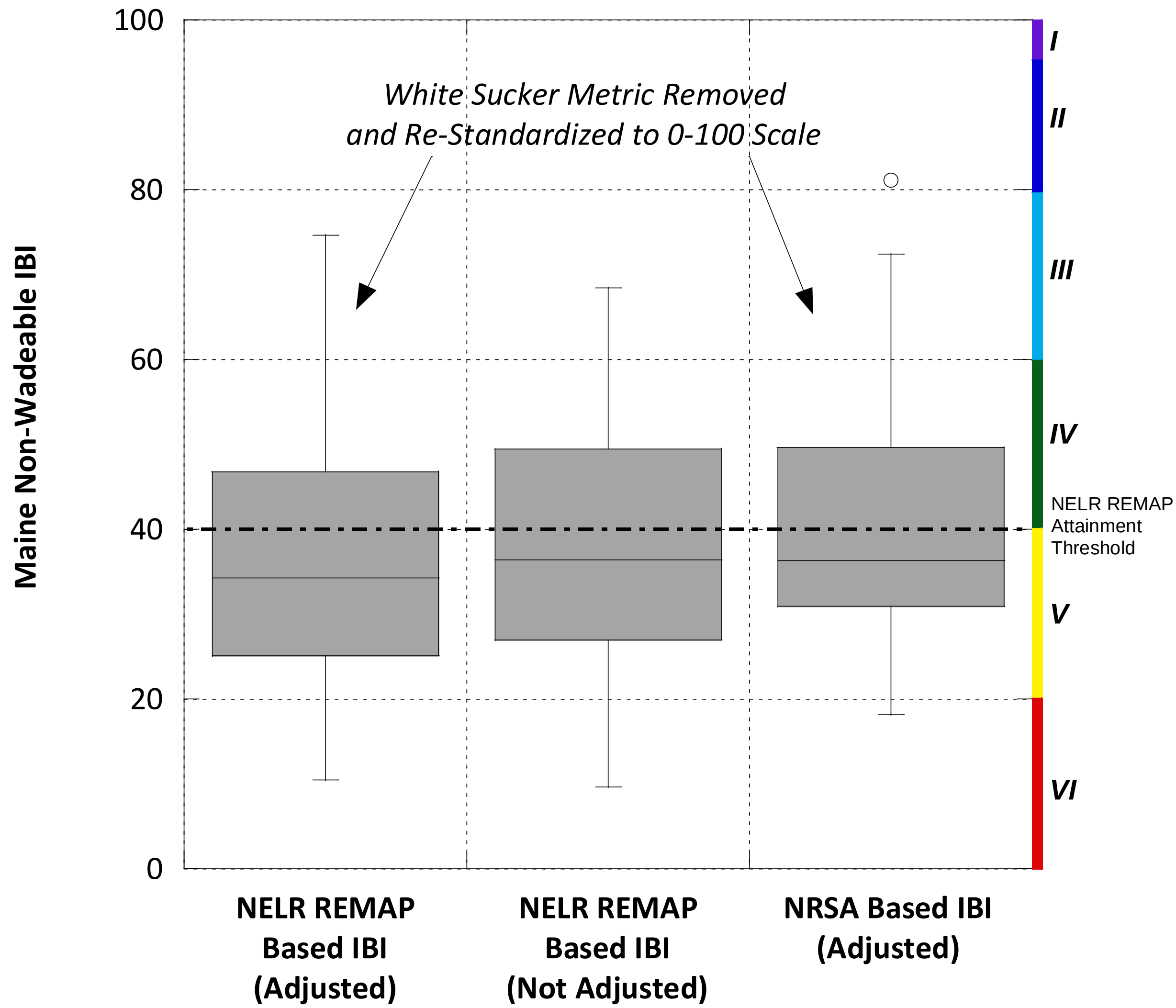


Figure 68. *Box-and-whisker plot comparisons of the NELR REMAP based IBI adjusted for the deletion of the adult white and longnose sucker biomass metric (right), not adjusted (center), and the NRSA based IBI (right). The NELR REMAP attainment threshold is indicated by the dashed horizontal line (IBI = 40) and the six corresponding BCG levels are indicated along the y2 axis.*

of biomass in the NRSA dataset and the need to delete this lone biomass based metric. The NELR REMAP based adjusted IBI that omits the adult white and longnose sucker biomass metric had lower median and 25th percentile values than the non-adjusted ME IBI, but had a higher maximum value. Omitting the adult white and longnose sucker metric “inflates” the adjusted ME IBIs especially when adult white and longnose suckers are either absent or in low abundance in a sample. This metric was one of the least correlated with the regional stressors that were examined in chapter 4, but it does show a strong relationship with the deep run habitats of riverine sites and it declines in impounded or otherwise modified habitats (see

Chapter 4). While some of the differences between the NELR REMAP based non-adjusted IBI and NRSA based IBI were due to the inherent differences between each method, some are due to the removal of this metric as evidenced by the comparisons of removing this metric within the NELR REMAP method. The contemporary approaches to developing regional IBIs (Whittier et al. 2007) would likely have omitted this metric, but it was included in the ME IBI and for reasons that relied more on life history attributes, an ability to discern important impacts that are not usually included in regional stressor analyses, and taxonomic completeness. These comparisons seem to bear out the need to include this important fish assemblage attribute. Given that the distances sampled for the NRSA varied considerably, the potential effect of this variable on the IBI was also examined (Figure 69). Based on a scatterplot of NRSA vs. NELR REMAP IBI scores and by four increments of distance sampled there was an inconsistent effect of this variable on IBI scores.

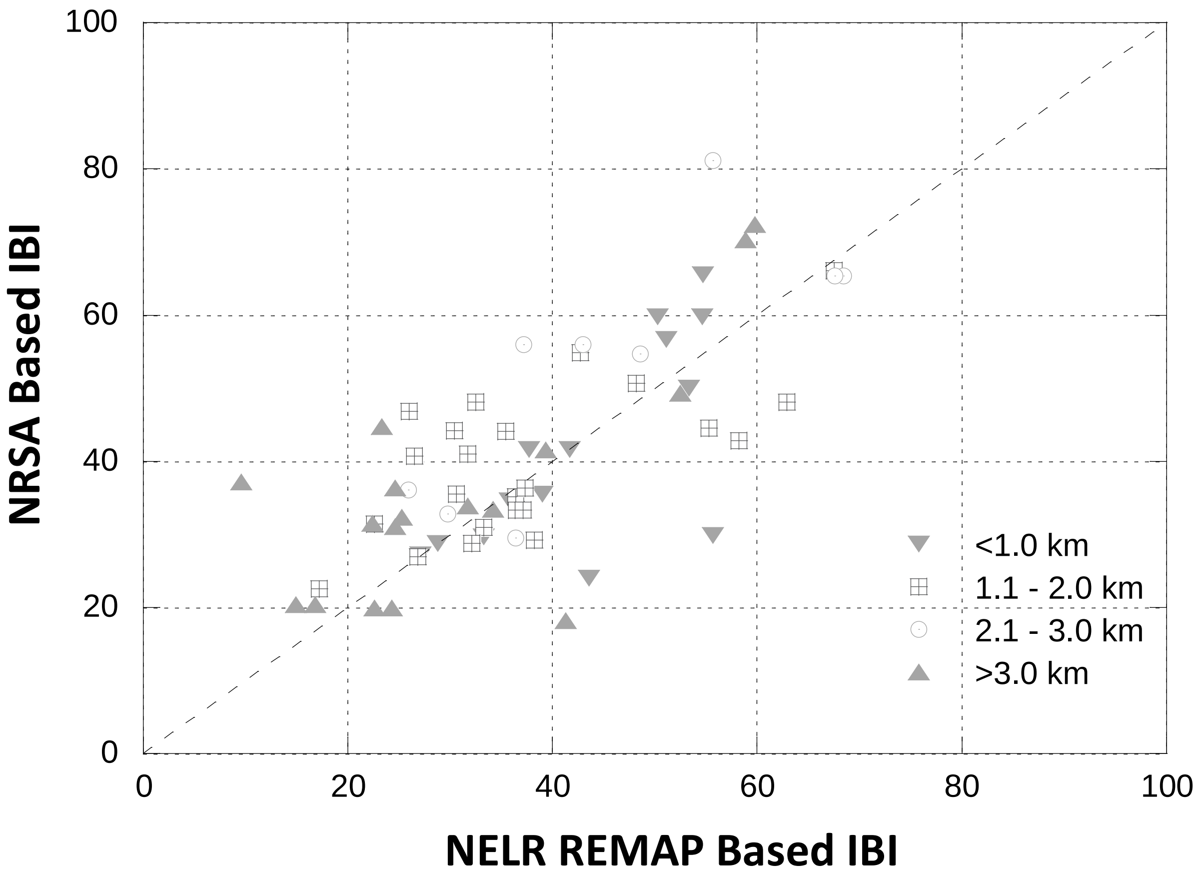


***Figure 69**. Scatterplot of the NELR REMAP based IBI vs. the NRSA based IBI at 64 non-wadeable New England river sites. Symbols differentiate four classes of sites based on 1.0 km increments NRSA fish sampling distance.*

Aquatic Life Condition Assessment

The comparison of condition assessment outcomes between the NELR REMAP and NRSA methods relied on the ME IBI thresholds that were used to develop the New England-wide estimates in Chapter 3. The accounting for ME IBI values that indicate non-attainment was set at the boundary between BCG Level 4 and 5 which is an IBI = 40. A scatterplot of the NRSA based IBI and NELR REMAP IBI (non-adjusted) revealed a general directional agreement, but an r^2 of only 0.53 (Figure 70), an indication of substantial quantitative departures between each method. In this case the comparison used the NELR REMAP based IBI without adjustment as the inclusion of biomass data is simply one of the fundamental differences between the two methods.

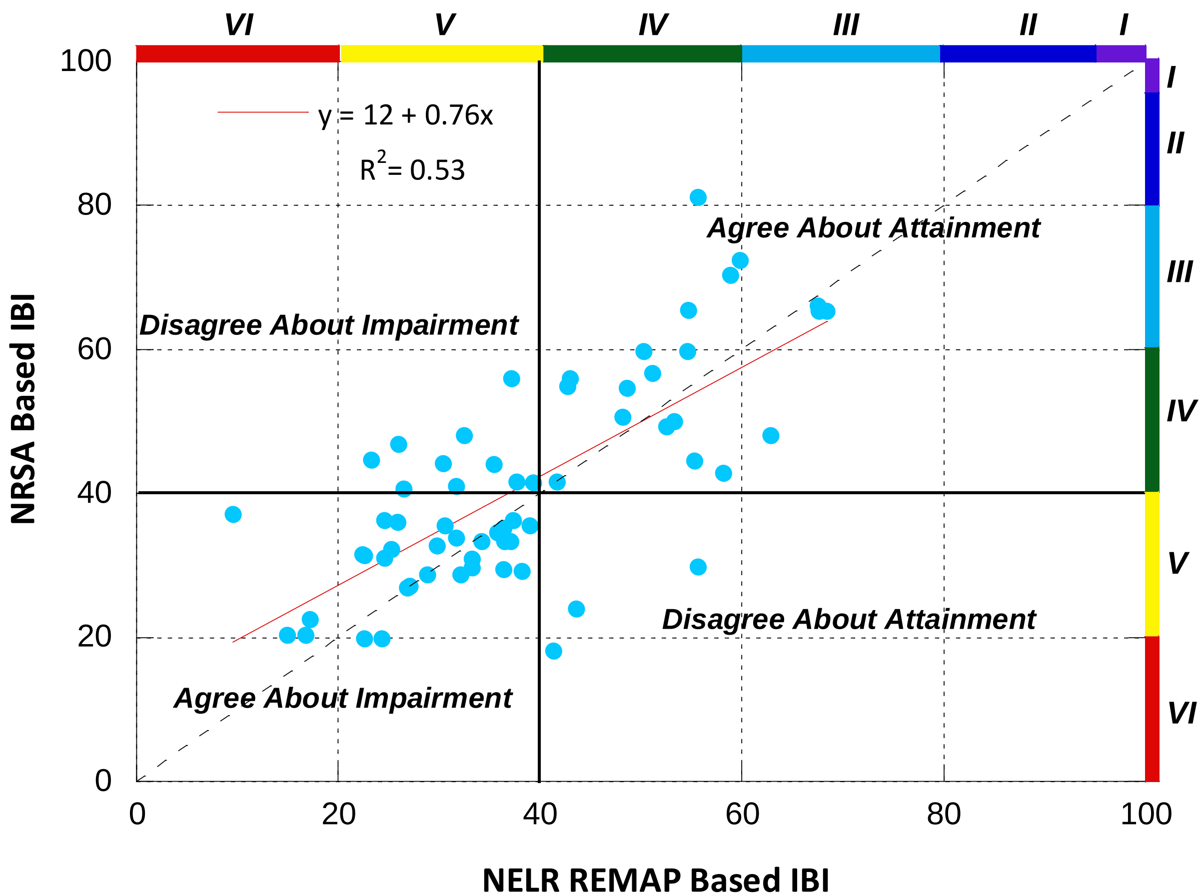


***Figure 70**. Scatterplot of NELR REMAP based IBI (not adjusted) and NRSA based IBI results at 64 New England non-wadeable river comparability sites. The NELR REMAP attainment threshold was used to draw quadrants of agreement and disagreement between the two IBIs about attainment and non-attainment of the NELR REMAP attainment threshold (IBI = 40). The six corresponding BCG levels are indicated along the x2 and y2 axis.*

The NELR REMAP results were used as the standard for agreement or disagreement with the NRSA results (Table 26). The NRSA agreed about attainment at 20 of 23 sites judged as attaining the threshold by NELR REMAP which assigned 3 additional sites as impaired. The NRSA agreed about impairment at 31 of 41 sites judged as impaired by NELR REMAP which assigned 10 additional sites to impaired status. The agreement about BCG level was less consistent with the same level assigned for 39 of 64 sites (61%). Almost all of the disagreements were at the boundary between a BCG Level 4 and 5 score. A scatterplot of the

***Table 26**. Table comparing the Maine IBI at NRSA sites vs. similarly located REMAP stations. NRSA samples always exclude white sucker biomass metric because no weights are collected and 11 metrics standardized to a 12 metric scoring range. REMAP sites have biomass data and IBIs represent data with the white sucker metric (top) or removed and standardized as with the NRSA data. Light blue shaded results are in agreement about the BCG level.*

NRSA Based IBI	**REMAP Based IBI (Not Adjusted)**					
BCG Level	Level 6: IBI <20	Level 5: IBI ≥20; <40	Level 4: IBI ≥40; <60	Level 3: IBI ≥60; <80	Level 2: IBI ≥80; <95	Level 1: IBI ≥95
Level 1: IBI ≥ 95	0	0	0	0	0	0
Level 2: IBI ≥80; < 95	0	0	1	0	0	0
Level 3: IBI ≥60; < 80	0	0	3	3	0	0
Level 4 IBI ≥40; < 60	0	10	12	1	0	0
Level 5: IBI ≥20; < 40	4	24	2	0	0	0
Level 6: IBI <20	0	3	1	0	0	0
NRSA Based IBI	**REMAP Based IBI (Adjusted)**					
BCG Level	Level 6: IBI <20	Level 5: IBI ≥20; <40	Level 4: IBI ≥40; <60	Level 3: IBI ≥60; <80	Level 2: IBI ≥80; <95	Level 1: IBI ≥95
Level 1: IBI ≥ 95	0	0	0	0	0	0
Level 2: IBI ≥80; < 95	0	0	1	0	0	0
Level 3: IBI ≥60; < 80	0	0	1	5	0	0
Level 4 IBI ≥40; < 60	0	11	11	1	0	0
Level 5: IBI ≥20; < 40	5	23	3	0	0	0
Level 6: IBI <20	0	2	1	0	0	0

NRSA based IBI and NELR REMAP IBI (non-adjusted) revealed a general directional agreement, but an r^2 of only 0.53 (Figure 70), an indication of substantial quantitative departures between each method. In this case the comparison used the NELR REMAP based IBI without adjustment as the inclusion of biomass data is one of the fundamental differences between the two methods.

Maine IBI Metric Comparisons

Comparisons between each of the 12 metrics of the ME IBI, fish relative abundance, and the top five species by numbers were visually examined by paired box-and-whisker plots.

Fish Relative Abundance

The comparable measures of relative abundance are numbers of fish/km in the aggregate and by species or species groupings. This can be an effective indicator of sampling sufficiency, but it does not always discriminate the distribution of fish species and life stages within a sample. Biomass (kg/km) would have been a more informative and at least complimentary indicator, but it was not measured by the NRSA. Aggregate relative numbers between NELR REMAP and NRSA samples overlapped, but the median and 75th percentiles were slightly higher for the NRSA (Figure 71). An observation reported by the NRSA field crews was that when the NRSA site included less diverse shoreline habitats (i.e., shallower inside bends, bedrock shoals, etc.) that numbers of commonly occurring, more tolerant Cyprindae and white suckers would frequently be at their highest. Such less diverse and monotonous habitats could be included in an NRSA site because the selection of the right or left shoreline was purely random and did not consider the "best habitats" as did the NELR REMAP protocol. A paired plot of the five most commonly occurring fish species (fallfish, spottail shiner, common shiner, golden shiner, and

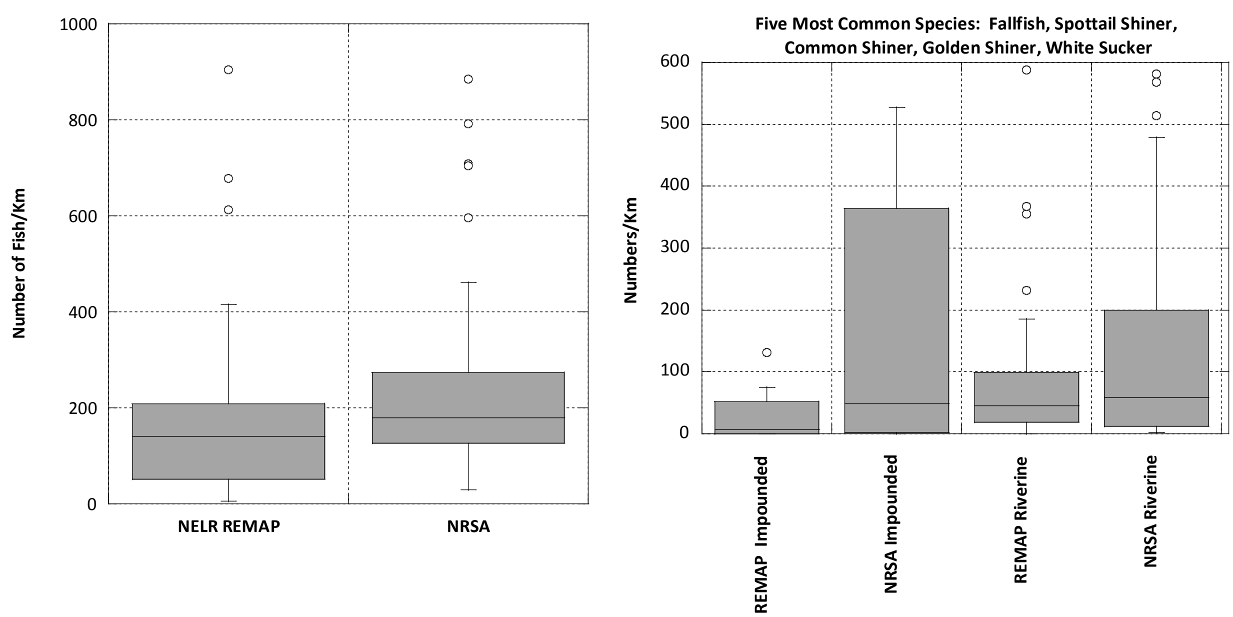


***Figure 71**. Box-and-whisker plots of the aggregate numbers of fish/km (left) and the numbers/km of the five most common fish species (right) between the NELR REMAP and NRSA comparison sites. The five most common species are further stratified by impounded and riverine sites.*

white sucker) in the NELR REMAP survey revealed significantly higher numbers of these species for the NRSA compared to lower numbers for the NELR REMAP (Figure 71). The differences suggest that a substantial number of NRSA sites produced samples dominated by one or more of these species. These are the most commonly occurring species in terms of the breadth of their distribution across New England rivers and they are also habitat generalists frequently found to be the predominant species in lesser habitat areas. The differences were greater at impounded sites thus reinforcing this assertion. As such this reflects a potential and perhaps unpredictable source of variation in the NRSA protocol by potentially irruptive species.

Native Species Richness

Differences in native species richness were slight with the minimum, median, and 25th percentile values being identical between NELR REMAP and the NRSA samples (Figure 72). However, some differences were evident and included a higher 75th percentile and statistical maximum for the NRSA although the latter was not higher than an outlier value for NELR REMAP. The higher 75th percentile and maximum is likely the result of where some NRSA sampling distances were two to four times that of the comparable NELR REMAP site. Increasing species richness with increased sampling distance is a well-known phenomenon.

Proportion of Native Cyprindae (less Fallfish)

The proportion of native Cyprindae (less fallfish) were generally comparable between NELR REMAP and NRSA samples with minor differences in a higher median value for the NRSA, but a higher maximum for the NELR REMAP samples (Figure 72). This metric precludes the potentially irruptive influence of fallfish by excluding this species.

Proportion of Native Salmonids

This metric is comprised of native Salmonidae in New England which includes brook trout and Atlantic salmon, the latter including landlocked populations – it excludes non-native rainbow trout and brown trout. Native salmonids were absent from most New England riverine sites due to a combination of modified thermal regimes and other stressors thus their presence was sporadic at the comparability sites. Despite this the NELR REMAP samples generally had higher values for this key and highly intolerant metric of the ME IBI (Figure 72). It seems likely that the differences were due to the NELR REMAP method being consistently more effective for Salmonids and the passive approach of the NRSA sampling method being less effective.

Proportion of Benthic Insectivores

The NRSA produced a higher proportion of benthic insectivores than the NELR REMAP samples in terms of the central tendencies of the results, but the NELR REMAP had a higher range of values (Figure 72). The absolute differences were minor with the 75th percentiles of the NRSA and NELR REMAP at approximately 8%.

Proportion of Blackbasses

This metric includes the proportion of smallmouth and largemouth bass both of which are introduced throughout New England. Both can act as opportunists especially in degraded coldwater habitats that have warmed enough to permit the ingress of these species. The NRSA

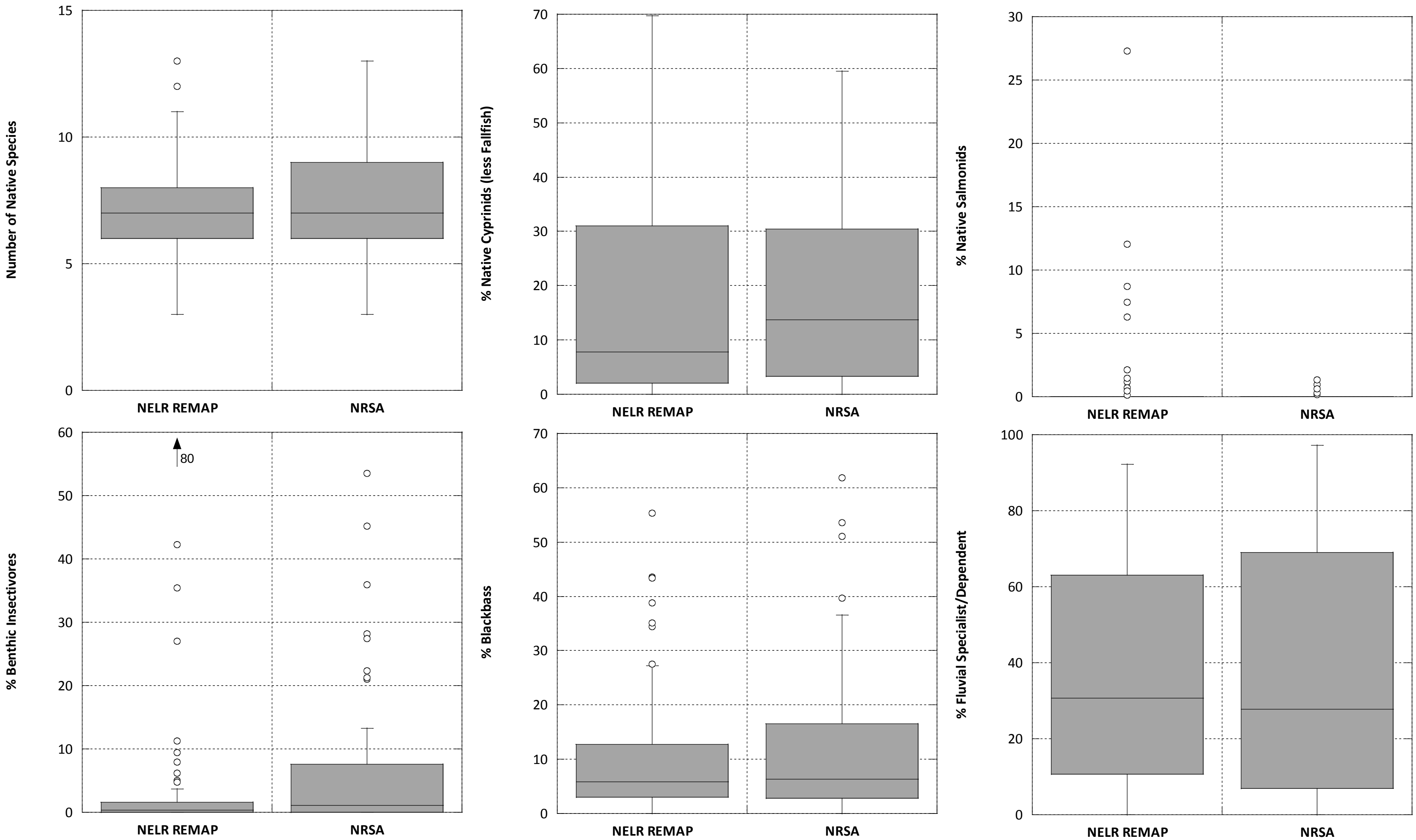


Figure 72*. Box-and-whisker plots comparing results for six metrics of the Maine IBI between the NELR REMAP and NRSA comparison sites; native species richness (upper left), % native Cyprindae (less Fallfish; upper middle), % native Salmonidae (upper right), % benthic insectivores (lower left), % Blackbasses (lower middle), and % fluvial specialist/dependent species (lower right).*

had a slight tendency to have higher proportions, but the median and 25th percentile values were identical (Figure 72). Given the distribution of blackbasses across most of the habitat types sampled by either method the comparable results are not surprising.

Proportion of Fluvial Specialist and Dependent Species
The results were generally comparable with the NRSA having a slightly wider range of extremes than the NELR REMAP (Figure 72).

Proportion of Macrohabitat Generalists
The NRSA samples produced a slightly higher proportion of macrohabitat generalists, but the range of values were essentially the same (Figure 73). The propensity of the NRSA to include lesser quality habitats might have contributed to this result as macrohabitat generalists would be more likely to be present in such habitats.

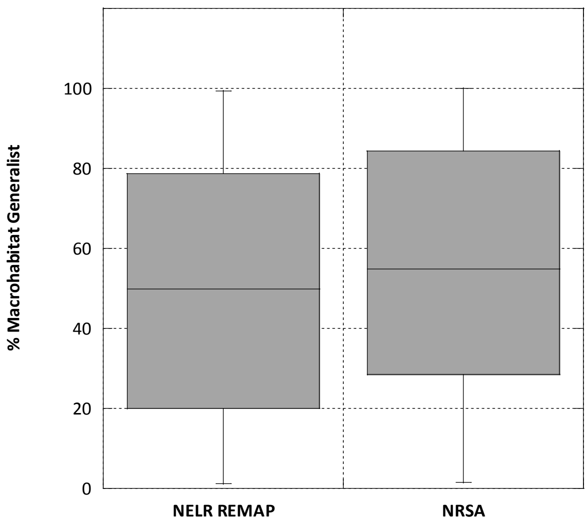
Number of Stenothermic Species
This metric includes obligate coldwater fish species. The results were the most comparable of any of the 12 IBI metrics between the NELR REMAP and NRSA samples with the results being nearly identical (Figure 73).

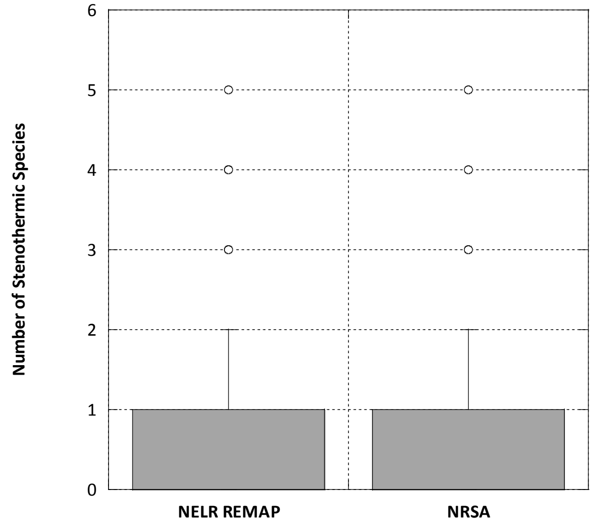
Number of Non-Guarding Lithophilic Species
This metric is comprised of true riverine fish species. Similar to the previous metric the results were nearly identical between the NELR REMAP and NRSA samples (Figure 73).

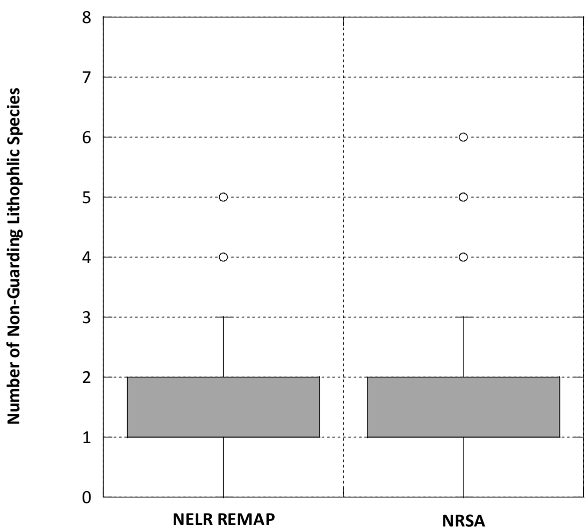
Number of Non-Indigenous Species
Non- indigenous species comprise a significant portion of the New England fish fauna at present, especially in the southern latitudes. The results between the NELR REMAP and NRSA samples were similar with only minor differences noted (Figure 73).

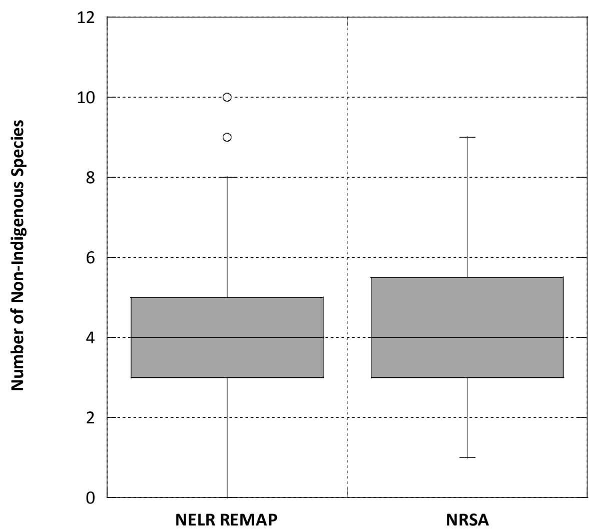
Percentage of Fish with DELT Anomalies
This metric includes the proportion of fish with deformities, erosions, lesions, and tumors (DELTs) and it is an indicator of sublethal stresses. The NELR REMAP samples produced higher DELT anomalies than the NRSA (Figure 73). The higher DELT anomalies yielded from the NELR REMAP samples may well be attributed to the recording of biomass data and the perhaps closer scrutiny of individual fish condition that process entails.

Discussion of Comparability Results
The analyses conducted herein suggested that there are important site-specific differences, but determining these would require additional analyses that were not conducted by this study. However, and with some notable exceptions, the NELR REMAP and NRSA produced comparable results at the assemblage assessment level on a regional basis. The differences that we observed would have less influence on a pass/fail or the “good-fair-poor” level of assessment of the NRSA as the detail along the BCG gradient is less important in such truncated assessment paradigms. It would seem feasible then to use the NRSA and NELR REMAP methods interchangeably at this level of regional assessment. It also exposes the limitations of this level









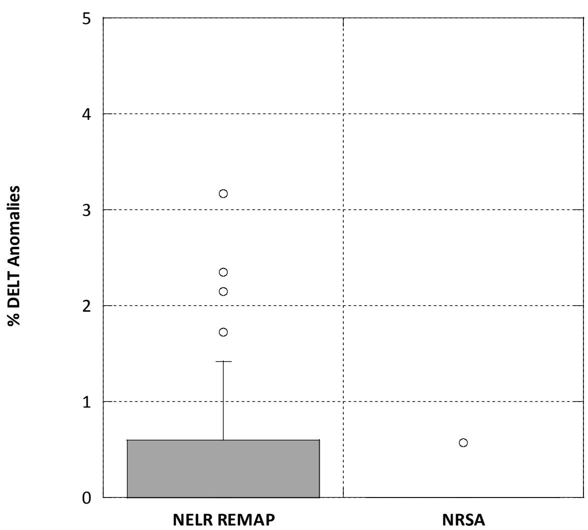


***Figure 73**. Box-and-whisker plots comparing results for five metrics of the Maine IBI between the NELR REMAP and NRSA comparison sites; % macrohabitat generalists (upper left), number of stenothermic species (upper right), number of non-guarding lithophils (middle left), number of non-indigenous species (middle right), and % DELT anomalies (lower left).*

of comparability because critical differences that would be important at a more detailed spatial scale or under a more refined and rigorous assessment scale where such differences would have mattered. The two metrics where differences were noted include the native Salmonid and %DELT metrics. The NELR REMAP method showed higher raw values and a greater frequency of those values which is an indication that the NRSA sampling method is vulnerable to under-rating these attributes at selected sites. Given the importance of native salmonids to discriminating between the upper BCG levels and %DELT to discriminating among lower BCG levels, the inability to reveal such differences when they exist would not translate to a more refined IBI along the scale of the BCG. The net effect is a methodology that is limited to pass/fail assessment and a reduced capacity to support refined aquatic life uses and biocriteria.

Another key difference is that the NRSA method by virtue of the site length could mask important site-specific differences that might occur within the NRSA sampling reach especially where site lengths approach the maximum of 4 km. An example of this is in the Connecticut River downstream from the Turners Falls dam which is affected by flow diversions to the Cabot hydropower project. Two NRSA sites were located within the 3+ miles of the bypass reach and four NELR REMAP were co-located within this reach in keeping with the convention of co-location two 1.0 km NELR REMAP sites within an NRSA site. In addition, the focus of the 2008-9 Connecticut River intensive survey was to assess possible local scale effects fǊ2m stressors such as the flow diversion for the Cabot hydropower project. River flows in an approximate 3.5 mile long reach of the Connecticut River are effectively modified with most of the flow being diverted into a canal that provides water to the Cabot hydroelectric generating station. A minimum flow of 120 cfs is maintained over the Turners Falls dam during low flow periods. The result is a very constricted wetted channel with the wider natural channel lacking flows that are comparable to a typical New England moderate-high gradient river of this size (Figure 74). As a result the habitat consisted almost entirely of pools with little or no flow velocity that was especially pronounced in the upper reach that is represented by the upstream most NELR REMAP site (RM 67.9). ME IBI, DIBI, and metric results for the two NRSA sites and the four overlapping NELR REMAP sites are shown in Table 27. The results between the longer NRSA sites and the shorter fixed distance of the first two NELR REMAP sites that were “embedded” within each of the two NRSA sites reveal contrasting ME IBI and DIBI scores and metric values. The NRSA site that started downstream from the Turners Falls dam and extended 4 km downstream into the bypass reach (RM 67.9) revealed BCG Level 5 (poor) as measured by both the ME IBI and DIBI. Three metrics reflected level 6 and included native species richness, alien species, and proportion of blackbasses. Other metrics were largely level 4 and 5, but there were no DELT anomalies recorded and a BCG Level 3 (good) result for benthic insectivores. By comparison, the first NELR REMAP site (RM 67.9) revealed level 6 (very poor) quality and the second NELR REMAP site was level 4 (fair) for the ME IBI. The ME IBI + DIBI was one BCG level better for each indicating a higher abundance of diadromous species in these samples. These two sites were sampled later in the summer than the NRSA site which could explain some of the difference between the ME IBI and DIBI. Metric values were much lower at the NELR REMAP site RM 67.9 which had the lowest ME IBI score in the entire Connecticut River during 2008-9 and all of New England. By contrast the results were much improved at the second

***Table 27**. Fish sampling results in and downstream from the Turners Falls bypass reach in the Connecticut River and in the vicinity of the Cabot hydropower project in 2009 showing Maine IBI and DIBI scores and Maine IBI metric results. Gray shaded NELR REMAP sites were "embedded" within the preceding NRSA site (site numbers are from NRSA, NELR REMAP, and Connecticut River intensive survey designations). Color shading in the cells corresponds to the BCG level for each result (see Table 24).*

River Mile	Site Number	Distance (Km)	Maine IBI (BCG)	Maine DIBI (BCG)	Native Species	Steno-therms	Alien Sp.	Non-guarding Lithophils	%Cyprini-dae	Native Salmon-ids	Benthic Insecti-vores	% Black-bass	Fluvial Special-ist	Macro-habitat General-ists	White/Long-nose Suckers[1]	%DELT
67.9 (NRSA)	FW08MA 021	3.81	34 (5)	39.5 (5)	3	1	6	2	27	0	21	51	29	68	NA	0
67.9 (REMAP)	FW08MA 021	1.0	10 (6)	25.1 (5)	2	0	3	0	0	0	9	44	9	62	0	2.4
66.9	CTR-46	1.0	57 (4)	74.4 (3)	5	1	2	2	18	10	19	13	26	17	22	0
66.1 (NRSA)	FW08MA 020	4.0	29 (6)	36.7 (5)	4	0	3	1	15	0	8	37	33	60	NA	0
66.1	CTR-47A1	1.0	22 (6)	34.3 (6)	6	0	5	2	52	0	4	27	12	85	0	0.4
65.5	CTR-46A	1.0	38 (5)	56.5 (4)	6	1	4	2	7	1	3	48	18	57	9	2.3

[1] This is a biomass based metric and it could not be calculated for the NRSA method.

***Figure 74**. Connecticut River downstream from the Turners Falls Dam showing the “bypass reach” (upper; RM 67.9 site) and the partial return of flows from the Cabot station bypass channel at the beginning of the RM 66.9 NELR REMAP site (lower). Photos taken on September 28, 2009.*

NELR REMAP site RM 66.9 which revealed BCG Level 4 (fair) for the ME IBI and BCG Level 3 (good) for the DIBI. This site was downstream from the partial return of flows from the Cabot station feed channel. The ME IBI metric results showed a lower number of native species, higher number of alien species, a higher proportion of macrohabitat generalists, and zero %DELT anomalies at the two NRSA sites. The NELR REMAP sites revealed DELT anomalies at three of the four sites with BCG Level 6 (very poor) values at two sites. Some of these differences may be due to the different shorelines sampled by each protocol (see Figure 67, lower panel) and the difference in flow between NELR REMAP sites RM 67.9 (FW08MA021) and RM 66.9 (CTR-46), the latter having higher flows due to the return of water from the Cabot station feed channel. The NRSA site was sampled in July 2009 under slightly higher flows over the Turners Falls dam which resulted in the bedrock along the west shoreline being covered by approximately 12-18 inches of water. This same area was dry in September (see Figure 74). The NELR REMAP site was located along the east shoreline which comprised the “best” bank when the sampling was conducted in late September 2009 (this would have been the case when the NRSA site was sampled in late July). This partially illustrates the consequence of the random selection of the NRSA transects vs. the “best” bank approach of the NELR REMAP protocol. Despite this the NELR REMAP results were poorer than the NRSA result due primarily to the lack of sufficient flow in the upper bypass reach. The results at the second NRSA site (RM 66.1) were less contrasting with the exception of a higher proportion of blackbass and macrohabitat generalists such that these reflected BCG Level 6 (very poor) for the BCG. DELT anomalies were also recorded at the NELR REMAP sites with zero observed in the NRSA sample. This latter observation is consistent with the results across the 64 NRSA sites and may reflect a method execution issue as opposed to the configuration of the sampling site. The comparison of the bypass reach sites showed varying BCG levels with the NRSA results tending to “homogenize” the overall condition as BCG Level 5 (poor) with the first NELR REMAP site (RM 67.9) showing BCG Level 6 (very poor) and the second NELR REMAP site BCG Level 4 (fair). When these two NELR REMAP sites are averaged the result is in closer agreement with the NRSA results. However, if the goal is to detect localized impacts from modifications to flow, habitat, and water quality this comparison suggests that the NELR REMAP protocol is more effective at revealing those issues.

NELR REMAP/NRSA Comparability Summary

The comparability of different biological assessment methods is an important, ongoing, and largely unresolved issue (Cao and Hawkins 2011; Cao et al. 2005). Most efforts to date have focused on examining raw data attributes and few if any have focused on assessment outcomes, which is what matters the most to the states and EPA in making choices about protocols when more than one is available to choose from. Dukerschein et al. (2013) is one of the few comparability studies that focused on assessment outcomes, in this case the application of two different methods of sampling fish assemblages in the upper Mississippi River. The results of their comparison was not unlike the NRSA/NELR REMAP comparison in that they concluded that the different methods could support broad pass-fail assessments, but anything more refined would be difficult with a more passive methodology. In our comparison the NELR REMAP method was better suited to detecting and characterizing local reach and longitudinal scale effects than the NRSA method. Simply put the variable and longer sampling site of the NRSA method can obscure important site-specific impacts by homogenizing these effects because of the longer site distance as was shown in the Turners Falls bypass reach in the Connecticut River. The NELR REMAP method also provides the opportunity to be consistent with the U.S. EPA (2013) critical technical elements of bioassessment level of rigor for spatial survey design which assigns a higher level of rigor to more spatially intensive monitoring approaches.

CHAPTER 6: CONSIDERATIONS FOR THE DEVELOPMENT OF TIERED USES AND BIOCRITERIA FOR NEW ENGLAND LARGE RIVERS

Overview

An expected outcome of this project is the description of a process for biocriteria including the development of reference condition, multimetric indices, and a template for tiered aquatic life uses for the non-wadeable rivers of New England using the Biological Condition Gradient (BCG; Davies and Jackson 2006) as a conceptual foundation. While a numeric index and interim threshold for a pass-fail framework was accomplished by the NELR REMAP project, further exploration of key issues underlying the development of biocriteria and tiered aquatic life uses is needed.

The key steps in this process include the following:

1. Natural Classification.
2. Reference Condition.
3. Biological condition gradient for major river classes.
4. Refined index development for each riverine class.
5. Derivation of thresholds for aquatic life use tiers stratified by riverine classes.
6. Development of supporting implementation tools.

Each of these steps is described herein by how they relate to what has been accomplished by NELR REMAP and what remains to be accomplished in the way of further refinements. Each step is also described in relation to how it would attain the highest level of technical rigor as described by *Biological Assessment Program Review: Assessing Level of Technical Rigor to Support Water Quality Management* (U.S. EPA 2013). While this document supports the evaluation of bioassessment programs mostly at the state program level, it has been used to evaluate bioassessment protocols at the regional level, hence it is used here to assess developments to date and to determine what steps remain to achieve the highest level of rigor for New England Large Rivers based on an examination of the development of 13 critical technical elements (Table 28; Appendix D).

Assessment of the NELR Bioassessment Protocol

The development of the current NELR Bioassessment Protocol has fully addressed the Index Period (Element 1) and Spatial Sampling Design (Element 2) critical technical elements. Each is sufficiently developed to support the derivation of biocriteria for the fish assemblage. The NELR protocol has also fully addressed Taxa and Taxonomic Resolution (Element 6), Sample Collection (Element 7), and Sample Processing (Element 8) *for a single assemblage (fish).* The equal development of a second assemblage is needed to attain the maximum score for these elements. Data management (Element 9) has been partially addressed and would require a more institutionalized process (as opposed to a project basis) to attain full development. Stressor Association (Element 12) was also partially addressed herein, but complete

***Table 28**. Summary definitions of the 13 critical technical elements across 3 disciplinary categories that reflect the level of rigor of a bioassessment program (after U.S. EPA 2013).*

	Technical Element	Definition
Biological Assessment Design	Index Period	A consistent time frame for sampling the assemblage to characterize and account for temporal variability.
	Spatial Sampling Design	Representativeness of the spatial array of sampling sites to support statistically valid inference of information over larger areas (e.g., watersheds, river and stream segments, geographic region) and for supporting water quality standards (WQS) and multiple programs.
	Natural Variability	Characterizing and accounting for variation in biological assemblages in response to natural factors.
	Reference Site Selection	Abiotic factors to select sites that are least impacted, or ideally, minimally affected by anthropogenic stressors.
	Reference Condition	Characterization of benchmark conditions among reference sites, to which test sites are compared.
Data Collection and Compilation	Taxa and Taxonomic Resolution	Type and number of assemblages assessed and resolution (e.g., family, genus, or species) to which organisms are identified.
	Sample Collection	Protocols used to collect representative samples in a water body including procedures used to collect and preserve the samples (e.g., equipment, effort).
	Sample Processing	Methods used to identify and count the organisms collected from a water body, including the specific protocols used to identify organisms and subsample, the training of personnel who count and identify the organisms, and the methods used to perform quality assurance/quality control (QA/QC) checks of the data.
	Data Management	Systems used by a monitoring program to store, access, and analyze collected data.
Analysis and Interpretation	Ecological Attributes	Measurable attributes of a biological community representative of biological integrity and that provide the basis for developing biological indices.
	Discriminatory Capacity	Capability of the biological indices to distinguish different increments, or levels, of biological condition along a gradient of increasing stress.
	Stressor Association	Relationship between measures of stressors, sources, and biological assemblage response sufficient to support causal analysis and to develop quantitative stress-response relationships.
	Professional Review	Level to which program data, methods, and procedures are reviewed by others.

development would be complete only when the other design and interpretation elements are more fully developed.

While the NELR REMAP and Maine Rivers projects partially addressed the remaining five elements by consequence of the initial BCG and IBI development, they are incomplete at this time. Additional exploration and development within each element is needed and if done in the appropriate sequence would be complimentary in terms of elevating the technical rigor and detail of each element. A project focused on each and guided by BCG principles would deliver a more robust and rigorous set of assessment tools and biocriteria.

Natural Variability

This pertains to the classification basis for an assemblage index such as exists in only a few places in New England, i.e., the Vermont wadeable streams IBI that distinguishes between cold and mixed water assemblages (Langdon 2001). The basis for natural classification affects the types and composition of metrics that comprise an IBI type of index. The ME IBI that we used is predicated on a moderate gradient cold water riverine fish assemblage which means the inclusion of metrics that are sensitive to reveal a gradient of response to stressors such as impacts to thermal and flow regimes. While this seems to have worked for the initial testing of the index in Maine, some exceptions were apparent and include naturally low gradient rivers. Besides gradient, these rivers are generally warmer and occur in conjunction with extensive wetland complexes. Thus it seems inappropriate to evaluate such rivers that have a different natural baseline with an IBI developed against a starkly different natural baseline. As the ME IBI was applied throughout New England it became apparent that some rivers may be outside the cold water paradigm of the index, thus a better thermal classification scheme needs to be developed and tested. Such is at least conceptually available from the Northeast Aquatic Habitat Classification (NEAHCS; Olivero and Anderson 2008) that includes stream and river size, gradient, and temperature regime. A similar scheme has been developed for Wisconsin streams and rivers and is currently being examined as the principal basis for an underlying natural classification scheme in support of biocriteria development (Figure 75).

One issue to resolve is the existing assessment of many sites in southern New England as being impaired when using the ME IBI. Without an accompanying relevant classification scheme, it is difficult to determine if the current fish assemblages are indeed impaired by the accumulation of human-induced disturbances that have substantially altered flow and thermal regimes. This begs the question about the "as naturally occurs" true natural potential of these rivers. If a cold water or transitional cold system is the "as naturally occurs" then dealing with degraded rivers becomes a different matter than if they are simply misclassified and the resulting assessment is inaccurate due to the application of the wrong baseline. The mapping of the NEAHCS classification at the NELR REMAP sites (see Ch. 3; Figure 22) shows that transitional cold water assemblages (which are consistent with the ME IBI) occur as far south as mid-Connecticut and Rhode Island. There were many more sites that were classified as transitional warmwater, but this observation raises the question of whether this is a natural phenomenon or an artifact of two centuries of human-induced changes to flow and thermal regimes. All of these observations and questions point to the need to better develop a classification scheme with IBI development specific to those classes to follow.

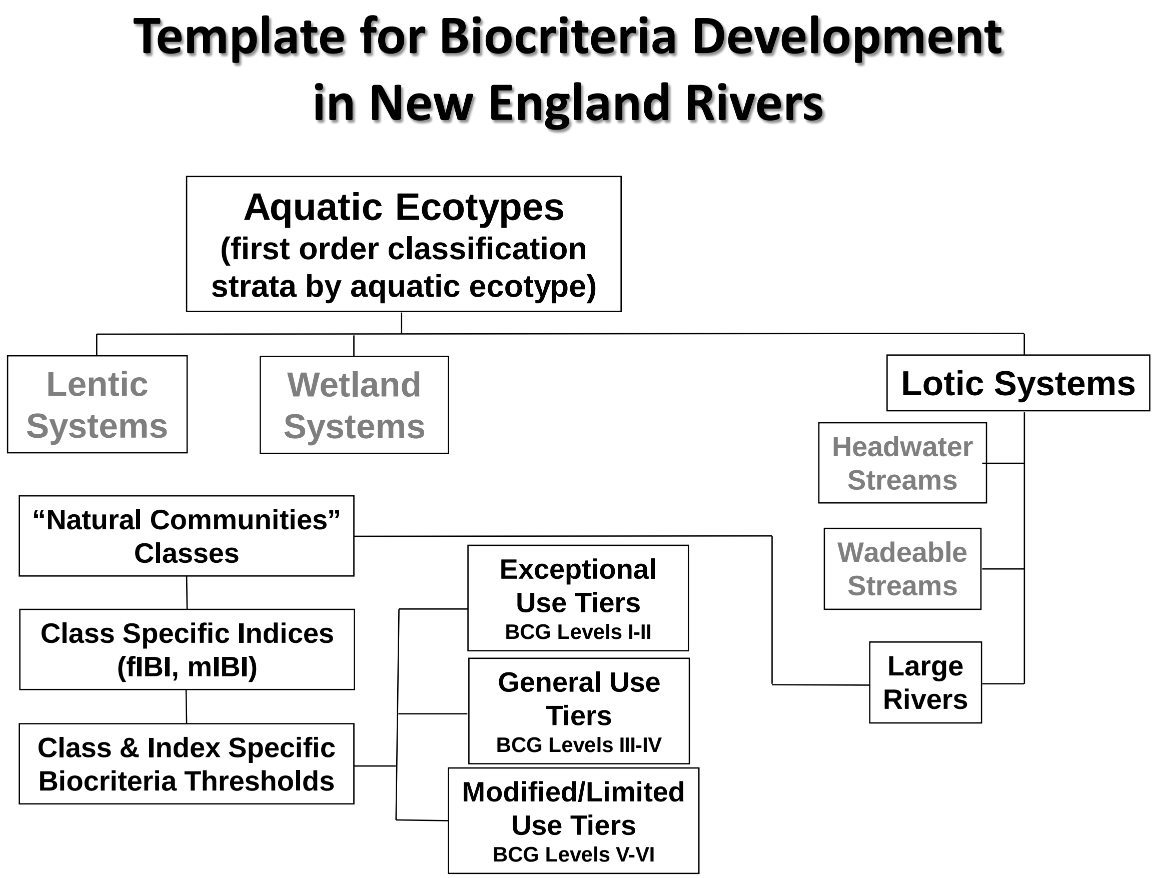


***Figure 75**. Template for the development of biocriteria and tiered aquatic life uses for New England Rivers based on a natural communities classification framework.*

Reference Site Selection and Reference Conditions

These two critical elements pertain to the criteria used to screen and select reference sites that are in turn used to derive reference conditions thus they are considered here in tandem. The Maine Rivers project included the selection of reference sites, but they were not used directly in the ME IBI development and derivation process. Instead, candidate metrics were evaluated against a habitat gradient and reference sites that were selected to exclude the occurrence of blackbasses and other introduced species. ME IBI assessment thresholds were assigned using a “desktop” BCG analysis and resulted in ranges of the ME IBI that correspond to the six different BCG levels. For the time being an IBI that scores at the minimum for BCG Level 4 was considered as meeting the interim goal of the CWA. This arrangement seemed to work for the ME IBI development and also herein for the NELR REMAP assessment against the stressors that were evaluated. While it did not directly reflect the traditional reference site and condition approaches that have been more widely employed (Stoddard et al. 2006), it did account for many of the same conceptual elements. Add to this that non-native and invasive species not only comprise a major negative impact on native fish assemblages, they are pervasive throughout New England in all except the most northern drainages in Maine. This begs the same question as with natural classification, are the current conditions reflective of human-induced changes or natural ones? With regard to non-native species their origin is clearly human-induced, but is their prominence at many locations an artifact of a natural or altered set of circumstances? At this point it does not matter if their presence is or is not irreversible, but rather is their presence an indication of the level of disturbance as reflected by the BCG? Thus re-examining the mechanics of reference site selection and the derivation of reference conditions are important to pursue and refine in the future.

Ecological Attributes

This critical element pertains to how well the composition of an IBI reflects the properties and characteristics of a BCG relevant to the natural classes that are at issue. In the case of the ME IBI the metrics were at least implicitly based on a BCG model for a moderate-high gradient cold water riverine fish assemblage. The derivation of the ME IBI was based on a more traditional IBI calibration approach that assumes a relationship with the BCG. We believe this is defensible so long as the metrics reflect critical BCG attributes and a sufficiently relevant geographical dataset that includes the full array of quality levels along the BCG is available. Again, this seemed to fit the Maine Rivers dataset, with perhaps the lack of true BCG Level 1 analogs being represented. Still, there was enough breadth in the range of IBI scores to justify these assertions. If other distinct natural riverine classes are identified the task of developing representative IBI metrics should follow a similar path.

Discriminatory Capacity

This critical element pertains to the statistical properties of an IBI and the capacity to distinguish multiple categories of condition. This capacity is essential to the development and implementation of tiered aquatic life uses (see Figure 75), but the ME IBI has not been tested in this manner. Instead, ranges of the IBI that correspond to BCG levels have been assumed using an estimation process. Techniques are available to test this attribute of an IBI (e.g. Fore et al. 1993) and would need to be accomplished to verify the score for this element.

Summary

The NELR REMAP protocol scores at 79.8% which is consistent with a Level 2 program (Appendix Table D-1). This means the protocol is suited for producing large scale trend assessments, which was accomplished via the NELR REMAP assessment. Adding an equally developed second assemblage would elevate the protocol to an upper Level 3. Elevating the level of rigor is an important step in the development of biological criteria and tiered aquatic life uses. Attaining a Level 4 technical rigor and having full TALU program support and implementation are *mutually inclusive.* The steps outlined in Appendix Table D-2 would need to be completed to attain a Level 4 technical rigor.

References

Angermier, P.L. and J.R. Karr. 1986. Applying an index of biotic integrity based on stream-fish communities: considerations in sampling and interpretation. N. Am. J. Fish. Mgmt. 6: 418-427.

Armitage, B.J. and E.T. Rankin. 2009. An assessment of threats to the biological condition of the Wabash River aquatic ecosystem of Indiana. Report to The Nature Conservancy, Indiana Chapter. Midwest Biodiversity Institute, Columbus, OH. 86 pp. http://www.midwestbiodiversityinst.org/publications?type=L

Armstrong, D.S., Richards, T.A., and Levin, S.B. 2011. Factors influencing riverine fish assemblages in Massachusetts: U.S. Geological Survey Scientific Investigations Report 2011–5193, 59 p. (also available at http://pubs.usgs.gov/sir/2011/5193).

Bain, M. B., and M. S. Meixler. 2008. A target fish community to guide river restoration. River Res. Applic. 24: 453–458.

Barbour, M.T. and others. 1997. Revision to rapid bioassessment protocols for use in rivers and streams: periphyton, benthic macroinvertebrates, and fish. EPA 841-D-97-002. U.S. EPA, Office of Water, 4503F. Washington, D.C. (Draft).

Bartsch, A. F. 1948. Biological aspects of stream pollution. Sewage Works Journal 20:292-302.

Bartsch, A. F., and W. M. Ingram. 1967. Stream life and the pollution environment. Pages 119–127, in Biology of water pollution (L. E. Keup, W. M. Ingram, and K. M. MacKenthun, Editors). U.S. Department of the Interior, Washington, D.C.

Bigelow, H.B. and W.C. Schroeder. 1953. Fishes of the Gulf of Maine. U.S. Dept. of Commerce, Bureau of Fisheries, Washington, DC.

Blocksom, K.A. and J.E. Flotemersch. 2004. Comparison of macroinvertebrate sampling methods for nonwadeable streams. Env. Mon. Assess. (xxx): 1-20.

Cao, Y. and C.P. Hawkins. 2011. The comparability of bioassessments: a review of conceptual and methodological issues. J. N. Am. Benthol. Soc. 30(3): 680–701.

Cao, Y., C.P. Hawkins, and A.W. Storey. 2005. A method for measuring the comparability of different sampling methods used in biological surveys: implications for data integration and synthesis. Freshwater Biology 50: 1105-1115.

Collette, B.B. and G. Klein-McPhee. 2000. Fishes of the gulf of Maine. Third edition. Smithsonian Institution Press. Washington. 748 pp.

Curry, R.A. 2007. Late glacial impacts on dispersal and colonization of Atlantic Canada and Maine by freshwater fishes. Quaternary Research 67(2): 225-233.

Daly, C. and G. Taylor. 1988. United States Average Monthly or Annual Precipitation, 1961-90. (Corvallis, OR: Spatial Climate Analysis Service at Oregon State University, 1998).

Daniels, R.A., K. Riva-Murray, D.B. Halliwell, D. Vana-Miller, and M.D. Bilger. 2005. An Index of Biological Integrity for northern mid-Atlantic slope drainages. Trans. Am. Fish. Soc. 131:1044–1060.

Danielson, T. 2006. Standard Operating Procedure Methods for Sampling Stream and Wetland Algae. Maine Dept. of Environmental Protection, Bureau of Land and Water quality, Division of Environmental Assessment, Augusta, ME. DEPLW0634. 18 pp.

Davies, S.P. and C.O. Yoder. 2010. Region I State Biological Assessment Programs review: Critical Technical Elements Evaluation (2006-2010). Report submitted to U.S. EPA, Region I, Boston, MA. Grant X7-97166801. 148 pp. (also found at http://www.epa.ohio.gov/dsw/document_index/psdindx.aspx)

Davies, S.P. and S.K. Jackson. 2006. The biological condition gradient: a descriptive model for interpreting change in aquatic ecosystems. Ecological Applications 16 (4): 1251-1266 + appendices.

Davies, S.P. and L. Tsomides. 2002. Methods for Biological Sampling and Analysis of Maine's Rivers and Streams. Maine Dept. of Environmental Protection, Bureau of Land and Water quality, Division of Environmental Assessment, Augusta, ME. DEP LW0387-B2002. 31 pp.

Davies, S.P. and L. Tsomides. 1997. Methods for biological sampling and analysis of Maine's inland waters. Maine Dept. of Environmental Protection, Bureau of Land and Water quality, Division of Environmental Assessment, Augusta, ME. 29 pp.

Davis, W.S. and T.P. Simon. 1995. Biological criteria and assessment: tools for water resource planning and decision making. Lewis Publishers, Boca Raton, FL. 415 pp.

Detenbeck, N.E., L. Hayes, C. Rosiu, J. Legros, D. Parsley, and A. Sherman. 2010. Distinguishing the effects of point source from those caused by upstream nonpoint source (NPS) inputs: Refinement of a watershed development index for New England. Presentation at 2010 New England Association of Environmental Biologists (NEAEB), Newport, RI. (found at http://www.epa.gov/aed/html/comm/posters.html).

Dukerschein, J.T., A.D. Bartels, B.S. Ickes, and M.S. Pearson. 2013. Are two systemic fish assemblage sampling programmes on the upper Mississippi River telling us the same thing? River Res. Applic. 29: 79-89.

Emery, E.B., R. Tewes, R. Argo, and C.O. Yoder. 2007. Development of a probability-based monitoring and assessment strategy for select mainstem large rivers within US EPA Region V. Report submitted to U.S. EPA, Assistance I.D. RM-83169201-0. Ohio River Valley Water Sanitation Commission, Cincinnati, OH. 99 pp. + appendices.

Emery, E. B., T. P. Simon, F. H. McCormick, P. A. Angermier, J. E. DeShon, C. O. Yoder, R. E. Sanders, W. D. Pearson, G. D. Hickman, R. J. Reash, J. A. Thomas. 2003. Development of a Multimetric Index for Assessing the Biological Condition of the Ohio River. Trans. Am. Fish. Soc. 132:791-808.

Flotemersch, J.E., J.S. Stribling, R.M. Hughes, L. Reynolds, M.J. Paul, and C. Wolter. 2010. Site length for biological assessment of boatable rivers. River Res. Applic. 27: 520–535.

Flotemersch, J.E. and K.A. Blocksom. 2004. Electrofishing in boatable rivers: does sampling design affect bioassessment metrics. Env. Mon. Assess. (xxx): 1-21.

Flotemersch, J. E., B. C. Autrey, and S. M. Cormier. 2001. Comparisons of boating and wading methods used to assess the status of flowing waters. U.S. Environmental Protection Agency, Cincinnati, Ohio, EPA/600/R-00/108.

Fore, L.S., J.R. Karr, and L.L. Conquest. 1993. Statistical properties of an index of biotic integrity used to evaluate water resources. Can. J. Fish. Aquatic Sci. 51: 1077-1087.

Gammon, J.R., Spacie, A., Hamelink, J.L., and R.L. Kaesler. 1981. Role of electrofishing in assessing environmental quality of the Wabash River, in Ecological assessments of effluent impacts on communities of indigenous aquatic organisms, in Bates, J. M. and Weber, C. I., Eds., ASTM STP 730, 307 pp.

Gammon, J.R. 1980. The use of community parameters derived from electrofishing catches of river fish as indicators of environmental quality. pp. 335-363 in Seminar on water quality management trade-offs (point source vs. diffuse source pollution). EPA-905/9-80-009.

Gammon, J.R. 1976. The fish populations of the middle 340 km of the Wabash River, Purdue Univ. Water Res. Research Cen. Tech. Rep. 86. 73 p.

Gammon, J.R. 1973. The effect of thermal inputs on the populations of fish and macroinvertebrates in the Wabash River. Purdue Univ. Water Res. Research Cen. Tech. Rep. 32. 106 pp.

Goldstein, R.M. and T.P. Simon. 1999. Toward a unified definition of guild structure for feeding ecology of North American freshwater fishes, pp. 97-122. *in* T.P. Simon (ed.), Assessing the Sustainability and Biological Integrity of Water Resources Using Fish Communities. CRC Press, Boca Raton, FL.

Halliwell, D.B. 2005. Introduced Fish in Maine. Maine DEP, Internet: http//www.lakesofmaine.org/library/fish/Halliwell (formerly MABP Series – Focus on Freshwater Biodiversity). 14 pp.

Halliwell, D.B, R.W. Langdon, R.A. Daniels, J.P. Kurtenbach, and R.A. Jacobson. 1999. Classification of freshwater fish species of the northeastern United States for use in the development of indices of biological integrity, with regional applications, pp. 301-338. *in* T.P. Simon (ed.), Assessing the Sustainability and Biological Integrity of Water Resources Using Fish Communities. CRC Press, Boca Raton, FL.

Hartel, K.E., D.B. Halliwell, and A.E. Launer. 2002. Inland fishes of Massachusetts. Massachusetts Audubon Press, Faunal Series No. 3, Lincoln, MA.

Hokanson, K. E. F. 1977. Temperature requirements of some percids and adaptations to the seasonal temperature cycle. J. Fish. Res. Bd. Can. 34, no. 10: 1524-50.

Hughes, R.M., S. G. Paulsen, and J.L. Stoddard. 2000. EMAP-surface waters: a multiassemblage, probability survey of ecological integrity in the U.S.A. Hydrobiologia 422/423: 429-443.

Hughes, R.M., P.R. Kaufmann, A.T. Herlihy, T.M. Kincaid, L. Reynolds, and D.P. Larsen. 1998. A process for developing and evaluating indices of fish assemblage integrity. Can. J. Fish. Aquatic Sci. 55: 1618-1631.

Hughes, R.M. and J.R. Gammon. 1987. Longitudinal changes in fish assemblages and water quality in the Willamette River, Oregon. Trans. Am. Fish. Soc., 116: 196-209.

Jackson, D.A. 2002. Ecological effects of *Micropterus* introductions: the dark side of black bass, pp. 221-231. *in* Phillip, D.P. and Ridgway, M.S. Black Bass: Ecology, Conservation, and Management. Amer. Fish. Soc. Symp. 31. Bethesda, MD. 724 pp.

Karr, J.R. 1981. Assessment of biotic integrity using fish communities. Fisheries 6(6): 21-27.

Karr, J. R., K. D. Fausch, P. L. Angermier, P. R. Yant, and I. J. Schlosser. 1986. Assessing biological integrity in running waters: a method and its rationale. Illinois Natural History Survey Special Publication 5: 28 pp.

Kovacs, T.G., P.H. Martel, and R.H. Voss. 2002. Assessing the biological status of fish in a river receiving pulp mill effluents. Environmental Pollution, 118: 123-140.

Lamouroux, N., N. L. Poff, and P. L. Angermeier. 2002. Intercontinental convergence of stream fish community traits along geomorphic and hydraulic gradients. Ecology, 83(7): 1792–1807.

Langdon, R.W., M.T. Ferguson, and K.M. Cox. 2006. Fishes of Vermont. Vermont Agency of Natural Resources, Dept. of Fish and Wildlife. Waterbury, VT. 320 pp.

Langdon, R.B. 2001. A preliminary Index of Biotic Integrity for fish assemblages of small coldwater streams in Vermont. Northeastern Nat. 8(2): 219-232.

Libby, A.D. 2013. Inland fishes of Rhode Island. Rhode Island Dept. of Env. Mgmt.

Lyons, John, R.R. Piette, and K.W. Niermeyer. 2001. Development, validation, and application of a fish-based index of biotic integrity for Wisconsin's large warmwater rivers. Trans. Amer. Fisheries Society: Vol. 130, No. 6, pp. 1077–1094.

Lyons, J., L. Wang, and T.D. Simonson. 1996. Development and validation of an index of biotic integrity for coldwater streams in Wisconsin. North American Journal of Fisheries Management 16:241–256.

Martin, E. H. and C. D. Apse. 2011. Northeast Aquatic Connectivity: An Assessment of Dams on Northeastern Rivers. The Nature Conservancy, Eastern Freshwater Program.

McCune, B. and J. B. Grace. 2002. Analysis of Ecological Communities. MjM Software Design, Gleneden Beach, Oregon, USA.

Mebane, C.A., T.R. Maret, and R.M. Hughes. 2003. An index of biotic integrity (IBI) for Pacific Northwest rivers. Trans. Am. Fish. Soc. 132: 239-261.

Midwest Biodiversity Institute (MBI). 2008. Quality Assurance Project Plan and Detailed Work Plan: Development and Assessment of Methods and Designs for the Biological Assessment of Non-Wadeable Rivers in New England. Submitted to U.S. EPA, ORD-AED and Region I (effective date June 30, 2008). Midwest Biodiversity Institute, Inc. Columbus, OH. 58 pp.

Midwest Biodiversity Institute (MBI). 2004. Region V state bioassessment and ambient monitoring programs: initial evaluation and review. Midwest Biodiversity Institute Tech. Rept. MBI/01-03-1. 36 pp. + appendices. (http://www.epa.gov/region5/water/wqb/wqb_r5mon.htm)

Miltner, R.M. and E.T. Rankin. 2009. Analysis of National Wadeable Stream Survey Results in Region V 2004-5. MBI Technical Report MBI/2009-03-1. Final Report (Grant X-96536801) to U.S. EPA, Region V, Water Division, Chicago, IL. 77 pp. http://www.midwestbiodiversityinst.org/index.php.

Mundahl, N.D. and T.P. Simon. 1999. Development and application of an index of biotic integrity for coldwater streams of the upper Midwestern United States, pp. 383-416. *in*

T.P. Simon (ed.), Assessing the Sustainability and Biological Integrity of Water Resources Using Fish Communities. CRC Press, Boca Raton, FL.

Moore, R.B., C.M. Johnston, K.W. Robinson and J. R. Deacon. 2004. Estimation of Total Nitrogen and Phosphorus in New England Streams Using Spatially Referenced Regression Models. Scientific Investigations Report 2004-5012. U.S. Geological Survey, Pembroke, New Hampshire: 2004.

Nelson, J.S. and 6 others. 2004. Common and Scientific Names of Fishes from the United States, Canada, and Mexico. Sixth Edition. American Fisheries Society Spec. Publ. 29. 386 pp.

Ohio Environmental Protection Agency. 2006. Methods for assessing habitat in flowing waters: using the qualitative habitat evaluation index (QHEI). Division of Surface Water, Ecological Assessment Section, Columbus, OH. 23 pp. (also found at http://www.epa.ohio.gov/dsw/document_index/psdindx.aspx).

Ohio Environmental Protection Agency. 1996. Ohio EPA's guide to DELT anomalies (deformities, erosions, lesions, and tumors). DSW-MAS-EAU. Working Copy. Columbus, OH. 19 pp.

Ohio Environmental Protection Agency. 1989a. Biological criteria for the protection of aquatic life. volume III: standardized biological field sampling and laboratory methods for assessing fish and macroinvertebrate communities, Division of Water Quality Monitoring and Assessment, Surface Water Section, Columbus, Ohio. (also found at http://www.epa.ohio.gov/dsw/document_index/psdindx.aspx)

Ohio Environmental Protection Agency. 1989b. Addendum to Biological criteria for the protection of aquatic life: volume II. users manual for biological field assessment of Ohio surface waters, Division of Water Quality Monitoring and Assessment, Surface Water Section, Columbus, Ohio. (also found at http://www.epa.ohio.gov/dsw/document_index/psdindx.aspx)

Ohio Environmental Protection Agency. 1987. Biological criteria for the protection of aquatic life: volume II. users manual for biological field assessment of Ohio surface waters, Division of Water Quality Monitoring and Assessment, Surface Water Section, Columbus, Ohio. (also found at http://www.epa.ohio.gov/dsw/document_index/psdindx.aspx)

Olivero, Arlene and Anderson, Mark. 2008. Northeast Aquatic Habitat Classification System. Boston. http://rcngrants.org/node/38.

Olsen, A.R. 2007. National Flowing Waters Assessment Survey Design: 2008-2009. Created 7/16/2007, Revised 8/14/07. Contact: Susan Holdsworth, Office of Water, OWOW

Monitoring Branch, USEPA (4503T), 1200 Pennsylvania Ave. NW., Washington, DC 20460. http://water.epa.gov/type/rsl/monitoring/riverssurvey/index.cfm.

Olsen, A.R., J. Sedransk, D. Edwards, C.A. Gotway, W.Ligget, S. Rathbun, K.H. Reckhow, L.J. Young. 1999. Statistical issues for monitoring ecological and natural resources in the United States. Environmental Monitoring and Assessment 54: 1–45.

Omernik, J. M. 1987**.** Ecoregions of the conterminous United States. Annals of the Association of American Geographers 77(1): 118-125.

Page, L.M., H. Espinoza-Perez, L.T. Findley, C.R. Gilbert, R.N. Lea, N.E. Mandarak, R.L. Mayden, and J.S. Nelson. 2013. Common and scientific names of fishes from the United States, Canada, and Mexico, 7th edition. American Fisheries Society, Special Publication 34, Bethesda, Maryland. 384 pp.

Rankin, E.T. and C.O. Yoder. 2011a. Improving Water Quality Standards and Assessment Approaches for the Upper Mississippi River: UMR Clean Water Act Biological Assessment Implementation Guidance: Development of a Biological Condition Gradient for Fish Assemblages of the Upper Mississippi River and Development of a "Synthetic" Historical Fish Community. MBI Technical Report/2011-5-2. Submitted to UMRBA WQTF. 24 pp. http://www.midwestbiodiversityinst.org/publications?type=L.

Rankin, E.T., R. Mueller, and C.O. Yoder. 2011b. Ecological Low Flow Protection Process for Ohio Streams and Rivers of the Lake Erie Basin. Report submitted to The Nature Conservancy, Columbus, OH. 49 pp. + appendices. http://www.midwestbiodiversityinst.org/publications?type=L.

Rankin, E.T. 1995. Habitat indices in water resource quality assessments, *in* W.S. Davis and T.P. Simon (Eds.), Biological Assessment and Criteria: Tools for Water Resource Planning and Decision Making, Lewis Publishers, Boca Raton, FL, 181-208.

Rankin, E. T. 1989. The qualitative habitat evaluation index (QHEI), rationale, methods, and application, Ohio EPA, Division of Water Quality Planning and Assessment, Ecological Assessment Section, Columbus, Ohio.

Rankin, E.T. , C.O. Yoder and R.M. Mueller. 2011. Ecological Low Flow Protection Process for Ohio Streams and Rivers of the Lake Erie Basin, Prepared by the Midwest Biodiversity Institute for the The Nature Conservancy in Ohio, 6375 Riverside Drive, Dublin, OH 43017. http://www.midwestbiodiversityinst.org/publications?type=L

Sanders, R.S., R.J. Miltner, C.O. Yoder, and E.T. Rankin. 1999. The use of external deformities, erosions, lesions, and tumors (DELT anomalies) in fish assemblages for characterizing aquatic resources: a case study of seven Ohio streams, pp. 225-248. *in* T.P. Simon (ed.), Assessing the Sustainability and Biological Integrity of Water Resources Using Fish Communities. CRC Press, Boca Raton, FL.

Scott, W.B. and E.J. Crossman. 1973. Freshwater fishes of Canada. Fisheries Research Board of Canada Bulletin 184. 966 pp.

Snook, H., S.P. Davies, J.T. Gerritsen, B.K. Jessup, R. Langdon, D. Neils, and E. Pizzuto. 2007. The New England Wadeable Stream Survey (NEWS): Development of Common Assessments in the Framework of the Biological Condition Gradient. Prepared for U.S. EPA Office of Water by U.S. EPA, New England Regional Lab, Chelmsford, MA. http://water.epa.gov/type/rsl/monitoring/streamsurvey/index.cfm.

Smith, C.L. 1985. The inland fishes of New York State. New York Dept. Environmental Conservation.

Stoddard, J.L., D.P. Larsen, C.P. Hawkins, R.K. Johnson, and R.H. Norris. 2006. Setting expectations for the ecological condition of streams: the concept of reference condition. Ecological Applications, 16(4): 1267–1276.

Tarquinio, E. 2011. Maryland Stream, River and Lake Sampling as Part of the National Aquatic Resource Surveys: Current Status and Future. Third Maryland Streams Symposium and Mid-Atlantic Volunteer Monitoring Conference, August 10-13, 2011; Carroll Community College, MD.

Tewes, R., E. Emery, J. Thomas, L.E. Hersha, E.T. Rankin, C.O. Yoder (editor). 2007. Evaluation and Development of Large River Biological Assessment Methods: Electrofishing Methods and Standardized Protocols for Region V. U.S. EPA Grant CP-96510501. Ohio River Valley Water Sanitation Commission, Cincinnati, OH. 110 pp.

U.S. EPA. 2013. Biological Assessment Program Review: Assessing Level of Technical Rigor to Support Water Quality Management. EPA 820-R-13-001. Office of Water, Washington, D.C. 144 pp.

U.S. EPA. 2011. National Aquatic Resource Surveys: An Update, January 2011. NARS Fact Sheet. U.S. Environmental Protection Agency, Office of Water, Washington, D.C.

U.S. EPA. 2009. National Rivers and Streams Assessment: Field Operations Manual. EPA-841-B-07-009. U.S. Environmental Protection Agency, Washington, DC. http://water.epa.gov/type/rsl/monitoring/riverssurvey/index.cfm

U.S. EPA. 2005. Use of biological information to better define designated aquatic life uses in state and tribal water quality standards. Office of Water, Washington, DC. EPA 822-R-05-001. 188 pp. http://www.midwestbiodiversityinst.org/publications?type=L

U.S. EPA. 1995. Biological criteria: technical guidance for streams and small rivers. EPA 822-B-94-001, U. S. EPA, Office of Water (WH 4304), Washington, D. C. 20460. 162 pp.

U.S. EPA. 1990. Biological Criteria, national program guidance for surface waters. U. S. EPA, Office of Water Regulations and Standards, Washington, D. C. EPA-440/5-90-004.

USGS. 2004. Chemistry of Stream Sediments and Surface Waters in New England. By Gilpin R. Robinson, Jr., Katherine E. Kapo, and Jeffrey N. Grossman U.S. Geological Survey, Open-File Report 2004-1026, Version 1.0, 2004, Online only. Chemistry of Stream Sediments and Surface Waters in New England

USGS. 2007. NHDPlus User's Guide. July 1, 2007. United States Geological Service. Washington DC.

Vander Zanden, M. J., J.D. Olen, J.H. Thorne, and N. E. Mandrak. 2004. Predicting occurrences and impacts of smallmouth bass introductions in north temperate lakes. Ecol. Applic. 14(1): 132-148.

Wang, L. , T. Brenden, P. Seelbach, A. Cooper, D. Allan, R. Clark Jr. and M. Wiley. 2008. Landscape based identification of human disturbance gradients and reference conditions for Michigan streams. Environmental Monitoring and Assessment 141: 1–17.

Warner, K. 2005. Smallmouth bass introductions in Maine: history and management implications. Fisheries 30(11): 20-26.

Whittier, T.R., D.B. Halliwell, and R.A. Daniels. 2001. Distribution of lake fishes in the northeast – IV. benthic and small water column species. Northeastern Naturalist 8(4): 455-482.

Whittier, T.R., D.B. Halliwell, and R.A. Daniels. 2000. Distribution of lake fishes in the northeast – II. the Cyprinidae (minnows). Northeastern Naturalist 7(2): 131-156.

Whittier, T. R., & Hughes, R. M. (1998). Evaluation of fish species tolerances to environmental stressors in lakes in the northeastern United States. *North American Journal of Fisheries Management*, *18*(2), 236-252.

Whittier, T.R., D.B. Halliwell, and S.G. Paulsen. 1997. Cyprinid distributions in Northeast U.S.A. lakes: evidence of regional-scale minnow biodiversity losses. Can. J. Fish. Aquat. Sci. 54: 1593–1607.

Whitworth, W.R. 1996. Freshwater fishes of Connecticut. 2nd Edition. Connecticut Geo. Nat. Hist. Surv. Bull. 114: 234 pp.

Yoder, C.O., R.F. Thoma, L.E. Hersha, E.T. Rankin, B.H. Kulik, and B.R. Apell. 2008. Maine Rivers Fish Assemblage Assessment: Development of an Index of Biotic Integrity for Maine

Rivers. MBI Technical Report 2008-11-2. Report to U.S. EPA, Region I, Boston, MA. 69 pp. http://www.midwestbiodiversityinst.org/publications?type=L

Yoder, C.O., B.H. Kulik, B.J. Apell, and J.M. Audet. 2006a. 2005 Maine Rivers Fish Assemblage Assessment: I. Northern Maine Rivers Results; II. Maine Rivers Fish Species Distribution Atlas; III. Toward the Development of a Fish Assemblage Index for Maine Rivers. MBI Technical Report 12-06-1. Report to U.S. EPA, Region I, Boston, MA. 71 pp. + appendices. http://www.midwestbiodiversityinst.org/publications?type=L

Yoder, C.O., B.H. Kulik, and J.M. Audet. 2006b. The spatial and relative abundance characteristics of the fish assemblages in three Maine Rivers. MBI Technical Report MBI/12-05-1. Grant X-98128601 report to U.S. EPA, Region I, Boston, MA.. 136 pp. + appendices. http://www.midwestbiodiversityinst.org/publications?type=L

Yoder, C.O. and 9 others. 2005. Changes in fish assemblage status in Ohio's non-wadeable rivers and streams over two decades, pp. 399-429. *in* R. Hughes and J. Rinne (eds.). Historical changes in fish assemblages of large rivers in the America's. American Fisheries Society Symposium 45: 399-429.

Yoder, C. O. and B. H. Kulik. 2003. The development and application of multimetric biological assessment tools for the assessment of impacts to aquatic assemblages in large, non-wadeable rivers: a review of current science and applications. Canadian Journal of Water Resources 28 (2): 1 - 28.

Yoder, C.O. and M.A Smith. 1999. Using fish assemblages in a state biological assessment and criteria program: essential concepts and considerations, pp. 17-56. *in* T.P. Simon (ed.), Assessing the Sustainability and Biological Integrity of Water Resources Using Fish Communities. CRC Press, Boca Raton, FL.