APPENDIX 4:

STREAM FISH C0MMUNITY ASSESSMENT STANDARD OPERATING PROCEDURES

STANDARD OPERATING PROCEDURE
BIOLOGICAL MONITORING

STREAM FISH COMMUNITY ASSESSMENT PROGRAM

NORTH CAROLINA
DEPARTMENT OF ENVIRONMENT
AND NATURAL RESOURCES
Division of Water Quality
Environmental Sciences Section
Biological Assessment Unit

August 1, 2006

Standard Operating Procedures
Stream Fish Community Assessment Program

Environmental Sciences Section
Biological Assessment Unit

NORTH CAROLINA DEPARTMENT OF ENVIRONMENT
AND NATURAL RESOURCES
Division of Water Quality

This report has been approved for release

(original signed by Jimmie R. Overton)
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Date: August 01, 2006

INTRODUCTION

It is the purpose of this manual to provide details on standard operating procedures of the Biological Assessment Unit of the Division of Water Quality (DWQ or Division) for the collection and analysis of stream fish community assessment data. Consistency in data collection and analysis is the cornerstone for evaluating biological integrity. The procedures provided are a synthesis of widely used methods and methods developed from the experience of personnel within the Unit. These methods have been shown to provide repeatable and useful data for water quality evaluation.

This document will be reviewed regularly and revised as necessary. The prior approved version (Version 3) was dated March 14, 2001 and contained both fish and fish tissue procedures. In January 2006, the decision was made to produce two separate documents because revision needs for the two programs were different. All current employees and new employees within the Unit will be provided with this document to serve as a guideline of the Unit's activities, methods, and procedures. Revisions to this document will be provided to each employee and it will be the responsibility of the Environmental Biologist III to insure that the procedures are current.

The standard operating procedures (SOP) and quality control procedures (QC) in this manual will be the basis for all stream fish community assessment monitoring and the subsequent data provided in memoranda and reports prepared by the Biological Assessment Unit. Deviations from these procedures for unusual sampling situations shall be documented in the appropriate report or memorandum.

SAFETY PROGRAM

The Biological Assessment Unit is required to sample throughout North Carolina at times and places where medical facilities may not be readily available. It is imperative that all employees are instructed in and follow safety precautions when using sampling equipment and hazardous materials. The Environmental Sciences Section has a Safety Committee which is responsible for maintenance and development of current safety procedures. The Committee also maintains the safety standard operating procedures document which all personnel should be familiar. All personnel involved in electrofishing activities should be trained in First Aid and CPR and should be familiar with standard electrofishing safety procedures.

Sampling conditions are the primary safety factor to be considered for field work. If any field conditions such as high flows or thunderstorms raise the question of whether a sample can be safely collected, then decisions should always be made with the safety of personnel of prime concern. This same concern for safety of staff must be of primary importance when scheduling the amount of time to be spent in the field. Long days combined with strenuous effort increase the probability of accidents occurring. "**Safety first**" must always be the rule.

Employees should promptly report on-the-job accidents to the Unit Supervisor. If an accident occurs during field operations, the first responsibility of the team leader is to get first aid treatment for the injured employee; their second responsibility is to promptly notify the Unit Supervisor. The Safety Committee maintains a written record of accidents.

STUDY PLANS

All investigations conducted by the Biological Assessment Unit will follow a written study plan including but not limited to the:

- **Introduction** - Identify the nature and history of the area being investigated and the person or agency requesting the study.
- **Objectives** - The purpose of the investigation.
- **Sampling Location Selection** – Location of the sampling points is of extreme importance in the initiation of stream fish community assessment monitoring. The variables in watersheds are many and should be considered in as much detail as possible before sites are selected to monitor any body of water. Land use (i.e., urban, rural, forested, agricultural, and industrial) should be considered when locating sample sites, because man-made activities significantly affect the amount of sedimentation, nutrients, and organic or inorganic compounds entering a given

segment of a river or stream. The location of permitted dischargers should be reviewed, using the database provided by the Division's Basinwide Information Management System. Discussion of the proposed study with regional office personnel can also provide additional information useful for determining sampling locations. Pre-study planning of this nature will enhance data analyses and interpretation after the collections have been made.

- **Methods** - Sampling techniques should be listed with reference to those described in this manual. Any deviation from these standard methods must be noted and described.
- **Analytical Requirements** - All physico-chemical variables to be collected and analyses that will be required should be noted.
- **Logistics** - Shall include estimates of manpower requirements, equipment needed, time requirements, methods of sample transport to laboratories, *etc*. The study plan must be submitted and approved by the Unit Supervisor prior to conducting the investigation.

A study is complete when a report or memorandum is sent to and approved by the appropriate level of management within the Division (typically the Environmental Sciences Section Chief). Each memorandum should contain these sections: an Introduction or Background, Sampling Sites, Methods, Results and Discussion, and Summary or Recommendations. Any figures, maps, and photographs needed to allow a reader to easily locate the sampling sites should also be included. When the report or memorandum is approved, a Biological Assessment Unit file number is assigned. Finally, the report or memorandum is filed in a Projects File that is organized by basin and subbasin.

STREAM FISH COMMUNITY ASSESSMENT PROGRAM

OBJECTIVES

North Carolina consists of 17 major river basins (Figure 1). Each of these basins is assessed every five years to support the Planning Section's Basinwide Water Quality Management Plans. The Division utilizes several water quality programs and tools to assess the quality of the state's waters. One of the more recently developed (and still developing) programs is the Stream Fish Community Assessment Program. The primary objective of this program is to provide fish community ratings for wadeable streams to the Basinwide Planning Unit for use support determinations and for the Section's Basinwide Water Quality Management Plans.

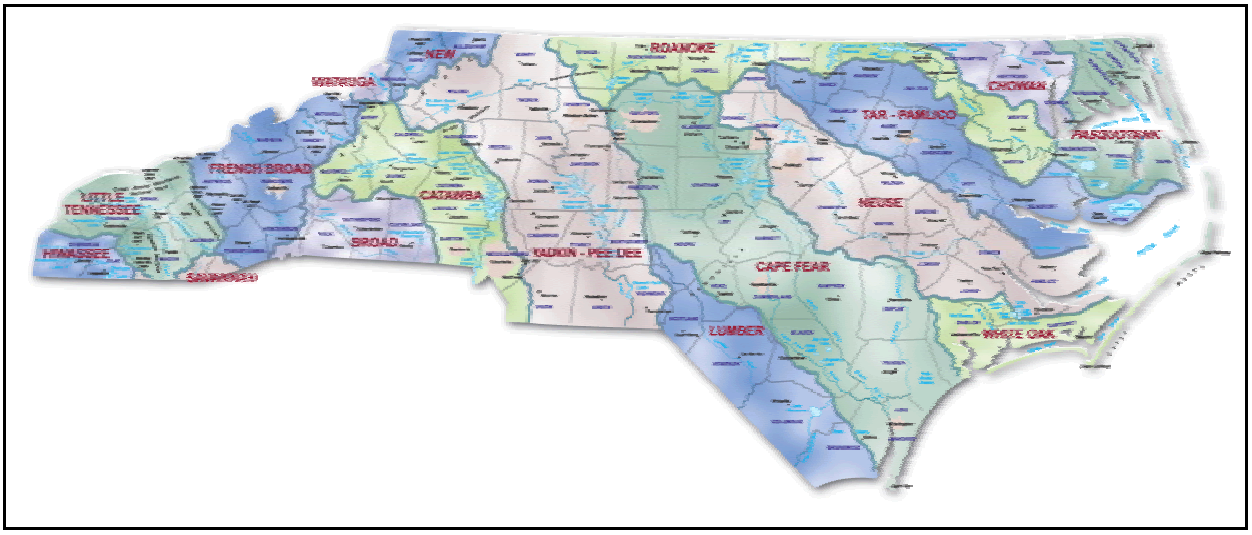


Figure 1. Major river basins of North Carolina.

Secondary objectives of the Program are to provide data suitable for supporting these DWQ activities:

- Planning Section
  - ❖ Biennial 303(d) and 305(b) reporting to EPA, including identification of areas of

impairment or degradation,
    - TMDL development,
    - Prioritization of restoration activities, and
    - Background information for Use Attainability studies such as trout survival and propagation waters, High Quality Waters, and Outstanding Resource Waters.
- Surface Water Protection Section
    - Identification of background levels of constituents for determination of NPDES permit limits, and
    - Identification of dischargers causing unacceptable impacts.
- Regional Offices
    - Background information to assist with water quality management activities in each region.

The Stream Fish Community Assessment Program was designed as an additional basinwide assessment tool and has been in existence since 1991. It's core mission is to sample a set of fixed sites on lower Strahler order wadeable creeks, streams, and rivers on a five-year rotating basis to support the DWQ's Basinwide Management Plan Program. To date, more than 1,000 samples from 700 sites have been assessed (Figure 2), primarily in the Piedmont and Mountains. Most of the stations are located at bridge crossings or other public accesses and are accessible by land. Nonwadeable and higher Strahler order rivers, estuaries, and reservoirs are not monitored. The program compliments other DWQ programs such as the Benthic Macroinvertebrate and Ambient Monitoring System programs which tend to focus monitoring efforts on larger waterbodies and watersheds.

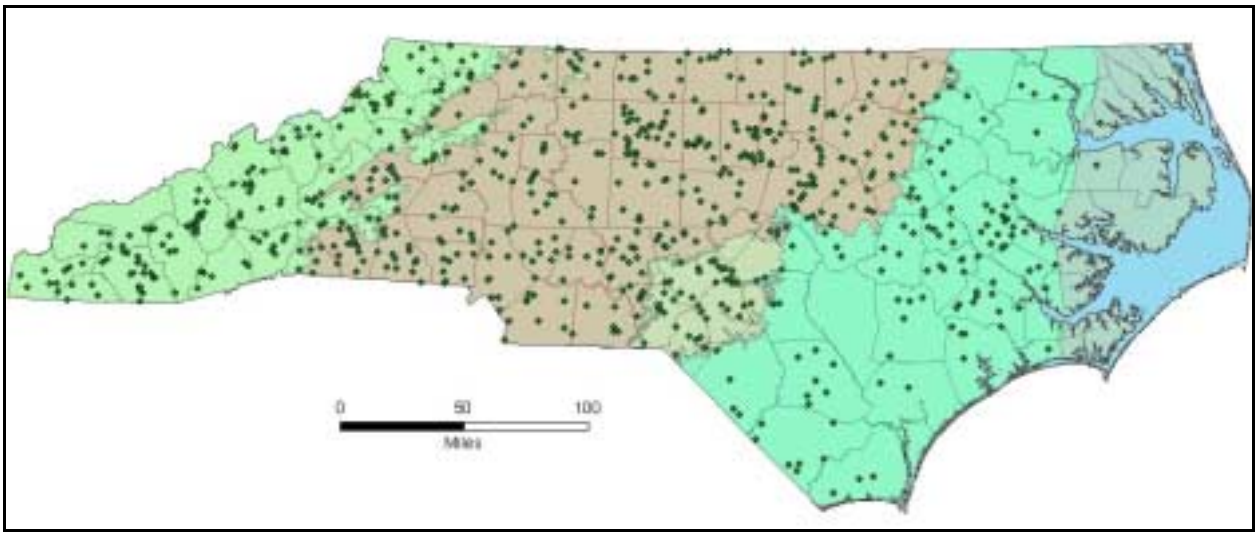


Figure 2. Stream fish community sampling sites, 1991 – 2005. Colored regions indicate Level III ecoregions and green dots indicate fish community sampling sites.

THE NORTH CAROLINA INDEX OF BIOTIC INTERGRITY

The Division has been monitoring the biological integrity of stream fish communities since the early 1990s. The biological monitoring tool that is used is referred to as the North Carolina Index of Biological Integrity (NCIBI). The NCIBI method was developed for assessing a stream's biological integrity by examining the structure and health of its fish community. The North Carolina Administrative Code defines *Biological Integrity* as: " . . . *the ability of an aquatic ecosystem to support and maintain a balanced and indigenous community of organisms having species composition, diversity, population densities, and functional organization similar to that of reference conditions*" (15A NCAC 02B .0200; NCAC 2004). The NCIBI is a modification of the Index of Biotic Integrity (IBI) initially proposed by Karr (1981) and Karr, *et al*. (1986).

The NCIBI incorporates information about species richness and composition, trophic composition, fish abundance, and fish condition. The NCIBI summarizes the effects of all classes of factors influencing

aquatic faunal communities such as water quality, energy source, habitat quality, flow regime, and biotic interactions. While any change in a fish community can be caused by many factors, certain aspects of the community are generally more responsive to specific influences. Species composition measurements reflect habitat quality effects. Information on trophic composition reflects the effect of biotic interactions and energy supply. Fish abundance and condition information indicates additional water quality effects. It should be noted, however, that these responses may overlap. For example, a change in fish abundance may be due to decreased energy supply or a decline in habitat quality, not necessarily a change in water quality.

The scores derived from this index are a measure of the ecological health of the waterbody and may not directly correlate to water quality. For example, a stream with excellent water quality, but with poor or fair fish habitat, may not be rated excellent with this index. However, a stream which rated excellent on the NCIBI should be expected to have excellent water quality.

APPLICATION OF THE NCIBI

The NCIBI is continually being refined for greater applicability to wadeable streams in North Carolina. Currently, the NCIBI is applicable **only** to **streams** that are **wadeable** from one shoreline across to the other and for a distance of 600 feet. The NICIBI is only applicable to wadeable streams in the Western and Northern Mountains (French Broad, Hiwassee, Little Tennessee, New, and Watauga River basins), the Inner Piedmont, Foothills, and Eastern Mountains (Broad, Catawba, Savannah, and Yadkin (exclusive of the Sand Hills) River basins); and the Outer Piedmont (Cape Fear, Neuse, Roanoke, and Tar River basins).

The delineations of the Mountains, Piedmont, and Sand Hills in these river basins are based upon a North Carolina State University Co-operative Extension Service map (*North Carolina Watersheds* by J. Fels published in 1997) (Figure 3) and Griffith, *et al*. (2002). More specifically, the Outer Piedmont includes:

- Cape Fear River Basin -- except for the streams draining the Sand Hills in Moore, Lee, and Harnett counties, the entire basin upstream of Lillington, NC;
- Neuse River Basin -- the entire basin above Smithfield and Wilson, NC, except for the south and southwest portions of Johnston County and the eastern two-thirds of Wilson County;
- Roanoke River Basin -- the entire basin in North Carolina upstream of Roanoke Rapids, NC and a small area between Roanoke Rapids and Halifax, NC; and
- Tar River Basin -- the entire basin above Rocky Mount, NC, except for the lower southeastern one-half of Halifax County and the extreme eastern portion of Nash County.

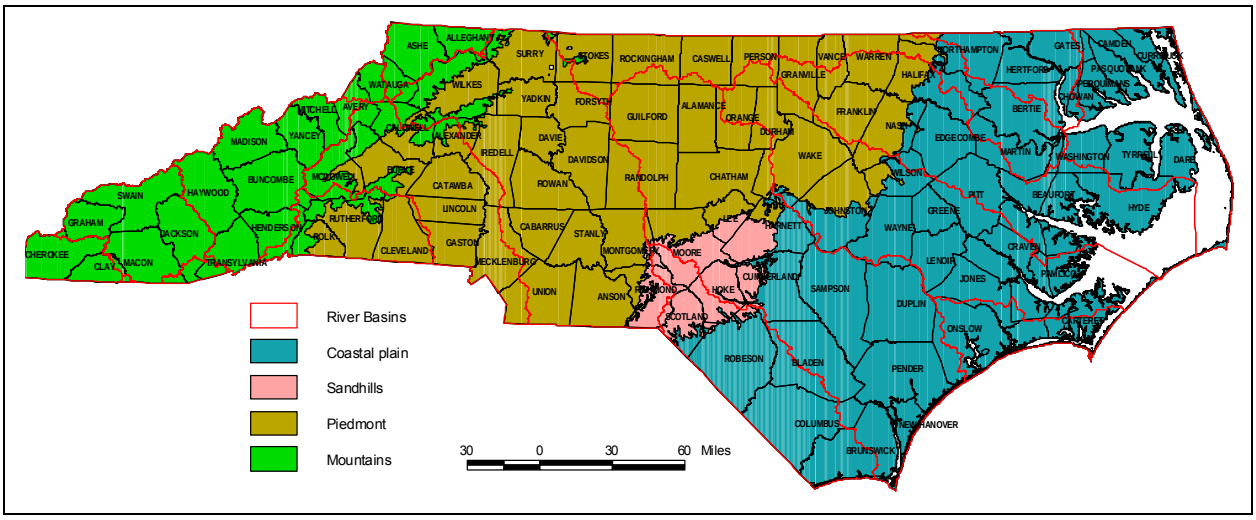


Figure 3. Physiographic regions and river basins in North Carolina.

The Index is undergoing revisions for the Upper Coastal Plain (Chowan, Neuse, Pasquotank, Roanoke, Tar, and White Oak River basins), the Lower Coastal Plain (Cape Fear and Lumber River basins), and the Sand Hills (Cape Fear, Lumber, and Yadkin River basins).

NCIBI QUALIFIERS

The North Carolina Index of Biological Integrity is only applicable if the methods of collection and data analyses described herein are strictly followed. The Index has not been tested using other collection techniques. Nonwadeable streams and larger rivers that must be sampled with a boat are not currently evaluated with the NCIBI. Neither are high elevation, cold water trout streams. Southern Appalachian trout streams are typically high gradient streams with plunge pools, *Rhododendron*- and Eastern hemlock-lined within a forested watershed, have cold water with low specific conductance, have a naturally low fish species diversity (usually brook trout, rainbow trout, or brown trout, blacknose dace, and mottled sculpin), have few tolerant fish, and support a reproducing population of one or more species of trout. Finally, young-of-year fish are excluded from all NCIBI calculations.

NCIBI ANALYSIS

The NCIBI incorporates information about species richness and composition, pollution indicator species, trophic composition, fish abundance, fish condition, and reproductive function by the cumulative assessment of 12 parameters or metrics (Tables 1 - 3). Each metric is designed to contribute unique information to the overall assessment. The values provided by the metrics are converted into scores on a 1, 3, and 5 scale. A score of 5 represents conditions commonly associated with undisturbed reference streams in the specific river basin or ecoregion. A score of 1, however, indicates that conditions deviate greatly from those typically observed in undisturbed streams of the region. All metrics for each of the three regions were calibrated using regional reference sites.

The scores for all metrics are then summed to obtain the overall NCIBI score, an even number between 12 and 60. The score is then used to determine the biological integrity class of the stream (i.e., Poor, Fair, Good-Fair, Good, or Excellent) (Karr 1981 , Karr, *et al.* 1986). A fish community rated Excellent is comparable to the best situations with minimal human disturbance; all regionally expected species for the habitat and stream size, including the most intolerant forms, are present along with a full array of size classes and a balanced trophic structure. Conversely, a fish community rated Poor deviates greatly from the reference condition. The number of fish is fewer than expected, usually fewer than expected number of species, an absence of intolerant species, and an altered trophic structure. Communities rated Good, Good-Fair, or Fair fall within this disturbance gradient.

Currently, if a fish community is rated Excellent, Good, or Good-Fair it is deemed to be Fully Supporting its Aquatic Life Use Support stream classification. If a fish community is rated Fair or Poor it is deemed to be Not Supporting its Life Use Support stream classification and the water quality standard is not being met. Waters that have an Excellent fish community rating are also eligible for reclassification to a Outstanding Resource Waters or to a High Quality Waters supplemental classifications.

NCIBI METRICS

These 12 metrics (Tables 1 – 3) are grouped into five categories with each metric designed to contribute unique information to the overall assessment:

1. Species richness and composition (Metric Nos. 1 and 3 - 5)
2. Indicator species (Metric Nos. 6 and 7)
3. Trophic function (Metric Nos. 8 - 10)
4. Abundance and condition (Metric Nos. 2 and 11)
5. Reproductive function (Metric No. 12)

Eight of the metrics involve species composition, pollution tolerance, and trophic composition. Table 4 lists, phylogenetically, the pollution tolerance ratings and trophic guild assignments of the freshwater fish found throughout North Carolina. Several of the species (for example, Paddlefish, American Shad, and Sauger) will not be encountered in streams that are sampled adhering to these procedures. Estuarine

species, extirpated species, and species found in nearby drainages of bordering states (but not in North Carolina) are not included. Revisions and updates to this table will be published periodically.

SPECIES RICHNESS AND COMPOSITION (Metric Nos. 1 and 3 - 5)
Distributional data for these four metrics were obtained from Menhinick (1991), Lee, *et al*. (1980), Biological Assessment Unit studies, North Carolina State Museum of Natural Sciences, Tennessee Valley Authority, and many other sources.

- **Metric No. 1. Number of Species**

The total number of species supported by a stream of a given size in a given region decreases with environmental degradation. In addition, some streams with larger watersheds or drainage areas can be expected to support more species than streams with smaller watersheds. In other instances, the number of species and the watershed size are not correlated. This metric is rated according to the river basin from which the sample was taken and, in the case of the Inner Piedmont, Foothills, and Eastern Mountains region, the drainage area size at the sampling point. Drainage area size is calculated from USGS 7.5 minute series topographic maps or from the Division's geographic information system, if not otherwise known (ambient database, USGS publications, or a USGS masterfile printout which gives drainage areas for many streams at given road crossings). This metric is a count of all the species in the sample.

- **Metric No. 3. Number of Species of Darters**
  Darters are sensitive to environmental degradation particularly as a result of their specific reproductive and habitat requirements (Page 1983, Kuehne and Barbour 1983). Darter habitats are degraded as a result of channelization, siltation, and reduced oxygen levels. The collection of fewer than the expected number of species of darters can indicate that some degree of habitat degradation is occurring. This metric is a count of all the species of *Etheostoma* and *Percina* in the sample (Table 4).

  As with Metric No. 1, the total number of species of darters supported by a stream of a given size in a given region decreases with environmental degradation. In addition, some streams with larger watersheds or drainage areas can be expected to support more species than streams with smaller watersheds. In other instances, the number of species and the watershed size are not correlated. This metric is rated according to the river basin from which the sample was taken and, in the case of the Inner Piedmont, Foothills, and Eastern Mountains region, the drainage area size at the sampling point.

- **Metric No. 4. Number of Species of Rockbass, Smallmouth Bass, and Trout (Western and Northern Mountains)**
  Rock Bass, Smallmouth Bass, and the three species of trout are particularly responsive to habitat degradation such as the filling in of pools with sediment and the loss of instream cover. This metric is a count of these five species in the sample. Stocked trout (characterized by pale colors and worn or deformed fins) are not counted.

- **Metric No. 4 Number of Species of Sunfish, Bass, and Trout (Inner Piedmont, Foothills, and Eastern Mountains)**
  Sunfish, black bass, and trout species are particularly responsive to habitat degradation such as the filling in of pools with sediment and the loss of instream cover. This metric includes *Lepomis* (all species), *Centrarchus macropterus*, *Ambloplites rupestris*, *Micropterus* (all species), and all three species of trout (Table 4). Stocked trout (characterized by pale colors and worn or deformed fins) are not counted.

- **Metric No. 4 Number of Species of Sunfish (Outer Piedmont)**
  Sunfish species are particularly responsive to habitat degradation such as the filling in of pools with sediment and the loss of instream cover. This metric includes *Lepomis* (all species), *Enneacanthus* (all species), *Centrarchus macropterus*, *Acantharchus pomotis*, and *Ambloplites cavifrons* (Table 4).

- **Metric No. 5 Number of Species of Cyprinids (Western and Northern Mountains)**

Many species of minnows are intolerant of habitat and chemical degradation and, because some of the species may have life spans up to six years, provide a multiyear integrated perspective. They also reflect the condition of the benthic community which may be harmed by sedimentation or by sediment contamination. In the Western and Northern Mountains, the Number of Species of Cyprinds (Minnows) is used as a substitute metric for the Number of Species of Suckers. This metric is a count of all the species within the family Cyprinidae in the sample (Table 4).

- **Metric No. 5. Number of Species of Suckers (Inner Piedmont, Foothills, and Eastern Mountains and Outer Piedmont)**

Many species of suckers are intolerant of habitat and chemical degradation and, because they are long lived, provide a multiyear integrated perspective. They also reflect the condition of the benthic community which may be harmed by sedimentation or by sediment contamination. This metric is a count of all the species within the family Catostomidae in the sample (Table 4).

INDICATOR SPECIES (Metric Nos. 6 and 7)

The tolerance ratings for these two metrics were derived from Karr, *et al*. (1986), Saylor and Scott (1987), from polling various university, federal, and state fisheries management personnel using the Delphi Technique (Zuboy 1981), Etnier and Starnes (1993), Jenkins and Burkhead (1993), Rohde, *et al*. (1994), and from Biological Assessment Unit data.

- **Metric No. 6 Number of Intolerant Species**

Intolerant species are those which are most affected by environmental perturbations and therefore should disappear, at least as viable populations, by the time a stream is rated as "Fair". Intolerant species also includes some species that have a very restricted zoogeographic distribution or are considered rare, endangered, or threatened. Of the approximately 219 species of freshwater fish found in North Carolina, 54 species are considered intolerant. This metric is a count of all intolerant species in the sample (Tables 4 and 5).

- **Metric No. 7 Percentage of Tolerant Individuals**

Tolerant species are those which are often present in a stream in low or moderate numbers but as the stream degrades, they can become dominant. Of the approximately 219 species of freshwater fish found in North Carolina, 21 species (and one hybrid) are considered tolerant. This metric is a percentage metric. The number of individuals of the tolerant species (Tables 4 and 5) is summed and divided by the total number of fish collected to obtain the percentage of tolerant fish in the sample.

TROPHIC FUNCTION (Metric Nos. 8 - 10)

These three trophic composition metrics are used to measure the divergence from expected production and consumption patterns in the fish community that can result from environmental degradation. The main cause for a shift in the trophic composition of the fish community, generally a greater proportion of omnivores and lesser proportion of insectivores than what is expected, is nutrient enrichment. However, in some instances, the percentage of insectivores, especially Redbreast Sunfish *Lepomis auritus*, may increase dramatically due to environmental degradation and nutrient enrichment. And where the herbivorous Central Stoneroller *Campostoma anomalum* is found, canopy removal, riparian alteration, and nutrient enrichment may lead to its dramatic increase.

The trophic guild data for these three metrics were derived from the literature (Lee, *et al*. (1980), Karr, *et al*. (1986), Plafkin, *et al*. (1989), Etnier and Starnes (1993), Jenkins and Burkhead (1993), Rohde, *et al*. (1994)), and from Biological Assessment Unit data.

- **Metric No. 8 Percentage of Omnivorous + Herbivorous Individuals**

  This metric is a percentage metric. The number of individuals of omnivores and herbivores (Table 4) is summed and divided by the total number of fish collected.

- **Metric No. 9 Percentage of Insectivores**

The number of individuals of insectivores (Table 4) is summed and divided by the total number of fish collected.

- **Metric No. 10 Percentage of Piscivores**
  The number of individuals of piscivores (Table 4) is summed and divided by the total number of fish collected. This metric was not used in the Western and Northern Mountains region because the metric failed to discriminate between the impaired and the reference sites and was not significantly correlated with the total NCIBI score. No substitute or alternative metrics were found suitable.

ABUNDANCE AND CONDITION (Metric Nos. 2 and 11)

- **Metric No. 2 Number of Fish**
  The total number of fish supported by a stream of a given size in a given region decreases with environmental degradation. However, in some instances, nutrient enrichment or environmental degradation may actually increase the number of fish supported by the stream. This metric is a count of all the fish in the sample.

- **Metric No. 11 The Percentage of Diseased Fish**
  This metric occurs infrequently, and in most instances, is absent entirely. The metric does occur below point sources and in areas where toxic chemicals are concentrated (e.g., Sanders, *et al.* 1999). This metric is: "*an excellent measure of the aesthetic value of game and nongame fish*" (Barbour, *et al.* 1999).

  DELT (Disease, fin Erosion, Lesions, and Tumors) may not be observed in streams the size of which are typically sampled because the worst (urban and industrial) streams are often not sampled. Neither are the larger streams and rivers where NPDES dischargers are typically located and which may have a greater DELT rate than the smaller streams. Generally, North Carolina fish are healthy.

  To rate this metric, the number of fish in the sample which have sores, lesions, skeletal anomalies (as evident externally), or diseased, damaged, or rotten fins is summed and divided by total number of fish collected to obtain the percentage of diseased fish. Fin or other external damage as a result of spawning should not be counted. Fish are considered to be in spawning condition when tubercles or breeding colors are evident.

  This metric was not used in the Western and Northern Mountains region because the metric failed to discriminate between the impaired and the reference sites and was not significantly correlated with the total NCIBI score. No substitute or alternative metrics were found suitable.

  Blackspot and Other Diseases
  Blackspot and yellow grub diseases are naturally occurring, common infections of fish by an immature stage of flukes. The life cycle involves fish, snails, and piscivorous birds. Although heavy, acute infections can be fatal, especially to small fish, fish can carry amazingly high worm burdens without any apparent ill effects (Noga 1996). Although some researchers incorporate the incidence of black spot and yellow grub into indices of biotic integrity (e.g., Steedman 1991), others, because of a lack of a consistent, inverse relationship to environmental quality, do not (e.g., Sanders, *et al*. 1999). The diseases are not considered in the NCIBI because it is widespread, affecting fish in all types of streams.

REPRODUCTIVE FUNCTION (Metric No. 12)

- **Metric No. 12 Percentage of Species with Multiple Age Groups**
  This metric was developed by the Division in 1989 as an indicator of the suitability of the habitat for reproduction. Other researchers have used proportion of individuals as hybrids, proportion of individuals as introduced species, simple lithophils (species of fish that spawn where the egg can develop in the interstices of sand, gravel, and cobble substrates without parental care), and

number of simple lithophils (Barbour, *et al*. 1999). This metric is strongly influenced by rare species (species represented by 1 or 2 fish) that are not reproducing in the stream. A community may be diverse but if a large proportion of the species are represented by only 1 or 2 fish per species, these rarer species may depress the metric value.

For each species, the total length distribution data are used to determine the presence of different age groups and, thus, the degree of reproductive success. This metric is calculated by first counting the total number of species present in the sample. Then, the total lengths of all the fish of each species are examined to determine whether or not all the fish of that species are of one or multiple age groups. Finally, the percentage of species with multiple age groups is determined by dividing the number of species with multiple age groups by the total number of species collected in the sample. Although some species are rare and some species have fewer age groups than others, <u>at least three individuals per species must have been collected</u> to determine the presence of multiple age groups within the population. In some instances, professional judgment may also be used to determine the reproductive success of a particular species.

Publications such as Carlander (1969 and 1977), Kuehne and Barbour (1983), Page (1983), Manooch (1984), Etnier and Starnes (1993), Jenkins and Burkhead (1993), and Rohde, *et al*. (1994) may also be consulted to determine length-age class relationships.

Table 1. Scoring criteria for the NCIBI for wadeable streams in the Western and Northern Mountains of the French Broad (including the Pigeon River), Hiwassee, Little Tennessee, New, and Watauga River basins with watersheds ranging between 3.1 and 161 mi^2.

No.	Metric		Score
1	**No. of species**		
	≥ 16 species		5
	12-15 species		3
	< 12 species		1
2	**No. of fish**		
	320-1,000 fish		5
	205-319 fish		3
	< 205 fish		1
	> 1,000 fish		3
3	**No. of species of darters**		
	French Broad & Little Tennessee River Basins	New River, Pigeon River, Watauga[1], & Hiwassee River Basins	
	≥ 4 species	≥ 3 species	5
	2 or 3 species	1 or 2 species	3
	0 or 1 species	0 species	1
4	**No. of species of Rock Bass, Smallmouth Bass, and trout**		
	≥ 2 species		5
	1 species		3
	0 species		1
5	**No. of species of cyprinids**		
	All basins, except Pigeon River Basin	Pigeon River Basin	
	≥ 8 species	≥ 6 species	5
	6 or 7 species	4 or 5 species	3
	≤ 5 species	≤ 3	1
6	**No. of intolerant species**		
	All basins, except New River Basin	New River Basin	
	≥ 3 species	≥ 5 species	5
	2 species	3 or 4 species	3
	0 or 1 species	0, 1, or 2 species	1
7	**Percentage of tolerant individuals**		
	≤ 2%		5
	2-10%		3
	> 10%		1
8	**Percentage of omnivorous + herbivorous individuals**		
	10-36%		5
	37-50%		3
	> 50%		1
	< 10%		1
9	**Percentage of insectivorous individuals**		
	55-85%		5
	40-54%		3
	< 40%		1
	> 85%		1
12	**Percentage of species with multiple age groups**		
	≥ 65% of all species have multiple age groups		5
	45-64% all species have multiple age groups		3
	< 45% all species have multiple age groups		1

[1]Tentative for the Watauga River basin; also includes *Cottus bairdi* (Mottled Sculpin) and *Noturus insignis* (Margined Madtom). The Watauga River Basin and the Toxaway River (Savannah River Basin) are the only river basins in North Carolina where these three benthic, insectivorous groups (darters, Mottled Sculpin, and Margined Madtom) are sympatric.

Table 2. Scoring criteria for the NCIBI for wadeable streams in the Inner Piedmont, Foothills, and Eastern Mountains of the Broad, Catawba, Savannah, and Yadkin River basins with watershed drainage areas ranging between 2.8 and 245 mi^2.

No.	Metric		Score
1	**No. of species**		
	where Y is the number of species in the sample and X is the stream's drainage area in mi^2:		
	$Y \geq 9.5*Log_{10}X+1.6$		5
	$4.8*Log_{10}X+0.8 \leq Y < 9.5*Log_{10}X+1.6$		3
	$Y < 4.8*Log_{10}X+0.8$		1
2	**No. of fish**		
	Mountains	Piedmont	
	≥ 300 fish	≥ 150 fish	5
	200-299 fish	100-149 fish	3
	< 200 fish	< 100 fish	1
3	**No. of species of darters**		
	where Y is the number of species of darters in the sample and X is the stream's drainage area in mi^2.		
	$Y \geq 1.6*Log_{10}X$		5
	$0.8*Log_{10}X \leq Y < 1.6*Log_{10}X$		3
	$Y < 0.8*Log_{10}X$		1
	If the drainage area is > 70 mi^2, then ≥ 3 species = 5, 2 species = 3, and 0 or 1 species = 1		
4	**No. of species of sunfish, bass, and trout**		
	≥ 3 species		5
	2 species		3
	0 or 1 species		1
5	**No. of species of suckers**		
	≥ 2 species		5
	1 species		3
	0 species		1
6	**No. of intolerant species**		
	Mountains	Piedmont	
	≥ 3 species	≥ 1 species	5
	1or 2 species	(no middle criteria or score)	3
	0 species	0 species	1
7	**Percentage of tolerant individuals**		
	Mountains	Piedmont	
	≤ 12%	≤ 25%	5
	13-25%	26-35%	3
	> 25%	> 35%	1
8	**Percentage of omnivorous + herbivorous individuals**		
	10-35%		5
	36-50%		3
	> 50%		1
	< 10%		1
9	**Percentage of insectivorous individuals**		
	60-90%		5
	45-59%		3
	< 45%		1
	> 90%		1
10	**Percentage of piscivorous individuals**		
	≥ 1.0%		5
	0.25-1.0%		3
	< 0.24%		1
11	**Percentage of diseased fish (DELT = diseased, fin erosion, lesions, and tumors)**		
	< 0.75%		5
	0.76-1.25%		3
	> 1.25%		1
12	**Percentage of species with multiple age groups**		
	Mountains	Piedmont	
	≥ 65% of all species have multiple age groups	≥ 55% of all species have multiple age groups	5
	45-64% all species have multiple age groups	35-54% all species have multiple age groups	3
	< 45% all species have multiple age groups	< 35% all species have multiple age groups	1

Table 3. Scoring criteria for the NCIBI for wadeable streams in the Outer Piedmont of the Cape Fear, Neuse, Roanoke, and Tar River basins ranging between 3.1 and 328 mi^2.

No.	Metric		Score
1	**No. of species**		
	≥ 16 species		5
	10-15 species		3
	< 10 species		1
2	**No. of fish**		
	≥ 225 fish		5
	150-224 fish		3
	< 150 fish		1
3	**No. of species of darters**		
	Cape Fear	Neuse, Roanoke, and Tar	
	≥ 2 species	≥ 3 species	5
	1 species	1 or 2 species	3
	0 species	0 species	1
4	**No. of species of sunfish**		
	≥ 4 species		5
	3 species		3
	0, 1, or 2 species		1
5	**No. of species of suckers**		
	Cape Fear	Neuse, Roanoke, and Tar	
	≥ 2 species	≥ 3 species	5
	1 species	1 or 2 species	3
	0 species	0 species	1
6	**No. of intolerant species**		
	Cape Fear	Neuse, Roanoke, and Tar	
	≥ 1 species	≥ 3 species	5
	no middle score	1 or 2 species	3
	0 species	0 species	1
7	**Percentage of tolerant individuals**		
	≤ 35%		5
	36-50%		3
	> 50%		1
8	**Percentage of omnivorous and herbivorous individuals**		
	10-35%		5
	36-50%		3
	> 50%		1
	< 10%		1
9	**Percentage of insectivorous individuals**		
	65-90%		5
	45-64%		3
	< 45%		1
	> 90%		1
10	**Percentage of piscivorous individuals**		
	≥ 1.4-15%		5
	0.4-1.3%		3
	< 0.4%		1
	> 15%		1
11	**Percentage of diseased fish (DELT = diseased, fin erosion, lesions, and tumors)**		
	≤ 1.75%		5
	1.76-2.75%		3
	> 2.75%		1
12	**Percentage of species with multiple age groups**		
	≥ 50% of all species have multiple age groups		5
	35-49% all species have multiple age groups		3
	< 35% all species have multiple age groups		1

Table 4. North Carolina freshwater fishes tolerance ratings, adult trophic guild assignments, and young-of-year (YOY) cut-off lengths (total length in millimeters). Common and scientific names follow Nelson, *et al*. (2004), except for *Scartomyzon*.

Family/ Species	Common Name	Tolerance Rating	Trophic Guild of Adults	YOY (< TL mm)
Petromyzontidae	**Lampreys**			
Ichthyomyzon bdellium	Ohio Lamprey	Intermediate	Parasitic	50
I. castaneus	Chestnut Lamprey	Intermediate	Parasitic	
I. greeleyi	Mountain Brook Lamprey	Intermediate	Non-feeding	40
Lampetra aepyptera	Least Brook Lamprey	Intolerant	Non-feeding	50
L. appendix	American Brook Lamprey	Intermediate	Non-feeding	40
Petromyzon marinus	Sea Lamprey	Intermediate	Parasitic	100
Acipenseridae	**Sturgeons**			
Acipenser brevirostrum	Shortnose Sturgeon	Intermediate	Insectivore	200
A. oxyrinchus	Atlantic Sturgeon	Intermediate	Insectivore	200
Polyodontidae	**Paddlefishes**			
Polyodon spathula	Paddlefish	Intermediate	Planktivore	200
Lepisosteidae	**Gars**			
Lepisosteus osseus	Longnose Gar	Tolerant	Piscivore	200
Amiidae	**Bowfins**			
Amia calva	Bowfin	Tolerant	Piscivore	200
Hiodontidae	**Mooneyes**			
Hiodon tergisus	Mooneye	Intermediate	Insectivore	100
Anguillidae	**Freshwater Eels**			
Anguilla rostrata	American Eel	Intermediate	Piscivore	100
Clupeidae	**Herrings and Shads**			
Alosa aestivalis	Blueback Herring	Intermediate	Insectivore	100
A. mediocris	Hickory Shad	Intermediate	Insectivore	100
A. pseudoharengus	Alewife	Intermediate	Insectivore	50
A. sapidissima	American Shad	Intermediate	Insectivore	100
Dorosoma cepedianum	Gizzard Shad	Intermediate	Omnivore	100
D. petenense	Threadfin Shad	Intermediate	Omnivore	100
Cyprinidae	**Carps and Minnows**			
Campostoma anomalum	Stoneroller	Intermediate	Herbivore	60
Carassius auratus	Goldfish	Tolerant	Omnivore	50
Clinostomus funduloides	Rosyside Dace	Intermediate	Insectivore	40
Ctenopharyngodon idella	Grass Carp	Tolerant	Herbivore	200
Cyprinella analostana	Satinfin Shiner	Tolerant	Insectivore	40
C. chloristia	Greenfin Shiner	Intermediate	Insectivore	40
C. galactura	Whitetail Shiner	Intermediate	Insectivore	50
C. labrosa	Thicklip Chub	Intolerant	Insectivore	40
C. lutrensis	Red Shiner	Tolerant	Insectivore	30
C. nivea	Whitefin Shiner	Intermediate	Insectivore	40
C. pyrrhomelas	Fieryblack Shiner	Intolerant	Insectivore	40
C. spiloptera	Spotfin Shiner	Intermediate	Insectivore	40
C. zanema	Santee Chub	Intolerant	Insectivore	40
C. sp. cf. *zanema*	"Thinlip" Chub	Intolerant	Insectivore	40
Cyprinus carpio	Common Carp	Tolerant	Omnivore	150
Erimonax monachus	Spotfin Chub	Intolerant	Insectivore	40
Erimystax insignis	Blotched Chub	Intermediate	Omnivore	40
Exoglossum laurae	Tonguetied Minnow	Intolerant	Insectivore	50
E. maxillingua	Cutlip Minnow	Intolerant	Insectivore	50
Hybognathus regius	Eastern Silvery Minnow	Intermediate	Herbivore	50
Hybopsis amblops	Bigeye Chub	Intermediate	Insectivore	50
H. hypsinotus	Highback Chub	Intolerant	Insectivore	40
H. rubifrons	Rosyface Chub	Intolerant	Insectivore	50
Luxilus albeolus	White Shiner	Intermediate	Insectivore	50
L. cerasinus	Crescent Shiner	Intermediate	Insectivore	50
L. chrysocephalus	Striped Shiner	Intermediate	Omnivore	50
L. coccogenis	Warpaint Shiner	Intermediate	Insectivore	50

Table 4 (continued).

Family/ Species	Common Name	Tolerance Rating	Trophic Guild of Adults	YOY (< TL mm)
Lythrurus ardens	Rosefin Shiner	Intermediate	Insectivore	50
L. matutinus	Pinewoods Shiner	Intolerant	Insectivore	50
Nocomis leptocephalus	Bluehead Chub	Intermediate	Omnivore	50
N. micropogon	River Chub	Intermediate	Omnivore	50
N. platyrhynchus	Bigmouth Chub	Intermediate	Omnivore	50
N. raneyi	Bull Chub	Intermediate	Omnivore	50
Notemigonus crysoleucas	Golden Shiner	Tolerant	Omnivore	75
Notropis alborus	Whitemouth Shiner	Intermediate	Insectivore	40
N. altipinnis	Highfin Shiner	Intermediate	Insectivore	40
N. amoenus	Comely Shiner	Intermediate	Insectivore	50
N. bifrenatus	Bridle Shiner	Intermediate	Omnivore	40
N. chalybaeus	Ironcolor Shiner	Intolerant	Insectivore	40
N. chiliticus	Redlip Shiner	Intermediate	Insectivore	40
N. chlorocephalus	Greenhead Shiner	Intermediate	Insectivore	40
N. cummingsae	Dusky Shiner	Intermediate	Insectivore	40
N. hudsonius	Spottail Shiner	Intermediate	Omnivore	50
N. leuciodus	Tennessee Shiner	Intermediate	Insectivore	50
N. lutipinnis	Yellowfin Shiner	Intermediate	Insectivore	40
N. maculatus	Taillight Shiner	Intolerant	Insectivore	40
N. mekistocholas	Cape Fear Shiner	Intermediate	Omnivore	40
N. micropteryx	Highland Shiner	Intolerant	Insectivore	40
N. petersoni	Coastal Shiner	Intermediate	Insectivore	40
N. photogenis	Silver Shiner	Intolerant	Insectivore	50
N. procne	Swallowtail Shiner	Intermediate	Insectivore	40
N. rubricroceus	Saffron Shiner	Intermediate	Insectivore	40
N. scabriceps	New River Shiner	Intolerant	Insectivore	40
N. scepticus	Sandbar Shiner	Intermediate	Insectivore	40
N. spectrunculus	Mirror Shiner	Intermediate	Insectivore	40
N. telescopus	Telescope Shiner	Intolerant	Insectivore	40
N. volucellus	Mimic Shiner	Intolerant	Insectivore	40
N. sp. cf. *chlorocephalus*	"Piedmont" Shiner	Intermediate	Insectivore	40
N. sp. cf. *rubellus*	"Rosyface" Shiner	Intolerant	Insectivore	40
Phenacobius crassilabrum	Fatlips Minnow	Intermediate	Insectivore	50
P. teretulus	Kanawha Minnow	Intolerant	Insectivore	50
Phoxinus oreas	Mountain Redbelly Dace	Intermediate	Herbivore	40
Pimephales notatus	Bluntnose Minnow	Tolerant	Omnivore	30
P. promelas	Fathead Minnow	Tolerant	Omnivore	30
Rhinichthys cataractae	Longnose Dace	Intermediate	Insectivore	50
R. obtusus	Western Blacknose Dace	Intermediate	Insectivore	50
Semotilus atromaculatus	Creek Chub	Tolerant	Insectivore	50
S. lumbee	Sandhills Chub	Intolerant	Insectivore	40
Catostomidae	**Suckers**			
Carpiodes carpio	River Carpsucker	Intermediate	Omnivore	100
C. cyprinus	Quillback	Intermediate	Omnivore	100
C. velifer	Highfin Carpsucker	Intermediate	Omnivore	100
C. sp. cf. *cyprinus*	(no common name)	Intermediate	Omnivore	100
C. sp. cf. *velifer*	(no common name)	Intermediate	Omnivore	100
Catostomus commersonii	White Sucker	Tolerant	Omnivore	100
Erimyzon oblongus	Creek Chubsucker	Intermediate	Omnivore	100
E. sucetta	Lake Chubsucker	Intermediate	Insectivore	100
Hypentelium nigricans	Northern Hog Sucker	Intermediate	Insectivore	100
H. roanokense	Roanoke Hog Sucker	Intermediate	Insectivore	100
Ictiobus bubalus	Smallmouth Buffalo	Intermediate	Omnivore	100
I. cyprinellus	Bigmouth Buffalo	Intermediate	Insectivore	100
I. niger	Black Buffalo	Intermediate	Insectivore	100
Minytrema melanops	Spotted Sucker	Intermediate	Insectivore	100
Moxostoma anisurum	Silver Redhorse	Intermediate	Insectivore	100
M. breviceps	Smallmouth Redhorse	Intermediate	Insectivore	100
M. collapsum	Notchlip Redhorse	Intermediate	Insectivore	100
M. carinatum	River Redhorse	Intermediate	Insectivore	100
M. duquesnei	Black Redhorse	Intermediate	Insectivore	100
M. erythrurum	Golden Redhorse	Intermediate	Insectivore	100
M. macrolepidotum	Shorthead Redhorse	Intermediate	Insectivore	100
M. pappillosum	V-Lip Redhorse	Intermediate	Insectivore	100

Table 4 (continued).

Family/ Species	Common Name	Tolerance Rating	Trophic Guild of Adults	YOY (< TL mm)
M. robustum	Robust Redhorse	Intolerant	Insectivore	100
M. sp. cf. *erythrurum*	Carolina Redhorse	Intermediate	Insectivore	100
M. sp. cf. *macrolepidotum*	Sicklefin Redhorse	Intermediate	Insectivore	100
Scartomyzon ariommus	Bigeye Jumprock	Intolerant	Insectivore	100
S. cervinum	Blacktip Jumprock	Intermediate	Insectivore	75
S. rupiscartes	Striped Jumprock	Intermediate	Insectivore	100
S. sp. cf. *lachneri*	"Brassy" Jumprock	Intermediate	Insectivore	100
Thoburnia hamiltoni	Rustyside Sucker	Intolerant	Insectivore	
Ictaluridae	**North American Catfishes**			
Ameiurus brunneus	Snail Bullhead	Intermediate	Insectivore	75
A. catus	White Catfish	Tolerant	Omnivore	100
A. melas	Black Bullhead	Tolerant	Insectivore	75
A. natalis	Yellow Bullhead	Tolerant	Omnivore	75
A. nebulosus	Brown Bullhead	Tolerant	Omnivore	75
A. platycephalus	Flat Bullhead	Tolerant	Insectivore	75
Ictalurus furcatus	Blue Catfish	Intermediate	Piscivore	100
I. punctatus	Channel Catfish	Intermediate	Omnivore	100
Noturus eleutherus	Mountain Madtom	Intermediate	Insectivore	40
N. flavus	Stonecat	Intermediate	Insectivore	40
N. furiosus	Carolina Madtom	Intolerant	Insectivore	40
N. gilberti	Orangefin Madtom	Intolerant	Insectivore	40
N. gyrinus	Tadpole Madtom	Intermediate	Insectivore	40
N. insignis	Margined Madtom	Intermediate	Insectivore	40
N. sp. cf. *leptacanthus*	Broadtail Madtom	Intolerant	Insectivore	40
Pylodictis olivaris	Flathead Catfish	Intermediate	Piscivore	150
Esocidae	**Pikes**			
Esox americanus americanus	Redfin Pickerel	Intermediate	Piscivore	100
E. masquinongy	Muskellunge	Intermediate	Piscivore	200
E. niger	Chain Pickerel	Intermediate	Piscivore	100
Umbridae	**Mudminows**			
Umbra pygmaea	Eastern Mudminnow	Intermediate	Insectivore	50
Salmonidae	**Trouts and Salmons**			
Oncorhynchus mykiss	Rainbow Trout	Intolerant	Insectivore	100
Salmo trutta	Brown Trout	Intermediate	Piscivore	100
Salvelinus fontinalis	Brook Trout	Intolerant	Insectivore	100
Aphredoderidae	**Pirate Perches**			
Aphredoderus sayanus	Pirate Perch	Intermediate	Insectivore	50
Amblyopsidae	**Cavefishes**			
Chologaster cornuta	Swampfish	Intermediate	Insectivore	25
Atherinopsidae	**New World Silversides**			
Labidesthes sicculus	Brook Silverside	Intermediate	Insectivore	50
Menidia beryllina	Inland Silverside	Intermediate	Insectivore	50
M. extensa	Waccamaw Silverside	Intolerant	Insectivore	50
Fundulidae	**Topminnows**			
Fundulus diaphanus	Banded Killifish	Intermediate	Insectivore	40
F. lineolatus	Lined Topminnow	Intermediate	Insectivore	40
F. rathbuni	Speckled Killifish	Intermediate	Insectivore	40
F. waccamensis	Waccamaw Killifish	Intolerant	Insectivore	40
Poeciliidae	**Livebearers**			
Gambusia affinis	Western Mosquitofish	Tolerant	Insectivore	20
G. holbrooki	Eastern Mosquitofish	Tolerant	Insectivore	20
Cottidae	**Sculpins**			
Cottus bairdii	Mottled Sculpin	Intermediate	Insectivore	50
C. carolinae	Banded Sculpin	Intermediate	Insectivore	50
C. caeruleomentum	Blue Ridge Sculpin	Intermediate	Insectivore	50

Table 4 (continued).

Family/ Species	Common Name	Tolerance Rating	Trophic Guild of Adults	YOY (< TL mm)
Moronidae	**Temperate Basses**			
Morone americana	White Perch	Intermediate	Piscivore	75
M. chrysops	White Bass	Intermediate	Piscivore	200
M. saxatilis	Striped Bass	Intermediate	Piscivore	175
Centrarchidae	**Sunfishes**			
Acantharchus pomotis	Mud Sunfish	Intermediate	Insectivore	50
Ambloplites cavifrons	Roanoke Bass	Intermediate	Piscivore	50
A. rupestris	Rock Bass	Intolerant	Piscivore	50
Centrarchus macropterus	Flier	Intermediate	Insectivore	50
Enneacanthus chaetodon	Blackbanded Sunfish	Intermediate	Insectivore	40
E. gloriosus	Bluespotted Sunfish	Intermediate	Insectivore	40
E. obesus	Banded Sunfish	Intermediate	Insectivore	40
Lepomis auritus	Redbreast Sunfish	Tolerant	Insectivore	50
L. cyanellus	Green Sunfish	Tolerant	Insectivore	50
L. gibbosus	Pumpkinseed	Intermediate	Insectivore	50
L. gulosus	Warmouth	Intermediate	Insectivore	50
L. macochirus	Bluegill	Intermediate	Insectivore	50
L. marginatus	Dollar Sunfish	Intermediate	Insectivore	50
L. microlophus	Redear Sunfish	Intermediate	Insectivore	50
L. punctatus	Spotted Sunfish	Intermediate	Insectivore	50
Lepomis sp.	Hybrid Sunfish	Tolerant	Insectivore	50
Micropterus coosae	Redeye Bass	Intermediate	Piscivore	100
M. dolomieu	Smallmouth Bass	Intolerant	Piscivore	100
M. punctulatus	Spotted Bass	Intermediate	Piscivore	100
M. salmoides	Largemouth Bass	Intermediate	Piscivore	100
Pomoxis annularis	White Crappie	Intermediate	Piscivore	75
P. nigromaculatus	Black Crappie	Intermediate	Piscivore	75
Percidae	**Perches**			
Etheostoma acuticeps	Sharphead Darter	Intolerant	Insectivore	40
E. blennioides	Greenside Darter	Intermediate	Insectivore	40
E. chlorobranchium	Greenfin Darter	Intolerant	Insectivore	40
E. collis	Carolina Darter	Intermediate	Insectivore	30
E. flabellare	Fantail Darter	Intermediate	Insectivore	30
E. fusiforme	Swamp Darter	Intermediate	Insectivore	30
E. gutselli	Tuckasegee Darter	Intermediate	Insectivore	40
E. inscriptum	Turquoise Darter	Intolerant	Insectivore	40
E. jessiae	Blueside Darter	Intolerant	Insectivore	40
E. kanawhae	Kanawha Darter	Intolerant	Insectivore	40
E. mariae	Pinewoods Darter	Intolerant	Insectivore	30
E. nigrum	Johnny Darter	Intermediate	Insectivore	30
E. olmstedi	Tessellated Darter	Intermediate	Insectivore	40
E. perlongum	Waccamaw Darter	Intolerant	Insectivore	30
E. podostemone	Riverweed Darter	Intolerant	Insectivore	30
E. rufilineatum	Redline Darter	Intermediate	Insectivore	40
E. serrifer	Sawcheek Darter	Intolerant	Insectivore	30
E. swannanoa	Swannanoa Darter	Intermediate	Insectivore	40
E. thalassinum	Seagreen Darter	Intolerant	Insectivore	40
E. vitreum	Glassy Darter	Intermediate	Insectivore	30
E. vulneratum	Wounded Darter	Intolerant	Insectivore	40
E. zonale	Banded Darter	Intermediate	Insectivore	40
Perca flavescens	Yellow Perch	Intermediate	Piscivore	80
Percina aurantiaca	Tangerine Darter	Intolerant	Insectivore	40
P. burtoni	Blotchside Logperch	Intolerant	Insectivore	40
P. caprodes	Logperch	Intermediate	Insectivore	40
P. crassa	Piedmont Darter	Intolerant	Insectivore	40
P. evides	Gilt Darter	Intolerant	Insectivore	40
P. gymnocephala	Appalachia Darter	Intolerant	Insectivore	40
P. nevisense	Chainback Darter	Intolerant	Insectivore	40
P. nigrofasciata	Blackbanded Darter	Intolerant	Insectivore	40
P. oxyrhynchus	Sharpnose Darter	Intolerant	Insectivore	40
P. roanoka	Roanoke Darter	Intolerant	Insectivore	30
P. sciera	Dusky Darter	Intermediate	Insectivore	40
P. squamata	Olive Darter	Intolerant	Insectivore	

Table 4 (continued).

Family/ Species	Common Name	Tolerance Rating	Trophic Guild of Adults	YOY (< TL mm)
Sander canadensis	Sauger	Intermediate	Piscivore	
S. vitreus	Walleye	Intermediate	Piscivore	
Elassomatidae	**Pygmy Sunfishes**			
Elassoma evergladei	Everglades Pygmy Sunfish	Intermediate	Insectivore	20
E. zonatum	Banded Pygmy Sunfish	Intermediate	Insectivore	20
E. boehlkei	Carolina Pygmy Sunfish	Intolerant	Insectivore	20
Sciaenidae	**Drums**			
Aplodinotus grunniens	Freshwater Drum	Intermediate	Insectivore	

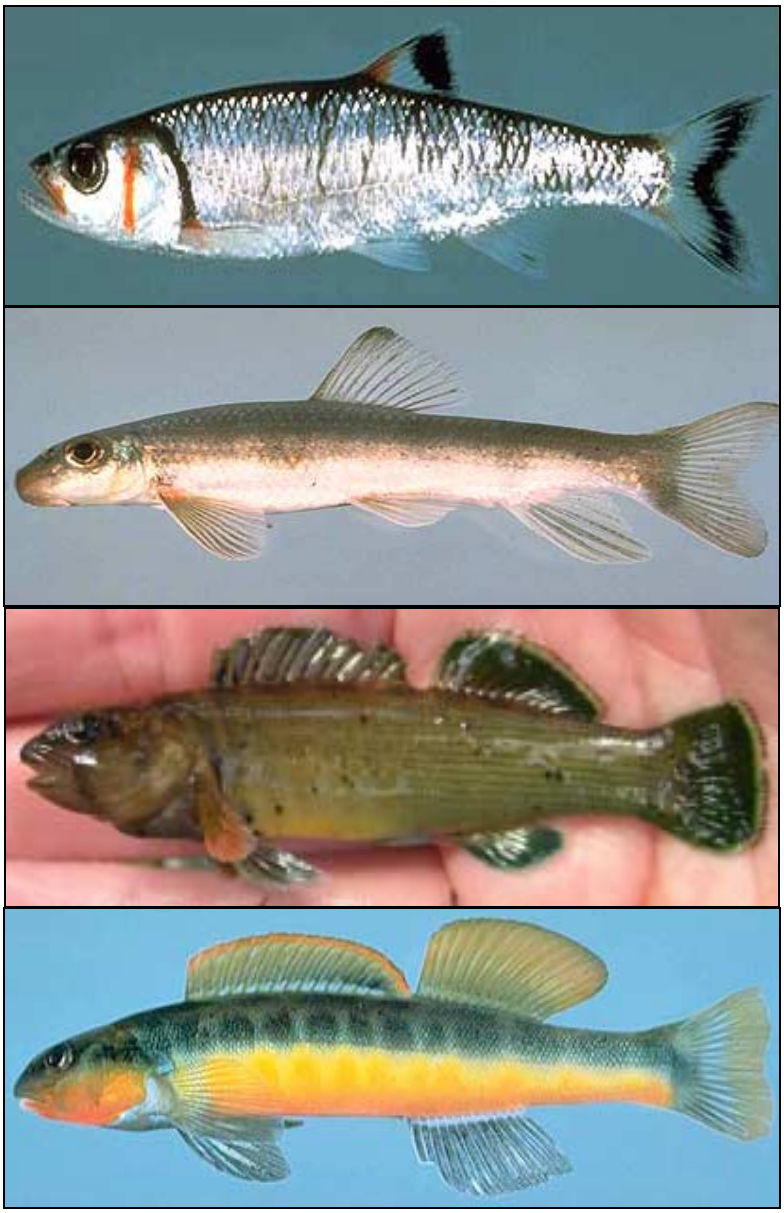


Table 5. Intolerant species of fish found in North Carolina.

Family/Species	Common Name	Family/Species	Common Name
Petromyzontidae	**Lampreys**	**Atherinopsidae**	**New World Silversides**
Lampetra aepyptera	Least Brook Lamprey	*Menidia extensa*	Waccamaw Silverside
Cyprinidae	**Carps and Minnows**	**Fundulidae**	**Topminnows**
Cyprinella labrosa	Thicklip Chub	*Fundulus waccamensis*	Waccamaw Killifish
C. pyrrhomelas	Fieryblack Shiner		
C. zanema	Santee Chub	**Centrarchidae**	**Sunfishes**
Erimonax monachus	Spotfin Chub	*Ambloplites rupestris*	Rock Bass
Exoglossum laurae	Tonguetied Minnow	*Micropterus dolomieu*	Smallmouth Bass
E. maxillingua	Cutlip Minnow		
Hybopsis hypsinotus	Highback Chub	**Percidae**	**Perches**
H. rubifrons	Rosyface Chub	*Etheostoma acuticeps*	Sharphead Darter
Lythrurus matutinus	Pinewoods Shiner	*E. chlorobranchium*	Greenfin Darter
Notropis chalybaeus	Ironcolor Shiner	*E. inscriptum*	Turquoise Darter
N. maculatus	Taillight Shiner	*E. jessiae*	Blueside Darter
N. micropteryx	Highland Shiner	*E. kanawhae*	Kanawha Darter
N. photogenis	Silver Shiner	*E. mariae*	Pinewoods Darter
N. scabriceps	New River Shiner	*E. perlongum*	Waccamaw Darter
N. telescopus	Telescope Shiner	*E. podostemone*	Riverweed Darter
N. volucellus	Mimic Shiner	*E. serrifer*	Sawcheek Darter
N. sp. cf. *rubellus*	Rosyface Shiner	*E. thalassinum*	Seagreen Darter
Phenacobius teretulus	Kanawha Minnow	*E. vulneratum*	Wounded Darter
Semotilus lumbee	Sandhills Chub	*Percina aurantiaca*	Tangerine Darter
		P. burtoni	Blotchside Logperch
Catostomidae	**Suckers**	*P. crassa*	Piedmont Darter
Moxostoma robustum	Robust Redhorse	*P. evides*	Gilt Darter
Scartomyzon ariommus	Bigeye Jumprock	*P. gymnocephala*	Appalachia Darter
Thoburnia hamiltoni	Rustyside Sucker	*P. nigrofasciata*	Blackbanded Darter
		P. nevisense	Chainback Darter
Ictaluridae	**North American Catfishes**	*P. oxyrhynchus*	Sharpnose Darter
Noturus furiosus	Carolina Madtom	*P. roanoka*	Roanoke Darter
N. gilberti	Orangefin Madtom	*P. squamata*	Olive Darter
N. sp. cf. *leptacanthus*	Broadtail Madtom		
		Elassomatidae	**Pygmy Sunfishes**
Salmonidae	**Trouts and Salmons**	*Elassoma boehlkei*	Carolina Pygmy Sunfish
Oncorhynchus mykiss	Rainbow Trout		
Salvelinus fontinalis	Brook Trout		

Table 6. Tolerant species of fish found in North Carolina.

Family/Species	Common Name	Family/Species	Common Name
Lepisosteidae	**Gars**	**Catostomidae**	**Suckers**
Lepisosteus osseus	Longnose Gar	*Catostomus commersonii*	White Sucker
Amiidae	**Bowfins**	**Ictaluridae**	**North American Catfishes**
Amia calva	Bowfin	*Ameiurus catus*	White Catfish
		A. melas	Black Bullhead
Cyprinidae	**Carps and Minnows**	*A. natalis*	Yellow Bullhead
Carassius auratus	Goldfish	*A. nebulosus*	Brown Bullhead
Ctenopharyngodon idella	Grass Carp	*A. platycephalus*	Flat Bullhead
Cyprinella analostana	Satinfin Shiner		
C. lutrensis	Red Shiner	**Poeciliidae**	**Livebearers**
Cyprinus carpio	Common Carp	*Gambusia affinis*	Western Mosquitofish
Notemigonus crysoleucas	Golden Shiner	*G. holbrooki*	Eastern Mosquitofish
Pimephales notatus	Bluntnose Minnow		
P. promelas	Fathead Minnow	**Centrarchidae**	**Sunfishes**
Semotilus atromaculatus	Creek Chub	*Lepomis auritus*	Redbreast Sunfish
		L. cyanellus	Green Sunfish
		Lepomis sp.	Hybrid Sunfish

INTEGRITY CLASS ASSIGNMENT

The scores for all 10 or 12 metrics are then summed to obtain the overall NCIBI score. Finally, the score (an even number between 12 and 60) is then used to determine the biological integrity class of the stream from which the sample was collected (Table 7).[1]

Table 7. Revised scores and classes for evaluating the fish community of a wadeable stream in select streams using the North Carolina Index of Biological Integrity.

River Basin	NCIBI Score	Integrity Class
French Broad, Hiwassee, Little Tennessee, New, and Watauga	58 or 60	Excellent
	48, 50, 52, 54, or 56	Good
	40, 42, 44, or 46	Good-Fair
	34, 36, or 38	Fair
	≤ 32	Poor
Broad, Catawba, Savannah, and Yadkin	54, 56, 58, or 60	Excellent
	48, 50, or 52	Good
	42, 44, or 46	Good-Fair
	36, 38, or 40	Fair
	≤ 34	Poor
Cape Fear, Neuse, Roanoke, and Tar	54, 56, 58, or 60	Excellent
	46, 48, 50, or 52	Good
	40, 42, or 44	Good-Fair
	34, 36, or 38	Fair
	≤ 32	Poor

[1] In the Western and Northern Mountains (French Broad, Hiwassee, Little Tennessee, New, and Watauga River basins), the NCIBI is based upon 10 rather than 12 metrics (Table 1). Using 10 metrics with each metric's criteria scored a 1, 3, or 5 and desiring to keep 60 as the maximum NCIBI Total Score, the total score was multiplied by 1.2 (60/50=1.2). Scores were rounded up or down to the nearest whole even number (e.g., 57.6 rounded up to 58; 50.4 rounded down to 50) .Using 10 metrics instead of 12 and following the conversions as described, the final Total NCIBI Scores of 54, 42, 30, and 18 are no longer possible. This slight flaw should not affect the usefulness and applicability of the 10 metric NCIBI for the Western and Northern Mountains

Total Score based upon 10 Metrics before Multiplier	Total Score based upon 10 Metrics after Applying a 1.2 Multiplier	Final Total Score after Rounding (if necessary)
50	60	60
48	57.6	58
46	55.2	56
44	52.8	52
42	50.4	50
40	48	48
38	45.6	46
36	43.2	44
34	40.8	40
32	38.4	38
30	36	36
28	33.6	34
26	31.2	32
24	28.8	28
22	26.4	26
20	24	24
18	21.6	22
16	19.2	20
14	16.8	16
12	14.4	14

OTHER WATER QUALITY INDICATORS

Although the North Carolina Index of Biological Integrity is the primary tool used in the Stream Fish Community Assessment Program, other water quality measurements (e.g., water temperature, dissolved oxygen, pH, specific conductance, and water clarity are also monitored at every site in accordance with the Intensive Survey Unit's SOP (NCDENR 2003). At each site, a non-regulatory stream and riparian habitat assessment is conducted (Appendices 5 and 6).

FIELD SAMPLING AND LABORATORY PROCESSING METHODS

SAMPLING SCHEDULE AND FREQUENCY

Sites that are part of the Basinwide Monitoring Program are sampled once every five years and, due to staffing constraints, usually between April and June. For example, basinwide sites in the Yadkin River Basin were sampled in 1996, 2001, and will be sampled again in 2006 (Figure 4). Watershed-specific special study sites that are designed to address a specific, short-term question (e.g., Use Attainability, impacts from a permitted discharger, watershed modifications, *etc*.) are usually sampled only once and may be sampled anytime between March and December.

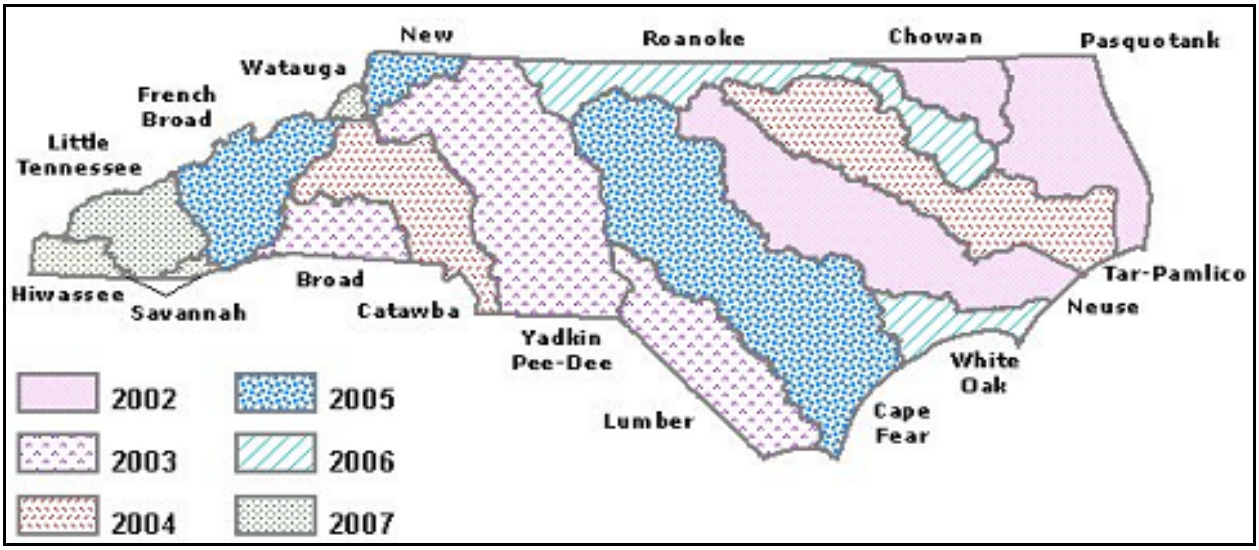


Figure 4. Basinwide planning schedule for North Carolina's 17 river basins, 2002 to 2007.

FISH COLLECTION LICENSES AND PERMITS

Collection permits are required to collect fish from North Carolina freshwater ecosystems and must accompany the field staff whenever collections are made. Annually, it is the responsibility of the Environmental Biologist III to insure that a Scientific Collection License and an Endangered Species Permit have been obtained from the North Carolina Wildlife Resources Commission's Division of Boating and Inland Fisheries and from the Division of Wildlife Management.

SITE LOCATIONS

Sites are established at publicly accessible, fixed locations (i.e., specific latitude and longitude), generally at bridge crossings. Lists of all the sites ever monitored, by river basin, may be found at: http://www.esb.enr.state.nc.us/IBIrate.htm. Locations and their geo-references were originally identified using USGS 7.5 minute topographic maps or Maptech Terrain Navigator ® software. Stations are strategically located to monitor a specific area of concern such as:

- overall water quality in a larger watershed,
- effect of point source discharges,
- effect of non-point sources of pollution (e.g., urban areas, animal operations, agriculture),
- effect of land use changes,
- waters of significant ecological, recreational, political, or municipal use, or

- waters that show an impairment due to unknown causes.

Sites that have been monitored between 1991 and 2005 were previously shown in Figure 2. Because this is a relatively new program, many of the current sites have been active for only 1 to 3 basinwide monitoring cycles. However, maintenance of many of these sites on a long-term basis is integral to identifying temporal patterns within a watershed and to gaining an understanding of the variability within the fish community. Consequently, requests from DWQ staff for station establishment and/or discontinuation will be assessed on the value gained from a long-term perspective. Requests for additional sampling of sites (usually a one-time sampling event within a watershed) are handled through special studies. Adjustments to site locations and sampling regimens may be made with sufficient reason, such as:

- safety concerns of field staff,
- changes to location accessibility,
- the reason for sampling is no longer valid (i.e., a discontinued discharge),
- the emergence of new water quality concerns, or
- resource constraints, particularly staff vacancies.

If any of these concerns arise, the Environmental Biologist III will meet with the BAU Supervisor to determine if it is appropriate for the site to be discontinued.

Sampling condition limitations are dictated by extremes in water clarity (turbidity), stream width and depth (too wide and deep), substrate (deep muck), precipitation (rainfall and electrical storms), aquatic macrophyte growths (excessive), flow (not flowing or too much flow), dangerous sampling conditions, time of day (lateness in the afternoon), *etc*.

A representative wadeable site of approximately 600 ft. is selected. Wadeable streams are those that can be safely waded by the sampling crew while wearing a backpack electrofisher unit and still allow the sampler and netter to reach all areas of the stream with the electrofishing probes and dipnet. When possible, the delineated reach should be located upstream from the bridge access area. If possible, personnel measuring the stream segment should avoid walking in the stream segment to avoid scaring fish out of the sample segment and to minimize habitat disturbance.

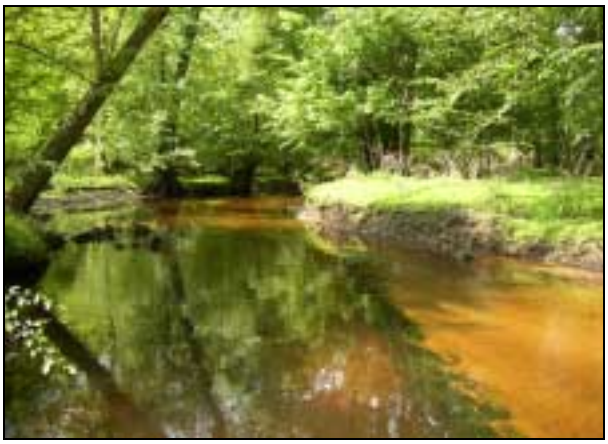
FIELD VARIABLES

The Stream Fish Community Assessment Program Samples Log Sheet (Appendix 1) is updated and a Stream Fish Community Assessment Program Field Data Sheet (Appendix 2) is completed whenever a sample is collected. Data that are recorded include: stream name, sample location, county, river basin, subbasin, latitude, longitude, drainage area, stream index number and classification (obtained from Basinwide Information Management System), habitat score, elevation, sample number, sample date, time, number of shocking units, duration of shocking, sampling personnel, location of sample reach, and use of a seine (yes or no). An example of a completed sheet is shown in Appendix 3. These data sheets

are kept in a folder in the field vehicle under the custody of the Environmental Biologist III or the Environmental Biologist II until returned to the ESS Building. After the sampling trip has been completed, samples are transported to the Fish Community Assessment Laboratory, located in the ESS building.

The sample information (sample number, waterbody, location, *etc*.) is recorded on the Log Sheet from NC DWQ Stream Fish Community Assessment Program Samples (Appendix 1). This log sheet tracks all the samples that have been collected for a particular year. The Environmental Biologist III assigns the Sample Number in numerical order. The first sample collected each year is Sample No. 1, the second sample is Sample No. 2, the third sample is Sample No. 3, etc. The sample numbers for 2005 took the form of 2005-01, 2005-02, 2005-03, *etc*. A sample number is assigned to a sample only after the sample has been collected. The log sheet and the field data sheets are stored in a 3-ring binder labeled "Field Data Sheets" in the Environmental Biologist III's office at the ESS Building.

Physical habitat and water quality data that are collected include specific conductance, dissolved oxygen, temperature, pH, habitat description, average stream width and depth, water clarity (e.g., clear, slightly turbid, turbid, tannin stained, or blackwater, *etc*.), and substrate. These data are also recorded on the Stream Fish Community Assessment Program Field Data Sheet (Appendix 2).



SAMPLE COLLECTION

Essential sampling equipment that should accompany the Staff when sampling are listed in Table 8.

Table 8. Field sampling equipment.

County, state, and topographic maps	Chest waders and rubber gloves
Digital camera and charger	Measuring boards
Appropriate identification keys and field guides	Data sheets, pens, pencils, and waterproof markers
Assorted jars and plastic buckets with lids	Formalin and 95 percent ethanol
GPS unit	Measuring chain, thread, tape measure, and flagging tape
Dipnets (1/8 in. mesh) and assorted sizes of seines	Identification labels, tags, and rubber bands
Backpack electrofishing units	First aid kit, cardiac resuscitation unit, and insect repellent
Electrofishing batteries and chargers	Large fish preservation containers
Electrofishing probes and replacement rings	Water quality instruments

The number of personnel required to efficiently and effectively sample a 600 ft. wadeable section of stream is listed in Table 9.

Table 9. Sampling personnel required to effectively sample streams of varying widths.

Stream width (m)	No. of electrofishers	No. of netters
≤ 3	1	1
3 to 10	2	2
10 to 15	2 or 3	2 or 3
> 15	3 or 4	3 or 4

Typically, one-half of the sampling crew is outfitted with backpack electrofishing units and the other half with dip nets and buckets.

Fish in the delineated stretch of stream are collected in a two-pass depletion technique using backpack electrofishing units and persons netting the stunned fish. Staff members collect samples by first moving in an upstream direction. After a short break, 5 to 10 minutes to allow the water to clear, sample collection is continued by staff members moving back downstream. All micro- and macrohabitats (riffles, pools, runs, snags, undercuts, deadfalls, quiescent leaf-covered substrates, *etc*.) should be thoroughly sampled. Electrofishing downstream into a seine should also be performed wherever there are significant riffles. Stunned fish are netted and placed into buckets with water that is frequently changed to minimize stress and mortality.

Details of the backpack electrofisher use and operation are given in the operator's manual and should be read carefully by all staff before using the equipment. Safety concerns require the wearing of chest waders and rubber gloves when the electrofishing unit is in operation.

After collection, all readily identifiable fish are examined for diseases, sores, lesions, fin damage, and skeletal anomalies, measured (total length to the nearest 1 mm), and then released. All data are recorded on the Stream Fish Community Assessment Program Field Data Sheet (Appendix 2). If a species is represented by multiple ages, a "Y" (for yes) is written in the margin of the data sheet across from the species name. If a species is not represented by multiple ages, a "N" (for no) is written. Deformed or diseased fish are also noted on the data sheet by circling the total length measurement of the affected fish. In addition, it is suggested that digital pictures be taken of any unusually deformed or diseased fish.

Once the first 50 specimens of a species are measured, the remaining fish of that particular species are just counted and released. All other fish (i.e., those fish that are not readily identifiable) are preserved in 10 percent formalin and returned to the laboratory for identification, examination, and total length measurement. If large (> 300 mm), unidentifiable fish are retained, the abdominal cavity should be injected with formalin soon after preservation or as soon as possible before the end of the sampling day.

SAMPLE IDENTIFICATION TAGS

Two sample identification tags (containing waterbody name, road crossing, county, date, and sample collection number) are completed and placed inside and attached outside every sample container (plastic bucket or jar). Because formalin is the only preservative used, it is understood by staff that the samples are preserved in formalin and labeling of the sample container as to containing formalin is not necessary. Collectors' names are not listed on the labels because that information has been previously recorded on the Fish Community Assessment-IBI Data Sheet (Appendix 2). It is not necessary to record on the data sheet or the sample identification tag what analysis is to be done on the sample because samples are only preserved and returned to the laboratory if the species level identification is to be performed in the laboratory.

FIELD WATER QUALITY MEASUREMENTS

Measurements made in the field include water temperature, specific conductance, pH, stream flow (low, normal, high), water clarity (clear, slightly turbid, turbid, tannin stained, or blackwater), and dissolved oxygen. Field measurements are discrete and are made *in situ* by field staff at the time of the station visit. The only acceptable exception is pH. Most field pH meters are not waterproof; therefore, pH is measured from a water sample within five minutes of sample collection. All field activities are to be performed in accordance with the Intensive Survey Unit's SOP (NCDENR 2003). In addition to the NC DWQ's Intensive Survey Unit's SOP sections cited in Table 10, the instruction manual for the appropriate meter should also be consulted.

Table 10. Field measurement method references and reporting levels. Adopted from the Intensive Survey Unit's SOP (NCDENR 2003).

Parameter	NC DWQ's Intensive Survey Unit's SOP & section[1]	EPA method	Reported to nearest
Dissolved oxygen	III.3	360.1	0.1 mg/L
pH	III.4	150.1	0.1 s. u.
Water temperature	III.1	170.1	0.1 ºC
Specific conductance	III.5.2	120.1	1 µmhos/cm

[1] Section numbers III.1 - III.5 refer to use of YSI combination meters and Fisher Scientific Accumet pH meters.

All field meters are to be inspected and calibrated before each sampling trip and at minimum at the end of each day used. Field staff should record calibration information on the Field Meter Calibration Sheet (Appendix 4). This calibration form, which was adopted from the NC DWQ's Intensive Survey Unit's SOP, is stored in a 3-ring binder labeled "Stream Fish Community Assessment Water Quality Meter Calibration Log" in the Stream Fish Community Assessment Program's Laboratory. Specific calibration procedures are documented in each meter's manufacturers' instruction manual. For specific conductance and pH, two-point calibrations should be performed. Dissolved oxygen meters should be calibrated using the air calibration method.

Standards should be selected so that they bracket the range of measurements expected that day. Conductance standards are prepared monthly by NC DWQ's Intensive Survey Unit staff and are shared with other ESS units. Standard concentrations calibrated against are 147 and 718 µmhos/cm. NC DWQ's Intensive Survey Unit also purchases traceable pH buffers (standards) and shares these with other ESS units. Meters currently in use require standards of 4.0, 7.0, and 10.0 s. u.

Meters should also be checked against standards periodically throughout the day and recalibrated if any of the following conditions occur:

- Physical shock to meter;
- Dissolved oxygen membrane is touched, fouled, punctured, or dries out;
- Unusual (high or low for the particular site) or erratic readings, or excessive drift;
- Extreme readings (e.g., extremely acidic or basic pH; dissolved oxygen saturation >120 percent); or
- Measurements are outside of the range for which the meter was calibrated.

HABITAT ASSESSMENT

A method has been developed by the Biological Assessment Unit to evaluate the physical habitats of a stream (Appendices 5 and 6). The narrative descriptions of eight (Mountain/Piedmont) or seven (Coastal Plain and Sand Hills) habitat characteristics, including channel modification, amount of instream habitat, type of bottom substrate, pool variety, riffle frequency, length and width, bank stability, light penetration, and riparian zone width, are converted into numerical scores. The total habitat score ranges between 1 and 100. Higher numbers suggest better habitat quality, but criteria have not been developed to assign impairment ratings.

SAMPLE HANDLING AND CUSTODY

Stunned fish are collected and temporarily stored in a bucket filled with stream water. Readily identifiable fish are counted and measured in the field and then released. If the sampling trip necessitates an overnight stay, samples are stored in the cargo portion of the field vehicle, which is kept locked whenever

staff members are away from the vehicle.

Samples are stored on bench space in the Fish Community Assessment Laboratory in the ESS Building until the fish have been properly preserved in formalin (usually 1-2 weeks or until the fish no longer are floating in the preservative). Once properly preserved, the sample can then be processed.

LABORATORY PROCESSING OF FISH SAMPLES

After the fish have been properly preserved in formalin (usually 1-2 weeks or until the fish no longer are floating in the preservative), the sample can be processed. The preservative is decanted under a hood (or other means providing appropriate ventilation) and discarded. The sample is rinsed with tap water several times and then allowed to soak in tap water for approximately one hour. The sample is sorted and each fish is identified to the **species** level and its total length measured to the nearest 1 millimeter. All laboratory-derived data are recorded on the Stream Fish Community Assessment Program Field Data Sheet (Appendix 3). Deformed or diseased fish are also noted on the data sheet by circling the total length measurement of the affected fish. If a species is represented by multiple ages, a "Y" (for yes) is written in the margin of the data sheet across from the species name. If a species is not represented by multiple ages, a "N" (for no) is written. Problematic identifications are verified by personnel from the North Carolina State Museum of Natural Science.

YOUNG-OF-YEAR CONSIDERATIONS AND ADJUSTMENTS

Young-of-year (YOY) fish may pose several challenges when applying the IBI metrics to a fish community sample (Angermeier and Karr (1986) and Angermeier and Schlosser (1987). Assessments made during the spring and early summer (April-June) tend to avoid these challenges. However, samples collected later in the summer and fall may contain an abundance of YOY fish. Individuals of a species who spawn in late summer or fall or from a late hatching cohort are not considered YOY when collected the following year (after January 1st) even though such individuals may be noticeably smaller than an earlier hatching cohort.

In some instances, depending upon the mildness of the winter and early spring, YOY fish (for example, redfin pickerel, creek chubsucker, bluegill, and redbreast sunfish), may already be present in samples collected during the spring. Assessments made in mid- to late June require careful attention and sometimes, professional judgment.

Efforts are made to not collect YOY fish, and, if collected, all YOY fish are excluded from all NCIBI calculations. Between July 1 and December 30, when most YOY may be collected, Table 4 should used as a guidance for the determination of YOY cut-off lengths. If a length for a particular species is not listed, best professional judgment or new knowledge of the life history of the species in North Carolina or the Southeast may be used for individuals collected where there may be doubt as to whether or not a fish is a YOY fish.

ACQUIRED DATA
All data are generated through the Stream Fish Community Assessment Program field activities and consequent laboratory analyses, with three exceptions:

- Geo-referenced (latitude and longitude) data are obtained from Maptech Terrain Navigator® software or from a Garmin GPS meter. These data are used in Geographic Information System mapping software and in describing the exact location from which a sample was collected.
- Watershed drainage areas for each site are obtained from the U. S. Geological Survey or from DWQ's geographical information system software/data layers.
- Species lists for each basin are compiled from up-to-date taxonomic keys listed in the Literature Cited and Suggested References section, from data previously collected by the Stream Fish Community Assessment Program, and from other researchers at universities and state and federal resource agencies. These data aid in the accurate identification of fish species by listing which species are typically found or are not found in a particular river basin. Species lists are available at http://www.esb.enr.state.nc.us/www.esb.enr.state.nc.us/Native and Introduced Freshwater Fish in North Carolina.2-1.htm.

COMPONENTS OF THE QA/QC PLAN
A detailed description of the Stream Fish Community Assessment Program Quality Assurance Project Plan can be found at http://www.esb.enr.state.nc.us/BAU.html.

The Environmental Biologist III or the Environmental Biologist II will be responsible for overseeing the collection of all stream fish community assessment program samples. Personnel from the Biological Assessment Unit will provide primary sampling assistance. Other experienced field biologists within the Environmental Sciences Section or other agencies may be used as needed.

Prior to sampling, a fish species list will be compiled of all the species known or suspected to occur within the basin or stream under study. Such a list is compiled from species distribution maps (Menhinick 1991 and amended with Biological Assessment Unit data and data from other regional fisheries researchers). The list will also show which species may be afforded protection at the federal or state level and which would require field identification and immediate release.

As discussed in the Sample Collection section, as many readily and easily identifiable fish are processed stream-side as possible. A fish whose specific identity is unknown, questionable, or disputed between the fisheries biologists is properly preserved for later laboratory identification.

Examples of a species or a specimen(s) that should be preserved are ones that:

- can not be readily and easily identified in the field;
- are not represented in the Reference Collection (a list of species in the Reference Collection is kept with the Reference Collection in the Fish Laboratory and should be consulted prior to sampling) ;
- are of known taxonomic value (e.g., a poorly understood or undescribed species (such as the Carolina redhorse) or rarely collected size classes of a species);
- represent a new distributional record; or
- may be a hybrid.

Additional suggested guidelines for when to preserve specimens may be found in Walsh and Meador (1998).

Random samples, identified in the laboratory, are re-processed for accurate and correct determinations of identity and presence or absence of multiple age classes. Because of the relatively limited icthyofauna within any specific river basin, the likelihood of misidentifications is not as great as is the case for other taxonomic groups (e.g., benthic invertebrates or phytoplankton). Consequently, each fisheries biologist is required to roll two dice after every 12 samples have been completed. The sample corresponding with the die number is re-identified and processed by another fisheries biologist for verification. Any misidentifications or inaccuracies in multiple age class determinations are resolved between the two

biologists. The data sheet from which the sample was chosen for verification is signed and dated by both biologists attesting to the accuracy and completeness of the sample.

A Reference Collection shall be maintained. Except for federally- and state-recognized rare, endangered, or threatened species (Table 11), the Reference Collection should include at least one specimen of every freshwater species found in the state. Species afforded the extra state or federal protection and which were collected accidentally (Incidental Take) shall be deposited in the North Carolina State Museum of Natural Sciences (NCSMNS). The Reference Collection shall be maintained and utilized for laboratory identifications of problematic species. Comparisons of such specimens or species may also be made to specimens in the NCSMNS. A list of species in the Reference Collection is kept with the Reference Collection in the Stream Fish Community Assessment Program's Fish Laboratory and should updated as needed.

Table 11. Phylogenetic listing of the state and federally protected endangered and threatened species (from LeGrand, *et al*. 2004).

Species	Common Name	State Status	Federal Status
Lampetra aepyptera	Least Brook Lamprey	Threatened	
L. appendix	American Brook Lamprey	Threatened	
Acipenser brevirostrum	Shortnose Sturgeon	Endangered	Endangered
Polyodon spathula	Paddlefish	Endangered	
Erimonax monachus	Spotfin Chub	Threatened	Threatened
Exoglossum maxillingua	Cutlip Minnow	Endangered	
Hybopsis rubifrons	Rosyface Chub	Threatened	
Luxilus chrysocephalus	Striped Shiner	Threatened	
Notropis mekistocholas	Cape Fear Shiner	Endangered	Endangered
Scartomyzon ariommus	Bigeye Jumprock	Threatened	
Thoburnia hamiltoni	Rustyside Sucker	Endangered	
Noturus flavus	Stonecat	Endangered	
N. gilberti	Orangefin Madtom	Endangered	
Menidia extensa	Waccamaw Silverside	Threatened	
Cottus carolinae	Banded Sculpin	Threatened	
Etheostoma acuticeps	Sharphead Darter	Threatened	
E. perlongum	Waccamaw Darter	Threatened	
Percina burtoni	Blotchside Logperch	Endangered	
P. caprodes	Logperch	Threatened	
P. sciera	Dusky Darter	Endangered	
Aplodinotus grunniens	Freshwater Drum	Threatened	
Elassoma boehlkei	Carolina Pygmy Sunfish	Threatened	

All specimens returned to the laboratory for identification which do not become part of the Reference Collection or of the Teaching Collection (a collection maintained to educate school groups, tours, or citizens at public fair and forums) will be donated to the NCSMNS. The State Ichthyologist (and staff) will serve as the qualified, independent fish taxonomic specialist(s). All specimens are verified for correctness of species identification prior to being incorporated into the NCSMNS Collection. Any misidentifications or other discrepancies by the Division fisheries biologists will be communicated back by the NCSMNS staff.

DATA MANAGEMENT

Field- and laboratory-generated data from a single sampling event are recorded on the same Stream Fish Community Assessment Program Field Data Sheet (Appendices 2 and 3). A vertical bar "**I**" is used to separate and distinguish field data (specimens identified, measured, and released in the field) from lab data (specimens identified and measured in the lab). This distinction is made so that staff members know and can keep track of which specimens were processed in the field and which specimens were returned to the laboratory.

Data are keyed by either the Environmental Biologist III or the Environmental Biologist II into the Stream Fish Community Assessment Program's Microsoft Access® 2000 database. Annually, this results in almost 1,500 records (~20 species per site X 75 sites sampled annually = 1,500 species records). The biologists review the data for completeness, data entry errors, unlikely or impossible values, *etc*. Copies of this database reside on the Environmental Biologist III's drive on the ESS server and on BAU's drive on the ESS server. Tape backups are run daily on the ESS servers. The database is updated on a as needed basis whenever samples are completed or whenever errors in previously entered data are identified.

All calculations that result in any data summaries as shown in the North Carolina Fish Community Reports (Appendix 8) are generated by programs in the Stream Fish Community Assessment Program's Microsoft Access® 2000 database.

LITERATURE CITED AND SUGGESTED REFERENCES

Angermeier, P. L. and J. R. Karr. 1986. Applying an Index of Biotic Integrity based on stream - fish communities: considerations in sampling and interpretation. N. Amer. J. Fish. Manage. 6:418 - 429.

Angermeier, P. L. and I. J. Schlosser. 1987. Assessing biotic integrity of the fish community in a small Illinois stream. N. Amer. J. Fish. Manage. 7:331 - 338.

Barbour, M. T., J. Gerritsen, B. D. Snyder, and J. B. Stribling. 1999. Rapid bioassessment protocols for use in streams and wadeable rivers: periphyton, benthic macroinvertebrates, and fish. 2nd edition. EPA 841 - B-99-002. U.S. Environmental Protection Agency. Office of Water, Washington, DC.

Carlander, K. D. 1969. Handbook of freshwater fishery biology, Vol. 1. Life history data on freshwater fish of the United States and Canada, exclusive of the Perciformes. Iowa State University Press, Ames, IA.

_____. 1977. Handbook of freshwater fishery biology, Vol. 2. Life history data on centrarchid fishes of the United States and Canada. Iowa State University Press, Ames, IA.

Etnier, D. A. and W. C. Starnes. 1993. The fishes of Tennessee. The University of Tennessee Press, Knoxville, TN.

Fausch, K. D., J. R. Karr, and P. R. Yant. 1984. Regional application of an index of biotic integrity based on stream fish communities. Trans. American Fish. Soc. 113: 39 - 55.

Fels, J. 1997. North Carolina watersheds map. North Carolina State University Cooperative Extension

Service. Raleigh, NC.

Gibson, G. R., Jr. (ed.). 1996. Biological criteria. Technical guidance for streams and small rivers. Revised edition. EPA 822-B-96-001. U.S. Environmental Protection Agency. Office of Water, Washington, DC.

Griffith, G., Omernik, J. and J. Comstock. 2002. Ecoregions of North Carolina. United States Environmental Protection Agency. Research and Development. NHEERL. Western Ecology Division. Corvallis, OR.

Hughes, R. M. 1995. Defining acceptable biological status by comparing with reference conditions. Pages 31 - 47. *in* Davis, W. S. and T. P. Simon, eds. Biological assessment and criteria: tools for water resource planning and decision making. Lewis Press, Boca Raton, FL.

_____., Kaufmann, P. R., 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. Aquat. Sci. 55: 1618 - 1631.

Hughes, R. M. and T. Oberdorff. 1999. Applications of IBI concepts and metrics to waters outside the United States and Canada. Pages 79 - 93. *in* Simon, T. P., ed. Assessing the sustainability and biological integrity of water resources using fish communities. CRC Press. Boca Raton, FL.

Jenkins, R. E. and N. M. Burkhead. 1993. Freshwater fishes of Virginia. Amer. Fish. Soc., Bethesda, MD.

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

_____, Fausch, K. D., Angermeier, P. L., Yant, P. R, and I. J. Schlosser. 1986. Assessing biological integrity in running water: a method and its rationale. Ill. Nat. Hist. Surv. Spec. Publ. 5.

Karr, J. R. and E. W. Chu. 1999. Restoring life in running waters. Better biological monitoring. Island Press, Washington, DC.

Kuehne, R. A. and R. W. Barbour. 1983. The American darters. Univ. Press of KY. Lexington, KY.

Lee, D. S., C. R. Gilbert, C. H. Hocutt, R. E. Jenkins, D. E. McAllister, and J. R. Stauffer, Jr. 1980, *et. seq*. Atlas of North American freshwater fishes. North Carolina State Museum of Natural History, Raleigh, NC.

LeGrand, H. E., McRae, S. E., Hall, S. P., and J. T. Finnegan. 2004. Natural Heritage Program list of the rare animal species of North Carolina. North Carolina Natural Heritage Program, Office of Conservation and Community Affairs, North Carolina Department of Environment and Natural Resources. Raleigh, NC.

Manooch, C. S, III. 1984. Fisherman's guide, Fishes of the southeastern United States. North Carolina State Museum of Natural History, Raleigh, NC.

Meador, M. R., T. F. Cuffney, and M. E. Gurtz. 1993. Methods for sampling fish communities as part of the National Water - Quality Assessment Program. U.S. Department of the Interior. U.S. Geological Survey. Water Resource Investigations Report 93 - 104. Raleigh, NC.

Menhinick, E. F. 1991. The freshwater fishes of North Carolina. North Carolina Wildlife Resources Commission. Raleigh, NC.

_____ and A. L. Braswell (eds). 1997. Endangered, threatened, and rare fauna of North Carolina. Part IV. A reevaluation of the freshwater fishes. Occas. Papers N.C. State Mus. Nat. Sci. and N.C. Biol. Surv. No. 11. Raleigh, NC.

NCAC. 2004. North Carolina administrative code. Effective August 1, 2004. Environmental Management Commission. North Carolina Department of Environment and Natural Resources. Division of Water Quality. Raleigh, NC.

NCDENR. 2003. Intensive Survey Unit standard operating procedures. North Carolina Department of Environment and Natural Resources. Division of Water Quality. Water Quality Section. Environmental Sciences Branch. Raleigh, NC. August 29, 2003.

Nelson, J. S., Crossman, E. J., Espinosa - Pérez, H., Findley, L. T., Gilbert, C. R., Lea, R. N., and J. D. Williams. 2004. Common and scientific names of fishes from the United States, Canada, and Mexico. American Fisheries Society, Special Publication 29, Bethesda, MD.

Noga, E. J. 1996. Fish disease. Diagnosis and treatment. Mosby - Year Book, Inc. St. Louis, MO.

Page, L. M. 1983. Handbook of darters. T. F. H. Publications, Inc. Neptune City, NJ.

Plafkin, J. L., M. T. Barbour, K. D. Porter, S. K. Gross, and R. M. Hughes. 1989. Rapid bioassessment protocols for use in streams and rivers: benthic macroinvertebrates and fish. Report No. EPA/444/4 - 89 - 001. U.S. EPA, Washington, DC.

Rohde, F. C., R. G. Arndt, D. G. Lindquist, and J. F. Parnell. 1994. Freshwater fishes of the Carolinas, Virginia, Maryland, and Delaware. The University of North Carolina Press. Chapel Hill, NC. 222 pp.

Sanders, R. E., R. J. Miltner, C. O. Yoder, and E. T. Rankin. 1999. The use of external deformities, erosion, lesions, and tumors (DELT anomalies) in fish assemblages for characterizing aquatic resources: a case study of seven Ohio streams. Pages 25 - 246. *in* Simon, T. P., ed. Assessing the sustainability and biological integrity of water resources using fish communities. CRC Press. Boca Raton, FL.

Saylor, C. and E. M. Scott. 1987. Application of the index of biotic integrity to existing TVA data. Tennessee Valley Authority, Norris, TN.

Simon, T. P. (ed.). 1999. Assessing the sustainability and biological integrity of water resources using fish communities. CRC Press. Boca Raton, FL.

_____. and J. Lyons. 1995. Application of the Index of Biotic Integrity to evaluate water resource integrity in freshwater ecosystems. Pages 245 - 262. *in* Davis, W. S. and T. P. Simon, eds. Biological assessment and criteria: tools for water resource planning and decision making. Lewis Press, Boca Raton, FL.

Smoger, R. A. and P. L. Angermeier. 1999. Effects of drainage basin and anthropogenic disturbance in relations between stream size and IBI metrics in Virginia. Pages 249-272. *in* Simon, T. P., ed. Assessing the sustainability and biological integrity of water resources using fish communities. CRC Press. Boca Raton, FL.

Steedman, R. J. 1991. Occurrence and environmental correlates of blackspot disease in stream fishes near Toronto, Ontario. Trans. American Fisheries Soc. 120: 494 - 499.

Walsh, S. J. and M. R. Meador. 1998. Guidelines for quality assurance and quality control of fish taxonomic data collected as part of the National Water-Quality Assessment Program. U.S. Department of the Interior. U.S. Geological Survey. Water Resource Investigations Report 98-4239. Raleigh, NC.

Warren, M. L., Jr., Burr, B. M., Walsh, S. J., Bart, H. L., Jr., Cashner, R. C., Etnier, D. A., Freeman, B. J., Kuhajda, B. R., Mayden, R. L., Robison, H. W., Ross, S. T. and W. C. Starnes. 2000. Diversity, distribution, and conservation status of the native freshwater fishes of the southern United States. Fisheries. 25: 7-29.

Yoder, C. O. and M. A. Smith. 1999. Using fish assemblages in a state biological assessment and criteria program: essential concepts and considerations. Pages 17-56. *in* Simon, T. P., ed. Assessing the sustainability and biological integrity of water resources using fish communities. CRC Press. Boca Raton, FL.

Zuboy, J. R. 1981. A new tool for fishery managers: the Delphi Technique. N. Amer. J. Fish. Manage. 1: 55-59.

Appendix 1. Stream Fish Community Assessment Program Samples Log Sheet.

NC DIVISION OF WATER QUALITY

STREAM FISH COMMUNITY ASSESSMENT PROGRAM SAMPLES LOG SHEET

YEAR _____

Sample No.	Waterbody	Location	County	Collection Date	Basin	Study

Appendix 2. Stream Fish Community Assessment Program Field Data Sheet.

NC DIVISION OF WATER QUALITY

STREAM FISH COMMUNITY ASSESSMENT PROGRAM FIELD DATA SHEET

UNIQUE SITE IDENTIFIER	SAMPLE NO.
STREAM	SAMPLE DATE
LOCATION	TIME
COUNTY	NO. OF SHOCKING UNITS
RIVER BASIN	DURATION (sec.)
SUBBASIN	SAMPLING PERSONNEL
LATITUDE	LOCATION OF REACH
LONGITUDE	SEINE USED ? (Y/N)
DRAINAGE AREA (mi.^{2})	SAMPLE IDENTIFIED BY
STREAM INDEX NO.	DATE SAMPLE IDENTIFIED
STREAM CLASSIFICATION	DATA ENTERED BY
HABITAT SCORE	DATE OF DATA ENTRY
ELEVATION (ft)	

SPECIFIC CONDUCTANCE (µmhos/cm)	AVG. STREAM WIDTH (m)
DISSOLVED OXYGEN (mg/L)	AVG. STREAM DEPTH (m)
TEMPERATURE (ºC)	WATER CLARITY (clear, turbid, blackwater)
pH	SUBSTRATE TYPE(s)
HABITAT DESCRIPTION	

Species	Total No.	Length	Length	Length	Length	Length	Length	Length	Length	Length

Appendix 2 (continued).

PAGE ___ OF ___

NC DIVISION OF WATER QUALITY

STREAM FISH COMMUNITY ASSESSMENT PROGRAM FIELD DATA SHEET

STREAM	SAMPLE NO.
SAMPLE LOCATION	SAMPLE DATE

Species	Total No.	Length	Length	Length	Length	Length	Length	Length	Length	Length

Appendix 3. Example of a completed Stream Fish Community Assessment Program Field Data Sheet. Note: this data sheet was the version used between 1996 and 2005.

BAT 08/1/05

PAGE 1 OF 2

H S = 97
C
9-29-43 Elev. = 840'

FISH COMMUNITY ASSESSMENT-IBI DATA SHEET

STREAM	Britten Creek	SAMPLE NO.	2005-81
SAMPLE LOCATION	NC 9	SAMPLE DATE	23 June 2005
COUNTY	Polk	TIME	1310 - 1445
RIVER BASIN	BRD	NO. OF SHOCKING UNITS	2
SUBBASIN	02	DURATION (sec.)	3567 + 3637 = 7204
LATITUDE	35 20 30	SAMPLING PERSONNEL	Arnett (NCSU), Crouch, DeBerardinis, Tracy
LONGITUDE	82 10 55		
DRAINAGE AREA (mi.²)	6.8		

Sampled 600 ft upstream beginning @ bridge (concrete arch)
- Beautiful stream w/ appearance
- just upstream from confluence w/ Green River (400 yds) 20' seine used

PHYSICAL DATA

SP. CONDUCTANCE (µS/cm)	35	AVG. STREAM WIDTH (m)	8 m
DISSOLVED OXYGEN (mg/L)	7.3	AVG. STREAM DEPTH (m)	0.2
TEMPERATURE (°C)	21.3	WATER CLARITY (clear, cloudy, turbid, blackwater)	
pH	6.4	SUBSTRATE TYPE (s)	Boulder / cobble / sand / gravel

HABITAT DESCRIPTION High gradient riffles / plunge pools / runs / large pool in lower end of segment
Small stream w/ abundant [illegible] ; not many [illegible] shiners - [illegible] gradient ?
- Spotfail Shiner

SPECIES COLLECTED

	Species	Total No.	Length	Length	Length	Length	Length	Length	Length	Length	Length	Length
Y-	Rosyhead chub	59	134	75	153	75	121	105	60	56	70	78
	" (+9)		100	83	68	74	80	74	60	103	72	62
	" (+8 @ ~25 cm)		60	100	70	85	108	104	88	87	93	74
	"		102	134	87	77	102	104	113	108	95	90
	"		68	58	53	68	69	70	67	78	74	73
Y-	Redbreast SF	36	180	123	87	116	124	110	92	149	129	129
	"		75	128	137	110	112	107	97	90	112	98
	"		80	80	120	93	123	118	108	72	90	112
	"		87	77	63	83	81	121				
Y-	Swallow shiner	41	72	72	84	71	73	70	78	60	82	78
	"		69	68	67	68	85	90	77	87	72	75
	"		70	88	64	86	84	71	68	68	73	68
	"		71	71	73	65	63	70	68	68	66	66
	"		63									
1 YOY Y-	Marginal Pet	28	78	119	105	107	105	116	112	127	112	110
	"		118	87	65	123	115	124	67	95	92	70
	"		105	63	120	130	116	110	62	75		
Y-	Bluegill	10	58	65	60	65	68	93	58	67	70	57
N-	N H Sucker	3	258	243	218							
Y-	Striped jumpkin	7	93	230	118	93	119	95	84			
Y-	Fairy/black shiner	97	88	97	89	64	83	78	71	88	79	83
	" (+47)		92	86	90	81	63	69	69	75	68	68
	"		58	74	59	59	74	94	64	62	76	70
	"		78	68	73	80	62	77	70	68	63	60
	"		65	58	67	78	80	75	78	89	49	64
Y-	Flat bullhead	13	153	95	105	154	163	160	117	138	112	98

Species — 45% E+OM for W.C. stories -

Flat bullhead - 3
Seagreen darter - 5
Fairy/black SH - 5

Redbreast darter - 5
Swallow SH - 6
Spotfail SH - 5

over →

Appendix 3 (continued).

PAGE 2 OF 2

FISH COMMUNITY ASSESSMENT-IBI DATA SHEET

STREAM	Brillow Creek	SAMPLE NO.	2006-81
SAMPLE LOCATION	NC9	SAMPLE DATE	23 June 2005

SPECIES COLLECTED

	Species	Total No.	Length	Length	Length	Length	Length	Length	Length	Length	Length	Length
N-	Redeye dace	6	65	62	63	60	60	59				
Y-	Spotfin shiner	68	74	75	108	125	105	83	78	84	78	80
	" (+8)		80	85	108	82	80	79	80	72	84	80
	"		74	111	69	114	100	87	96	94	89	84
	"		83	83	74	74	77	78	78	74	82	81
w/ white bar	Bluntnose shiner	6	54	49	46	42	42	41				
N-	Northern creek chub	1	68									
—	LMB	—	47mm	60mm	50mm	(4 yoy @ 41-50mm)						
Y-	Sharpnose dace	30	72	71	70	61	61	62	72	48	52	55
	"		55	48	67	65	70	40	70	66	66	70
	"		67	64	58	61	60	54	55	53	53	45
↑	Fish bulkhead		72	87	100							
↑	N. hudsonius		78	80	78	82	78	75	78	78	77	75
	"		72	75	73	75	73	75	73	77	74	75
N-	Etheostoma simoterum	1	59	(+6 yoy @ ~25mm)								
—	Creek chub	—	(yoy @ 28mm)									
	11/15 sp =	73%										

Appendix 4. Field meter calibration sheet.

Water Quality Monitoring Field Meter Calibration Sheet

Collector(s): _____

Study: _____

Sampling Location: _____

Meter(s) Make/Model: _____ / _____

Meter(s) Serial #/ ID: _____ / _____

	Date yy/mm/dd	Time 24hr hh:nn	Initials
Pre-Sampling Calibration			
Post Sampling Meter Check			

Miscellaneous (Does not apply to YSI 85 or Accumet 61 meters

	Battery Level (V)	Stirrer Working
Pre-Sampling Calibration		Y / N
Post Sampling Meter Check		Y / N

Battery Ranges - Surveyor: internal- 7.2-7.5V, external- 11-13V; Quanta: 4.0-4.5V

Dissolved Oxygen (mg/L)

	Temp. °C	% Saturation	DO Table Value	Barometric Pressure mmHg[1]	Altitude ft.	Corrected DO from Table[2]	Initial Meter Reading	Calibrated Meter Reading
Pre-Sampling Calibration								
Post Sampling Meter Check								
Within ± 0.5							Y / N	

[1]B.P. = 760 -2.5 (A/100); A = local altitude above sea level in feet

[2]On DO Table move one block right for every 55 ft. of altitude above sea level

Specific Conductance (µS/cm* at 25°C)

	Standard 1 147 / 718 / 24820/_____		Standard 2 147 / 718 / 24820/_____	
	Initial Meter Reading	Calibrated Meter Reading[3]	Initial Meter Reading	Calibrated Meter Reading[3]
Pre-Sampling Calibration				
Post Sampling Meter Check				
Within ± 10%	Y / N		Y / N	

10% ranges: 147 = 132-162; 718 = 646-790; 24820 = 22338-27302

*Quanta reads in mS/cm; move decimal 3 places right for µS/cr

[3]Does not apply to YSI 85

pH (SU)

	Buffer #1- 7.0		Buffer #2- 4.0 / 10.0			Check Buffer - 7.0
	Initial Meter Reading	Calibrated Meter Reading	Initial Meter Reading	Calibrated Meter Reading	Slope Efficiency[4]	Meter Reading
Pre-Sampling Calibration						
Within ± 0.1						Y / N
Post Sampling Meter Check						
Within ± 0.2	Y / N		Y / N			

[4]Does not apply to Hydrolab or Quanta

Comments: _____

FMC-20060616

Keep original on file for 5 years

Appendix 5. Habitat assessment field data sheet -- Mountain/Piedmont streams.

3//06 Revision 6

Habitat Assessment Field Data Sheet
Mountain/Piedmont Streams

Biological Assessment Unit, DWQ **TOTAL SCORE**

Directions for use: The observer is to survey a **minimum of 200 meters** of stream, preferably in an **upstream** direction starting above the bridge pool and the road right-of-way. The segment which is assessed should represent average stream conditions. To perform a proper habitat evaluation the observer needs to get into the stream. To complete the form, select the description which best fits the observed habitats and then circle the score. If the observed habitat falls in between two descriptions, select an intermediate score. A final habitat score is determined by adding the results from the different metrics.

Stream_____Location/road:_____(Road Name_____)County_____
Date_____**CC#**_____Basin_____Subbasin_____
Observer(s)____ Type of Study: ☐ Fish ☐ Benthos ☐ Basinwide ☐ Special Study (Describe) _____
Latitude _____Longitude _____Ecoregion: ☐ MT ☐ P ☐ Slate Belt ☐ Triassic Basin
Water Quality: Temperature_____^{0}C DO _____mg/l Conductivity (corr.) ____µmhos/cm pH ____

Physical Characterization: Visible land use refers to immediate area that you can see from sampling location - include what you estimate driving thru the watershed in watershed land use.

Visible Land Use: _____%Forest _____%Residential _____%Active Pasture _____
% Active Crops _____%Fallow Fields _____% Commercial _____%Industrial _____%
Other - Describe:___

Watershed land use : ☐ Forest ☐ Agriculture ☐ Urban ☐ Animal operations upstream

Width: (meters) Stream_____ Channel (at top of bank)_____ **Stream Depth**: (m) Avg__Max __
☐ Width variable ☐ Large river >25m wide
Bank Height (from deepest part of riffle to top of bank-first flat surface you stand on) (m)_____
Bank Angle: _____º or ☐ NA (Vertical is 90º, horizontal is 0º. Angles > 90º indicate slope is towards mid-channel, < 90º indicate slope is away from channel. NA if bank is too low for bank angle to matter.)

☐ Channelized Ditch
☐ Both banks undercut at bend
☐ Recent overbank deposits
☐ Buried structures
☐ Excessive periphyton growth
☐ Green tinge
☐ Deeply incised-steep, straight banks
☐ Channel filled in with sediment
☐ Bar development
☐ Exposed bedrock
☐ Heavy filamentous algae growth
☐ Sewage smell

Manmade Stabilization: ☐ N ☐ Y: ☐ Rip-rap, cement, gabions ☐ Sediment/grade-control structure ☐Berm/levee
Flow conditions : ☐ High ☐ Normal ☐ Low
Turbidity: ☐ Clear ☐ Slightly Turbid ☐ Turbid ☐ Tannic ☐ Milky ☐ Colored (from dyes)
Good potential for Wetlands Restoration Project? ☐ YES ☐ NO Details_____
Channel Flow Status

Useful especially under abnormal or low flow conditions.

A. Water reaches base of both lower banks, minimal channel substrate exposed ☐
B. Water fills >75% of available channel, or <25% of channel substrate is exposed ☐
C. Water fills 25-75% of available channel, many logs/snags exposed ☐
D. Root mats out of water ☐
E. Very little water in channel, mostly present as standing pools ☐

Weather Conditions:_____**Photos:** ☐ N ☐ Y ☐ Digital ☐ 35mm
Remarks:_____

Appendix 5 (continued).

I. Channel Modification Score

A. channel natural, frequent bends..... 5
B. channel natural, infrequent bends (channelization could be old)..... 4
C. some channelization present..... 3
D. more extensive channelization, >40% of stream disrupted..... 2
E. no bends, completely channelized or rip rapped or gabioned, etc..... 0

☐ Evidence of dredging ☐ Evidence of desnagging=no large woody debris in stream
☐ Banks of uniform shape/height Remarks_____ Subtotal____

II. Instream Habitat: Consider the percentage of the reach that is favorable for benthos colonization or fish cover. If >70% of the reach is rocks, 1 type is present, circle the score of 17. Definition: leafpacks consist of older leaves that are packed together and have begun to decay (not piles of leaves in pool areas). Mark as **R**are, **C**ommon, or **A**bundant.

___**Rocks**__**Macrophytes**__**Sticks and leafpacks**__**Snags and logs**___**Undercut banks or root mats**

	Amount Of Reach Favorable For Colonization Or Cover			
	>70% Score	40-70% Score	20-40% Score	<20% Score
4 or 5 types present.....	20	16	12	8
3 types present.....	19	15	11	7
2 types present.....	18	14	10	6
1 type present.....	17	13	9	5
No types present.....	0			

☐ No woody vegetation in riparian zone. Remarks_____ Subtotal_____

III. Bottom Substrate (silt, sand, detritus, gravel, cobble, boulder) Look at entire reach for substrate scoring, but only look at riffle for embeddedness, and use rocks from all parts of riffle-look for "mud line" or difficulty extracting rocks.

Score

A. substrate with good mix of gravel cobble and boulders
1. embeddedness <20% (very little sand, usually only behind large boulders)..... 15
2. embeddedness 20-40%..... 12
3. embeddedness 40-80%..... 8
4. embeddedness >80%..... 3

B. substrate gravel and cobble
1. embeddedness 20%..... 14
2. embeddedness 20-40%..... 11
3. embeddedness 40-80% 6
4. embeddedness >80%..... 2

C. substrate mostly gravel
1. embeddedness <50%..... 8
2. embeddedness >50%..... 4

D. substrate homogeneous
1. substrate nearly all bedrock..... 3
2. substrate nearly all sand 3

E. substrate nearly all detritus..... 2
F. substrate nearly all silt/ clay..... 1

Remarks_____Total____

IV. Pool Variety Pools are areas of deeper than average maximum depths with little or no surface turbulence. Water velocities associated with pools are always slow. Pools may take the form of "pocket water", small pools behind boulders or obstructions in large high gradient streams, or side eddies.

A. Pools present Score

1. Pools Frequent (>30% of 100m area surveyed)
   a. variety of pool sizes..... 10
   b. pools same size (indicates pools filling in)..... 8
2. Pools Infrequent (<30% of the 100m area surveyed)
   a. variety of pool sizes..... 6
   b. pools same size..... 4

B. Pools absent..... 0

Remarks_____Total____

☐ Pool bottom boulder-cobble=hard ☐ Sandy-sink as you walk ☐ Silt ☐ Some pools over wader depth
Remarks_____Page Total_____

Appendix 5 (continued).

V. Riffle Habitats

Definition: Riffle is area of reaeration-can be debris dam, or narrow channel area.

	Frequent Score	Infrequent Score
A. well defined riffle and run, riffle as wide as stream and extends 2X width of stream....	16	12
B. riffle as wide as stream but riffle length is not 2X stream width	14	7
C. riffle not as wide as stream and riffle length is not 2X stream width	10	3
D. riffles absent......	0	

Channel Slope: ☐ Typical for area ☐Steep=fast flow ☐ Low=like a coastal stream Total_____

VI. Bank Stability and Vegetation (FACE UPSTREAM)

	Left Score	Right Score
A. Banks stable		
1. no evidence of erosion or bank failure(except outside of bends), little potential for erosion....	7	7
B. Erosion areas present		
1. diverse **trees**, shrubs, grass; plants healthy with good root systems.....	6	6
2. few trees or small trees and **shrubs**; vegetation appears generally healthy.....	5	5
3. parse **mixed** vegetation; plant types and conditions suggest poorer soil binding.....	3	3
4. mostly **grasses**, few if any trees and shrubs, high erosion and failure potential at high flow..	2	2
5. no bank vegetation, mass erosion and bank failure evident.....	0	0

Remarks_____Total____

VII. Light Penetration Canopy is defined as tree or vegetative cover directly above the stream's surface. Canopy would block out sunlight when the sun is directly overhead. Note shading from mountain, but not use to score this metric.

	Score
A. Stream with **good** shading with some breaks for light penetration	10
B. Stream with **full canopy** - breaks for light penetration absent.....	8
C. Stream with **partial** shading - sunlight and shading are essentially equal.....	7
D. Stream with **minimal** shading - full sun in all but a few areas.....	2
E**. No shading**.....	0

Remarks_____Total____

VIII. Riparian Vegetative Zone Width

Definition: Riparian zone for this form is area of natural vegetation adjacent to stream (can go beyond floodplain).
Definition: A break in the riparian zone is any place on the stream banks which allows sediment or pollutants to directly enter the stream, such as paths down to stream, storm drains, uprooted trees, otter slides, etc.
Dominant vegetation: ☐ Trees ☐ Shrubs ☐ Grasses ☐ Weeds/old field ☐ Exotics (kudzu, etc.)

FACE UPSTREAM

	Left Bank (score)	Right Bank (score)
A. Riparian zone **intact** (no breaks)		
1. width > 18 meters.....	5	5
2. width 12-18 meters.....	4	4
3. width 6-12 meters.....	3	3
4. width < 6 meters.....	2	2
B. Riparian zone **not intact** (breaks)		
1. breaks rare		
a. width > 18 meters.....	4	4
b. width 12-18 meters.....	3	3
c. width 6-12 meters.....	2	2
d. width < 6 meters.....	1	1
2. breaks common		
a. width > 18 meters.....	3	3
b. width 12-18 meters.....	2	2
c. width 6-12 meters.....	1	1
d. width < 6 meters.....	0	0

Remarks_____Total____

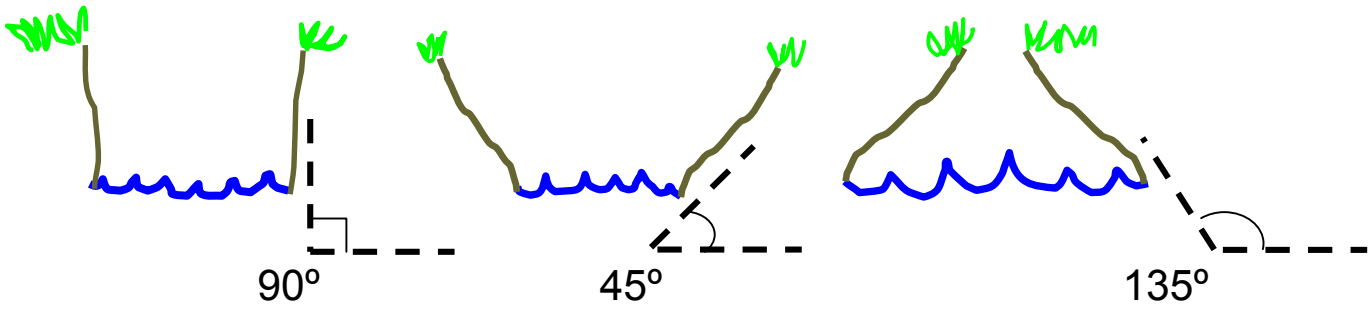
Page Total_____

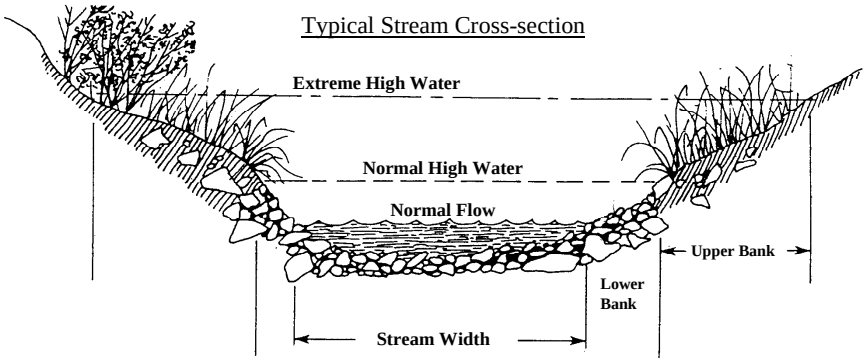
☐ Disclaimer-form filled out, but score doesn't match subjective opinion-atypical stream. **TOTAL SCORE**_____

Appendix 5 (continued).

Supplement for Habitat Assessment Field Data Sheet

Diagram to determine bank angle





This side is 45º bank angle.

Site Sketch:

Other comments:_____

3/01 Revision 6

Habitat Assessment Field Data Sheet
Coastal Plain Streams

Biological Assessment Unit, DWQ **TOTAL SCORE**

Directions for use: The observer is to survey a **minimum of 100 meters** with 200 meters preferred of stream, preferably in an **upstream** direction starting above the bridge pool and the road right-of-way. The segment which is assessed should represent average stream conditions. To perform a proper habitat evaluation the observer needs to get into the stream. To complete the form, select the description which best fits the observed habitats and then circle the score. If the observed habitat falls in between two descriptions, select an intermediate score. A final habitat score is determined by adding the results from the metrics.

Stream_____Location/road:_____(Road Name_____)County_____
Date_____**CC#**_____Basin_____Subbasin_____
Observer(s)____ Type of Study: ☐ Fish ☐ Benthos ☐ Basinwide ☐ Special Study (Describe) _____
Latitude _____Longitude _____Ecoregion: ☐ CA ☐ SWP ☐ Sandhills ☐ CB
Water Quality: Temperature_____^{0}C DO _____mg/l Conductivity (corr.) ____µmhos/cm pH ____

Physical Characterization: Visible land use refers to immediate area that you can see from sampling location - include what you estimate driving thru the watershed in watershed land use.

Visible Land Use: _____%Forest _____%Residential _____%Active Pasture _____
% Active Crops _____%Fallow Fields _____% Commercial _____%Industrial _____%
Other - Describe:___

Watershed land use : ☐ Forest ☐ Agriculture ☐ Urban ☐ Animal operations upstream

Width: (meters) Stream_____ Channel (at top of bank)_____ **Stream Depth**: (m) Avg__Max __
☐ Width variable ☐ Large river >25m wide
Bank Height (from deepest part of channel (in riffle or run) to top of bank): (m)_____
Bank Angle: _____º or ☐ NA (Vertical is 90º, horizontal is 0º. Angles > 90º indicate slope is towards mid-channel, < 90º indicate slope is away from channel. NA if bank is too low for bank angle to matter.)

☐ Channelized Ditch
☐ Both banks undercut at bend
☐ Recent overbank deposits
☐ Buried structures
☐ Excessive periphyton growth
☐ Green tinge
☐ Deeply incised-steep, straight banks
☐ Channel filled in with sediment
☐ Bar development
☐ Exposed bedrock
☐ Heavy filamentous algae growth
☐ Sewage smell

Manmade Stabilization: ☐ N ☐ Y: ☐ Rip-rap, cement, gabions ☐ Sediment/grade-control structure ☐Berm/levee
Flow conditions : ☐ High ☐ Normal ☐ Low
Turbidity: ☐ Clear ☐ Slightly Turbid ☐ Turbid ☐ Tannic ☐ Milky ☐ Colored (from dyes)
Good potential for Wetlands Restoration Project? ☐ YES ☐ NO Details_____
Channel Flow Status

Useful especially under abnormal or low flow conditions.

A. Water reaches base of both lower banks, minimal channel substrate exposed ☐
B. Water fills >75% of available channel, or <25% of channel substrate is exposed ☐
C. Water fills 25-75% of available channel, many logs/snags exposed ☐
D. Root mats out of water ☐
E. Very little water in channel, mostly present as standing pools ☐

Weather Conditions:_____**Photos:** ☐ N ☐ Y ☐ Digital ☐ 35mm
Remarks:_____

Appendix 6 (continued).

I. Channel Modification

	Score
A. Natural channel-minimal dredging.....	15
B. Some channelization near bridge, or historic (>20 year old), and/or bends beginning to reappear	10
C. Extensive channelization, straight as far as can see, channelized ditch.....	5
D. Banks shored with hard structure, >80% of reach disrupted, instream habitat gone.....	0

Remarks_____Total____

II. Instream Habitat: Consider the percentage of the reach that is favorable for benthos colonization or fish cover. If >50% of the reach is snags, and 1 type is present, circle the score of 16. Definition: leafpacks consist of older leaves that are packed together and have begun to decay (not piles of leaves in pool areas). Mark as **R**are, **C**ommon, or **A**bundant.

____**Sticks**____**Snags/logs**____**Undercut banks or root mats**____**Macrophytes**____**Leafpacks**

AMOUNT OF REACH FAVORABLE FOR COLONIZATION OR COVER

	>50% Score	30-50% Score	10-30% Score	<10% Score
4 or 5 types present.....	20	15	10	5
3 types present.....	18	13	8	4
2 types present.....	17	12	7	3
1 type present.....	16	11	6	2

No substrate for benthos colonization and no fish cover.....0

☐ No woody vegetation in riparian zone

Remarks_____Total____

III. Bottom Substrate (silt, clay, sand, detritus, gravel) look at entire reach for substrate scoring.

	Score
A. Substrate types mixed	
1. gravel dominant.....	15
2. sand dominant.....	13
3. detritus dominant.....	7
4. silt/clay/muck dominant.....	4
B. Substrate homogeneous	
1. nearly all gravel.....	12
2. nearly all sand	7
3. nearly all detritus.....	4
4. nearly all silt/clay/muck.....	1

Remarks_____Total____

IV. Pool Variety Pools are areas of deeper than average maximum depths with little or no surface turbulence. Water velocities associated with pools are always slow.

	Score
A. Pools present	
1. Pools Frequent (>30% of 100m length surveyed)	
a. variety of pool sizes.....	10
b. pools same size (indicates pools filling in).....	8
2. Pools Infrequent (<30% of the 100m length surveyed)	
a. variety of pool sizes.....	6
b. pools same size.....	4
B. Pools absent	
1. Deep water/run habitat present.....	4
2. Deep water/run habitat absent.....	0

Remarks_____Total____

Page Total_____

Appendix 6 (continued).

V. Bank Stability and Vegetation

FACE UPSTREAM

	Left Bank Score	Right Bank Score
A. Banks stable or no banks, just flood plain		
1. no evidence of erosion or bank failure(except outside of bends), little potential for erosion....	7	7
B. Erosion areas present		
1. diverse **trees**, shrubs, grass; plants healthy with good root systems.....	6	6
2. few trees or small trees and **shrubs**; vegetation appears generally healthy.....	5	5
3. parse **mixed** vegetation; plant types and conditions suggest poorer soil binding.....	3	3
4. mostly **grasses**, few if any trees and shrubs, high erosion and failure potential at high flow..	2	2
5. no bank vegetation, mass erosion and bank failure evident.....	0	0

Remarks_____Total____

VI. Light Penetration (Canopy is defined as tree or vegetative cover directly above the stream's surface. Canopy would block out sunlight when the sun is directly overhead).

	Score
A. Stream with **good** shading with some breaks for light penetration	10
B. Stream with **full canopy** - breaks for light penetration absent.....	8
C. Stream with **partial** shading - sunlight and shading are essentially equal.....	7
D. Stream with **minimal** shading - full sun in all but a few areas.....	2
E**. No shading**.....	0

Remarks_____Total____

VII. Riparian Vegetative Zone Width

Definition: A break in the riparian zone is any area which allows sediment to enter the stream. Breaks refer to the near-stream portion of the riparian zone (banks); places where pollutants can directly enter the stream.

	Left Bank Score	Right Bank Score
A. Riparian zone **intact** (no breaks)		
1. width > 18 meters.....	5	5
2. width 12-18 meters.....	4	4
3. width 6-12 meters.....	3	3
4. width < 6 meters.....	2	2
B. Riparian zone **not intact** (breaks)		
1. breaks rare		
a. width > 18 meters.....	4	4
b. width 12-18 meters.....	3	3
c. width 6-12 meters.....	2	2
d. width < 6 meters.....	1	1
2. breaks common		
a. width > 18 meters.....	3	3
b. width 12-18 meters.....	2	2
c. width 6-12 meters.....	1	1
d. width < 6 meters.....	0	0

Remarks_____Total____

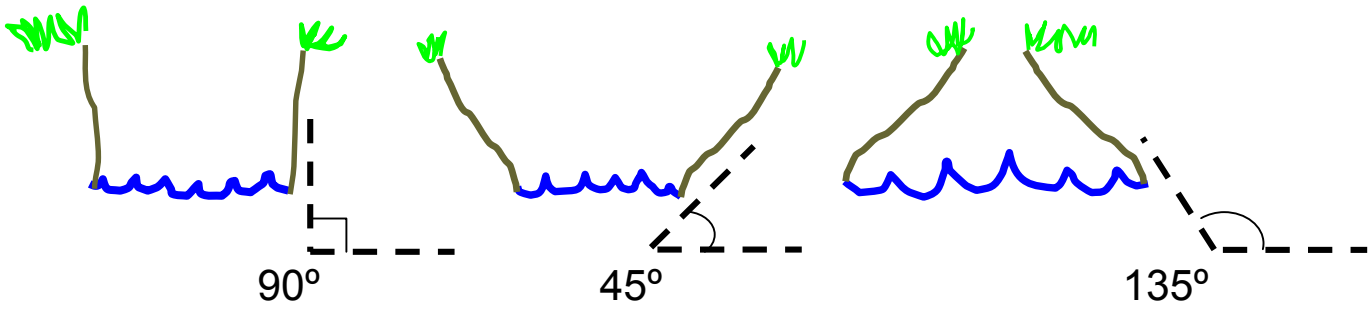
Page Total_____

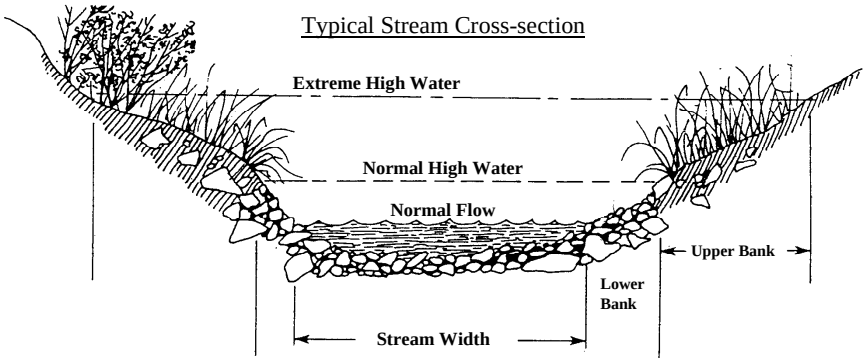
☐ Disclaimer-form filled out, but score doesn't match subjective opinion-atypical stream. **TOTAL SCORE**_____

Appendix 6 (continued).

Supplement for Habitat Assessment Field Data Sheet

Diagram to determine bank angle





This side is 45º bank angle.

Site Sketch:

Other comments:_____

Appendix 7. Stream Fish Community Assessment Program Data Entry Log Sheet.

NC DIVISION OF WATER QUALITY

STREAM FISH COMMUNITY ASSESSMENT PROGRAM DATA ENTRY LOG SHEET

YEAR ____

Sample No.	Waterbody	Date Identified	Date Data Entered	Date Data Checked	Date Data "Clean"

Appendix 8. Example of a North Carolina Fish Community Report.

2/12/2005 11:57:30 AM

North Carolina Fish Community Report

Collection No.	2005-81
Data Entry	BT
Date	6/23/2005

Waterbody	Britten Cr	No. Secs.	7204	
Station	NC 9	No. Units	2	
County	Polk	CPUE	5.6	
Latitude	352030	Width	8	m
Longitude	821055	Depth:	0.2	m
Ecoregion	P	Drainage	6.8	mi2
Basin	BRD	Elevation	840	ft
Subbasin	2			

- [x] Reference Site
- [x] Basin Site
- [ ] Special Study

Stream Index No.	9-29-43
Stream Classification	C
	True

Water Quality and Habitat

Temperature	21.3	°C
Specific Conductance	35	µS/cm
Dissolved Oxygen	7.3	mg/L
pH	6.4	s.u.
Clarity	Clear	
Total Habitat Score	97	

NCIBI Score 54 **NCIBI Rating** Excellent

Family	Scientific Name	Trophic Status	Tolerance	Multiple Age	No.	Exotics
Catostomidae	Hypentelium nigricans	Insectivore	Intermediate	No	3	☐
Catostomidae	Scartomyzon rupiscartes	Insectivore	Intermediate	Yes	7	☐
Centrarchidae	Lepomis auritus	Insectivore	Tolerant	Yes	36	☐
Centrarchidae	Lepomis macrochirus	Insectivore	Intermediate	Yes	10	☐
Cyprinidae	Cyprinella pyrrhomelas	Insectivore	Intolerant	Yes	97	☐
Cyprinidae	Nocomis leptocephalus	Omnivore	Intermediate	Yes	59	☐
Cyprinidae	Notemigonus crysoleucas	Omnivore	Tolerant	No	1	☐
Cyprinidae	Notropis hudsonius	Omnivore	Intermediate	Yes	68	☐
Cyprinidae	Notropis scepticus	Insectivore	Intermediate	Yes	41	☐
Cyprinidae	Notropis sp. cf. chlorocephalus	Insectivore	Intermediate	Yes	6	☐
Ictaluridae	Ameiurus platycephalus	Insectivore	Tolerant	Yes	13	☐
Ictaluridae	Noturus insignis	Insectivore	Intermediate	Yes	28	☐
Percidae	Etheostoma olmstedi	Insectivore	Intermediate	No	1	☐
Percidae	Etheostoma thalassinum	Insectivore	Intolerant	Yes	30	☐
Percidae	Percina crassa	Insectivore	Intolerant	No	6	☐

NCIBI Metrics

Metric	Value	Score
Number of Species:	15	5
Number Fish:	406	5
Number Darter Sp:	3	5
No. Sunfish,Bass,Trout:	2	3
Number Sucker Sp:	2	5
Number Intolerant Sp.	3	5
Percent Tolerant Fish:	12	5
Percent Omni + Herb:	32	5
Percent Insectivores:	68	5
Percent Piscivores:	0.00	1
Percent Diseased Fish:	0.00	5
Percent Sp Multiple Ages:	73	5

Exotics

Number Exotic Fish	0
Number Exotic Species	0

Notes

Collectors = Averett (NCSU), Crouch, DeBerardinis, Tracy. Substrate = boulder, cobble, sand, gravel. Sampled 600 ft. above concrete arch bridge, beginning at bridge. No seine used. High gradient riffles, plunge pools, runs, some Podostemum, large pool in lower end of segment. Small stream with abundant Flat Bullhead, Spottail Shiner; not many Piedmont Shiners collected -- too high gradient? Piedmont Shiner with white fins. Large Striped Jumprock. Y-O-Y = Margined Madtom, Largemouth Bass, Creek Chub; Largemouth Bass and Creek Chub represented only by Y-O-Y. Beautiful stream and riparian areas. Just upstream (~ 400 yards) of its confluence with the Green River. Located in the Southern Inner Piedmont Level IV Ecoregion. New regional reference site.

Appendix 9. Web Links

Digital Pictures of Fish – EFISH, the Virtual Aquarium, the Department of Fisheries and Wildlife Sciences, Virginia Polytechnic Institute and State University -- http://www.cnr.vt.edu/efish/

NCDWQ Basinwide Assessment Reports -- http://www.esb.enr.state.nc.us/bar.html

NCDWQ Basinwide Planning -- http://h2o.enr.state.nc.us/basinwide/

NCDWQ Biological Assessment Unit -- http://www.esb.enr.state.nc.us/BAU.html

NCDWQ Intensive Survey Unit Standard Operating Procedure -- http://www.esb.enr.state.nc.us/ISUwww/isgsop.pdf

NCDWQ Stream Fish Community Assessment Program Raw Data -- http://www.esb.enr.state.nc.us/NCIBI.htm

NCDWQ Stream Fish Community Assessment Program NCIBI Scores and Ratings -- http://www.esb.enr.state.nc.us/IBIrate.htm

NCDWQ Water Quality Standards -- http://h2o.enr.state.nc.us/admin/rules/documents/rb080104.pdf

NC Division of Water Quality -- http://h2o.enr.state.nc.us/contact.html

Native and Exotic Freshwater Fish in North Carolina -- http://www.esb.enr.state.nc.us/Native and Introduced Freshwater Fish in North Carolina.2-1.htm