SAVEM—SAMAB's Southern Appalachian Volunteer Monitoring Program

Index of Biotic Integrity (IBI) Monitoring Protocol

Following are suggested fish biomonitoring methods for wadable streams anywhere along the Appalachian chain. This will include most streams draining watershed areas of up to 70 sq. mi., and sometimes more. Large streams with extensive unwadable areas will normally require the use of an electrofishing boat, which will be beyond the means of most groups. Such streams will normally be monitored only by, or with the participation of, state or federal agencies.

Experience suggests that, at least under certain conditions, monitoring of the fish assemblage should be supplemented or even replaced by standard benthic macroinvertebrate monitoring methods. In the upper Little Tennessee River watershed of North Carolina and Georgia, the conditions are as follows:

- Watershed drainage area less then 4 sq. mi.
- Average gradient over 100 ft./mi.
- Elevation above 2,800 ft.
- Site located upstream of natural barrier to fish movement.

The key factor here is to segregate those sites where natural fish diversity is too low to permit application of an Index of Biotic Integrity based solely on fish. The specifics of watershed area, gradient and elevation will vary from region to region.

Monitoring of benthic macroinvertebrates will always be a useful supplement to fish monitoring. Where time and cost permit, both types of monitoring should be employed, especially in areas where prior monitoring data is scarce or non-existent. Since benthic macroinvertebrate monitoring methodologies are well developed and largely standardized, they will not be described here.

The suggestions which follow should be helpful in designing a stream fish monitoring program in most places. If an Index of Biotic Integrity (IBI) or similar criteria have not been established for your area, be advised that development of criteria may involve hundreds of samples. However, this advice is not intended to discourage sampling. If sampling is done properly, biomonitoring application of data may be made many years into the future. As a general rule, if you have the opportunity to get baseline data, do so. One of the greatest weaknesses of most biomonitoring programs is the absence of historical data.

<u>Equipment</u>:

All that follows presumes the availability of a minimal amount of equipment, including:

- At least one, and preferably two, backpack electrofishers.
- One or more seines with 1/8 to ¼ inch mesh. The most useful length will be 20-25 ft., depth can be 4-6 ft. For smaller streams a shorter seine, say 10 ft. long, will be useful.
- Several dipnets with 4-6 ft. handles, same mesh size.
- Buckets
- Aquarium nets for dipping fish out of buckets.

Personnel:

It is essential to have at least one person on the crew who is capable of sight identification of all expected fish species (and of recognizing an unexpected species when it appears). This does not necessarily imply a professional ichthyologist with a doctorate (in fact, some such individuals are not capable of in-the-hand field IDs). In most watersheds (remember we are not in the Amazon Basin), biomonitoring surveys may be directed by persons who are best described as "parataxonomists," whose knowledge of fishes is largely circumscribed by their home watersheds.

The requisite experience may be acquired in one season through fieldwork with a professional, supplemented by access to texts, taxonomic keys, and occasional access to professional help and perhaps museum specimens. (If the day-to-day services of a professional ichthyologist are not available, it is a good idea to set up a reference collection of local fish.)

The indispensable characteristic of a crew leader is not technical knowledge of fishes, but a "feel" for the stream environment. Some (not all) aquatic biologists have this, as do many sport anglers and some aquarium hobbyists, canoeists or others who hang around streams for their own reasons. If a person is highly motivated to direct a stream biomonitoring project in the field, chances are that this individual has the requisite "feel."

While efficiency of sampling will increase in proportion to the number of professionals on the crew, the rest of the crew may be made up entirely of volunteers with no experience in biomonitoring. A modicum of physical ability is required; the need for such ability increases with the size, gradient, and other characteristics of the stream. The safety and comfort level of volunteers are paramount considerations here. Most crew leaders learn good judgement in these matters without incurring fatalities in field crews.

When sampling streams at the upper limit of feasibility in terms of size and gradient, make sure that your crew includes at least three individuals of reasonable height and weight, with good motor skills and "water legs." (Most canoeists, anglers or backcountry hikers will have water legs.)

In a pinch, a two-person crew may sample very small streams. A third individual will nearly always prove to be an asset, and more are better up to some limit. On large or very diverse sites, having a few extra crew members may facilitate some individuals taking breaks during the work day. The need for crew members increases with stream size (also with other factors, such as number of fish expected), up to nine individuals may profitably employed in some situations. Larger crews may be employed where one of the objectives is demonstration or education, but no increase in efficiency should be expected. And remember, every one of those bodies has to be somewhere at every moment. Especially on small streams a too large crew can be a disadvantage.

Site selection:

Select monitoring sites with a clear purpose in mind. If the idea is to assess the contribution of a particular tributary stream to a watershed, select a site 0.25 mi. (smallest streams) to 0.5 mi. upstream of the mouth. If the idea is to assess the effect of a particular tributary, pollution source, land use change, restoration effort, or natural change in the stream it may be useful to select two paired sites, one each above and below the point of impact. Be sure to select sites far enough upstream and downstream of the presumed change to allow for mixing of waters (in the case of downstream sites) and to reduce the probability of "casual visits" by fish not actually resident in the reach to be monitored. Be sure that the sample does not bracket any major tributaries.

Once the monitoring reach is selected, walk the stream for a good distance above and below the logical access point. Two miles is not too much to walk if there are no major tributaries, point sources, impoundments, etc. From this reach, try to pick a "typical" section for your sample. This should be a section that reflects the proportion of conditions in the sample reach as a whole , e.g. depth, substrate composition, proportion of habitat types, riparian zone characteristics, etc. For example, if a third of the reach flows through forested land, and two thirds through open pasture with no shade, then roughly two thirds of the sample should be laid out in unshaded areas.

If there are any special habitats, try to include them in the sample, even though they represent a very small percentage of the total habitat. This is likely to increase your species count. A common example is muddy backwaters, partially isolated from the main flow. Other examples might be an unusually powerful riffle, cascade area or torrent or side channels around islands or bars. (Note that side channels may have different species, especially those more characteristic of smaller streams, even if the physical habitat does not differ in any significant way from other parts of the stream.)

It is especially important to have full representation of the 3 major habitat types—riffle, run, and pool. Unless one of these habitats is absent or very scarce, you should try to do the equivalent of three standard subsamples (300 sq. ft. of water surface) in each of these habitat types. To achieve the best sample, it will be helpful to learn how to distinguish variations within habitat types (i.e. shallow "flats" vs. well developed pools, riffles dominated by gravel vs. those with boulders or bedrock ledges, etc.) A special case is very deep pools (over safe or feasible wading depth). Where the area of deep water is relatively small, such pools may sometimes be fished with two electrofishers by reaching probes into the deepest areas and drawing fish into dip net range. Otherwise, if very deep pools are a relatively infrequent feature, they may be passed over in favor of more workable pools, on the assumption that species composition will not be radically different.

Criteria for size of sample/amount of effort:

The following rules of thumb are recommended to insure an adequate sample:

- The length of the sample reach, from the downstream to the upstream end, should be at least 15 times the average width of the stream.
- Sample reach should include at least two complete riffle/pool sequences (except where riffles or pools are very infrequent or absent). Where these habitat features are not obvious, if they are absent due to anthropogenic causes, it may be helpful to try to imagine where riffles or pools were naturally.
- Sample reach should include at least two bends. This may not be possible in channelized streams or in some naturally straight, high gradient reaches. In the latter case, the riffle/pool criterion should be strictly followed.
- Include all discernible habitat types.
- Set a minimum investment of time. In most cases 20 minutes of actual electrofishing time (using the timer on the primary electrofisher) should be adequate to get a representative sample if the work is well planned.) Actual work time (elapsed time) is

immaterial, as it may vary with the number of fish caught, difficulty of the site, size and capability of crew, etc.

- Except in very poor habitats, strive for a minimum number of individual fish, so that statistical inferences can be made with confidence. I usually try to collect at least 200 fish—sometimes less in very small streams.
- Sample should include all "expected" species (or if an expected species is not present, there should be a reasonable hypothesis for its absence—for example, a new pollution source). In the beginning, this will require dependence on pre-existing sources of information (reference works, other investigators), which may refer only to the watershed, not specific sites, but with time, investigators will develop their own lists of expected species at different sites. Where criteria for expected species are weak or non-existent, pay special attention to the following point on species depletion.
- Species depletion occurs when no more species new to the sample are encountered. Classical species depletion requires the completion of at least three subsamples in each of the major habitat types without adding any species to the list. With experience and familiarity with expected species at a site, this rule may be followed less rigorously but new investigators, or those working in unfamiliar watersheds, should adhere to it strictly. (Species depletion also serves as a surrogate for habitat differentiation. If one does not have a practiced eye for subtle differences in habitat, but species depletion is achieved, it may be assumed that all significant habitats are being sampled.)
- Numerical depletion runs (i.e., repeated sampling of a sample reach until no more fish appear), as advocated in some procedural manuals is, in my opinion, an inefficient use of time (and will be impossible on large streams). So long as good procedure is followed with respect to species depletion, and assuming the objective is not to estimate the total fish population, less inclusive, but standardized methods will be adequate. The same applies to the setting of block nets to close off a sampling site so fish do not enter or leave. In my opinion, it is not an effective use of time and may result in unacceptable fish mortality.

Planning the sample:

How the sample is planned depends on the width of the stream. In streams that can be closed off completely or almost completely with a 20-25 ft. seine, sample planning can consist simply in selecting a downstream and upstream terminus for the sample and trying to sample the entire area of stream between these points.

Wider streams may require more careful planning. In some cases, it may still be feasible to strive for complete coverage of a given length of stream. However, since the wider the stream, the greater the length of the total sample, this may lead to an unfeasible investment of time. In this case, one should walk the sample reach before sampling, making careful note of all significant habitat factors (riffle/run/pool, depth, substrate type, current velocity, riparian vegetation, and special features such as logs, root wads, backwaters, etc.) While one does develop an "eye" for this sort of thing, in the beginning it will not be amiss to make a sketch map and plot each subsample, striving for proportional representation of each significant habitat feature. (For example, if 25% of the banks are undercut, then 25% of the sample effort should include such areas.)

In all cases, on large streams or small, allow for the possibility of extra effort if needed. For example, if at the completion of the anticipated sample effort, the sample does not contain a minimum number of individual fish, or species depletion has not been achieved, there should be room to continue upstream without distorting the sample.

Sampling:

The following instructions assume some familiarity with electrofisher operation.

Sampling is normally conducted in an upstream direction; so that once fish are identified, they may be released downstream to avoid double counting of individuals. The basic technique for sampling is the shocker-seine run. The following description will apply to shocker-seine runs in large streams, with a full crew of nine individuals. It is assumed that the reader can make the necessary adjustments for smaller sites/crews. The crew may include the following individuals:

- 2 electrofisher operators
- 2 or 3 seine setters (occasionally more on difficult sites)
- 2 or more dipnetters
- 1 or 2 bucket carriers

It is assumed that at least one of these individuals is capable of doing field identification of fish. This individual also needs to be familiar with the common disease, parasites, and anomalies found in fish. (Normally these do not need to be precisely identified, but most IBI's do incorporate a count of the proportion of fish with disease or anomalies). One person—not the individual doing identification—needs to be responsible for keeping accurate and legible notes. Since this will occur after each subsample is collected, this function can be served by any of the above individuals.

Where there is appreciable current, the effort begins with two individuals, one on each end of the seine, setting the seine across the current, with some slack so that it forms a "bag" to retain the fish. There should be a wide area of "apron" on the stream bottom. This requires that the seine poles be placed with the foot upstream and the pole inclined so that the top is downstream of the foot. It is best to position the seine in the air, then aggressively bring the two poles down at the same time. Where the current is strong, it may be helpful for the seine operators to cross one foot over the pole and place it on the lead line.

In case of very strong current, or even in its absence, if the substrate is very irregular, one or more persons may stand on the lead line, with their feet as wide apart as is comfortable. This will serve both to take some of the strain off the individuals holding the poles and to minimize the passage of fish under the net. A similar effect may be achieved through the placement of rocks on the lead line.

The shocker operators then proceed upstream to the upper limit of the subsample. (Note the distance from the seine, for later use in calculating the area of water actually sampled. The width of the sample will normally be equivalent to the length of the seine.)

They proceed downstream toward the seine, trying to maintain the same speed, while operating the electrofishers. When they reach the seine, a single pass through the seine with the electrofishers is advisable, particularly where large fish are expected.

When the electrofishers are turned off and the electrodes are removed from the water, the two persons holding the seine poles sweep them forward and up, retaining the fish in the "hammock" formed by the seine. They may be assisted by one or more individuals grasping the lead line (and removing any rocks which may have been placed.)

The electrofisher operators are accompanied by two or more dipnetters who attempt to capture any fish they see. When choices must be made, they should favor large fish (which are more likely to be rare species), vigorous, fast swimming fish, benthic species which may get stuck under rocks or any fish which looks unusual. Polarized sunglasses will be helpful for all concerned, but are particularly important for dipnetters.

One or two individuals carrying buckets with small amounts of water should accompany the dipnetters, striving to be accessible but out of the way. Netted fish should be placed in the buckets as soon as possible, to avoid losses but also to minimize mortality occasioned by crowding in nets and repeated exposure to the electric current.

Once the seine is out of the water, professional crews will normally concentrate fish in a small "pot" in the middle of the seine. However, with volunteer crews, assuming part of the objective is to educate, it will be preferable to carry the seine to shore, lay it down and then remove the fish. (The technique of concentrating the fish in the seine may be used in very wide streams, or where the banks are steep and brushy.)

Get all fish in buckets, with an adequate amount of water, as rapidly as possible, to reduce mortality.

Once all fish are in buckets, they may be removed with an aquarium net for identification, counting, and demonstration purposes. They are then transferred to a second bucket for release downstream. With practice, you will develop techniques of handling fish which increase efficiency and reduce handling. Remember that, especially with small fish, actual handling is far more stressful than either shocking or netting.

Certain situations require special techniques:

- Where currents are irregular, or where there are deep-water refuges, experienced electrofisher operators will develop strategies for surrounding and pursuing fish that take advantage of natural features. In such cases the criteria of downstream, parallel movement by the electrofisher operators and dipnetters may be suspended. Application of these techniques may leave the seine crew "stranded" for appreciable periods. However, where possible, such subsamples should be completed as described above for "normal" subsamples.
- Small, shallow streams are often better sampled with a single electrofisher.
- Large streams require special attention to the shoreline. A standard effort would be three 100 ft. long shoreline samples. Shoreline sampling is done without the seine, and usually with one electrofisher. The electrofisher operator proceeds upstream along the shoreline, maintaining the electric field within 3 ft. of the shoreline. Dipnetters and bucket carriers follow, capturing fish as they are seen.
- Methods similar to that described for shoreline samples may be applied in certain special habitats. For example, in backwaters without appreciable current or in narrow chutes or side channels, a seine may be superfluous.

- Large pools with relatively unobstructed substrates are often best fished by haul seining without the electrofisher. Normal procedure is to start at one side of the stream, or at the middle of the pool, and haul the seine up onto a beach (or to lift it just short of the shore). Two people are required for this operation, but it will be helpful to have another individual follow the seine in case of snagging, plus one or more individuals posted on shore to help bring the seine in. Strive to keep the lead line on the bottom at all times, up to the last minute!

Identification and what to include:

As mentioned, field identification is a skill that must be developed. It may be prudent to do a series of "unofficial" samples in order to get practice, before embarking on a monitoring program, or to accompany professionals in the field, if possible. And even the most experienced professional should always carry a small supply of formalin and jars, so that any completely unanticipated or unidentifiable fish can be preserved for purposes of later identification and documentation.

In general, all fish captured are identified and recorded. "Visual" identification of fish seen, but not actually captured, is permissible with appropriate caution, particularly for purposes of completing a species list. An example from experience in North Carolina may be helpful.

Among the most difficult fish to capture are the several species of redhorse suckers (*Moxostoma* spp.). These large, highly mobile fish may evade even an experienced crew. However, they are often visible and if seen clearly are not likely to be mistaken for other fish. Since one of the IBI metrics for some streams is "number of sucker species," inclusion or exclusion of redhorses could be critical. Were I to observe, but not capture, redhorses in a sample, I would include "redhorse" as one species in the sample. Depending largely on visibility, I might not attempt to count them, and I would certainly not attempt to differentiate among the species of redhorse in the water.

Two groups of fish may be excluded from the sample:

- Young-of-the-year (YOY) are not normally counted in IBI samples for several reasons, but particularly because they are differentially catchable. That is, our nets may retain YOY of some of the larger species, or species which spawn early in the year and thus reach catchable size sooner, while other species pass through the mesh. One of the principal reasons for concentrating biomonitoring work in the early summer is to avoid confusion resulting from YOY. Even so, it will be useful to have an idea as to the size each species is expected to reach in its first year, so that decisions may be made. While YOY are not normally counted, if a species is found only as YOY (note that identification of very young fish may be difficult) it should be noted and, if abundant, may be included in species count.
- Recently stocked fish should be identified and noted, but not normally included in the sample. This situation arises most often with respect to trout. An experienced operator will learn to distinguish between the appearance of a recently stocked trout and one which, if not hatched in the stream, has spent some months there. Fish of any species which have obviously acclimated to the environment being sampled, and particularly if they are judged to have passed a year or more there, should probably be included in the sample, even if there is no evidence of reproductive success.

Permits and permissions:

All states (and some other jurisdictions, such as National Forests, Indian Reservations, etc.) will normally require a scientific collection permit for use of an electrofisher, even though there may be not intent to “take” fish. Some jurisdictions also require either a scientific collection permit or a sport-fishing license to capture fish with nets only. If your group includes a bona fide scientist, it should not be difficult to obtain the necessary permit, which may be free of charge or relatively costly.

Obtaining the necessary permits may be a difficult hurdle for groups composed entirely of volunteers and parataxonomists. In such cases, it may be possible to get authorization from a recognized scientific collector to operate under his or her permit.

Note that almost all permits come with reporting criteria, which should be followed, even though this may seem tedious. Apart from the legal requirement, sharing data can only lead to good things.

It is also important to have proper access permission. In some situations, such as access from bridges or in streams heavily used for public recreation, access may be taken for granted. However, it is never a bad idea to know whose land you are on, and it is a must if you intend to cross private property to reach the stream. (And remember that in most states, private property includes the bottom of wadable streams.)

Thus, an essential part of the biomonitoring task is to precede the season by making inquiries, and perhaps visiting the county courthouse to determine land ownership and obtain the necessary permissions. It is also a good idea to invite landowners to witness, or participate in the proceedings, and to provide them with information and results afterward.