Sampling and Use of the Stream Condition Index (SCI) for Assessing Flowing Waters: A Primer

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A Brief Description of Contents

1.1 Historical Background

The response of benthic macroinvertebrate communities to human point source pollution began receiving attention in Florida during the late 1950's. In 1958, Bill Beck, biologist with the Florida State Board of Health, wrote a series of "Biological Letters", where he introduced the concept of using invertebrates as biological indicators, especially for demonstrating the effects of excess organic matter on streams and lakes (the saprobium index concept). What became known as "Beck's Biotic Index" was developed by sampling invertebrates at control sites located upstream of point source discharges and observing which sensitive taxa were eliminated at sites downstream of the effluent sources (Beck 1954). Concurrently, there typically was a dramatic increase in abundance of tolerant taxa, such as "bloodworms" (certain species of chironomid midges) as illustrated in Figure 1.

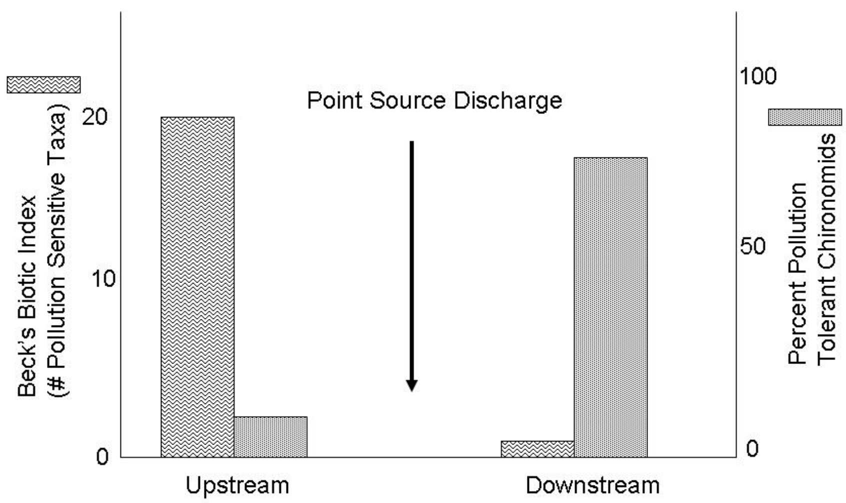


Figure 1. Typical macroinvertebrate response to organic loading associated with primary wastewater treatment typical in the 1950s and 1960s.

In the early 1970's and 1980's, benthic invertebrates were routinely sampled via multi-plate artificial substrate samplers (Hester-Dendys). Hester Dendy samplers are incubated in the receiving waters for 28 days, a minimum period of time for colonization by a representative benthic community (Figure 2). The Shannon-Weaver diversity index, a biological metric derived from information theory, became a popular method to communicate complicated biological results. The Shannon-Weaver diversity index is based upon a combination of the taxa richness at a site and the equitably of the distribution of abundance of individuals. Low diversity scores represent conditions where a few pollution tolerant organisms are very abundant, to the exclusion of other taxa. This index is specified in the Florida Administrative Code as a measure of biological integrity (Rule 62-302.530 FAC). It generally has been applied by comparing site-specific control sites to nearby test sites.

Figure 2. Photo of Hester-Dendy samplers used for determining the Shannon-Weaver diversity index.

In 1992, EPA promulgated the concept of “rapid bioassessment”. Regional expectations (generally eco-regions) for biological communities were established by sampling “minimally disturbed” reference sites. Metrics, defined as measures of biological health which respond in a predictable manner to human disturbance, were calculated from the raw reference site data. Next, a distribution of the reference site metric values was calculated, and a percentile (typically, the lower 25th percentile for metrics which decrease in response to human disturbance) was selected to represent the expectations for that metric in a minimally disturbed condition. A variety of metrics would then be combined into a dimensionless index. This was accomplished by assigning points to individual metrics based on their relative similarity to the reference condition, and summing the points.

The current Stream Condition Index was built upon the 1990s concepts. The main improvement in the present index is the use of a human disturbance gradient to determine effective metrics and then determining impairment thresholds by using a Biological Condition Gradient (BCG) approach. The BCG employs a group of experts to individually review species level data and determine the site’s ecological status (see section 1.4). Further discussion of the present Stream Condition Index occurs below.

1.2 Bioassessment Theory

To successfully manage ecosystems, a basic understanding of the system's biological components is critical. The biota respond to a wide variety of cumulative factors, both natural and anthropogenic (Figure 3). As the organisms integrate these factors over time, a characteristic community structure emerges. When human actions adversely affect a system, the biological population will change, leading to an impaired or imbalanced community. For example, pollution sensitive taxa will disappear, food webs are disturbed, taxa richness and diversity usually decrease, and undesirable nuisance species may dominate.

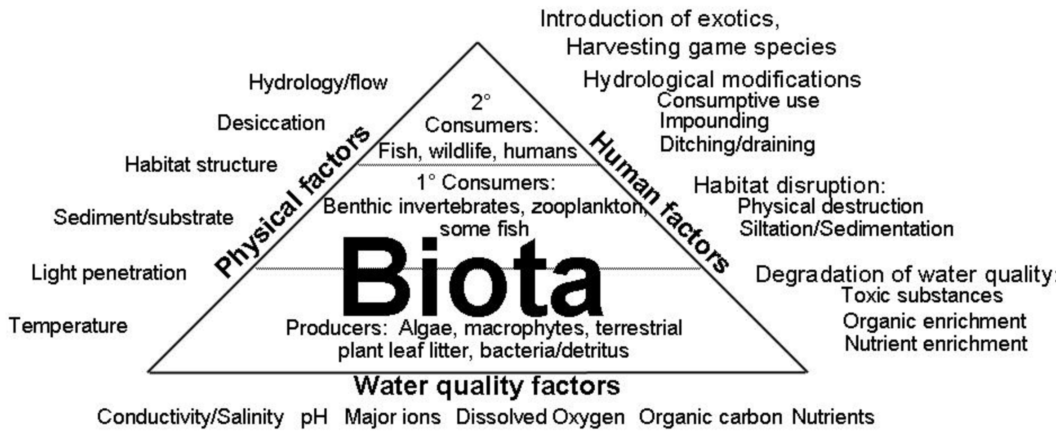


Figure 3. Many factors affect biological community composition. To conclude that human factors are primarily responsible for biological degradation, reasonable knowledge of the influence of natural factors is essential.

To accurately determine when humans have negatively affected a biological community, one must be familiar with the structure and function of natural, or "reference" systems in a given geographical region (Griffith et al. 1994; Figure 4). First, it is important to establish the normal or typical range of certain key measures of community health at these reference systems, often thought of as “biological integrity”. Karr defined biological integrity as the ability of an aquatic ecosystem to support and maintain a balanced, adaptive community of organisms having:

- species composition,
- diversity,
- and functional organization comparable to that of natural habitats within a region.

Measures (or attributes) of ecosystem health that respond predictably to human influence are termed **metrics**. Metrics from reference sites are compared with the same metrics from an unknown or "test" system to determine unacceptable departures from the expected condition, associated with human impairment. To be scientifically defensible, the systems being compared should be similar except for potential human influences (compare streams to streams, not streams to a system with lake-like conditions). Additionally, one or more natural stressors (*e.g.*, flood, drought, low substrate diversity, periodic natural low dissolved oxygen, etc) may affect sampling sites, even those sites with minimal disturbance from humans. These natural stressors should be reasonably understood and controlled for in the sampling design to more

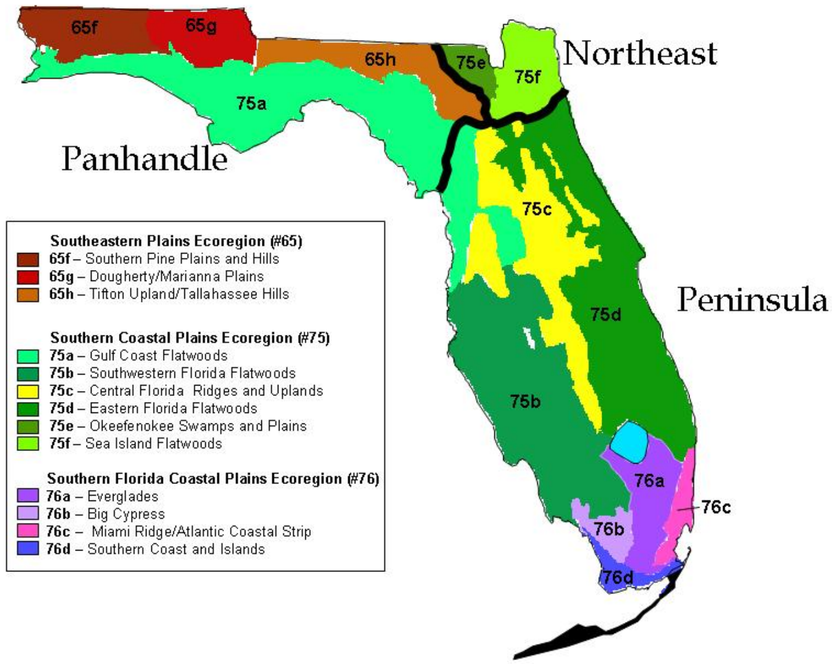


Figure 4. Sub-ecoregions of Florida, which were aggregated into 3 bioregions, based on multivariate measures of taxonomic similarity.

conclusively determine when human actions have caused biological degradation (see below in "Section 1.5, How objectives affect SCI sampling decisions").

1.3 Development of Stream Condition Index Metrics using the Human Disturbance Gradient

DEP has utilized a Human Disturbance Gradient approach to allow for the objective selection of metrics (Fore *et al.* 2007). The Human Disturbance Gradient is composed of four factors:

- The Landscape Development Intensity Index (Brown and Vivas 2006)
- Habitat Assessment scores (DEP SOPs)
- Hydrologic Modification Index
- Water column ammonia concentration

These components, described in detail by Fore *et al.* (2007), were converted into a dimensionless index, with low values denoting low disturbance and increasing values associated with more intense human influences. The index was subsequently used as the x-axis for testing a wide variety of biological attributes associated with the measurement of ecological integrity (Figure 5). Figure 6 depicts the absolute value of correlation coefficients (Spearman's r) for a variety of biological attributes against the HDG. Once an attribute is demonstrated to respond predictably to human influence, it is termed a **metric.** The 10 selected attributes metrics were chosen to:

- represent as many attribute categories as possible;
- provide meaningful and predictable assessment of human effects;

- avoid redundancy if several correlated metrics were providing similar information.

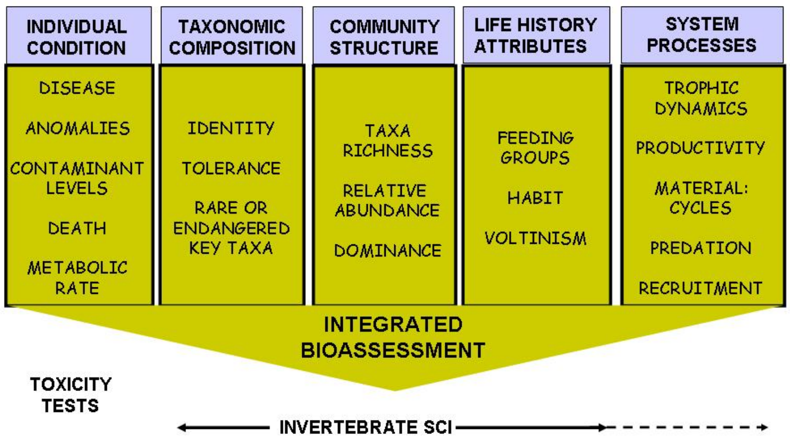


Figure 5. Major attribute categories, and example metrics, for determining biological integrity.

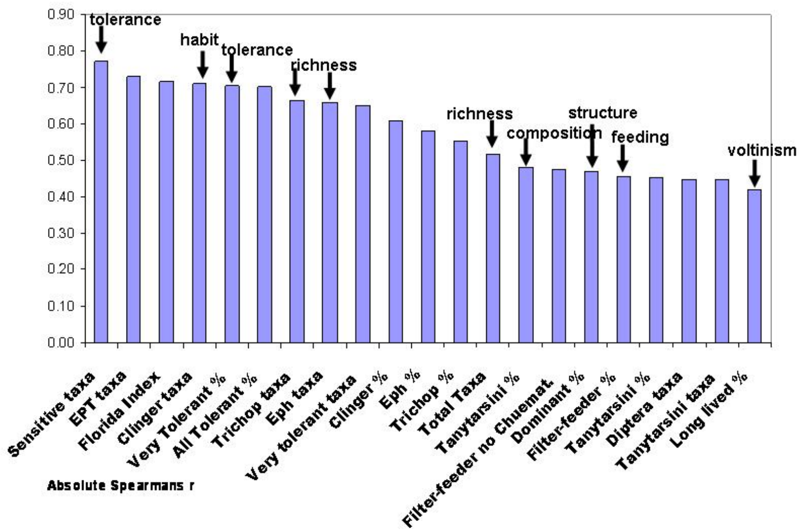


Figure 6. Correlation between various metrics and the Human Disturbance Gradient. Arrows indicated metrics selected for the SCI, and associated attribute group.

The following is a brief description of the metrics, divided into several metric types.

Taxonomic richness

Total taxa richness (the number of different types of organisms present) and the richness of the Trichoptera (caddisflies) and Ephemeroptera (mayflies) has historically been shown to decrease with human disturbance. Figure 7 depicts the response of the number of Ephemeroptera metric to human disturbance, which is similar to the response of the

Trichoptera taxa and total taxa metrics. These three measures were chosen since each metric may respond differently, depending on the type of disturbance (*e.g.*, mayflies are more sensitive to metals, certain caddisflies may be more sensitive to flow disruption). Plecoptera (stoneflies) are not found throughout the state and were therefore determined not to be a consistent metric. However, they are still evaluated as "sensitive" taxa (see below) where found.

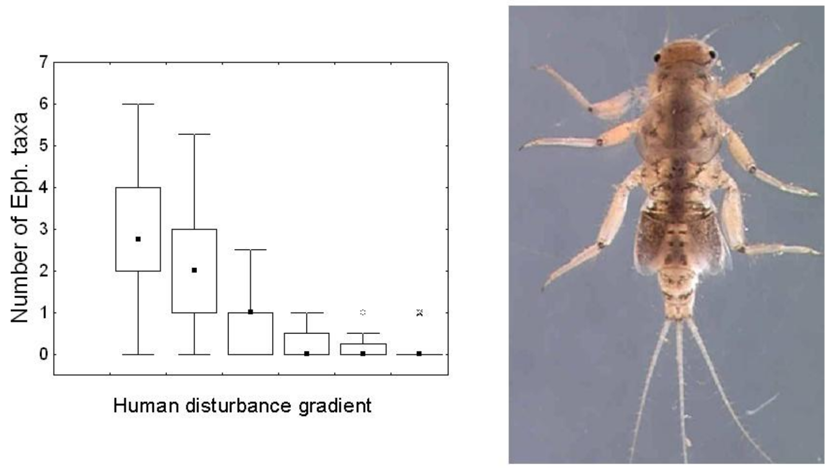


Figure 7. Response of the Ephemeroptera metric to the HDG.
The photo is of Tricorythodes, a sensitive mayfly.

Feeding group
Disruption of food webs has long been associated with human influence, especially organic pollution. Of the functional feeding group measures, the relative abundance of filterers or suspension feeders (percentage of filterer individuals) had the highest correlation and most consistent relationship with the HDG (Figure 8). Filter feeders extract nutrients by straining food particles from the water column. If the water flow or quality of the organic matter in the water is compromised, a reduction in filter feeders will occur.

Voltinism
Voltinism refers to the number of distinct reproductive cycles for a given organism that may take place in a year. Long-lived taxa included semi-voltine insects and non-insects that require greater than one year to complete their life cycles. Long-lived taxa richness would be expected to decrease if a disturbance event (*e.g.*, sporadic illegal dumping, periodic pulses of chemicals from rain events) occurred at a site within a year of sample collection (Figure 9).

Habit
Clingers are those taxa morphologically adapted to hold onto substrates during routine flow conditions and would be expected to decline as humans alter a stream's hydrograph (*e.g*,. channelization), especially during abrasive events caused by high stormwater inputs

from impervious surfaces. Clinger taxa richness was highly correlated with the HDG (Figure 10).

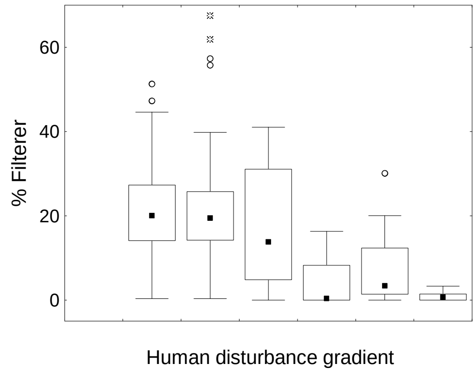


Figure 8. Response of the % filter-feeder metric to the HDG. The photo is of a net-spinning caddisfly.

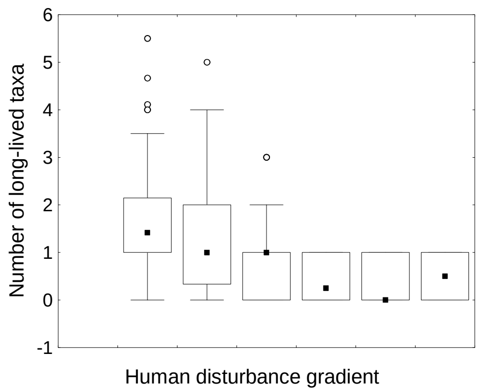


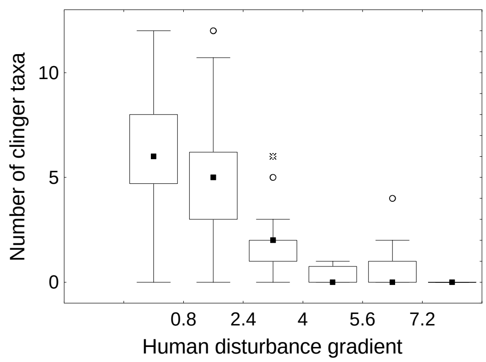
Figure 9. Response of the long-lived taxa metric to the HDG. The photo is of a mollusk, the threatened “purple bank climber”.

Community structure

Substantial shifts in proportions of major groups of organisms, compared to reference conditions, may indicate degradation. The percent dominant taxon, which increases in conditions where a few pollution tolerant organisms are very abundant, to the exclusion of other taxa, was selected as a metric. Tanytarsini midges are sensitive to disturbance, so the % Tanytarsini metric was included in the SCI as the best available measure of the chironomid assemblage.

Sensitivity and Tolerance

Lists of sensitive and very tolerant macroinvertebrates were established by analyzing the responses of individual species to the HDG (Fore 2004). The number of taxa selected as sensitive equaled around 12% of the taxa tested, and the number of very tolerant taxa was approximately 10% of the taxa tested.



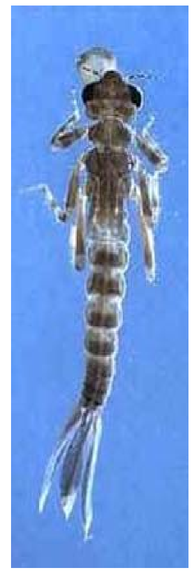


Figure 10. Response of the clinger taxa metric to the HDG. The photo is of a damselfly larvae.

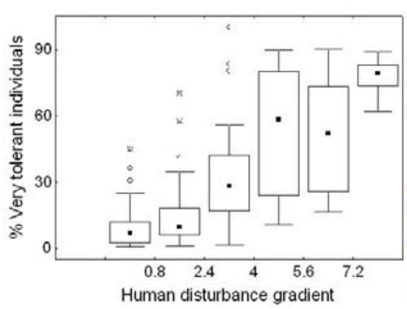


Figure 11. Response of the % Very Tolerant metric to the HDG. Photos include lunged snails, tolerant midges, and leeches.

Many sensitive species belonged to the Ephemeroptera, Trichoptera or Odonata; several chironomids were also included. All the Plecoptera were included as sensitive taxa. The number of sensitive taxa and the percent very tolerant individuals were highly correlated with the HDG (Figures 11 and 12).

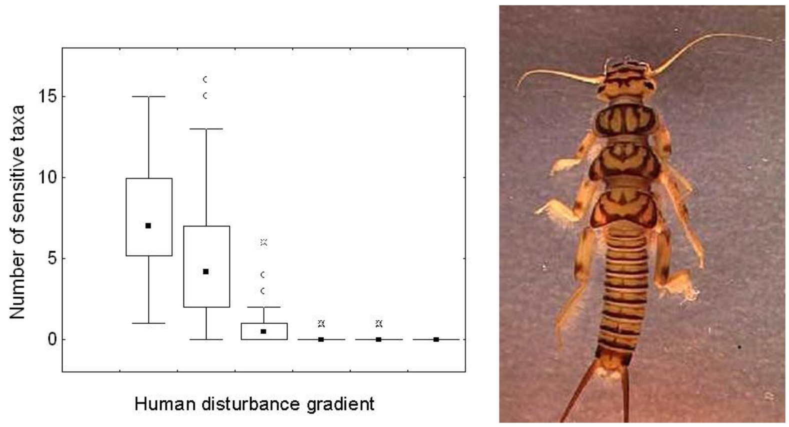


Figure 12. Response of the number of sensitive taxa metric to the HDG. The photo is of a plecopteran (stonefly).

1.4 Establishing an SCI Impairment Threshold using the Bio-Condition Gradient

The U.S. EPA has outlined a tiered system of aquatic life use designation, along a Biological Condition Gradient (BCG), that illustrates how ecological attributes change in response to increasing levels of human disturbance. The BCG is a conceptual model that assigns the relative health of aquatic communities into one of six categories, from natural to severely changed (Figure 13). It is based in fundamental ecological principles and has been extensively verified by aquatic biologists throughout the U.S.

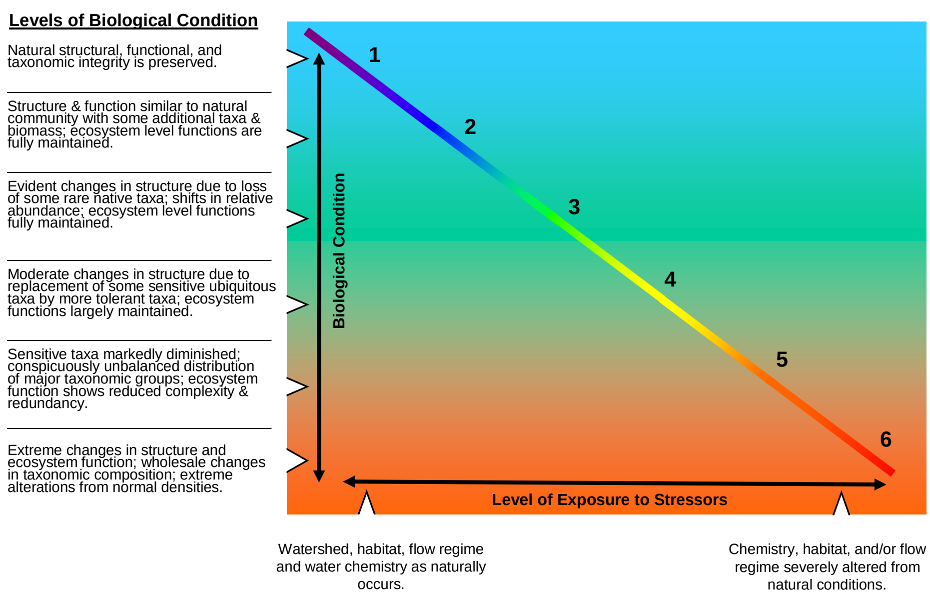


Figure 13. EPA's Biological Condition Gradient predicts that biological health will decline in response to increasing levels of stress.

The BCG utilizes ten biological attributes of aquatic systems which predictably respond to increasing pollution and human disturbance. While these ten attributes are measurable, some are not routinely quantified in monitoring programs (*e.g*., rate measurements such as productivity), but may be inferred via the community composition data (*e.g*., abundance of taxa indicative of organic enrichment).

The attributes are:

1. Historically documented, sensitive, long-lived or regionally endemic taxa
2. Sensitive and rare taxa
3. Sensitive but ubiquitous taxa
4. Taxa of intermediate tolerance
5. Tolerant taxa
6. Non-native taxa
7. Organism condition
8. Ecosystem functions
9. Spatial and temporal extent of detrimental effects
10. Ecosystem connectance

The gradient represented by the BCG has been divided into 6 levels (tiers) of condition that were defined via a consensus process (Davies and Jackson 2006) using experienced aquatic biologists from across the U.S.:

1. Natural or native condition;
2. Minimal changes in structure of the biotic community and minimal changes in ecosystem function;
3. Evident changes in structure of the biotic community and minimal changes in ecosystem function;
4. Moderate changes in structure of the biotic community with minimal changes in ecosystem function;
5. Major changes in structure of the biotic community and moderate changes in ecosystem function;
6. Severe changes in structure of the biotic community and major loss of ecosystem function.

A panel of 22 experienced aquatic biologists in Florida was convened to apply and calibrate the general BCG model to benthic macroinvertebrates in Florida streams (FDEP BCG workshop as described in Fore *et al*. 2007), and later, the workshop findings were used to define the threshold below which the SCI score is associated with a biological impairment. The panel was provided with macroinvertebrate species lists and metrics for 15 reference streams and 30 test samples, equally divided into Florida's three bioregions (Panhandle, Northeast, and Peninsula). The panel was NOT given the Stream Condition Index scores associated with any of these sites, or any physical, chemical, or habitat descriptions of the sites (other than the Bioregion location). Decisions were based on biological community composition data collected via the SCI method. Detailed descriptions of the above attributes and tiers were provided to the panel prior to the meeting and on the day of the meeting, a presentation reiterated these definitions accompanied by extensive discussion. A correlation between the experts BCG ranking with SCI scores is shown in Figure 14. Note that these impairment decisions are independent of the Human Disturbance Gradient approach that established the SCI metrics.

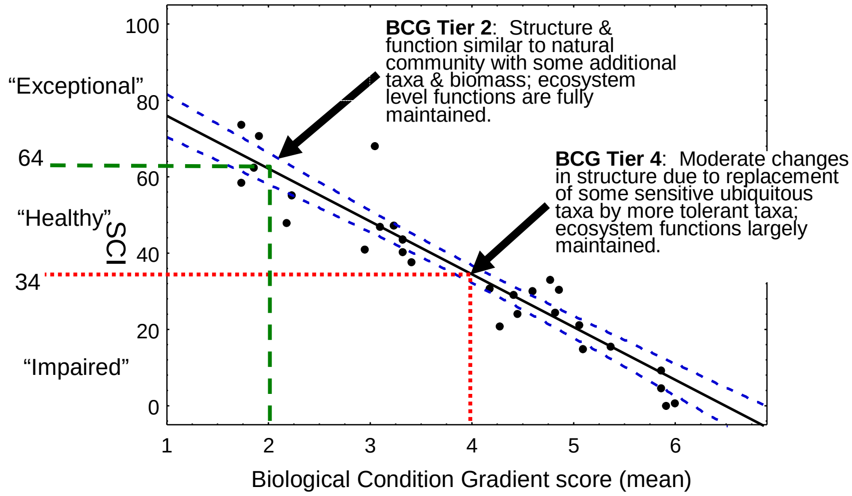


Figure 14. Linking the Biological Condition Gradient Model to SCI thresholds. The central tendency of the expert group was that "impairment" occurred below BCG Tier 4, and that Tier 2 and above represented minimally disturbed reference conditions.

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1.5 *How Objectives Affect SCI Sampling Decisions and Interpretation of Results*

It was mentioned previously that biota respond to natural and human stressors alike (Figure 3). It is imperative the study objectives associated with each SCI sample are clearly articulated and that efforts are taken to control for confounding factors that may interfere with the appropriate interpretation of the SCI scores. Although there may be multiple factors to consider, the main three issues to be aware of during an SCI study are:

- existing and antecedent flow conditions,
- habitat conditions at a given site, and
- water quality, especially human degradation of water quality (such as exceeding water quality standards).

Potential uses of the SCI, in context of DEP program decisions, are mentioned below and interaction of these components and their effects on score interpretation is discussed.

Water Quality Investigations for the Total Maximum Daily Load Program

One objective for a TMDL study is to determine if water quality issues are adversely affecting biological health. To list a waterbody on the verified TMDL list, DEP must reasonably demonstrate the pollutant responsible for poor SCI scores. Since water flow significantly affects stream biota, the investigator must first determine if the existing and antecedent flow conditions were appropriate for sampling. It may seem obvious, but aquatic organisms WILL DIE if a site goes dry. If desiccation has occurred within the past 6 months of a sampling event, the recent dry conditions, not water quality, will dominate the invertebrate response. The SCI SOP mandates “a minimum” wait of 3 months after a dry site has begun flowing again, before considering SCI sampling. To ensure the desiccation was not an issue, waiting an additional time period could easily be warranted. Similarly, stream organisms are rheophyllic (“flow loving”). If the water velocity is minimum (standing water, stagnant conditions), it will adversely affect the assemblage of organisms, even if water quality is excellent. Therefore, sampling for TMDL purposes should be conducted during periods (including antecedent conditions) of at least minimal (> 0.05 m/sec) water velocity. Controlling for these water flow issues (not sampling during inappropriate conditions) will help minimize the influence of desiccation and water velocity on the SCI results.

Additionally, habitat conditions significantly affect macroinvertebrate communities. Since the objective of a TMDL study is to isolate water quality factors causing degradation, efforts should be taken to establish sites where habitat is not a substantial factor limiting potential biological health. This would mean establishing sampling sites (where possible) in stream reaches with adequate substrate diversity and abundance, intact stream morphology (no or minimal channelization effects), adequate flow, and decent riparian buffer zones. Note that deleterious sediment input is defined as a pollutant, so the habitat smothering may be an issue in cases where sediment is the pollutant of concern.

Specific conductance (or conductivity) is a water quality parameter worthy of special discussion. Elevated conductivity at a site may be due to its proximity to natural saline conditions (at tidally influenced systems) or due to human pollution. The SCI was designed for freshwater streams, and as such, it would not be appropriate to use the tool where conductivity is naturally elevated (near estuarine areas). However, if a human discharge has artificially elevated a stream site’s conductivity, the SCI may be used to document the resulting adverse community response. One must take care to assess the source of the conductivity when deciding the appropriateness of using the SCI.

In conclusion, if flow and habitat limitations are controlled for during a TMDL study, and sufficient water quality data are collected, a reasonable case for the water quality factor(s) responsible for any observed biological degradation is possible.

Point Source Studies

Typically, point source studies involve an evaluation of the effluent quality and whether existing permit limits are sufficient to maintain surface water quality standards (62-302.530 F.A.C.) and prevent degradation of the biological communities in the receiving waters. An upstream-downstream SCI study is routinely employed, emphasizing the control of important variables between the control and test sites, enabling influence of the discharge to be assessed. Therefore, selection of similar substrates for sampling from similar areas of water velocity would be important to determine if characteristics of the effluent can be associated with any longitudinal changes in the SCI scores. If reductions in the SCI scores occur between the control and test sites, the magnitude of the change should be assessed, as well as potential categorical shifts (e.g., “exceptional” to “healthy”, “healthy” to “impaired”).

Studies to Determine Effectiveness of Best Management Practices (BMPs)

Previous studies on the effectiveness of forestry best management practices, using the SCI as a tool, followed a typical Before-After-Control-Impact design. This design may be applicable to other BMP studies. Stream reaches were selected where neither flow, habitat, or water quality were limiting to aquatic communities. An upstream "control site" and a downstream "test site" were established, and both were sampled (with replicates) prior to the onset of the human activities (conducted with BMPs). Sampling continued at the same control and test sites after the potentially damaging human activities (with mitigating BMPs) had taken place and SCI scores were compared, both pre- and post- disturbance (see Figure 15). In this particular case, Analysis of Variance indicated that no significant differences between the control and test sites had occurred after the forestry activities, demonstrating that the BMPs were effective in protecting stream biota .

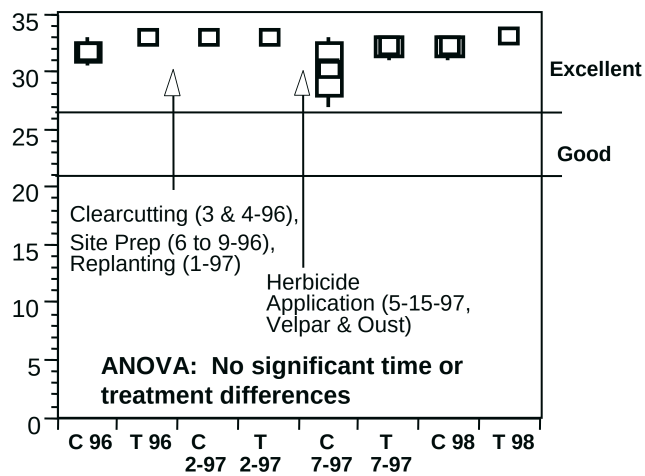


Figure 15. SCI results of a Before-After-Control-Impact study assessing the effectiveness of forestry Best Management Practices. "C" and "T'" mean control and test sites, respectively. Note that a different version of the SCI was used during the time period, but the concepts apply to the current SCI.

Stream Restoration Studies

The objectives for a stream restoration study may be to determine if one or all the following factors have been improved or mitigated in a manner that adequately supports aquatic communities:

- Stream morphology
- Habitat
- Water supply to the stream and in-stream water velocity
- Water quality

The investigator should measure each of the important variables over time, along with conducting SCI sampling. This will enable a demonstration that the restoration activities can be successfully linked with a positive biological response (improving the SCI score as the desired environmental endpoint). Past studies of reclaimed streams in mining areas have suggested that all four factors listed above need to be adequately addressed to ensure a positive biological response. It is important that data collected as part of a restoration study not be indiscriminately used for unintended purposes (e.g., placing a waterbody on the TMDL verified list when habitat, not water quality, was the limiting issue).

Minimum Flow and Levels Studies

As mentioned above, sufficient water flow is critical to stream biological community health. Biological communities will be negatively affected when humans adversely modify watershed hydrology or artificially reduce water inputs to a stream (leading to extended dry or stagnant conditions). However, care must be taken to distinguish between effects of natural droughts and the similar effects caused by human reductions in water quantity. Also, if a study design calls for using SCI sampling after stream desiccation (within 3 months) or during periods of stagnant water velocities (not recommended for the general SCI SOP), it is important that the resulting data (probable SCI failures) not be misinterpreted as water quality issues (see section 1.8 on maintaining associated data with the SCI below).

Ambient Monitoring (Status and Trends) Program

The Integrated Water Resource Monitoring program (IWRM, aka Ambient) is a monitoring program designed to determine the quality of Florida's fresh surface and ground waters at a large scale, using two differing approaches. The first (Trend network) is a fixed-point monitoring program that is designed to determine changes in water quality over time at 75 set locations around the state. The sampling locations were selected to capture the quality of waters that flow into the state, and at the bottom of watershed basins (determined using a Hydrological Unit Code, (HUC)). Rivers, streams, and one spring are monitored as part of the program. Samples are collected monthly at all surface water Trend sites.

The second component of the program uses a random stratified (probabilistic) sampling network, also called the Status monitoring network. The objective of the Status network is to provide an estimate of water resource conditions within the state for surface and ground waters. Because of the extent of aquatic resources within the state, no one sampling network could adequately sample all waters in Florida each year due to logistical and practical limitations. The probabilistic design was selected to balance resources, provide a scientific and statistically sound platform, and provide coverage of waters at a reasonable scale. A subsample of the water resource is selected, collected and analyzed during a specified sampling window referred to as an "index period". The design is based on a set geographic boundary, or "reporting unit" that follows the watershed boundary. In the current design, the Status network follows the TMDL boundaries.

The SCI tool was adopted as part of the TMDL Impaired Waters Rule listing process and was incorporated into use by the IWRM program in both the Status and Trend monitoring networks in 2004.

To assist samplers in making the decision whether to sample SCI at a particular site for the IWRM program, the following rules have been developed:

- Do not sample if conductivity at the bottom of the river/stream exceeds 600 µhmos, where indicative of estuarine influence. However, do collect the SCI in non-tidal freshwater environments that have conductivity above 600 µhmos, as it could be indicative of pollutants.

- Do not sample if a site is currently dry, or has been dry within 3 months prior to the site visit.
- Do not sample if water level is elevated, resulting in unreachable habitat (however, a water quality sample is collected). The SCI sample is not collected until the water level has returned to a “normal” stage or until the habitats are accessible.
- Do not sample in Ecoregion 76 (south of Lake Okeechobee).
- Do not sample in lake-like systems (e.g., portions of the St. Johns River), since the SCI was designed for streams.

The assumption was made at the onset of the use of the SCI tool in the IWRM program that it applied to Class III freshwaters. Many of the Class III waters within the central and southern region of the state have been hydrologically altered or have been created for the primary purpose of flood control. Canals and ditches that are connected to waters of the state are currently included for sampling because they must meet Class III standards according to existing designated use rules. It is possible for a canal with adequate habitat and flow to “pass” the SCI, however it is likely that the majority of these canals and ditches have poor habitat and highly modified hydrology, usually resulting in a “failed” SCI. In these cases, the poor SCIs are indicative of actual resource conditions in the basin. It is also true that human activities in some regions exert a higher negative influence than others (see Figure 16). For example, one would expect higher SCI scores in region 65 and 75a (panhandle), which are associated with minimal disturbance, than from the majority of 75 and 76, where human alterations are common.

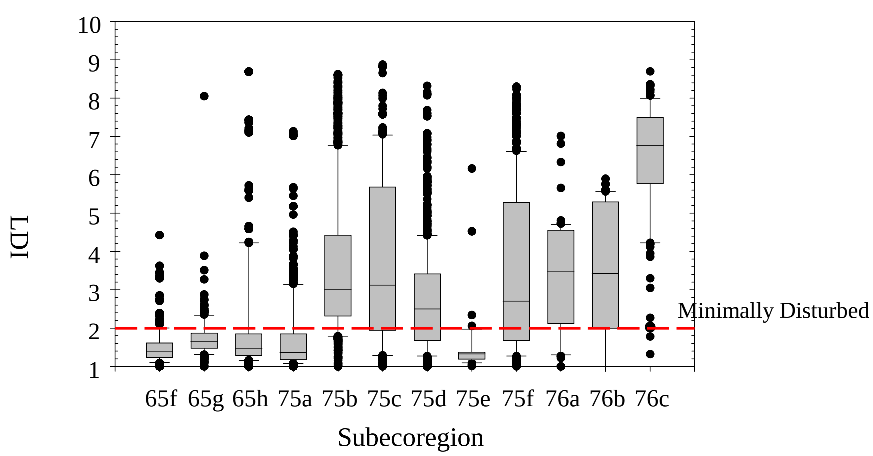


Figure 16. Landscape Development Intensity Index (Brown and Vivas 2006) scores for Florida’s 12 sub-ecoregions based on 5570 stations statewide. An LDI of less than 2 is associated with minimally disturbed conditions.

Due to the random design of the probabilistic network, samples are collected only where the site is specifically selected, based on a 1:100,000 scale map. This results in sites being selected in areas that are possibly not optimum habitats, but should be representative of the stream or river resources in the reporting unit. The objective, as stated above, is to characterize the condition of waters within a region. The intent is not to characterize any specific stream or river. Therefore, when results are reported, they pertain only to the estimate of condition of representative resources within the basin. Note, however, that SCI failures in canals or ditches with altered hydrology and minimal habitat are likely due to these factors, and may not necessarily be related to water quality issues

1.6 Water Level and the SCI

All scientific methods have limitations that must be understood to effectively use the method for making valid decisions. As previously mentioned, aquatic organisms will die if a site goes dry. Wait a minimum of 3 months after a dry site has begun flowing again before considering sampling. Typically, SCI sampling is performed within approximately 0.5 m of the water's surface (the arm length of an average sampler). It is imperative that the sampler be confident that the "reachable" habitat in the top 0.5 m has been inundated with water for a minimum of one month (28 days) prior to sampling to allow time for stream organisms to colonize the formerly exposed habitats. The one month period was previously found to be the minimum incubation time for artificial substrate Hester-Dendy samplers to become colonized. As an example, Figure 17 depicts a recent increase in water level which would limit a sampler's ability to collect organisms from the previously wetted and colonized substrate. When conditions such as these are encountered, the sampler must have sufficient knowledge and training to abort SCI sampling. Understanding hydrographs from streams in the general area to be sampled (not every stream has a gauge) is extremely valuable for determining when sampling is, or is not, appropriate (Figure 17). Smaller streams typically have more spikes in their hydrograph, where the water level rises quickly and significantly but then returns to "normal" levels within days (Figure 18 and 19). A valid SCI sample can be collected when the formally colonized habitats may be reached; however, it is important that samplers exercise caution to make sure the habitats they select have been appropriately inundated.

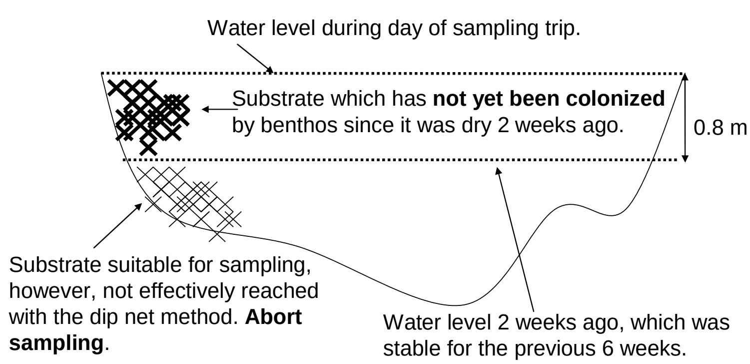
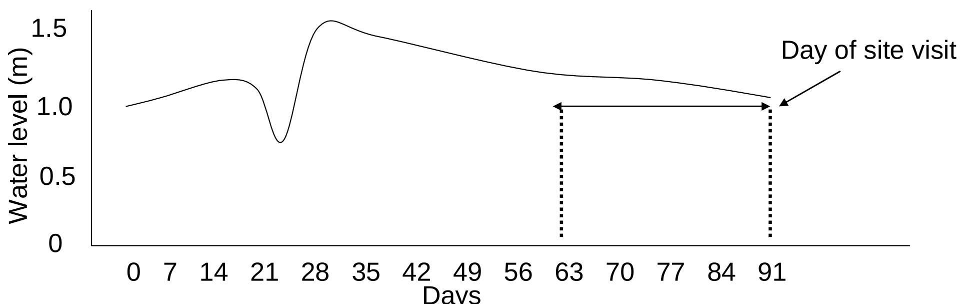


Figure 17. Schematic cross section of a stream showing recent increase in water levels indicating the SCI sampling should not be conducted.

All SCI samplers must fully understand how water levels affect their ability to collect a valid, meaningful SCI sample, and abort sampling when conditions are not suitable. The following examples will help illustrate this concept.

Example 1, TMDL Sampling

A sampler is collecting SCI data to determine if water quality degradation is sufficient to list a waterbody on the verified TMDL list. When arriving at the site, the sampler determines that almost all of the productive habitats (roots, snags, leaf packs) are exposed to air, due to extremely low water levels. Hydrographs from nearby streams indicate these low water conditions have occurred for the past few months. Water velocity in the stream is non-existent. Should the sampler collect the SCI? No, the conditions are such that the lack of inundated habitat and no velocity are the dominant factors affecting the stream biota. Collecting the SCI and attributing the low scores to poor water quality is not scientifically defensible, as factors other than water quality were highly influential.



Hydrograph showing the reachable substrates have been inundated more than one month. This is a good time to sample SCI.

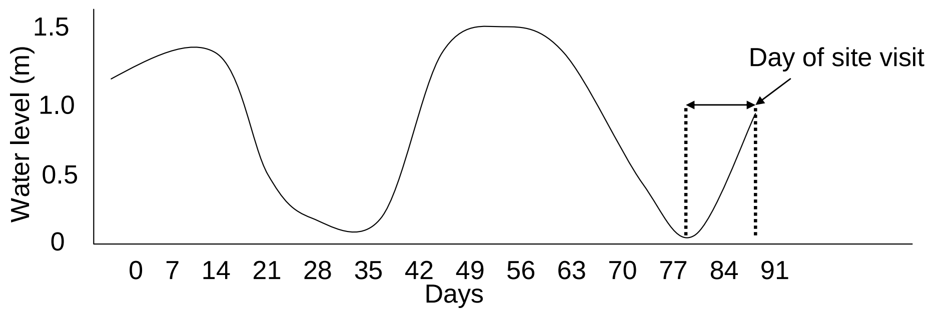


Figure 18. Recent increase in water level. Showing reachable substrates have been inundated for less than 2 weeks. Abort sampling.

Example 2, MFL Sampling

A study is attempting to establish a relationship between water quantity in a stream and the SCI scores with the hypothesis that more water yields higher scores. Water levels in the stream rose by one meter during the past week. The sampler notes that terrestrial vegetation is currently under water and reachable substrates in the top 0.5 m (snags, limerock) have no “slimy” feel. Should the sampler collect the SCI? No, the recent increase in water level means the organisms have not yet colonized the accessible substrates. Sampling under these conditions would erroneously produce data indicating reductions in SCI scores with increased water delivery.

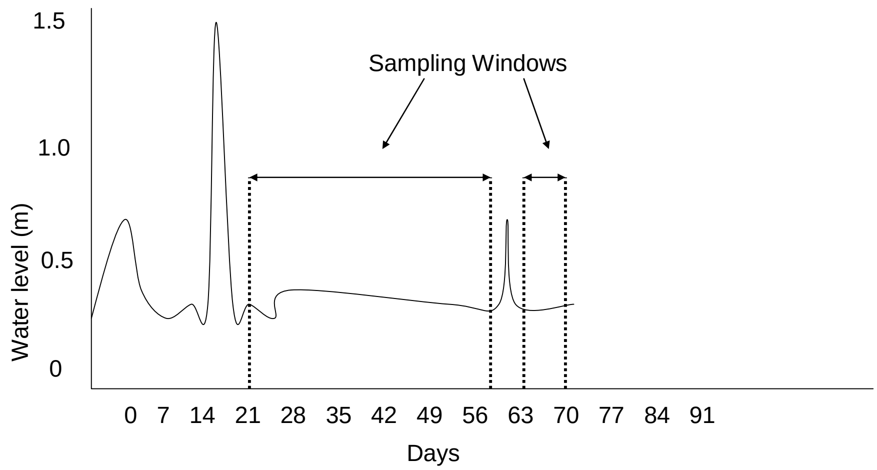


Figure 19. Hydrograph showing times when substrates are reachable.

Example 3, TMDL Sampling

Due to heavy rains, a stream has water levels up to two meters over its banks into the riparian floodplain. This condition has occurred for four weeks. The sampler notes that there is no access to the actual stream channel due to the water depth, but some habitat in low velocity backwaters of the floodplain can be reached. Should the sampler collect the SCI? No, the actual stream can not be sampled. The few organisms that may have colonized the low velocity backwaters of the floodplain would not be representative of the actual stream health. Again, collecting the SCI and attributing the low scores to poor water quality is not scientifically defensible, as factors other than water quality were highly influential. Note: in a large stream or river where the water lever has risen significantly (greater than 0.5 m) but not out of its banks, an SCI can be collected as long as the proper incubation period has occurred (minimum of 28 days).

1.7 SCI Samplers must exercise Best Professional Judgment

Only an experienced, qualified SCI sampler can make the difficult field decisions necessary for proper application of the method. Field staff must be absolutely confident they fully understand the objectives of the sampling to enable these necessary field decisions. Samplers should NOT be burdened by undo pressures to sample when conditions are not appropriate for the method (e.g., there should NOT be a binding contract that stipulates collecting a specific number of samples by a certain date, even if conditions are not appropriate).

1.8 Maintaining Linkages between the SCI and Important Associated Data

Because so many factors affect aquatic biota and the SCI results, it is imperative that all associated data (flow conditions, habitat scores, etc.) be linked to the SCI results, so that a determination may be made that each sample was, or was not, consistent with the study objectives. Indiscriminate use of SCI scores in the absence of these associated data will invariably result in inappropriate or incorrect environmental decisions. It is the responsibility of the staff and managers analyzing the data and making environmental decisions to fully understand the complexities associated with the SCI scores and use the data appropriately.

1.9 Overview of SCI Sampling Process

Fundamental to SCI sampling is the selection of the best available habitats, in the optimal flow, to collect the indicator organisms in the areas they typically inhabit. This was the manner by which all the reference and potentially disturbed sites for calibrating the SCI were sampled. If the "healthy" organisms are not found in their optimal living quarters (best habitat and flow) one may conclude that some disturbance (human or natural) was responsible for their absence. A pristine stream, if not sampled according to the SCI protocol (*e.g*., if one erroneously sweeps only sand or low velocity backwaters), will assuredly fail the SCI. Conversely, if the very best habitat and flow conditions are sampled in accordance with the SOP in a damaged system, the SCI result will accurately reflect the level of disturbance. Therefore, training and ethics of SCI samplers is very important. A biologically healthy site, if sampled poorly, will fail the SCI. A disturbed site with an impaired community will also fail, even when sampled with a bias toward the best available habitats, due to environmentally relevant reasons, not a sampling artifact. Samplers must thoroughly understand the concepts associated with the Standard Operating Procedures (SOPs), and consistently follow the SOPs in order to prevent sampling errors (see FT 7000 at: http://www.dep.state.fl.us/labs/qa/sops.htm.).

Sampling Site Selection and System Classification

First, the study objectives must be clearly understood, and a 100 m segment of stream that is appropriate to address the objectives should be selected as a sample site. For purposes of site selection, it is important to understand there are variations in a stream's flow, habitat and biota as it moves through the landscape, and this variability has great implications for the proper application of the SCI. Within a single reach of a stream or river, there are areas of higher and lower densities and diversities of macroinvertebrates. These differences occur both on the local scale (*i.e*., different qualities of in-stream habitats; snags vs. muck in a 100 m section) and the landscape scale (*i.e*., different flow regimes and habitat diversity over a 5-10 mile section of stream).

For defining our inherent biological expectations associated with the SCI, stream or river segments that generally had flow (except during seasonal droughts) and typical "stream" habitats were selected. In other words, the SCI should be applied to streams that have similar and comparable characteristics to those streams used in the calibration set. Comparing biological communities from "swamp-like", lake-like or tidally affected segments of streams to the biological expectations established for "typical" streams is not scientifically reasonable. Proper classification of the system type one is attempting to sample is another fundamental concept for appropriate application of the SCI. However, it should be noted that the SCI may be used to assess human alterations in habitat and hydrology via a logically designed study.

Over a short distance, a stream may change from a system with a well defined natural channel, good flow, and an abundance of habitats to a forested swamp with little to no defined channel and very little perceivable flow. If the system is behaving like a swamp, and not a flowing stream in that specific area, one would not expect the swamp-like segment to perform well on the SCI. Conversely, if a study is attempting to assess the detrimental effects of stream impoundment, it may be appropriate to sample a former stream segment which has been hydrologically modified to resemble a lake. Also, consider large rivers that become very wide with dramatic reductions in flow as they transition toward an estuarine situation. This area of the "river" may actually be acting more like a flow-through lake. Sampling these types of areas may result in inappropriate SCI failures, because of incorrect system classification (comparing "apples" to oranges").

Thus, atypical areas not representative of the stream reach should generally be avoided when using the SCI as an indicator of biological integrity, unless the study objectives dictate otherwise. For example, unless the study objectives are to determine adverse habitat effects of road construction, sampling right at or under a bridge (usually disturbed by channelization) should be avoided, as this area would not be representative of the stream reach.

Appropriate Antecedent Hydrologic Conditions

Water levels should be examined as outlined in Section 1.6 above to determine if conditions are appropriate for the purpose of the study. Samplers should be careful to consider how long habitats in the top 0.5 m of the surface have been inundated. If the habitats have been recently dry, they should not be sampled. This is most important when sampling large rivers where water levels can rise over 0.5 m without being easily observed. For larger systems, data from stage height recorders are typically available and the resulting hydrograph should be carefully examined to determine when conditions are appropriate for sampling. Samplers need to develop intimate familiarity with the hydrology of streams in their regions.

Optimal Habitat Selection

Once it's been decided the hydrologic conditions are suitable for the objectives of the study, the sampler must identify the best available habitats where the macroinvertebrates actually reside. This is accomplished by performing the habitat assessment procedures to determine the types and quantity of substrates present (see FT 3000, found at: http://www.dep.state.fl.us/labs/qa/sops.htm.).

The dip net sweeps are apportioned by determining the number of productive habitats (roots, woody debris, leaf material, macrophytes or rock) present with a surface area greater than 2 m^2 (see SOP). When targeting specific substrates to sample in particular areas of the stream (best available habitats), samplers should keep in mind how macroinvertebrates use the substrates. It is important to "think like a bug". Some examples are:

- The invertebrate taxa important for calculating many of the SCI metrics (*e.g.*, sensitive taxa, Trichoptera, Ephemeroptera, filter feeders) are rheophyllic, meaning they prefer areas with higher water velocity, which also often translates into areas with higher concentrations of dissolved oxygen and food availability. Therefore, leaf packs that are in the main flow are preferred over leaf mats, which tend to be associated with lower velocity. Additionally, snags, roots, macrophytes and rocks in the flow are better habitats than the snags, roots, etc. in lesser flow or backwater areas.

- Organisms use the substrate as refuge from predators (e.g., fish, other invertebrates) and as a place to feed. Fine fibrous roots are preferred substrates, since they have more surface area and therefore more areas to hide, when compared with larger diameter roots. Similarly, snags with softer, deteriorating bark have more hiding places and attachment points for organisms (e.g., net spinning caddisfly filter feeders, hellgrammites) than fresh, smooth snags (e.g., cypress knees). This makes the deteriorating snag with many crevices a much preferred habitat. Similarly, jagged rocks with a rough architecture (i.e., with nooks and crannies) are preferred over smooth rocks.
- Since aquatic organisms need to live in the water, habitats that are constantly inundated with water are preferred over ones that go dry. For example, samplers should focus on the types of aquatic macrophytes that can survive long periods of inundation rather than those species which typically may be exposed to air for long periods. When terrestrial plants are seen submerged in a stream, it is a "dead giveaway" that the water level at a site has recently increased, and depending on the magnitude of the increase, aborting the trip should be given serious consideration.

Sampling Technique

Another important aspect of the SCI concerns the sampler's ability to actually remove the organisms from the substrates and properly collect them into the dip net. Samplers absolutely must provide sufficient agitation of substrates to dislodge the organisms, and ensure that all organisms are captured (into the net) without loss.

Based on this guiding principle, here are important sampling technique issues to be aware of:

- The opening of the dip net should always be placed perpendicular to the flow and the net should be placed downstream of any agitation so that organisms flow into the net.
- When agitating the substrate, the material (water, detritus, plus organisms) should be directed into the mouth of the dip net, using hands or a brush (scrub INTO the net, not parallel to it). Similarly, the substrates should be agitated very close to (or inside of) the dip net to avoid loss of organisms. For example, roots, remove-able rocks and snags, and submersed macrophytes should be agitated inside the bag of the dip net, where large snags, rocks, macrophytes, and sand should be sampled as close to the dip net opening as possible.
- Leaf pack material should be placed directly into the net and the organisms dislodged "one leaf at a time" before discarding excess leaves. It is critical

that there be NO LOSS of organisms during any field reduction of leaf material.

- It is important to vigorously shake and scrape all surfaces of the habitats at least 3 times, while having the net situated in a manner such that no organisms are lost.
- When sampling sand, penetrate the sand with fingers, to approximately 2 cm deep, and using a pulling motion, draw the organisms from the sand into the waiting dip net. Feel for partially buried bivalves and ensure they are placed in the net.
- For leaf mats, only sample the top 2 cm to avoid the anoxic layers below.
- Large rivers can be sampled from the bow of a boat (best for reachable snags in deep areas) or by wading along the shoreline. When wading large rivers, be particularly sure that the sampled habitats are in areas of adequate water velocity (not in a backwater area) and have been sufficiently inundated.

Field Sorting as a Training Tool

Field sorting at reference sites is a useful activity for a “sampler-in-training” to learn whether their selection of habitats and dip netting techniques are effective for capturing macroinvertebrates. After a sampler chooses a particular habitat and samples it, they should bring the contents of the net to the stream bank and ,using a white tray, sort through the material searching for organisms. Before sorting, the material in the net should be thoroughly rinsed with site water to eliminate turbidity. During sorting, only a small amount of material should be placed into the tray with about a centimeter of site water, so that approximately half of the white background is visible. Samplers should systemically search the tray and ,using forceps and pipettes, remove organisms for additional examination with a hand lens. Samplers need to become familiar with the basic orders and families of aquatic macroinvertebrates, as outlined in DEP SOP Table LT9700-1. Although there are many comprehensive taxonomic guides, a useful field book for beginners is “A Guide to Common Freshwater Invertebrates of North America”, by J. Reese Voshell, Jr., published in 2002 by the McDonald and Woodward Publishing Company, Blacksburg, Virginia. During field sorting, the sampler should compare the relative diversity of taxa found in individual sweeps taken from various habitats and flow regimes. This type of systemic examination will provide immediate feedback regarding the degree of success associated with the sampler’s field decisions.

Apprenticeship

Because of the complexities mentioned above, DEP recommends that experienced SCI samplers contribute to the training of novice staff. The goal of the training is to produce SCI samplers able to demonstrate the necessary critical thinking skills and sampling technique required by the SOP. Training should consist of numerous field visits (minimum of 12) at a variety of sites (starting at reference sites, followed by disturbed sites) and different water levels, where novice staff receives instruction from the experienced staff on the concepts presented here. As training progresses, the novice staff should gradually demonstrate the required best professional judgment and sound sampling technique (see Section 1.11 for training checklists). Once training has been completed, a field audit to assess a sampler's ability to adhere to the SOPs may be scheduled.

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1.10 SCI Training Materials, Training Requirements, and Checklists

See: www.dep.state.fl.us/labs