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

FDEP
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Contents

Contents 2

1. Introduction 3
2. How Objectives Affect SCI Sampling Decisions and Interpretation 9
3. Water Level and the SCI 30
4. Overview of SCI Sampling Process 36
5. SCI Training 41
6. SCI Data Usability 42
7. SCI Training Materials, Training Requirements, and Checklists 46
8. Literature Cited 46

1. Introduction

The Stream Condition Index (SCI) is a biological assessment procedure that measures the degree to which flowing fresh waters support a healthy, well-balanced biological community, as indicated by benthic macroinvertebrates. The BioRecon is a companion tool to the SCI, which provides screening level information. For both the SCI and BioRecon, the principles in this document must be followed for successful application of the methods. In addition to the concepts presented in this document, samplers, data analysts, and resource managers who use the SCI (or BioRecon) must also read DEP SOP SCI 1000 and BRN 1000, for the training, quality assurance, sampling, laboratory, and index calculation Standard Operating Procedures. Furthermore, those wishing to implement and interpret the SCI must also read and understand "Development of Aquatic Life Use Support Attainment Thresholds for Florida's Stream Condition Index and Lake Vegetation Index" (DEP-SAS-003/11).

Because multiple natural and anthropogenic factors affect biological results (**Figure 1**), it is critical that SCI users fully understand the method to ensure any conclusion regarding potential human environmental effects are scientifically defensible.

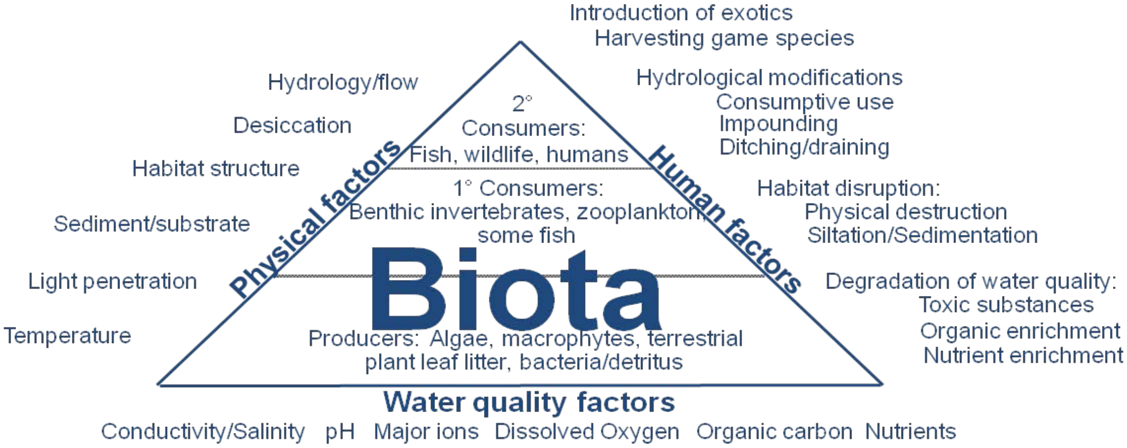

Figure 1. Many factors affect biological community composition. To conclude that human disturbance is primarily responsible for biological degradation, reasonable knowledge of the influence of natural factors is essential.

1.1. SCI Samplers Must Exercise Best Professional Judgment

Only an experienced, qualified SCI sampler is able 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 and objective (*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). Additionally, samplers must collect sufficient meta-data (*e.g*., water level, habitat, photographs) to document, and assist with determining the usability of a given SCI result for a particular purpose or objective (see section 6 below).

1.2. Maintaining Linkages between SCI and Important Related Data

Because many factors affect aquatic biota and the SCI results, it is imperative that all associated data (flow conditions, habitat scores, other physical factors affecting a site, etc.) be linked to the SCI results, so that a determination may be made that each sample is, or is not, consistent with the study objectives. There is a high likelihood that indiscriminate use of SCI scores, in the absence of these associated data, will 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 to use the data appropriately. Samplers must also assist data analysts and managers in the determination that SCI results are, or are not, appropriate for a given objective.

1.3. Summary of the Development of SCI Metrics

DEP considered a diverse array of community attributes (**Figure 2**) and used the Human Disturbance Gradient approach to objectively select metrics that respond to human influences in a predictable manner (Fore *et al.* 2007). The Human Disturbance Gradient is composed of four factors:

- The Landscape Development Intensity Index (Brown and Vivas 2004);
- Habitat Assessment scores (DEP SOP FS 3000);
- Hydrologic Modification Index; and
- 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 Human Disturbance Gradient was used as the x-axis for testing a wide variety of biological attributes associated with the measurement of ecological integrity (**Figure 2**). **Figure 3** 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; and

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

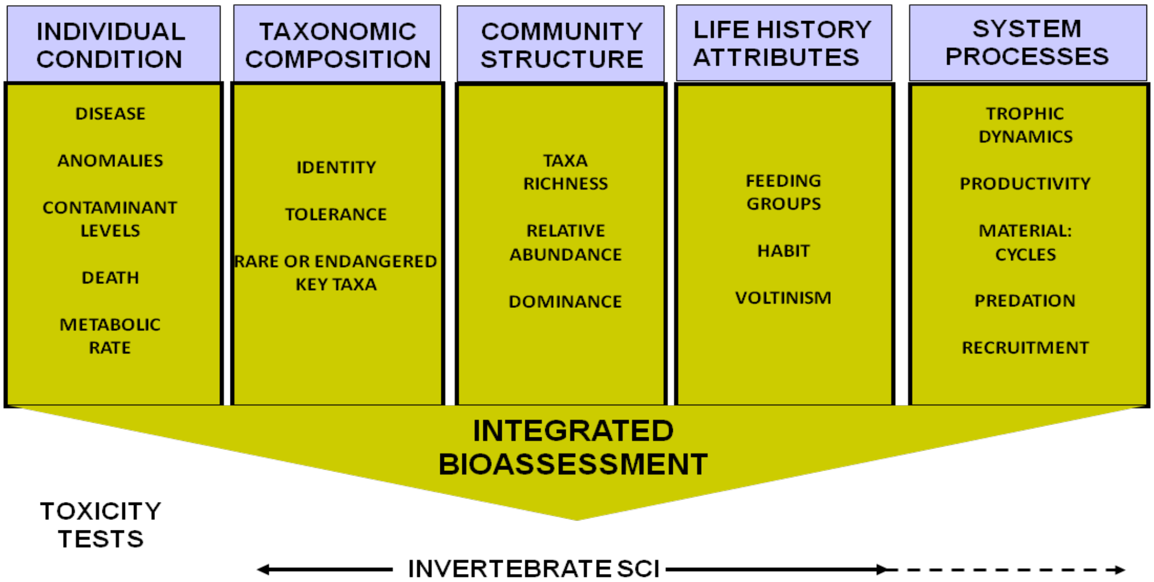

Figure 2. Major attribute categories, and example metrics, for determining biological integrity.

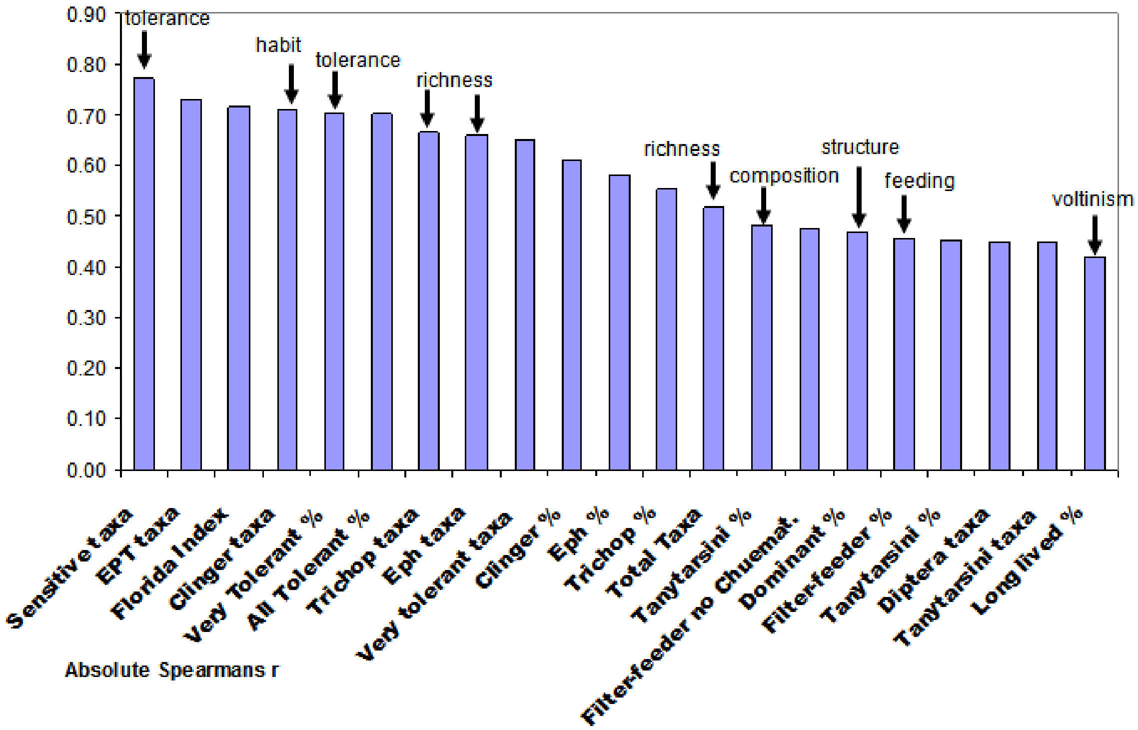

Figure 3. Correlation between various metrics and the Human Disturbance Gradient. Arrows indicated metrics selected for the SCI, and associated attribute group.

1.4. The 10 SCI Metrics

A description of the ten metrics is presented here. When aggregated into the SCI, these provide a comprehensive and robust assessment of stream biological health.

- ***Percent Tanytarsini***

Tanytarsini midges are sensitive to disturbance, so the metric was included in the SCI as the best available measure of the chironomid assemblage, which is an important group in stream invertebrate communities.

- ***Number of Sensitive Taxa***

Lists of sensitive and very tolerant macroinvertebrates were established by analyzing the responses of individual species to the HDG (Fore *et al.* 2007a). The number of taxa selected as sensitive equaled around 12% of the taxa tested. Many sensitive species belonged to the Ephemeroptera, Trichoptera or Odonata; several chironomids were also included. All the Plecoptera were included as sensitive taxa (**Figure 4**).

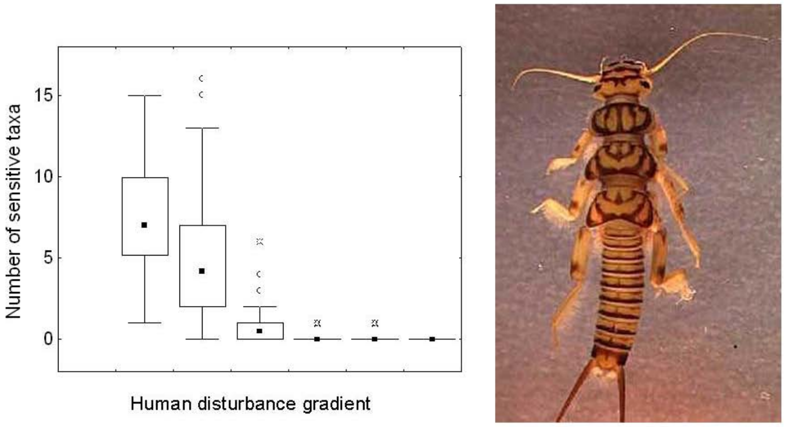

Figure 4. Response of the number of sensitive taxa metric to the HDG. The photo is of a plecopteran (stonefly).

- ***Percent Very Tolerant Taxa***

The number of very tolerant taxa was approximately 10% of the taxa tested. The percent very tolerant individuals were highly correlated with the HDG (**Figure 5**).

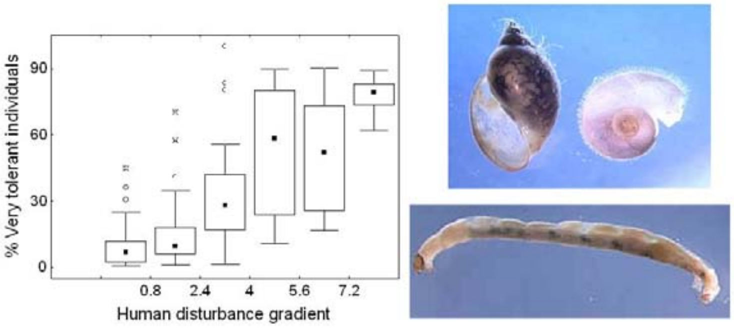

Figure 5. Response of the percent very tolerant metric to the HDG. Photos are of lunged snails and tolerant midges.

- ***Number of Total Taxa***

- ***Number of Ephemeroptera Taxa***

- ***Number of Trichoptera Taxa***

The number of different types of organisms present and the richness of the Trichoptera (caddisflies) and Ephemeroptera (mayflies) have historically been shown to decrease with human disturbance. **Figure 6** depicts the response of the number of Ephemeroptera taxa 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).

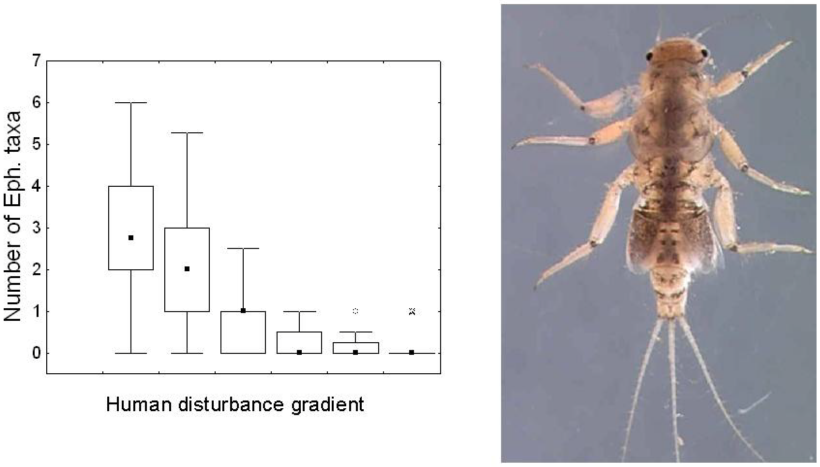

Figure 6. Response of the Ephemeroptera metric to the HDG. The photo is of *Tricorythodes*, a sensitive mayfly.

- ***Percent Dominant Taxon***

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.

- ***Percent Filterers or Suspension Feeders***

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 7**). 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.

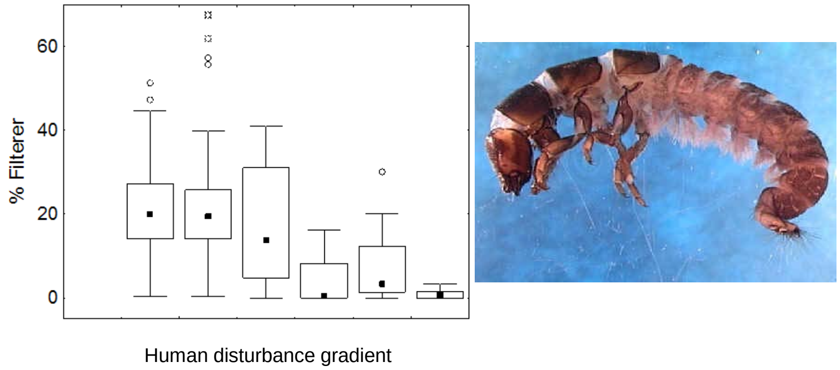

Figure 7. Response of the percent filter-feeder metric to the HDG. The photo is of a net-spinning caddisfly.

- ***Number of Long-Lived Taxa***

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 8**).

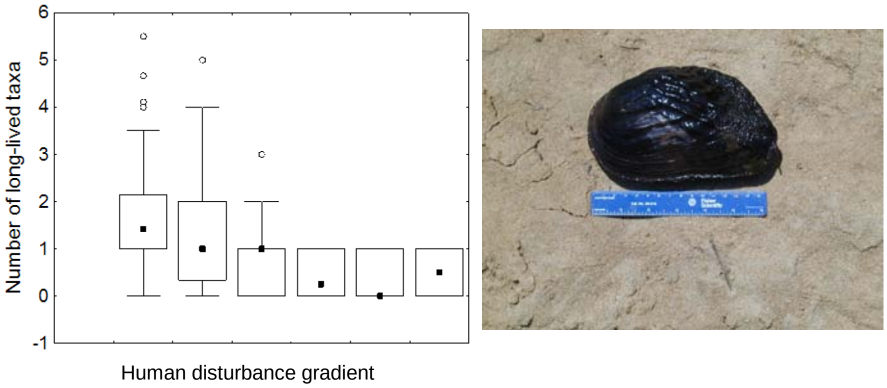

Figure 8. Response of the long-lived taxa metric to the HDG. The photo is of a mollusk, the threatened purple bank climber.

- ***Number of Clinger Taxa***

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 9**).

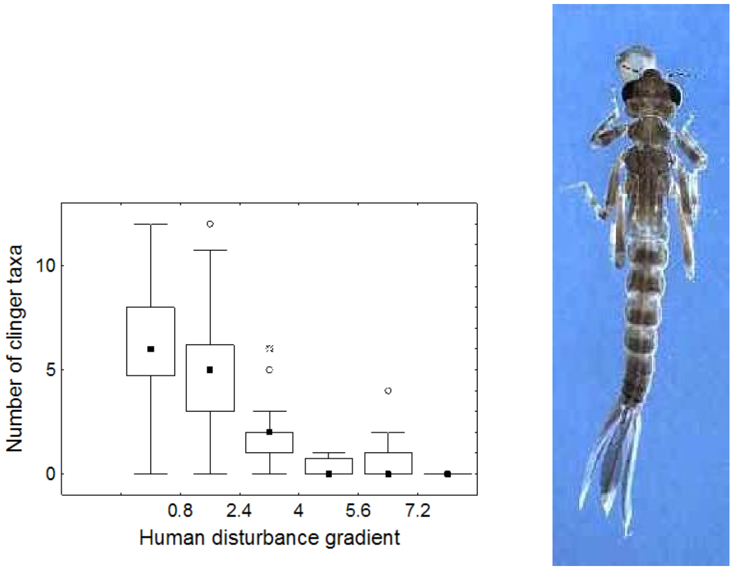

Figure 9. Response of the clinger taxa metric to the HDG. The photo is of a damselfly larvae.

2. How Objectives Affect SCI Sampling Decisions and Interpretation

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 criteria).

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

2.1. Water Quality Investigations for the Watershed Assessment/Total Maximum Daily Load Program and for Determining a Causative Pollutant

The objective for an impaired waters assessment/causative pollutant identification study is to decide if water quality issues are adversely affecting biological health. To list a waterbody on the verified list of impaired waters, 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, may dominate the invertebrate response. The SCI SOP contains a provision that SCI sampling be postponed for 6 months after a site has gone *completely* dry (with no refugia for organisms) and then has begun flowing again, before sampling (although additional information on this issue, including use of site specific information, is provided in section 3 below). This wait will help ensure that desiccation was not the most prominent factor influencing the SCI score. Similarly, stream organisms are rheophyllic ("flow loving"). If water velocity is very low (standing water, stagnant conditions), it is very likely to adversely affect the assemblage of organisms, even if water quality is excellent. Therefore, sampling for impaired waters assessments/causative pollutant identification purposes shall be conducted during periods when water velocity has been 0.05 m/sec or greater for at least 28 days (one month). 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 an impaired waters assessment /causative pollutant identification 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 means that the investigator must establish sampling sites (where possible) in stream reaches with adequate substrate diversity and availability, intact stream morphology (little or no artificial channelization), adequate flow, and optimal riparian buffer zones**. Figure 10** shows an example of a site where habitat and hydrology are significant adverse influences, meaning that an alternate site in the stream segment should be selected for SCI sampling, if one is available. If the entire stream reach has habitat and hydrological limitations and funding for restoration is not available, reclassification to a Class III-Limited category should be considered (as described in Chapters 62-302.400 and 62-302.800, F.A.C. Note that deleterious sediment input may result in habitat smothering, and that restoration would involve upland erosion control and other Best Management Practices.

Figure 10. A site where habitat and hydrology significantly and adversely influence the biological community, meaning that an alternate site in the stream segment should be selected for SCI sampling (if possible) during an impaired waters assessment /pollutant identification study. If habitat and hydrological limitations occur throughout the entire stream reach, and funding for restoration is not available, reclassification to a Class III-Limited category should be considered.

Specific conductance (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 (*e.g.*, at tidally influenced systems) or due to human sources. The SCI was designed for freshwater streams, and as such, it would not be appropriate to use the tool where conductivity is naturally elevated *(e.g.*, near estuarine areas). However, if a human discharge has artificially elevated a stream's conductivity, the SCI may be used to document the resulting potential adverse community response. One must take care to assess the source of the conductivity when deciding the appropriateness of the SCI.

In conclusion, if flow and habitat limitations are controlled for during an impaired waters assessment /causative pollutant identification study, and sufficient water quality data are collected, the water quality factor(s) responsible for any observed biological degradation are more effectively identified.

2.2. Point Source Studies

Point source studies involve an evaluation of 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. To assess the influence of the discharge, an upstream-downstream SCI study is routinely employed, while controlling for important variables (*e.g.,* habitat) between the upstream (control) and downstream (test) sites. Selection of similar habitats from areas of similar water velocity is important to determine if the effluent is associated with any changes in the SCI scores. Additionally, the use of replicate sampling stations, for both control and test sites, will better characterize the variability of the biological data. If reductions in the SCI scores occur between the control and test sites, the intra-site variability and the magnitude of the change should be assessed, as well as potential categorical shifts.

2.3. Best Management Practices (BMPs) Effectiveness Studies

Previous studies on the effectiveness of forestry best management practices, using the SCI, 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 11**). 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 .

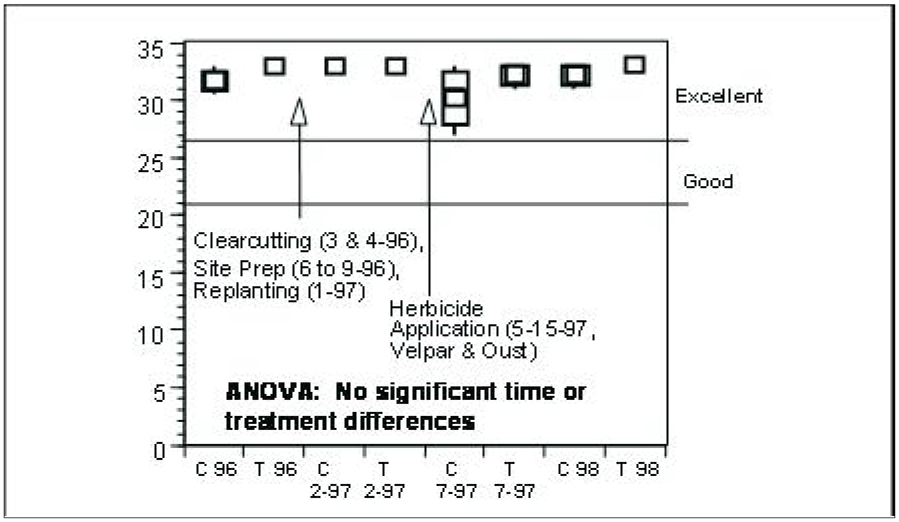

Figure 11. SCI results of a Before-After-Control-Impact study assessing the effectiveness of forestry Best Management Practices. "C" and "T" mean control and test sites, respectively. Note that a different version of the SCI was used during the time period, but the concepts apply to the current SCI.

2.4. Stream Restoration Studies

The objectives for a stream restoration study are 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 ; and
- Water quality.

The investigator shall measure each of these 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 verified list impaired waters list when habitat, not water quality, was the limiting issue).

2.5. 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 (e.g., within 6 months) or during periods of stagnant water velocities (not generally appropriate for conducting the SCI SOP), it is important that the resulting data (probable SCI failures) not be misinterpreted as water quality issues (see section on maintaining associated data with the SCI below).

2.6. Integrated Water Resource Monitoring (Status and Trends) Program

The Integrated Water Resource Monitoring program (IWRM, aka Status and Trends) 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 76 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. Water samples are collected monthly at all surface water Trend sites, and the SCI is conducted annually.

The second component of the program uses a random stratified (probabilistic) sampling network, 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 a “sampling period.” The design is based on a set geographic boundary, or reporting unit (“zone”) that follows the Water Management District boundaries.

The SCI tool was adopted as part of the 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 the system is not functioning as a stream or river (it is more like a lake, estuary, wetland, marsh, prairie, canal, ditch, etc.);
- Do not sample if the system is currently dry or disconnected, or has been completely dry within 6 months prior to the site visit. If this cannot be determined with confidence, do not sample;
- Do not sample if flood conditions exist and water levels are > 0.5 meters above normal;
- Do not sample if the system is tidally influenced (regardless of conductivity values);
- Do not sample if the system is a spring run with conductivity values ≥ 600 µmhos;
- Do not sample if the average velocity is ≤ 0.05 m/s or has been < 0.05 m/s in 28 days prior to the site visit. If this cannot be determined with confidence, do not sample;
- Do not sample if conditions are unsafe; and
- Do not sample in the South Florida Bioregion (south of Lake Okeechobee).

The assumption was made at the onset of the use of the SCI tool in the IWRM program that it applied to all 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, even if they are connected to waters of the state, are currently excluded for sampling because they are not functioning as a stream or river.

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 zone. 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 zone.

For the Status Network, the designated water quality sampling point must remain within the 100-meter Habitat Assessment (HA) and SCI stretch, but the stretch may be positioned upstream (water quality sampling point at the zero marker) or downstream (water quality sampling point at the 100 marker) as necessary in order to provide the most representative stretch.

For the Trend network, samplers are permitted to orient the 100 meter stretch upstream or downstream from the water quality sample collection point as necessary in order to provide the most representative habitat stretch for the system. The designated water quality sampling point does not have to reside within the 100-meter stretch, but it must be no farther than 200 meters away from the HA stretch. However, if moving the HA stretch 200 meters away still does not meet the acceptable criteria for performing the HA/SCI, do not perform the HA/SCI.

2.7 Use of the SCI and Measures of Floral Health to Assess Achievement of Nutrient Standards

This section describes the evaluations conducted by DEP to assess whether a stream attains the narrative nutrient criterion in Rule 62-302.530(47)(b), F.A.C., pursuant to the provisions in Rule 62-302.531(2)(c), F.A.C. This approach evaluates water chemistry and biological (flora and fauna) and physical information from the waterbody to determine if nutrient concentrations are causing an imbalance in flora or fauna in a given stream. Because of the complexity associated with nutrient enrichment effects, no single assessment tool is adequate to evaluate all potential impacts, and instead, a weight-of-evidence evaluation must be conducted.
This section discusses:

- The nutrient enrichment conceptual model for streams;
- The process for numerically interpreting the narrative nutrient criterion in streams;
- Available procedures for evaluating the floral community in the stream, including chlorophyll *a* levels, periphyton abundance and species dominance (as measured using the Rapid Periphyton Survey [RPS]), and nuisance macrophyte distribution (as measured using the Linear Stream Vegetation Survey [LVS]);

- Evaluating the faunal community in the stream using the Stream Condition Index (SCI) or BioRecon;
- Efficiently collecting the information during one sampling event; and
- Examples of a weight-of-evidence approach for determining achievement of nutrient criteria.

2.7.1 Nutrient Enrichment Conceptual Ecological Model for Streams

Nutrients are naturally present in aquatic systems and are necessary for the proper functioning of biological communities. Nutrient effects on aquatic ecosystems are moderated in how they are expressed by many natural factors (*e.g.*, light penetration, hydraulic residence time, presence of herbivore grazers and other food web interactions, and habitat considerations). As a result, determining the appropriate protective nutrient regime is largely a site-specific undertaking, requiring information about ecologically relevant responses.

To evaluate ecosystem health, it is important to acknowledge that adverse nutrient enrichment effects follow a conceptual ecological model (**Figure 12**). When anthropogenic nutrient loading or concentrations exceed a system's assimilative capacity, the primary response consists of changes to the primary producer communities (periphyton, phytoplankton, or vascular plants), and excess production of plant biomass. In turn, this enhanced floral biomass could lead to habitat loss (*e.g.*, from excess periphyton smothering or nuisance plant biomass accumulation), food web alterations (*e.g.*, dominance of taxa that thrive in nutrient/organic matter enriched conditions), and/or low dissolved oxygen (DO) from decomposition of plant biomass or respiration. This chain of events is ultimately reflected in meaningful biological endpoints, such as excessive algal mats, excess water column chlorophyll *a*, excess nuisance vascular plant growth, and/or failing Stream Condition Index (SCI) scores. These adverse biological endpoints constitute imbalances of aquatic flora and/or fauna.

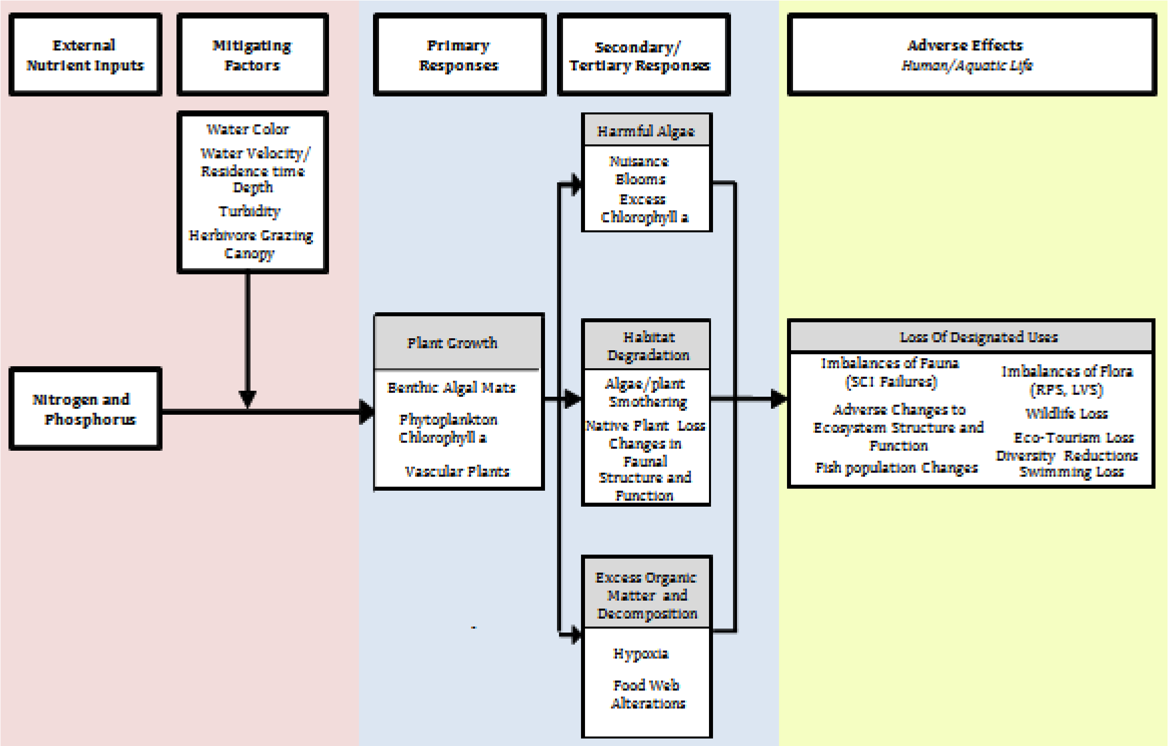

Figure 12. Simplified nutrient enrichment conceptual model used to assess potential adverse effects of nutrients on aquatic life and human uses in streams. Relationships between nutrients and biological responses are highly influenced by site-specific and mitigating factors.

Conversely, if data show that biological health is fully supported in an aquatic system (no adverse responses consistent with the ecological model), it may be concluded that the associated nutrient regime is inherently protective of the waterbody, and the narrative nutrient criterion is achieved.
When conducting nutrient studies, it is important that sampling locations and other environmental conditions (canopy cover, habitat, water depth and flow, etc.) are determined to be representative of the system and that water quality data be collected in the same waterbody segment as the biological monitoring stations. The stations shall be established in a manner consistent with the study design concepts described in the document titled *Development of Type III Site Specific Alternative Criteria (SSAC) for Nutrients* (DEP-SAS-004/11).

2.7.2 The Process for Numerically Interpreting the Narrative Nutrient Criterion in Streams

The narrative nutrient criterion in Rule 62-302.530(47)(b), F.A.C., states that “in no case shall nutrient concentrations of a body of water be altered so as to cause an imbalance in natural populations of aquatic flora or fauna.” The method for numerically interpreting this narrative nutrient criterion, on a site-specific basis, is provided in Rule

62-302.531, F.A.C, using a hierarchical process (**Figure 13**). This hierarchical scheme specifies a prioritization for determining the numeric nutrient criteria that apply to a given waterbody. Beginning at the top of **Figure 13,** if there is a Total Maximum Daily Load (TMDL), Site Specific Alternative Criterion (SSAC), Water Quality Based Effluent Limitation (WQBEL), or other DEP-approved action involving nutrients for a waterbody (*e.g*., Reasonable Assurance derived values), one of these would be the applicable nutrient criteria. If not, values based on cause-effect relationships between nutrients and biological response (*i.e*., springs and lakes) would apply. If no cause-effect relationship has been established, such as is the case for Florida streams, reference-based Nutrient Thresholds, used in conjunction with biological information, become the applicable interpretation of the narrative nutrient criterion. If none of the above are available, then the narrative nutrient criterion continues to apply to the waterbody, which could be numerically interpreted on a site-specific basis.

If a site specific interpretation has not been established for a stream, reference stream-based Nutrient Thresholds (**Table 1, Figure 14**), *combined with information on aquatic flora and fauna*, are used to interpret the narrative nutrient criterion.

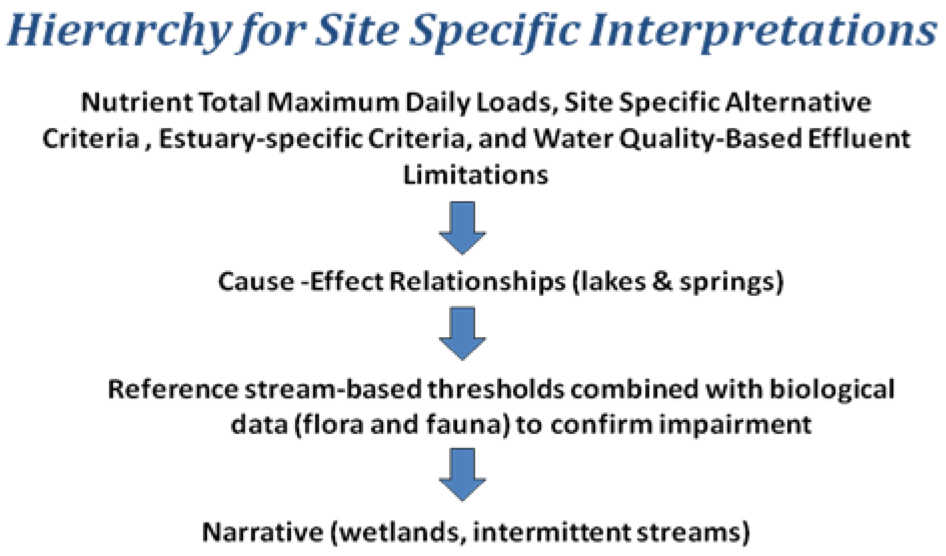

Figure 13. The hierarchy for numerically interpreting the narrative nutrient criterion.

Table 1. Reference stream-based Nutrient Thresholds.

Nutrient Region	Total Phosphorus Threshold	Total Nitrogen Threshold
Panhandle West	0.06 mg/L	0.67 mg/L
Panhandle East	0.18 mg/L	1.03 mg/L

North Central	0.30 mg/L	1.87 mg/L
Peninsula	0.12 mg/L	1.54 mg/L
West Central	0.49 mg/L	1.65 mg/L
South Florida	No numeric nutrient threshold. The narrative criterion in paragraph 62-302.530(47)(b), F.A.C., applies.	

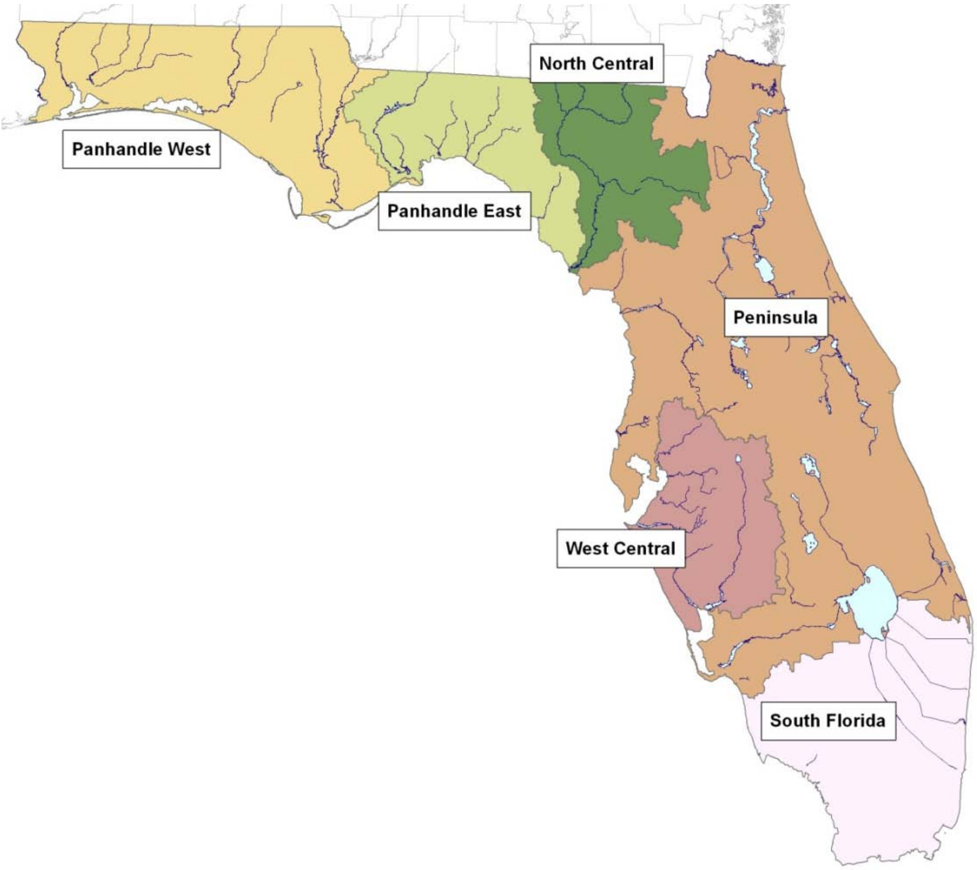

Figure 14. Map of stream Nutrient Regions.

These stream Nutrient Thresholds, which are the same as those promulgated by EPA (http://water.epa.gov/lawsregs/rulesregs/florida_index.cfm) are based on:

- The 90th percentile data distribution of rigorously verified, minimally disturbed streams in the Panhandle West, Panhandle East, North Central, and Peninsula regions; and
- The 75th percentile data distribution of biologically healthy streams (those with an average score of >40 on the Stream Condition Index [SCI]) in the West Central Region.

Note that stream Nutrient Thresholds do not apply in South Florida.

It is important to recognize that the method for deriving the Nutrient Thresholds directly results in the expectation that some of Florida's biological healthy streams will exceed the Thresholds. It is for this reason, and because nutrient thresholds may not be appropriate for every Florida waterbody, that Rule 62-302.531, F.A.C., contains a provision, described below, that allows use of *biological information to fully evaluate achievement of the criterion*.

For streams, if a site specific interpretation pursuant to paragraph 62-302.531(2)(a) (TMDL, SSAC, Level II WQBEL or RA Plan) has not been established, Nutrient Thresholds are used to interpret the narrative nutrient criterion in combination with biological information. The narrative nutrient criterion in paragraph 62-302.530(47)(b), F.A.C., shall be interpreted as being achieved in a stream segment if:

- Information on chlorophyll *a* levels, algal mats or blooms, nuisance macrophyte growth, and changes in algal species composition do not indicate an imbalance in flora or fauna; AND EITHER
- The average score of at least two temporally independent SCIs performed at representative locations and times is 40 or higher, with neither of the two most recent SCI scores less than 35 (*i.e*., no faunal imbalances), OR
- The Nutrient Thresholds (expressed as annual geometric means) in **Table 1** are not exceeded more than once in a three year period.

In cases where the Nutrient Thresholds are exceeded but there are no imbalances in both aquatic flora (phytoplankton, periphyton, vascular plants) AND fauna (invertebrate community), the narrative criterion is achieved. DEP has developed techniques to measure both flora and fauna, as described below, to determine when there is, or is not, an imbalance. Many of these evaluations can be conducted during the SCI sampling exercise. DEP continues to explore methods for gathering this type of biological information in aquatic systems. To stay informed of DEP bioassessment efforts, visit our website at: http://www.dep.state.fl.us/water/bioassess/index.htm
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2.7.3 Evaluating Stream Flora: Attached Algal Communities

DEP conducted a comprehensive study of stream periphyton in Florida in an attempt to formulate a multi-metric index for assessing human disturbance, including nutrient effects (see Development of a Stream Diatom Index). Preliminary analysis indicated that the best potential metrics were percent sensitive diatom cells, percent tolerant diatom cells, percent diatom cells that prefer high oxygen, percent cells that prefer oligotrophic conditions, and van Dam's weighted index for trophic status. These metrics were transformed into a dimensionless index, the Stream Diatom Index (SDI). Unfortunately, further analysis showed that the SDI was most highly correlated with pH. When the data were categorized according to pH, a *relationship between SDI and human* *disturbance was not observed*. Based on these findings, DEP determined that the SDI is not appropriate for use as biocriteria due to its poor correlation with human disturbance and its strong association with pH, meaning that other methods for assessing stream floral health were needed.

DEP has also investigated using a modification of EPA's Rapid Periphyton Survey (RPS) to quantify the extent (coverage) and abundance (thickness) of attached algae (periphyton) and found that the RPS was an *effective tool to demonstrate a lack of abundance of nuisance or problematic algal growth*. In subsection 62-303.200(10), F.A.C., a nuisance species is defined as "...species of flora or fauna whose noxious characteristics or presence in sufficient number, biomass, or areal extent may reasonably be expected to prevent, or unreasonably interfere with, a designated use of those waters."

To conduct the RPS method, a trained biologist surveys a 100 meter segment of a stream or river by establishing 11 transects across the waterbody at 10 meter intervals, and determining the presence and thickness of algae at 9 points along each transect, for a total of 99 sampling points. Additionally, a densiometer measurement to determine canopy coverage is taken near the center of each transect. The following ranks are used to quantify algal thickness:

- "0" = algae are absent (a rough surface with no algae).
- "1" = algae less than or equal to 0.5 mm OR no algae visible but surface is slimy (including muck and biofilm),
- "2" = greater than 0.5 mm to 1 mm,
- "3" = greater than 1 mm to 6 mm,
- "4" = greater than 6 mm to 20 mm,
- "5" = greater than 20 mm to 10 cm, and
- "6" = algae greater than 10 cm.

In DEP's experience, if a high percentage of the stream is covered by relatively thick algae (RPS rank $\geq$ 4), there may be adverse effects on the stream. Therefore, if the percentage of sampled points with a thickness rank of 4-6 is 20% or greater, the biologist collects a composite sample of the dominant groups of periphyton in the stream segment for lab identification of the dominant algal taxa. If autecological information is available for the dominant taxa, this is also qualitatively evaluated. Two determine persistence, two temporally independent RPS are routinely conducted. Rank 4-6 periphyton growth is naturally observed at minimally disturbed Nutrient Benchmark streams and at streams that have healthy SCI scores (> 40). Nutrient Benchmark streams had Landscape Development Intensity Index (LDI) values < 2, no point source discharges, optimal habitat, and were subjected to a comprehensive field evaluation by DEP scientists to ensure that they were minimally disturbed by humans. DEP is currently conducting a comprehensive analysis of RPS data to determine how the RPS correlates with water chemistry, the Stream Condition Index (SCI), and independent measures of human disturbance such as the LDI. Preliminary analysis of RPS data collected at 467 RPS sites statewide, including RPS collected at the Nutrient Benchmark streams used to derive stream Nutrient Thresholds and at other biological healthy sites, yields the information described below.

RPS rank 4-6 coverage at Nutrient Benchmark streams ranged from 0% to 66%, with a mean value of 6% and a 90th percentile value of 25%. RPS rank 4-6 coverage at all biologically healthy sites (as indicated by stream condition index scores > 40), ranged from 0% to 91%, with a mean value of 8% and a 90th percentile value of 32%. Therefore, if a stream exhibits RPS rank 4-6 percent coverage between the mean percent observed at these minimally disturbed and healthy sites (6-8%) and the associated 90th percentile values (25-32%), this would be considered an indication of no imbalance of flora. DEP also considers the persistence of periphyton coverage during this evaluation (see examples in section 2.7.8 below).

Both DEP and EPA used a change point in RPS algal coverage (expressed as persistent coverage of RPS rank of ≥ 4) as a line of evidence to link nitrate enrichment to adverse biological effects when deriving the springs nitrate criterion (**Figure 15**).

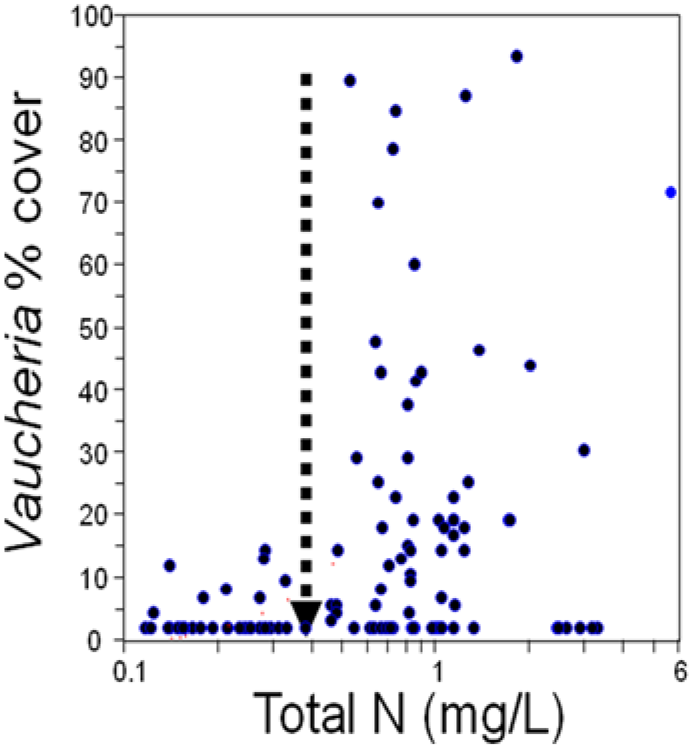

Figure 15. The relationship between persistent *Vaucheria* percent cover (coverage of RPS rank of ≥ 4) was a line of evidence used to develop the 0.35 mg/L nitrate-nitrite criterion in Florida springs.

2.7.4 Evaluating Stream Flora: Vascular Plant Communities

Another line of evidence to determine if streams are healthy is relative lack of nuisance macrophyte growth by certain vascular plant taxa that may interfere with designated uses of a waterbody. The Linear Vegetation Survey (LVS) is a rapid assessment tool for evaluating ecological condition based on vascular plants. To employ the LVS method, a trained biologist surveys a 100 meter segment of a stream, divides the stretch into 10 meter sampling units, and identifies the plant species present to the typical high water mark, including submersed, floating, and emergent plants. The sampler then determines the dominant or co-dominant species by estimating the 1 or 2 species with the largest areal extent. It is also possible to have no dominance or co-dominance, in which case, no dominant is assigned. Note that quantitative mapping of vascular plant communities is also part of the Habitat Assessment process.

To assess the community composition (including sensitive taxa), the LVS takes into account each plant species' Coefficient of Conservatism (C of C) score (assigned by expert botanists as part of a DEP initiative in 2011) that indicates the plant's specific habitat requirements. Plants with higher C of C scores have the most fidelity to high quality, unaltered sites, while a plant with a score of 0 displays no such fidelity, and includes invasive exotic species listed by the Florida Exotic Pest Plant Council (FLEPPC). For a list of C of C scores, FLEPPC taxa, and other vascular plant attributes relevant to the LVS, see DEP SOP LVI 1000 Appendix LVI 1000-1.

FDEP reviewed the available LVS results for the minimally disturbed reference streams used by EPA for nutrient criteria development in Florida. Because many streams naturally have very little or no vegetation, interpretation of LVS data requires that a minimum of 2 m^2 of macrophyte coverage be present throughout a 100 m stream reach. DEP evaluated LVS data from 58 reference sites and found that 19 of these sites had sufficient plant growth (> 2 m^2) to further evaluate the LVS data. Based on these data, if a site's average C of C score is greater than or equal to 2.5, the plant community composition may be considered to be part of the reference site distribution. Similarly, this analysis showed that if frequency of occurrence of FLEPCC exotics at a site is less than or equal to 25% of the total plant occurrences, the site may be considered to be part of the reference site distribution. Therefore, if a stream exhibits a C of C score $\geq$ 2.5 and the frequency of occurrence of FLEPCC exotics is $\leq$ 25% of the total plant occurrences, this would be considered an indication of no imbalance of flora. However, it is important to acknowledge that invasive exotic species can occur even in the absence of nutrient impacts. Care should be taken to avoid incorrectly concluding that any occurrence of exotic plants is an imbalance of flora caused by nutrients.

2.7.5 Evaluating Phytoplankton/Chlorophyll a Data

During development of the Impaired Waters Rule (Chapter 62-303, F.A.C.), DEP solicited expert input on ecological effects of chlorophyll *a* on streams from a panel of Florida scientists. This expert panel concluded that annual average stream chlorophyll *a* values exceeding 20 ug/L constitutes evidence that aquatic life uses were not fully supported. DEP uses this value, expressed as an annual average not to be exceeded, as an impairment threshold in Chapter 62-303 F.A.C. However, neither the expert panel nor a review of stream chlorophyll *a* literature was able to identify a stream chlorophyll *a* value below 20 ug/L that definitively did, or did not, support aquatic life uses.

DEP also uses the presence of persistent phytoplankton blooms as an indicator of floral imbalances. An unacceptable phytoplankton bloom would consist of a situation where an algal species, whose noxious characteristics or presence in sufficient number, biomass, or areal extent may reasonably be expected to prevent, or unreasonably interfere with, the designated use of a waterbody. DEP evaluates the autecological information for the dominant bloom species, in conjunction with the associated chlorophyll *a* and the persistence of the bloom, as a line of evidence when assessing imbalances of flora.

As part of the analysis, DEP also compares measured chlorophyll a values to the range of chlorophyll *a* concentrations that were observed at minimally disturbed Nutrient Benchmark streams and at streams that have healthy SCI scores (≥ 40). Nutrient Benchmark streams had Landscape Development Intensity Index (LDI) values ≤ 2, no point source discharges, optimal habitat, and were subjected to a comprehensive field evaluation by DEP scientists to ensure that they were minimally disturbed by humans.

Annual geometric mean chlorophyll a at the 94 Nutrient Benchmark streams used to derive stream Nutrient Thresholds ranged from 0.2 ug/L to 17 ug/L, with a mean value of 2.0 ug/L and a 90th percentile value of 3.2 ug/L. Annual geometric mean chlorophyll a at 274 biologically healthy sites (as indicated by stream condition index scores > 40), ranged from 1.0 ug/L to 19 ug/L with a mean value of 2.1 ug/L and a 90th percentile value of 3.5 ug/L.

If a stream exhibits annual geometric mean chlorophyll a concentrations between the mean observed at these minimally disturbed and healthy sites (2.0-2.1 ug/L) and the associated 90th percentile values (3.2-3.5 ug/L), this would be considered a clear indication of no imbalance of flora. Conversely, if a stream exhibits annual geometric mean chlorophyll *a* values that exceed 20 ug/L, this is evidence of imbalances of flora. Streams with annual average chlorophyll values between 3.5 ug/L and 20 ug/L are evaluated on a site specific basis. DEP also considers the residence time, flow, climatological conditions, and size of the stream/river (i.e., stream order) when considering chlorophyll *a* expectations within this range.

DEP additionally assesses chlorophyll *a* using a temporal trend test (a Mann's one-sided, upper-tail test for trend, with a 95% confidence interval) in conjunction with the chlorophyll *a* impairment threshold. The observation of a statistically significant increase in chlorophyll *a* in a stream is another line of evidence used by DEP to determine floral imbalances.

2.7.6 Evaluating Stream Fauna: SCI

As discussed earlier, the Stream Condition Index (SCI) is a biological assessment procedure that measures the degree to which flowing fresh waters support a healthy, well-balanced biological community, as indicated by benthic macroinvertebrates. Ten metrics that quantitatively describe stream community structure and function are summarized as a dimensionless index (the SCI), which scores between 0 and 100. DEP established a protective threshold SCI score based on a combination of the reference site data distribution and EPA's Biological Condition Gradient approach (see Development of Aquatic Life Use Support Attainment Thresholds for Florida's SCI and LVI – DEP, October 24, 2011).

DEP and EPA have concluded that a balanced faunal community is achieved if the average score of at least two temporally independent SCIs, performed at representative locations and times, is 40 or higher, with neither of the two most recent SCI scores less than 35. Attainment of the SCI threshold is an indication that the faunal community of the stream is not being adversely affected by nutrients to the extent that there is a loss in designated use. However, failure of the SCI threshold also does not mean that the stressor causing the loss of designated use is nutrients. Evaluation of other factors, as indicated by the nutrient enrichment model in 2.7.1 (including nutrient concentrations and floral communities) is useful information that could indicate nutrients are a factor. While the stressor may not be known, a failed SCI does indicate that fauna is not well-balanced.

The BioRecon is a companion tool to the SCI with six metrics and a total score of zero to ten. A BioRecon score of 4 is equivalent to an SCI score of 40 (see Stream Condition Index (SCI) Report). While BioRecon results may be considered as additional information, BioRecon should not be used as the only evidence (in the absence of SCI and other measures of floral health) to demonstrate attainment or nonattainment of biological health.

2.7.7 Efficiently Conducting the RPS, LVS, Habitat Assessment (HA), and SCI

As adopted by reference in the Quality Assurance Rule (Chapter 62-160, F.A.C.), the Standard Operating Procedures (SOPs) for conducting the RPS, LVS, HA, and SCI methods are available at http://www.dep.state.fl.us/water/sas/sop/sops.htm:

- RPS: DEP SOP FS 7230;
- LVS : DEP SOP FS 7320;
- HA: DEP SOP FT 3100; and
- SCI: DEP SOP SCI 1000.

Note that these SOPs are quite extensive, and that some include training and proficiency testing requirements and reference to the concepts presented elsewhere in this document for establishing study objectives, including considerations for the selection of representative sites and environmental conditions (e.g., section 2.1 above). All these procedures require that an appropriate 100 meter section of stream be measured, with flags placed in 10 meter intervals. The following guidance provides a method to most efficiently conduct these SOPs in a semi-simultaneous manner:

1. Collect any water chemistry (*e.g.,* nutrients, chlorophyll *a*, metered parameters) upstream of where biological sampling will occur prior to disturbing sediments;
2. Beginning at the downstream-most flag, measure the nine points for algal presence/thickness and record, as per the RPS SOP.
3. From this downstream-most flag, walk towards the next 10 meter flag and simultaneously perform the habitat mapping procedure while observing aquatic vascular plants (if present). Record both sets of observations according to the respective SOPs.
4. Continue steps 2 and 3 above until the upstream-most (100 m) flag is reached. If there is > 20% coverage of RSP rank 4-6, collect a representative sample of the algae for taxonomic identification. The RPS and LVS are now complete.
5. Complete the HA SOP based on the observations made during the habitat mapping exercise.
6. Conduct the SCI, sampling the observed habitats according to the SCI SOP. The data collection needed to evaluate the health of the flora and fauna is now complete.

These are current methods for evaluating flora and fauna in Florida streams. Measuring biology continues to be an evolving science. As improved methods are developed to evaluate information on chlorophyll *a*, algal mats or blooms, and excessive nuisance macrophyte growth, those should be integrated into the assessment.

2.7.8 Examples of a Weight-of-Evidence Approach for Determining Achievement of Nutrient Criteria

As noted in the sections, there are currently no quantitative endpoints for the RPS and LVS. To evaluate whether a stream achieves the narrative nutrient criterion, the investigator must compile water chemistry data (e.g., Total Nitrogen [TN], Total Phosphorus [TP], chlorophyll *a*, and ancillary parameters such as color, turbidity, DO, pH, conductivity, and temperature, etc.) and a minimum of two of each of the following: RPS, LVS (if appropriate), HA, and SCI. Taken together, these data are used as multiple lines of evidence to decide whether a stream is healthy, with acceptable levels of nutrients. Examples of how DEP evaluates these multiple lines of evidence are provided in **Table 2** and discussed below.

Table 2. Examples of RPS, LVS, chlorophyll *a*, HA, and SCI data used to illustrate a multiple lines-of-evidence approach used by DEP for determining whether or not a stream exhibits imbalances of flora or fauna. In these examples, TP, TN, or both nutrients **exceed the regional Nutrient Threshold values**.

Measure		**Sample #**	**Stream[1]**					
			1	**2**	**3**	**4**	**5**	**6**
RPS (% Rank 4-6)		**1**	21	45	4	8	3	26
		2	2	65	7	15	0	37
LVS	**Avg. C of C**	**1**	N/A	2.6	1.9	N/A	3.5	1.8
		2	N/A	3.2	0.5	N/A	4.2	2.4
	FLEPPC %	**1**	N/A	12	45	N/A	0	31
		2	N/A	4	74	N/A	0	26
Chlorophyll (µg/L as annual geo. mean)		**Year 1**	17.2	1.1	Non-Detect	4.5	3.5	1.3
		Year 2	22.1	2.1	Non-Detect	1.2	4.1	1.1
Habitat Assessment		**1**	121	109	105	133	81	110
		2	113	102	98	126	77	107
SCI		**1**	45	44	39	67	22	42
		2	39	33	29	58	31	39

[1] In these examples, TP, TN, or both nutrients exceed the regional Nutrient Threshold values.

In Stream 1, although the RPS data showed a pulse of periphyton (which consisted of the non-problematic alga, *Oedogonium*), it was not persistent, meaning the RPS results were acceptable (see section 2.7.3). No plants were found in the water. However, an increasing trend was observed in annual chlorophyll *a* (using a Mann's one-sided, upper-tail test for trend, with a 95% confidence interval), and the chlorophyll values exceeded those typically observed in healthy streams. Although the SCI score was currently acceptable and habitat was not limiting, DEP concluded that the chlorophyll issue, following the conceptual model in section 2.7.1, was sufficient to judge that this stream has impaired flora. It is likely that the increased organic matter enrichment associated with the excess phytoplankton (as indicated by the chlorophyll) would eventually lead to faunal imbalances.

Stream 2 was characterized by significant algal smothering, as demonstrated through the RPS results. Taxonomic identification showed the algae community to be dominated by *Lyngbya*, a known nuisance species. Although the vascular plant community, as assessed using the LVS, was within the range of reference streams, and chlorophyll *a* was non-problematic, the algal growth resulted in aquatic habitat smothering (a component of the HA), which likely led to the failing SCI score. DEP concluded that the RPS results, coupled with a poor habitat smothering score, was evidence that stream 2 has impaired flora, which in turn caused impaired fauna. These responses are consistent with the nutrient enrichment model in section 2.7.1.

Although periphyton and chlorophyll *a* were not issues in stream 3, the HA and LVS results showed that the invasive exotic vascular plant, *Hydrilla* was excessively abundant, leading to imbalances of flora. An increase in *Hydrilla* abundance was associated with reduced substrate diversity and failing SCI scores, meaning the elevated nutrient levels were associated with imbalances in flora and fauna, consistent with the nutrient enrichment model in section 2.7.1. This situation is complicated because invasive exotic plants can be observed even without nutrient enrichment. It is important to review other information, including the levels of nutrients in the waterbody that could contribute to species proliferation. In this circumstance, DEP concluded that excess nutrients exacerbated the floral community imbalances as evidenced by the LVS results.

Stream 4 is a minimally disturbed reference stream that was at the upper 98th percentile of the data distribution used to establish the regional Nutrient Threshold. The measures of both flora and fauna showed normal, healthy conditions, meaning that nutrient levels associated with the site are acceptable and the narrative nutrient criterion is achieved.

Floral measures at stream 5 were non-problematic, despite nutrient concentrations that exceeded the regional Threshold values. No primary or secondary nutrient responses, as described by the nutrient enrichment model in section 2.7.1, were observed, but the SCI indicated impaired fauna. The SCI results, combined with higher levels of nutrients, lead to the conclusion that the narrative nutrient criterion is not achieved. In this case however, habitat assessment results indicated artificial channelization, poor substrate diversity and availability, and a compromised riparian buffer zone. Observations also indicated extensive hydrologic modifications in the drainage basin. These habitat and hydrologic factors were evaluated as part of a TMDL process, prior to initiating a TMDL. After an evaluation of all stressors (through a stressor identification study), habitat and hydrologic improvements were found to be the stressors affecting stream health, and not nutrient concentrations. DEP would then evaluate this stream under a site specific structure described in Rule 62-302.531, F.A.C.

In stream 6, both the RPS and LVS results suggested the early warnings of nutrient enrichment, with persistent periphyton coverage and changes in the vascular plant community, even though chlorophyll *a* and SCI results were acceptable. The periphyton community was dominated by *Vaucheria*, a known nuisance species, while the vascular plants, *Alternanthera philoxeroides* and *Panicum repens* (two FLEPPC exotics), were moderately abundant. Habitat assessment results indicated moderate smothering by the periphyton and a reduction in substrate diversity associated with the exotic plant growth. DEP concluded that this was sufficient evidence of floral imbalances, which if allowed to continue without intervention, would also result in faunal imbalances, as predicted by the nutrient enrichment model in section 2.7.1.

In conclusion, the information presented in this section, including the examples, provide a multiple lines of evidence approach to numerically interpret the narrative nutrient criterion, on a site-specific basis, as provided for in Rule 62-302.531(2)(c), F.A.C.

2.8 Use Attainability Analyses and Class III-Limited Reclassifications

Chapters 62-302.400 and 62-302.800, F.A.C., describe the requirements to reclassify Class III waterbody to a Class III-Limited waterbody. For any downgrade, a Use Attainability Analysis (UAA) must be conducted, as explained in the document, *Process for Reclassifying the Designated Uses of Florida Surface Waters* (DEP-SAS-001/10). The Class III - Limited category is restricted to waters shown to be wholly artificial or altered through dredging to the extent the physical characteristics of the waterbody limit its ability to support aquatic life use.
The following is a summary of these requirements.

A petitioner must present appropriate and scientifically defensible water quality, biological, hydrological, and habitat studies and analyses, as well as environmental, social, and economic studies to demonstrate that:

- None of the uses being removed are existing uses;
- The uses to be removed would not be attained by implementing effluent limits required by Sections 301(b) and 306 of the Federal Clean Water Act in conjunction with implementation of cost-effective and reasonable best management requirements for nonpoint source pollution control;
- The proposed reclassification is clearly in the public interest;
- Water quality standards in downstream waters will be fully protected; and
- One or more of the criteria from Paragraph 62-302.400(11)(c), F.A.C., apply. This portion of the rule, based on 40 CFR 131.10(g), allows removal of a designated use that is *not* an existing use, as defined in § 131.3, or establishment of sub-categories of a use if the State can demonstrate that attaining the current designated use is not feasible because:

1. Naturally occurring concentrations of substances prevent the attainment of the use;

2. Natural, ephemeral, intermittent or low flow conditions or water levels prevent the attainment of the use, unless these conditions may be compensated for by the discharge of sufficient volume of effluent discharges without violating State water conservation requirements to enable uses to be met;

3. Human caused conditions or sources of pollution prevent the attainment of the use and cannot be remedied or would cause more environmental damage to correct than to leave in place;
4. Dams, diversions, or other types of hydrologic modifications preclude the attainment of the use, and it is not feasible to restore the waterbody to its original condition or to operate such modification in a way that would result in the attainment of the use;
5. Physical conditions related to the natural features of the waterbody, such as the lack of a proper substrate, cover, flow, depth, pools, riffles, and the like, unrelated to water quality, preclude attainment of aquatic life protection uses; or
6. Controls more stringent than those required by sections 301(b) and 306 of the Federal Clean Water Act would result in substantial and widespread economic and social impact.

Because a downgrade to the Class III-limited category is restricted to physically and hydrologically altered waterbodies, biological sampling should occur in areas representative of these human stressors. Consequently, the waterbody segment will likely be habitat limited and the hydrologic modifications may be such that the waterbody is frequently dry or not flowing. Although both of these conditions would likely prevent the waterbody from supporting a healthy biological community, it is appropriate to sample SCI and other biological measures under these obviously stressed conditions because it is consistent with the study objectives (see example in section 3 below).

3. Water Level and the SCI

All scientific methods have limitations that must be understood to effectively use the technique for making valid decisions. As previously mentioned, aquatic organisms will die if a site goes dry. For pollutant identification studies (including nutrient studies), wait a minimum of 6 months after a completely dry site has begun flowing again before considering sampling, unless site specific information is available that indicates a particular stream invertebrate community recovers more quickly than 6 months (*e.g.*, 3-4 months). In these situations, sampling may occur after the suitable time period has elapsed that allows for biological recolonization from the desiccation event. For example, if the investigator has the necessary taxonomic skills, exploratory dip net sweeps may be conducted and the organisms field identified to determine the degree of recovery from the dry conditions. If it appears that a typical stream community is present, the investigator may commence sampling, documenting the reasons behind the decision.

Routinely, SCI sampling can be performed only 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 this top 0.5 m has been fully inundated with water for a minimum of one month (28 days) prior to sampling (**Figure 16)**. If water level at a site increases into this reachable zone to the extent that the sampler is not confident that the accessible habitats were inundated, the sampler should **wait a minimum of 28 days** to allow time for stream organisms to colonize the formerly exposed habitats. Similarly, if a site did not go completely dry, but stopped flowing and contained a series of disconnected pools for some time period, samplers should wait at least 28 days after flow has commenced (with a minimum water velocity of > 0.05 m/sec), before sampling. Note that it may take longer than 28 days for organisms to re-inhabit previously dry substrates and for rheophyllic organisms to recolonize stagnant reaches, depending on site characteristics. Avoid sampling sites after these low water level/stagnant events until sufficient time passes to eliminate these water level effects as confounding factors in interpreting the SCI results. If there is doubt about a particular sampling event, samplers should communicate with data analysts to flag the SCI results as potentially being affected from these water level issues.

As an example**, Figures 17 and 18** depict a recent increase in water level that 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 (not every stream has a gauge) and extrapolating that information to the study stream is extremely valuable for determining when sampling is, or is not, appropriate. Smaller streams typically have more spikes in their hydrographs, where the water level rises quickly and significantly but then returns to "normal" levels within days (**Figure 19**). A valid SCI sample can be collected when the formerly colonized habitats may be reached; however, it is important that samplers exercise caution to make sure that the substrates selected for sampling have been appropriately inundated.

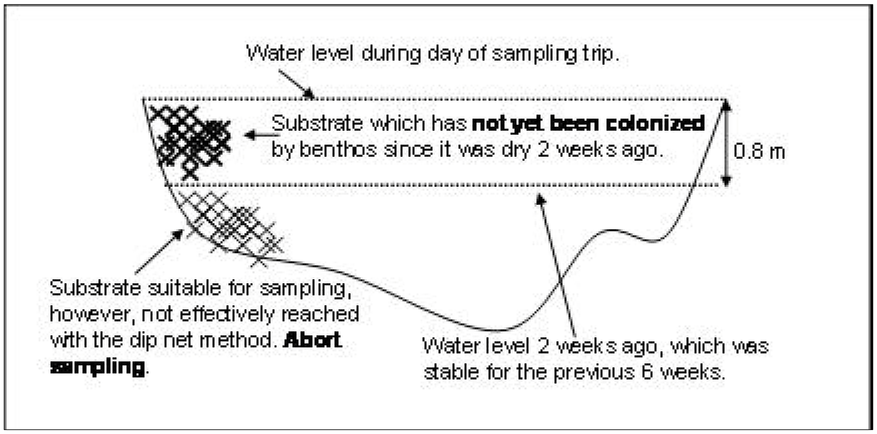

Figure 16. Schematic cross section of a stream showing recent increases in water levels indication the SCI sampling should not be conducted.

In streams with natural geomorphology and sinuosity, the habitats targeted for SCI sampling act as refugia from scouring during flood events, and sampling may be conducted when inundated substrates are reachable. However, in channelized systems there may be significant scouring after routine rain events. Depending on the study objectives, sampling may be conducted after a rain event in a channelized system to demonstrate the adverse effects of hydrological modification on the system.   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.

Figure 17. Stream water level two weeks prior to scheduled SCI sampling.

Figure 18. Stream level on day SCI was scheduled to be performed. Note that reachable substrates were dry on the previous photo. SCI sampling should be aborted when these conditions occur.

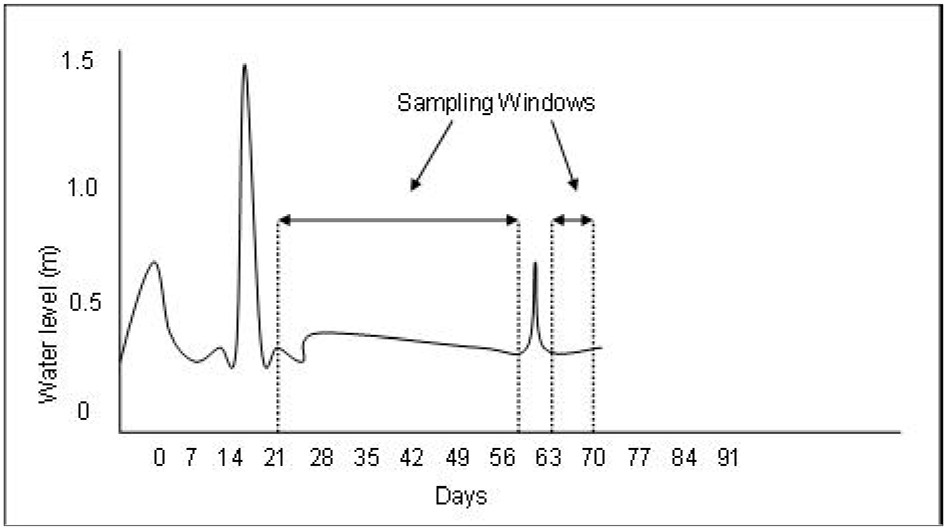

Figure 19. Hydrograph showing times when substrates are reachable.

Example 1, TMDL Sampling

A sampler is collecting SCI data to determine if water quality degradation (in this case, nutrient enrichment) is sufficient to list a waterbody on the verified list of impaired waters. 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 (see **Figure 20**). Hydrographs from nearby streams indicate these low water conditions have occurred for the past few months. Water velocity in the stream is not measureable (0.00 m/sec). Should the sampler collect the SCI? No, the conditions are such that the lack of inundated habitat and adequate 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.

Figure 20. Photo of low water conditions resulting in the desiccation of the majority of viable stream habitats. SCI sampling for TMDL/pollutant identification studies should not be conducted under these conditions.

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.

Example 3, Impaired Waters Assessment/Pollutant Identification Sampling

Due to heavy rains, a stream has water levels two meters over its banks into the riparian floodplain (see **Figures 21 and 22** for photos of typical and flooded conditions). 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 cannot 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).

Figure 21. Typical flow conditions at a sampling site.

Figure 22. Flow conditions at the same site shown in **Figure 21**, associated with a high water event, including floodplain inundation, when SCI sampling would not be appropriate.

Example 4, Sampling for Use Attainability Analyses

An entity is proposing to sample a series of severely hydrologically modified systems, including canals and urban ditches, as part of a Use Attainability Analysis to reclassify the systems into the Class III-Limited category, as described in Chapters 62-302.400 and 62-302.800, F.A.C. Although the canals do not typically go dry, the hydrologic modifications are such that the canals are characterized by consistently low water velocity (< 0.05 m/sec), except for during flood events. Can the canals be sampled for SCI under typical conditions? Yes, because the defined objective is to determine if the hydrologic modifications (and corresponding habitat limitations) have interfered with the development of a healthy community, the investigator may sample during the typical velocity conditions, even if they are below 0.05 m/sec. Similarly, the urban ditches in the study rarely contain water for more than 3 or 4 months, and are usually dry during the remainder of the year. Again, because the study objectives are to determine the influence of the hydrologic modifications, it is appropriate for the investigator to sample the ditches after they contain water for the typical maximum amount of time, even if that is only 3 or 4 months.

4. 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 human-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, but due to environmentally relevant reasons, not as a sampling artifact. Samplers must thoroughly understand the concepts associated with the Standard Operating Procedures (SOPs), and consistently follow the SOPs, and operate in an ethical manner to prevent sampling errors (see DEP SOP SCI 2100.

4.1. 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 DEP's inherent biological expectations associated with the SCI, stream or river segments that generally had flow (with velocities ranging from 0.05 m/sec to 0.4 m/sec except during seasonal droughts or floods) 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 data 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 defensible. 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 that has been hydrologically modified to resemble a lake, because such sampling is consistent with the study objectives. 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 very low velocity, flow-through lake, or may be tidally influenced. Sampling these types of areas may result in inappropriate SCI failures, because of incorrect system classification (comparing "apples" to "oranges"). Example photos of sites unsuitable for the SCI method are provided in **Figures 23, 24, and 25**.

Figure 23. Example of swamp conditions present, indicating that the SCI is inappropriate at this site.

Figure 24. Example photo of lake conditions present, indicating that the SCI is inappropriate at this site.

Figure 25. Example photo of tidal influence, meaning SCI is a not an appropriate tool.

Thus, atypical areas not representative of the stream reach should 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 directly adjacent to or under a bridge (usually disturbed by channelization) should be avoided, as this area would not be representative of the stream reach.

4.2. Appropriate Antecedent Hydrologic Conditions

Water levels should be examined as outlined above to determine if conditions are appropriate for the purpose of the study (see section 3 above). Samplers must 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 must develop intimate familiarity with the hydrology of streams in their regions and learn to extrapolate the information from available hydrographs to nearby streams to determine if antecedent hydrological conditions were appropriate.

4.3. 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/water/sas/sop/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 effective concentrations of dissolved oxygen and food availability. Therefore, leaf packs (leaves caught on snags above the substrate) that are in the main flow are preferred over leaf mats (leaves on the bottom), which tend to be associated with lower velocity and potential anaerobic conditions. 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 substrates as refugia 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.

4.4. 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 <u>at least</u> 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. The applicability of the SCI to large rivers is described in the **Stream Condition Index (SCI) Report**. Although some portions of rivers may have un-wadeable water depths, the SCI sampling is conducted only on the reachable habitats (top 0.5 m) in the system, thus minimizing effects of depth.
- When sampling streams or 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.

5. SCI Training

5.1. 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 must become familiar with the basic orders and families of aquatic macroinvertebrates, as outlined in DEP SOP SCI 2230. 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.

5.2. Apprenticeship

Because of the complexities mentioned above, DEP requires that novice sampling staff undergo a systematic training /apprenticeship program. 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 shall 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 (who have passed the SCI audit) on the concepts presented here. As training progresses, the novice staff should gradually demonstrate the required best professional judgment and sound sampling technique (see below for training checklists). Once training has been completed, a field audit with the Standards and Assessment Section to assess a sampler’s ability to adhere to the SOPs may be scheduled.

6. SCI Data Usability

The intent of this section is to provide a procedure for how Stream Condition Index (SCI) data will be used for DEP environmental decisions.

Determining if data are usable for a particular purpose is a complex task, requiring a logical and balanced evaluation of many factors. The procedural components of the SCI assessment must be performed by staff with sufficient scientific expertise and demonstrated proficiency, as mandated by Rule 62-160, F.A.C. Additionally, the following must be considered during a biological data usability determination:

- Understanding the purpose for the bioassessment sampling, including specific project objectives, and determining the extent to which the bioassessment data set fulfills the objectives of the project or Program. The environmental conditions associated with the sample (*e.g*., climatic, hydrologic, site location, habitat, etc.) must be consistent with the study objectives;

- Evaluating laboratory and field quality control measures and other supporting data, including the determination of the pattern, frequency, and magnitude of any quality control deficiencies associated with the results. This also may involve evaluating corroborative data (*e.g*., performance tests, data from other sampling entities);
- Determining the relationship between the bioassessment result, the associated decision or action level (*e.g*., water quality criteria), and the minimum detectable difference associated with the method; and
- Determining the reasonable cause for a poor bioassessment score (*e.g*., water quality, hydrology, and/or habitat) and ensuring that the data are appropriately used to address the causative factor(s).

Determining the Extent to Which the Bioassessment Data Set Fulfills the Objectives of the Project

Designing a sampling strategy that focuses on answering specific environmental questions is critical in the bioassessment process, so that confounding variables may be controlled for to the degree possible. Data collected to evaluate one environmental stressor may not be suitable for determining the influence from other stressors. The Department shall examine the purpose of the data collection, the associated potential confounding variables, and ensure that the results are used in a manner consistent with the study objectives.

> *Example: DEP scientists design a study to evaluate the effects of water withdrawls on the invertebrate community of a stream. SCIs were collected at typical water levels prior to the withdrawls to establish background SCI scores. Subsequent to significant consumptive water use, SCIs were collected during extremely low water levels. Although the stream had high SCI scores prior to the consumptive water use, numerous SCI failures were noted after the water withdrawls. In this scenario, the SCI failures observed during the low water levels should not be included in Impaired Waters Rule (IWR) listing decisions, because these samples were collected during conditions inconsistent with the objectives of IWR studies.*

Evaluating Staff Capability, Quality Control Measures, and Other Supporting Data

Data must be collected by qualified samplers, using the appropriate DEP Standard Operating Procedures (SOPs), following the concepts outlined in this SCI Primer. The SCI must be performed by individuals that have passed the proficiency test described in DEP SOP SCI 1300. Samplers must conduct the assessment per DEP SOP SCI 1100, following other guidelines outlined in this Primer. SCI scores must be calculated in accordance with SOP SCI 2100.

Quality control information will be systematically evaluated and assessed against the objectives of the study before a usability decision will be made. For example, the purpose for which bioassessment data are collected can vary widely, and may include such diverse activities as: initial screening or scoping studies, assessing waters for IWR purposes, or determining whether a stream created as part of an Environmental Resource Permit mitigation project has been successful. A quality control failure that may be tolerated for a screening study would not be acceptable for IWR purposes or declaring the success or failure of a restoration project.

As applicable to the data usability assessment process, any record associated with a reported sample result or set of sample results may be audited, per Chapter 62-160.240 and 62-160.340, F.A.C. Both original ("raw") and reduced or summarized versions of data records may be inspected to determine the acceptability of results, based on an evaluation the sample data and associated quality control records.

If any aspect of the assessment appears erroneous or suggests that the assessment was not made according to the SOPs (*e.g*., excessive family-level identifications, sampling conducted during extreme water levels not in accordance with the sampling objectives), the Department will further investigate the credibility of the bioassessment results. This may involve follow up audits of samplers or analysts and potential data rejection.

> *Example: An SCI was conducted in a small stream in a National Forest as part of a probabilistic sampling network. Sampling was conducted during a drought year, and the stream's water level was low enough that the majority of the habitat was exposed to air. Very few invertebrates were present in the sample (less than 150 individuals, however, the lab failed to qualify the sample with "x") and the stream received a failing SCI score. When the results were considered for IWR listing purposes (Chapter 62-303, F.A.C.), data users examined the rainfall and hydrograph data from the area and determined, from these data and from site photos, that the severely low water level did not allow for a representative assessment of the stream's invertebrate community. Further, the minimum required number of individual organisms (two aliquots of 150) were not identified in the laboratory (and the sample was not properly qualified with "x"). Therefore, these SCI results were not used for IWR listing purposes.*

> *Example: A county conducts SCI sampling twice annually on a river of local interest. During the first three years of the program, the average SCI scores were 42, 55, and*

48 (all passing scores). On the fourth year, a new consultant (including the sampling team and laboratory) conducted the SCIs, and the river received a failing score of 25. Upon investigation, data users noted that no one on the sampling team had passed the SCI proficiency test and that a high proportion of taxa identified in the laboratory were to the family level only (not to the level required by the SOP). Because the samplers did not demonstrate the required expertise and the laboratory SOPs were not properly followed, the year 4 SCI data were not considered usable, and DEP worked with the county to correct the deficiencies.

Determining the Relationship Between the Bioassessment Result, the Water Quality Criterion, and the Minimum Detectable Difference Associated with the Method

As in all scientific measurements, there is a quantified level of uncertainty associated with bioassessment results, known as the Minimum Detectable Difference (MDD). When SCIs are compared along a longitudinal or temporal gradient, differences in scores greater than the MDD (plus or minus 13 points) are considered to be statistically reliable.

Example: Staff from the Florida Fish and Wildlife Conservation Commission Invasive Plant Management program conducted an herbicide treatment in a spring-fed river to control the invasive exotic plant, Hydrilla verticillata. The average SCI score before control efforts was 24, and the average score nine months after control efforts was 50 (an increase of 26 points). Because the difference in SCI scores was greater than the MDD (statistically reliable), the management actions were considered successful.

Determining the Reasonable Cause for a Bioassessment Failure

Failure of the SCI indicates that the stream does not meet the Clean Water Act goal of biological integrity, as measured by community structure and function, but the reason for the unacceptable condition must still be explained. For IWR purposes, the pollutant causing the biological degradation must be identified prior to developing a TMDL. Although a stream could have a failing SCI due to water quality problems (*e.g.*, toxic substances or excess nutrients), it is possible that habitat disruption, hydrologic alterations, or other physical disturbances are significant stressors. If factors other than water quality are determined to be the cause of the SCI failure, DEP will address those factors through avenues other than the TMDL program.

Example: A stream fails the SCI, but data indicate the stream is not impaired for any water quality parameter. DEP biologists determine that the stream habitat assessment score was less than 80, and that the system had been artificially channelized in the 1960s. In this scenario, pollutant reduction is not required, but *physical restoration activities or reclassification of the waterbody would be potentially appropriate actions.*

Summary of SCI Data Usability

To determine appropriate actions associated with bioassessment results, DEP will review and evaluate the following information:

- The purpose for collecting the bioassessment data and the degree to which the study fulfilled the objectives;
- The documented quality control measures and other supporting data, as well as the pattern, frequency, and magnitude of any quality control deficiencies associated with the results;
- The relationship between the results, the water quality criterion, and the Minimum Detectible Difference associated with the method; and
- A reasonable determination of the cause of the bioassessment failures.

From this evaluation, DEP will determine how the data can be used by the relevant Department programs. Biological health usability assessments will evaluate the above factors relative to DEP program or project objectives, and the follow the principles characterized in this guidance document to draw an “overall conclusion” concerning the usability of the data set consistent with the processes and examples provided in this document.

7. SCI Training Materials, Training Requirements, and Checklists

See: http://www.dep.state.fl.us/water/bioassess/training.htm

8. Literature Cited

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