

SAP Review: A Non-predictive Algal Index for Complex Environments (Also known as 'Algal Stream Condition Index' or ASCI), Theroux et al. in prep

January 2019

Charge Question 1: Comment on the adequacy of the provisional ASCI to serve as a statewide bioassessment index applicable to most wadeable streams across California, specifically with respect to data, statistical approaches, evaluation of performance, and soundness of findings.

The number of sites sampled, variables analyzed, and resulting amount of data used in the analyses are sufficient for developing a statewide tool for bioassessment. The number of sites used in the California Algal Stream Condition Index (ASCI) development and testing was about the same (n≈2000) as the National Rivers and Streams Surveys (NRSA) of the United States Environmental Protection Agency (USEPA).

The California ASCI was developed and tested with state-of-the-science statistical methods. The reference site approach for establishing expected condition is well-accepted. Both O/E and multimetric indices (MMI) approaches were tested. Modeling expected metric scores for reference condition to account for site-specific natural variability was tested. Testing "all-subsets" of metric combination to select the best metrics for use in MMIs is relatively novel, thorough, and valid. Calibration and validation data sets were used to evaluate performance of O/E and MMIs with a broad suite of performance statistics.

Performance statistics for the ASCI were high enough to indicate the ASCI is sufficiently accurate and sensitive to human disturbance for use in a monitoring program. The ASCI metrics for diatom, hybrid, or soft-bodied algal (SBA) MMIs were as high as the California Stream Condition Index (CSCI) using benthic macroinvertebrates (Mazor et al. 2016). The ASCI performance statistics were higher than MMI performance statistics developed with the NRSA periphyton data (Tang et al. 2016). The diatom and hybrid ASCI compared favorably with the diatom MMI developed for Maine streams (Danielson et al. 2012).

The soundness of the findings are in line with the scientific rigor commonly used for developing indices of biological condition for stream assessments. Data quantity, statistical methods, and index performance have produced MMIs that distinguish reference from highly disturbed sites and respond to stressors (pollution and habitat alteration) and human disturbance as well as others used in ecological assessment. There is statistical error in those assessments, and there is statistical error in responses of MMIs to pollutants, but that error is sufficiently low to provide reliable and valuable information for managing streams.

Given the adequacy of the provisional ASCI to serve as a statewide bioassessment index for wadeable streams, there may be ways to improve both performance and soundness of the ASCI. We describe these recommendations concerns of the Science Advisory Panel (SAP) in the following sections. Some of these recommendations can be addressed in the short term and conclusions from that information can be included in a revision of this draft review. The SAP will provide guidance on how to address other concerns. The SAP is confident that these concerns can be addressed satisfactorily for application of the ASCI as a valuable tool for assessing biological condition of California streams.

Charge Question 2: Between the Diatom, Hybrid, and Soft Bodied Algae ASCIs, which one do the SAP members think works best for determining water quality impacts to biointegrity? What about impacts due to biostimulatory substances and/or conditions? Why?

Conceptually, application of the hybrid ASCI provides a more complete assessment of the biological condition of algae than either the diatom or SAB ASCIs. Performance of all three MMIs is sufficiently high for application (based on comparisons with other bioassessment indices being used by states), but performance of the hybrid MMIs is somewhat greater than diatom and SAB MMIs.

A key question is whether the added cost of analyzing samples of SAB is worth the conceptual improvement and increased MMI performance. One of the presentations included in the December 2018 stakeholder responses listed costs of diatom and SAB analyses at \$1200 per sample, and projected DNA analyses at \$300/sample. Diatom sample analysis should cost in the range of \$300-400 per sample in a routine monitoring program of the scale of the California project in which taxonomy has been well documented (as taxonomy should be in an ongoing routine monitoring program).

If these cost estimates are accurate, the SAP questions whether that \$700-800 difference in sample analysis cost is worth the conceptual and statistical performance improvements gained with adding SAB to diatom assays for a hybrid MMI. Additional information is needed to evaluate tradeoffs in conceptual and statistical performance. Do stakeholders value the additional information in the hybrid versus diatom or SAB MMIs to justify part of the cost difference? Could diatom MMI performance be improved more cost effectively than a hybrid ASCI if more diatom samples were collected and assayed, efforts in diatom sample assay were increased, or if relative abundance was used for metric calculation than presence/absence of taxa? What if you used the money for other things, even sampling more streams for both algae and macroinvertebrates?

One argument for application of the hybrid diatom and SAB analysis is the extra cost will only be necessary until a DNA method is developed, tested, and accepted. Depending upon accurate estimates how long that will take, a lot can happen scientifically and policy-wise that could justify reinvention of parts of the ASCI application that would make this argument mute. For example, are the DNA analyses cheaper than updated microscopic analyses? Why be constrained to algal taxa in DNA analyses of periphyton? Why be constrained by taxa versus genes and functional attributes?

The SAP will review other papers to try to understand this cost and benefit tradeoff before the next meeting. We will incorporate that information and SCCWRP responses to these questions in our review. Answering some of these questions could be addressed with analyses of existing data beyond the time of our review. Answering some of these questions may require policy decisions.

Charge Question 3: Do the measures of performance (i.e., the accuracy, precision, responsiveness, and sensitivity) of the ASCIs indicate that they are adequate for use in most Wadeable streams in California?

According to similar ASCI values (central tendency and variability) for reference conditions across ecoregions in California, that ASCI is adequate for use in MOST Wadeable streams in California. Issues in streams where the ASCI is suspected to be inaccurate can be addressed in management processes that make sure that the ASCI is appropriate for the specific stream being managed. The broad applicability in diatom assessments around the world attest to the ability of ASCI to be adequate for MOST Wadeable streams in California. Simpler, single-metric assessments are used in many European countries. Given

concern about variation in flow duration and frequency in California streams, diatom MMIs perform remarkably well in the intermittent streams of Kentucky (Wang et al. 2005).

More detailed performance assessments for the ASCIs in different ecoregions would also help to evaluate applicability for use in most types of wadeable streams in California. Currently we have comparisons of reference values of MMIs across ecoregions, box plots of metric sensitivity, and comparisons of sites falling in different percentile ranges of reference condition. The latter analysis shows potential issues variation in relative sensitivity to human disturbance in different ecoregions. A more thorough set of performance statistics (e.g. as in Table 8 of Theroux et al.) would help with that evaluation.

If there are differences in MMI sensitivity to human disturbance among regions, corrective action should be considered. Tang et al. (2016) found the best MMI performance called for accounting for natural variability by modeling metrics by ecoregion and for using different metrics in different ecoregions. The assessment of benthic invertebrates with an MMI in the NRSA also used different metrics in different ecoregions. Although we have conceptual issues with redefining MMIs (and thus measure of biological condition) with use of different metrics in different ecoregions (Tang et al. 2016), we would expect differences in reference condition as a result of natural variation among regions (and even sites within regions). We would also expect differences in sensitivity of biological condition to human disturbance to vary naturally among regions, sites within regions, and even seasonally. Natural variability in climate and geology determine many physical, chemical and biological factors in streams that more directly regulate algal species composition, physiology, and biomass (Biggs 1996, Stevenson 1997), and therefore measure of biological condition and their response to human disturbance.

In addition to concern about the ASCI being applied in most streams in California, the temporal window in which the ASCI has highest performance and limitations on that performance should be addressed in the future. This temporal window should consider flow duration and frequency attributes, as well as other physical and chemical conditions that vary seasonally.

The applicability of the ASCI to assist in biostimulatory management issues could be improved by evaluating the performance of the ASCI under different biostimulatory conditions. How do nitrogen, phosphorus, or NP co-limitation affect applicability of the ASCI? How does organic carbon enrichment affect applicability of the ASCI? How does channelization affect the ASCI? Do algae care whether the substratum is rock or concrete in channelized streams? Some of these questions can be addressed by further analyses of ASCI performance in different nutrient conditions (indicated by concentrations, ratios, and maybe even loads) with existing data. Application of experiments using simple devices like nutrient diffusing substrata could prove causal characterizations of species and assemblage responses to stressors, which could be used in testing ASCI performance as well as refining species traits and metrics.

Charge Question 4: Are there additional performance evaluations or refinements to the index that are essential and that can be done with available data?

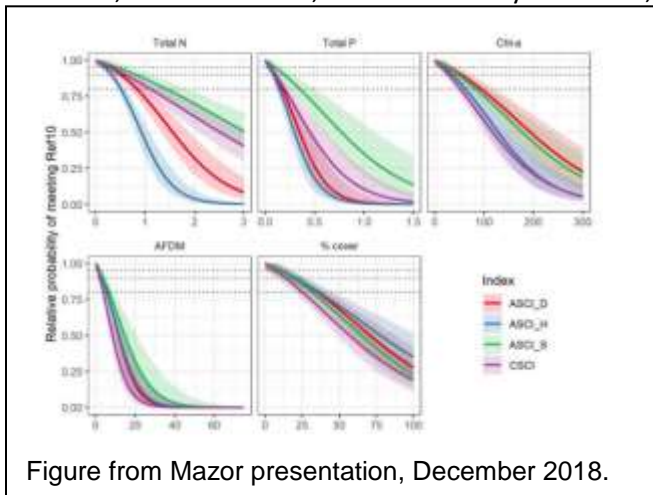
For sake of the scientific integrity of the ASCI, some additional analyses are warranted. Some of these were mentioned above. Others are mentioned in this section and below.

A more thorough analysis of why O/E approaches and modeled metric approaches did not improve ASCI performance are warranted.

Several potential issues are associated with the algal O/E. We know that high within-site variability (either temporal or sampling variability) is associated with periphyton sampling. However, another, potentially more interesting effect is that the diatom community tends to increase in richness in response to stress, particularly by biostimulatory substances. The rationale for this response is species membership is limited in low resource habitats to species adapted to these low resource habitats, and that increased resources make the habitat available to invasion by other species. Therefore, as habitats become more enriched, more species invade the habitat. Competitive exclusion of low resource species may occur in high resource conditions. As a result of the balance in these two processes, we often observe an increase in species numbers and evenness in counts of algae along a resource gradient (Stevenson 2014). Be aware, this apparent increase in species richness may not reflect loss of low nutrient native taxa or represent true taxa richness. For an O/E model, this increase in species numbers observed in counts with nutrient pollution could reduce the strength of the association between the metric value and a stressor gradient. Species presence/absence of algae also varies greatly among reference streams and temporally (potentially within the sampling window). It might be interesting to dig into this a bit.

Accounting for natural variability among sites, either by ecoregions or modeling metrics, should improve metric and O/E or MMI performance. Why did it not improve ASCI performance? In the National Lakes Assessment, simple MMI modeling improved MMI performance, but this was not always evident in measures of MMI sensitivity to human disturbance (Stevenson et al. 2013). This has been attributed to the likely covariation among natural and human factors across the landscape (Ellis and Ramankutty 2009), and therefore in watersheds (Schoolmaster 2013, Stevenson et al. 2013). A lack of understanding this covariation among natural and human factors and its effect on MMI performance, could lead to poor causal foundation and artificial inflation of MMI sensitivity to human disturbance. Again, it would be interesting to dig into this a bit.

There was some concern about why the ASCIs were not as sensitive to biostimulatory substances as the CSCI. Theoretically, algae should be more sensitive to inorganic nutrients than fauna because they are directly affected by them. Figure 7 (Theroux et al.) shows CSCI being more sensitive than algal MMIs to nutrients, sands and fines, and conductivity. However, Mazor et al. show ASCIs were generally more sensitive than the CSCI to nutrients, but not other indicators of biostimulatory substances (chl a, AFDM). Certainly, the amount of organic material (chl a, AFDM) in a habitat affects algae in that habitat, but it also affects invertebrates. Again, this should be analyzed a bit further to reconcile these contradictions.



Both the SAP and stakeholders were concerned about the relationship between metrics used in the MMIs and biological condition. Many of the metrics refer to stressor-style metrics and are not clearly related to elements of biological condition, i.e. the degree of natural structure and function of biological assemblages (Davies and Jackson 2006). Two approaches should be considered to improve this situation. Better explain how the selected metrics are related to your definition of biological condition. Consider selecting metrics that better reflect biological condition even though you may sacrifice some performance on the MMIs. We could make the argument that measures of biological condition should be related as much to our conceptual characterization of biological condition as possible, while still retaining a sensitive indicator of ecological change.

Charge Question 5: Are there any caveats or cautions that should be exercised when using the ASCIs to assess biological condition?

Caveats and cautions for using ASCIs to assess biological condition have been addressed above or under the next charge question about stakeholder concerns.

Charge Question 6: Are there technical ways to address stakeholder concerns?

Most stakeholder concerns should be addressed by SAP comments and recommended analyses above, and by further revisions to the ASCI if warranted.

Taxonomic variability. We address the issue of taxonomic variability in algal assays under stakeholder concerns, because it has been brought up by stakeholders more than the SAP. But the SAP is well aware of the issue. Here, the taxonomic variability that we are referring to is the variability in what bench taxonomists call taxa when they are observed microscopically in samples. Awareness of this issue has existed for a long time, but we it has reached a greater importance in the US as periphyton assessment projects have become more common and geographically extensive in the US. These issues have been addressed in Europe, and results have been published that help address the issue in the US.

So, how important is taxonomic variability for the scientific credibility and performance of ecological assessments? Little has been done to really understand the effect of taxonomic variability on algal assessments of biological condition, and relatively little has been done to characterize taxonomic variability among bench taxonomists. We have known about taxonomic variability for all taxa in ecological assessments over the long span of our careers, and have recognized that that source of variability is included in the variability of our assessments. So, for the ASCI, taxonomic variability exists among bench taxonomists, but the ASCI still distinguishes effects of water chemistry and human activities on biological condition accurately, and as accurately other biological assessment tools. The same has been observed in other assessment projects around the world (Kahlert et al. 2009, Stevenson unpublished data) and over the century in which bioassessments have been conducted, so the California situation is not unique.

The basics of why biological assessment tools can perform well with taxonomic variability is assessments are based on metrics and not just the identity of the species in samples. With metrics produced by

species identity in samples and species traits, as long as species traits of misidentified taxa are similar and are not biased, metric accuracy is not affected. This is clearly shown in Kahlert et al. (2009), where similarity in species composition of repeated counts of the same sample by different taxonomists can be low, but similarity in metric values resulting from different taxonomists is high.

So why be concerned? Improvements in taxonomic consistency could increase performance of biological indices, especially O/E indices that rely mostly on species identification and not species traits. Issues with taxonomic consistency also cause questions about scientific credibility of a tool that have answers, but they require explanations. Any time that there is need for more explanations, there is more opportunity for obfuscation and clouding of scientific credibility.

Diatom taxonomists in the US have started a website for the diatoms of the US (diatoms.org) which helps with taxonomic consistency. Together with diatom ecologists, this group is: developing training materials and tests for taxonomic certification; maintaining a standardized list of diatom taxa and taxonomic references for those taxa; documenting good practices for taxonomic analyses in ecological assessments; and working with Europeans to learn how taxonomic inconsistency has been addressed in programs that in many cases, have been going on for decades, and are now a key element in the European Union's Water Framework Directive. We expect this effort to quench concern about taxonomic consistency in biological assessments.

Application of ASCIs in range of policy options. Without listing all the potential policy options, the SAP can envision application of ASCIs in a wide range of policies that address stakeholder concerns.

Additional traits should be developed to improve the ASCI. Some new traits for algae were developed for taxa with the California dataset by Paul et al. and used in Theroux et al. Development of additional traits may be warranted with caution that causal foundations for species traits be carefully considered. This should include developing metrics that better balance loss of attributes of biological condition to complement MMIs that had a predominance of increasing metrics.

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