

Bay Area Macroinvertebrate Bioassessment Information Network (BAMBI)***Issue Paper # 2:******Standardization of Quality Assurance and Quality Control
Procedures for Benthic Macroinvertebrate Rapid Bioassessment Projects*****General Background Information**

Developing and implementing Quality Assurance and Quality Control (QA/QC) procedures in benthic macroinvertebrate rapid bioassessment projects are integral steps in creating a scientifically valid bioassessment program. Throughout the San Francisco Bay Area, municipal stormwater programs (Marin, Contra Costa, Alameda, Santa Clara and San Mateo Counties, and the City of Vallejo) and the Regional Water Quality Control Board (Regional Board) have designed water quality monitoring and watershed assessment programs to include rapid bioassessment techniques. A majority of these programs have used or are currently using the California Stream Bioassessment Procedure (CSBP), a standardized protocol for assessing biological and physical habitat conditions of wadeable streams in California. A previous BAMBI issue paper (i.e., BAMBI Issue Paper #2) discussed the need for standardized field and laboratory protocols in the Bay Area. Additionally, the development and consistent implementation of standardized bioassessment quality assurance and quality control (QA/QC) procedures are needed. Implementing standardized QA/QC protocols can assure the collection of reliable and defensible data.

Objectives

The purpose of this paper is to begin identifying and reviewing issues regarding the standardization of field and laboratory QA/QC procedures used in macroinvertebrate bioassessment projects in the Bay Area. Descriptions of these issues will aid in reaching a goal of developing guidance on standardized field and laboratory QA/QC procedures for those Bay Area agencies collecting BMI data. Due to the current use of the CSBP in a large majority of Bay Area bioassessment projects, this paper provides a description of, and generally refers to QA/QC procedures relative to the CSBP, as opposed to other field and laboratory protocols (e.g., EMAP, RIVPACS). However, most discussion points identified in the paper are general in scope and may have relevance to the standardization of QA/QC procedures for most rapid bioassessment protocols.

The following is a list of objectives for this issue paper:

1. Review the variety of bioassessment field and laboratory QA/QC procedures that could be used by BAMBI participants;
2. Identify high priority field and laboratory QA/QC procedures that should be implemented in Bay Area bioassessment projects; and,

3. Suggest next steps to BAMBI participants in implementing elements identified in objective #2.

The California Stream Bioassessment Procedure (*Excerpt from the CDFG's Aquatic Bioassessment Laboratory QAPP*):

In 1993 California initiated the first step in developing a state-wide bioassessment program by introducing the California Stream Bioassessment Procedure (CSBP). The CSBP is a standardized protocol for assessing biological and physical/habitat conditions of wadeable streams in California, and is a regional adaptation of the national Rapid Bioassessment Protocols outlined by the U.S. Environmental Protection Agency (EPA 841-D-97-002). The CSBP is a cost-effective tool that utilizes measures of a stream's benthic macroinvertebrate (BMI) community and its physical/habitat characteristics to determine the stream's biological and physical integrity. Biological and physical assessment measures integrate the effects of water quality over time, are sensitive to multiple aspects of water and habitat quality and can provide the public with a familiar expression of ecological health. A large majority of agencies in the Bay Area are using, or have used the CSBP when conducting rapid bioassessments.

Now in its third edition, the CSBP is recognized as California's standard protocol for conducting rapid bioassessments and forms the basis of California's effort to develop a statewide bioassessment program. The CSBP can be used to detect aquatic impacts from point and non-point sources of pollution and for assessing ambient biological condition. The sampling unit is an individual riffle or riffles within a reach of stream depending on the type of sampling design used. The CSBP should only be used when sampling wadeable, running water streams with available riffle habitats. It is important that BMI's are collected when streams are at base flow, as high flows can dramatically alter local community composition and can thus produce unrepresentative results. There are approved modifications of this procedure for narrow (< 1m) streams, wadeable streams with sand or mud bottoms and channelized streams. There are also procedures for lentic or still water environments. Additional information on the CSBP can be found at www.dfg.ca.gov/cabw/cabw/csbp.html or by contacting the California Department of Fish and Game's Aquatic Bioassessment Laboratory at (916) 358-0316.

CSBP Field Protocols

Point Source Sampling Design

Point source sampling design is used when discernable perturbations, impacting structures or discharges into the stream with point sources of pollution are apparent. The sampling units are individual riffles within the affected section of stream and an upstream unaffected section. At least one riffle in the unaffected section should be sampled and one or more riffles in the affected section, depending on the amount of detail that is required on downstream recovery. The riffles used for sampling BMIs should have relatively similar gradient, substrate and physical/habitat characteristics and quality. One sample will be collected from three (3) randomly chosen transects in each riffle selected.

Non-point Source Sampling Design

Non-point sources sampling design is used when no obvious perturbations or discharges into the stream with non-point sources of pollution are apparent. This sampling design is appropriate for assessing an entire stream or large section of stream. The sampling units are riffles within a reach of stream. The stream reach must contain at least five (5) riffles within the same stream order and relative gradient. One sample will be collected from each upstream third of three (3) randomly chosen riffles.

CSBP Laboratory Protocols

The CSBP utilizes a single standardized laboratory procedure for both point and non-point sampling. In brief, sample contents are rinsed in a 0.5 mesh sieve and placed into a gridded tray where they are spread evenly. Grids are then randomly selected and BMIs are picked from the debris and identified to a standard taxonomic level. BMIs from additional grids are identified until 300 organisms have been identified.

The CSBP laboratory methods are described in the California Department of Fish and Game Aquatic Bioassessment Laboratory's (ABL) Quality Assurance Project Plan (QAPP). In addition, the California Aquatic Bioassessment Laboratory Network (CAMLnet), and group of laboratories devoted to developing consistent laboratory procedures, has developed a standard of taxonomic effort (STE) and Standard Taxonomic List (STL) for bioassessment projects using the CSBP (August 22, 2002 version).

Quality Assurance and Control Elements

A variety of QA/QC elements have been developed and are currently available for implementation into Bay Area rapid bioassessment projects. In the proceeding pages, QA/QC procedures that can be used during macroinvertebrate bioassessment studies using the CSBP are described. BAMBI may chose to suggest that some, if not all of the following procedures be included in rapid bioassessment studies within the Bay Area in the future. The inclusion of QA/QC procedures will assure that the highest quality of data is maintained, thus creating a more robust regional data set that can be used for further analyses (e.g., IBI development).

Field BMI QA/QC Procedures

BMI Collection (CSBP) Quality Assurance – The CSBP is designed to produce random, unbiased BMI samples from riffle habitats, with the goal of collecting organisms that are representative of the entire stream reach. The following BMI collection QA procedures are suggested by the CDFG's Aquatic Bioassessment Laboratory to reach this goal:

- Most sampling reaches should contain riffles that are at least 10 meters long, one meter wide and have a homogenous gravel/cobble substrate with swift water velocity. (Note: There are approved modifications of the CSBP when these conditions do not exist. Methods to sample narrow streams, wadeable streams with muddy bottoms and channelized streams will be discussed at the BAMBI meeting).

- As described in BAMBI Issue Paper #1 – *Standardization of Field and Laboratory Procedures*, field crews should be adequately trained in the use of the BMI sampling procedures described in the CSBP, and field personnel should review the CSBP before each field season. During the training, crew members should practice collecting BMI samples as described in the CSBP. The two (2) ft² area upstream of the sampling device should be delineated using the measuring tape or a metal grid and the collection effort should be timed. Practice repeatedly until each crew member has demonstrated sampling consistency. Throughout the sampling season, assure that effort and sampling area remain consistent by timing sampling effort and measuring sampled area for approximately 20% of the sampling events. The results should be discussed immediately and need not be reported.

Physical/Habitat Assessment Quality Assurance- As part of the CSBP, a physical/habitat assessment is typically conducted using the methodology developed by the United States Environmental Protection Agency (EPA). Using this methodology, physical/habitat parameters are assessed using a ranking system ranging from optimal to poor condition. BAMBI recognizes that this rapid ranking system is qualitative and inherently subjective, relying heavily on visual evaluations. Consequently, BAMBI and/or the CDFG's ABL may suggest that more quantitative physical/habitat assessments be included in Bay Area rapid bioassessment projects in the future (Note: BAMBI may chose to develop QA/QC procedures for more quantitative physical/habitat assessments if implemented in the future). That said, the following procedures are suggested by the CDFG's ABL and are aimed at helping standardize individual observations to reduce differences in scores when using the EPA physical/habitat assessment methodology presented in the CSBP:

- Field crews should be adequately trained in the use of the EPA physical/habitat assessment procedures, and field personnel should review these procedures before each field season.
- At the beginning of each field season, all crew members should conduct a physical/habitat assessment of two practice stream reaches. Assess the first stream reach as a team and discuss in detail each of the 10 physical/habitat parameters described in the EPA procedure. Assess the second stream reach individually and when members are finished, discuss the 10 parameters and resolve discrepancies.
- Crews or individuals assessing physical/habitat quality should frequently mix personnel or alternate assessment responsibilities. At the end of each field day, crew members should discuss habitat assessment results and resolve discrepancies.
- The Project Supervisor should randomly pre-select 10 - 20% of the stream reaches where each crew member will be asked to assess the physical/habitat parameters separately. The discrepancies in individual crew member scores should be discussed and resolved with the Project Supervisor.

Field Equipment QA/QC

All field equipment used while conducting BMI rapid bioassessments should be inspected prior to each sampling event. Additionally, field instruments that require regular calibration (i.e. dissolved oxygen, pH and conductivity meters, probes, etc..) should be tested before each sampling event. Additional information regarding calibration can be found in the owners manual for each instrument.

Laboratory QA/QC Procedures

Various BMI taxonomy laboratories are currently used by Bay Area agencies conducting BMI rapid bioassessments. As part of the California Aquatic Macroinvertebrate Laboratory Network (CAMLNet), each participating laboratory should be aware of, if not follow ABL recommended QA/QC procedures in an effort to maintain the highest possible data quality. BAMBI may chose to recommend that some, if not all of the following procedures are implemented by laboratories identifying BMIs for Bay Area agencies conducting rapid bioassessment projects:

Subsampling QC (Remnant Evaluation) -All remnant samples from every project should be re-examined by a contracted taxonomist when subsampling (i.e. picking) is completed. These samples should be examined for organisms that may have been overlooked during subsampling. The number of unpicked BMI's (if any) and their identity should be recorded for each sample. For subsamples containing 300 or more organisms, the remnant sample should contain fewer than 10% of the total organisms subsampled. The remnant should contain fewer than 30 organisms for samples containing fewer than 300 organisms. If these criteria are not met, then corrective action should be initiated.

Taxonomic Identification QC – Following completion of taxonomic identification by a contracted laboratory, at least 10 percent of all samples should be evaluated by another laboratory (e.g., ABL) for taxonomic accuracy and accuracy of specimen counts. At the QC laboratory, identifications and counts should be compared with the original identifications. Any discrepancies should be checked to verify that the QC laboratory is not responsible for the error. A final report describing the any discrepancies should then be sent to the contracting agency and corrections to results, including metrics, should be made. Attached is an example of QA/QC results received from the CDFG's ABL.

Options for Improvement

The previous section provided a brief overview of important elements related the standardization of field and laboratory quality assurance and control procedures. Although it is suggested that BAMBI continue to take direction from the ABL regarding these issues, it is recommended that participants prioritize the these issues and focus future resources on providing guidance to BAMBI participants on selected topics. A discussion of resource allocation for the development of guidance information for BAMBI participants should take place at the next BAMBI workgroup meeting.

Future Steps to Improve Standardization

1. BAMBI participants should discuss and select high priority elements related to the standardization of rapid bioassessment field and laboratory quality control procedures that should be implemented in Bay Area projects; and,
2. Guidance development by BAMBI on priority elements identified by BAMBI participants in Future Step #1 (above).

**California Department of Fish and Game
Aquatic Bioassessment Laboratory**

Guidance Document for Interpretation of QA/QC Results

“Comparative Taxonomic Listing of all Submitted Samples”

The main document is a sample by sample listing of the taxa identified by the original laboratory and by the ABL. This listing includes counts of each taxon made by each lab and lists all cases where the ABL found more than one taxon in a vial. Each taxonomic list is identified by the original taxonomist/ taxonomic laboratory and the Sample ID number. This list allows for a side-by side comparison of all taxa on a per-vial basis.

This list can be sent to the original labs for their internal review and for them to evaluate whether there are taxa that they would like to recheck or contest. There should be opportunities for the labs involved to reach a consensus on final determinations. However, this process should not be included as a part of the QA/QC reporting because it can add an indefinite amount of time to the project. Labs can go back and forth several times while resolving a taxonomic identification, involving external specialists and literature review; some discrepancies may never be resolved completely.

“Listing of Taxonomic and Enumeration Discrepancies”

The second document provides a detailed grouping of all discrepancies into several categories. The category, “Disputed ID”, indicates instances in which the two labs do not agree on the identification of a particular taxon. “Relative Taxonomic Effort Discrepances” indicates cases where the original taxonomic determination is less or more precise than that of the QC lab. Although these differences in taxonomic effort are not as obvious as disagreements over identification, they can have a very strong impact on metrics calculations and often make up the majority of differences in the taxa lists of different labs.

Small amounts of difference in both categories of discrepancies are normal in this type of analysis. However, careful review of this document can help identify patterns of errors or consistent patterns of identification to lower or higher levels of taxonomic precision. Any such patterns should be brought to the attention of the original laboratory with the ultimate goal of promoting consistent levels of taxonomic resolution among labs.

In addition to taxonomic discrepancies, we evaluate differences in enumeration by the two labs. Small differences are a common occurrence in QC analysis and should not be a cause for concern unless the discrepancies are large. In our analysis, counting discrepancies are considered to be major or minor based on the following rules:
a) if the original count is greater than 20 and the difference is greater than 25% it is considered to be a major discrepancy, b) if the original count is between 5 and 20 and the

difference is greater than 50% it is considered to be a major discrepancy, c) if the original count is less than 5 all counting discrepancies are considered to be minor.

“Comparative Metrics”

The third document, presents a comparison of six common bioassessment metrics calculated from each of the two taxonomic lists in this analysis. This comparison of metrics provides an assessment of the degree of impact that different taxonomic lists have on final metrics calculations. This comparison is essential for two reasons: because small differences in taxonomic determinations can have large impacts on final metrics; conversely, many small differences in determinations may result in very insignificant differences in bioassessment metrics. The calculations in this table include differences in counts as well as taxonomic differences.

“Summary of Taxonomic and Enumeration Discrepancies”

The final document, is a summary table of all the discrepancies listed in the second document. Since it also lists the number of taxa identified (this number is taken from the list generated by the QC laboratory) it can be used to quickly determine the proportion of taxa that differ between the two lists for each sample. This version does not include information about counting discrepancies.

Documents two and four provide different types of information relevant to the evaluation of the quality of taxonomic determinations of the original laboratories.

Continuing Review of Problem Taxa

There are two main goals of an external QA/QC program: 1) to assess the quality of taxonomic data and its impacts on bioassessment metrics, and 2) to assure that taxonomic data from different sources can be included in a common database. The first goal is achieved when the initial QA/QC report is produced and the impact on bioassessment metrics is compared. The second goal is achieved when all taxonomic discrepancies have been resolved to the satisfaction of both labs. This can take time and should not interfere with the completion of the first goal.

It should be expected that most original labs will want to review the results of the QC analysis and have the opportunity to discuss any differences of opinion. For many reasons taxonomic identifications often require repeated exchanges between the taxonomic labs before the two can agree on a final determination. Occasionally, disputes may need to be resolved by sending material to an outside taxonomic specialist, or may not be able to be resolved.

Comparative Taxonomic Listing of all Submitted Samples

| Taxonomist | Sample # | Vial # | Original ID | Original Count | ABL Count | ABL ID |
|-------------------|-----------------|---------------|--------------------|-----------------------|------------------|-------------------|
| | RB-466 | | | | | |
| | | 1 | Agabus | 4 | 4 | Agabus |
| | | 2 | Bezzia/ Palpomyia | 1 | 1 | Bezzia/ Palpomyia |
| | | 3 | Orthoclaadiinae | 84 | 84 | Orthoclaadiinae |
| | | 4 | Tanypodinae | 1 | 1 | Tanypodinae |
| | | 5 | Tanytarsini | 5 | 5 | Tanytarsini |
| | | 6 | Simulium | 33 | 33 | Simulium |
| | | 7 | Baetis | 172 | 171 | Baetis 1 |
| headless-discard | | 8 | Hydroptila | 1 | 1 | Hydroptila |
| | | 9 | Gumaga | 3 | 3 | Gumaga |
| | | 10 | Nematoda | 1 | 1 | Nematoda |
| | | 11 | Naididae | 1 | 1 | Naididae |
| | | 12 | Tubificidae | 3 | 3 | Tubificidae |
| | | 13 | Megadrili | 1 | 1 | Megadrili |

IP 2

Summary of Taxonomic and Enumeration Discrepancies

| Sample # | Total Taxa | Taxonomic Discrepancies | | | | | | Counting Discrepancies | | | |
|----------|------------|-------------------------|-------------|---------------------------------------|--------------|----------|----------|------------------------|--------------|----------|----------|
| | | Disputed ID | | Taxonomic Precision Relative to QC | | | | Major | | Minor | |
| | | <i>f</i> * | <i>n</i> ** | More precise | Less precise | <i>f</i> | <i>n</i> | <i>f</i> | <i>d</i> *** | <i>f</i> | <i>d</i> |
| RB-466 | 13 | - | - | - | - | - | - | - | - | 1 | 1 |
| RB-470 | 17 | - | - | - | - | 1 | 1 | - | - | 1 | 1 |
| RB-473 | 17 | - | - | - | - | 1 | 1 | - | - | 1 | 2 |
| RB-481 | 29 | 1 | 1 | 1 | 24 | 2 | 3 | - | - | 2 | 2 |
| RB-484 | 20 | - | - | 1 | 3 | 1 | 1 | - | - | - | - |
| RB-497 | 21 | 2 | 2 | 1 | 1 | 1 | 1 | - | - | 2 | 3 |
| RB-501 | 12 | - | - | - | - | 1 | 1 | - | - | 1 | 1 |
| RB-510 | 14 | - | - | 1 | 3 | 1 | 1 | - | - | 1 | 1 |
| RB-517 | 14 | 2 | 2 | - | - | - | - | - | - | 2 | 2 |
| RB-519 | 19 | - | - | 1 | 26 | 1 | 1 | - | - | 1 | 1 |
| RB-523 | 18 | - | - | 1 | 2 | - | - | - | - | 1 | 1 |
| RB-527 | 26 | 2 | 2 | 4 | 10 | - | - | - | - | 2 | 3 |
| RB-532 | 27 | 1 | 1 | 3 | 13 | - | - | - | - | 4 | 4 |
| RB-535 | 22 | - | - | 1 | 1 | - | - | - | - | 3 | 4 |
| RB-537 | 20 | 2 | 3 | 1 | 1 | 1 | 1 | - | - | 1 | 1 |
| RB-544 | 26 | - | - | 1 | 1 | 2 | 2 | - | - | 4 | 5 |
| RB-545 | 23 | 2 | 3 | - | - | - | - | - | - | 4 | 4 |
| RB-548 | 23 | 2 | 3 | 3 | 3 | - | - | - | - | 5 | 9 |

* = the frequency of occurrence of the discrepancy, in number of samples

** = the number of organisms affected (by QC Lab count)

*** = the sum total of (absolute value of) differences in counts

IP 2

Listing of Enumeration Discrepancies

| Sample # | Vial # | Original ID | # Counted | | Difference (Original - QC) |
|----------|--------|------------------|-----------|-----|-------------------------------|
| | | | Original | QC | |
| RB-466 | 7 | Baetis | 172 | 171 | 1 |
| RB-470 | 9 | Simulium | 88 | 89 | -1 |
| RB-473 | 17 | Megadrili | 2 | 4 | -2 |
| RB-481 | 10 | Paraleptophlebia | 48 | 47 | 1 |
| | 16 | Argia | 5 | 6 | -1 |
| RB-497 | 4 | Tanytarsini | 17 | 19 | -2 |
| | 5 | Simulium | 22 | 21 | 1 |
| RB-501 | 11 | Naididae | 167 | 168 | -1 |
| RB-510 | 7 | Baetis | 67 | 66 | 1 |
| RB-517 | 6 | Orthocladinae | 58 | 57 | 1 |
| | 10 | Baetis | 17 | 18 | -1 |
| RB-519 | 13 | Hyaella azteca | 27 | 26 | 1 |
| RB-523 | 9 | Glutops | 4 | 3 | 1 |
| RB-527 | 9 | Baetis | 29 | 27 | 2 |
| | 16 | Baumannella | 3 | 2 | 1 |
| RB-532 | 8 | Baetis | 91 | 90 | 1 |
| | 9 | Ephemerella | 13 | 12 | 1 |
| | 16 | Malenka | 8 | 7 | 1 |
| | 20 | Taenionema | 9 | 8 | 1 |
| RB-535 | 8 | Ameletus | 39 | 38 | 1 |
| | 18 | Neohermes | 3 | 2 | 1 |
| | 19 | Neohermes | 3 | 1 | 2 |
| RB-537 | 4 | Tanypodinae | 1 | 2 | -1 |
| RB-544 | 3 | Prosimulium | 100 | 98 | 2 |
| | 7 | Baetis | 89 | 88 | 1 |
| | 8 | Caenis | 20 | 19 | 1 |
| | 17 | Argia | 15 | 14 | 1 |

Minor Counting Discrepancies

IP 2

Listing of Taxonomic Discrepancies

| Sample | Vial # | Original ID | Final ID | QC Final ID | Taxonomic level | # Organisms | Comments |
|---|--------|----------------|----------|------------------|-----------------|-------------|----------|
| RB-470 Probable sorting error | 9 | Simulium | | Naididae | Phylum | 2 | |
| Original ID less precise | 13 | Sperchontidae | | Sperchon | | 1 | |
| RB-473 Probable sorting error | 17 | Megadrili | | Orthocladinae | Phylum | 2 | |
| Original ID less precise | 12 | Sperchontidae | | Sperchon | | 1 | |
| RB-481 Disputed ID | 7 | Baetis | | Paraleptophlebia | Family | 1 | |
| Probable sorting error | 16 | Argia | | Orthocladinae | Order | 1 | |
| Original ID less precise | 8 | Dipheter | | Dipheter hageni | | 1 | |
| Original ID more precise | 18 | Sperchontidae | | Sperchon | | 2 | |
| RB-484 Terrestrial Organism | 19 | Hyaella azteca | | Hyaella | | 24 | |
| Original ID less precise | 2 | Coleoptera | | Terrestrial | | 1 | |
| Original ID more precise | 9 | Sperchontidae | | Sperchon | | 1 | |
| Original ID more precise | 10 | Hyaella azteca | | Hyaella | | 3 | |