

**Southern California Bight 2013
Regional Marine Monitoring Survey
(Bight'13)**

**Macrobenthic (Infaunal)
Sample Analysis
Laboratory Manual**

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Prepared for:
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June 2013

**SOUTHERN CALIFORNIA BIGHT
2013 REGIONAL MONITORING PROGRAM**

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INTRODUCTION

This document describes laboratory procedures for the analysis of macrobenthic (infaunal) samples collected for the Southern California Bight 2013 Regional Marine Monitoring Program (Bight '13). The procedures are based on existing practices in Publicly Owned Treatment Works (POTW) monitoring programs within the region and those employed during the 1994 Southern California Bight Pilot Project (SCBPP) and previous Southern California Bight Regional Monitoring Programs (Bight '98, Bight '03, and Bight '08). Some modifications have been made to ensure data comparability, facilitate coordination of quality control steps during the Bight '13 infaunal survey, and meet the requirements of the Bight '13 Information Management Plan. It is the responsibility of each participating laboratory's supervisor to assure that:

- The detailed procedures described in this manual are followed during sample processing and analysis,
- All Quality Control (QC) steps are implemented,
- Data submissions conform to the stipulated standards,
- Schedules are met for sample analysis, QC, data submission
- Copies of all records, forms, and documents generated in the process are securely maintained on file until all aspects of the survey and resulting reports are completed.

All stages of infaunal sample processing and analysis, following receipt in the laboratory of samples from the field, including QC and data submission are described in this manual. In overview, the process consists of the following tasks and activities (Figure 1) which are described in sections as indicated below:

- 1) **Sample Treatment and Storage:** The sample is washed free of fixative and transferred to an alcohol solution for processing and/or storage (**Section 1**),
- 2) **Sorting:** All organisms and plastic debris are removed from the grunge contained in the sample and sorted into taxa lots and debris to facilitate subsequent taxonomic analysis (**Section 2**)
- 3) **Taxonomic Analysis:** All specimens in the samples are identified to the lowest practical level, most often species, and counted (**Section 3**),
- 4) **Data Submission:** Resulting data are loaded to an electronic data file compliant with this manual and the Bight '13 Information Management Plan and submitted to the project Information Management Officer (**Section 4**).
- 5) **Quality Control:** QC is required for steps 2 and 3 (**Section 5**) to ensure data consistency. QC for step 2 involves re-sorting a minimum of 10% of the grunge from each sample. QC for step 3 consists of reanalyzing 10% of the samples processed by each laboratory and required taxonomist participation at Southern California Association of Marine Invertebrate Taxonomists (SCAMIT) workshops that identify and resolve Bight '13 taxonomic problems. Results of this process are used to determine whether the measurement quality objectives (MQOs) established for each step are met.
- 6) **Record keeping and Procedural responsibilities** are described in **Section 6**. Examples of forms to be used during processing and QC are presented in **Section 8**.

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It is essential that all participating taxonomists have the expertise and experience necessary to assure that Bight'13 macrofaunal data meet standards set during previous regional surveys. Qualification criteria for taxonomists who did not analyze macrofaunal samples for previous Bight surveys are described in **Appendix A**.

In addition, taxonomists are required to participate in the series of workshops jointly sponsored by Bight'13 and the Southern California Association of Marine Invertebrate Taxonomists (SCAMIT) focusing on taxonomic problems arising during analysis of the Bight'13 samples. These workshops culminate in a synoptic review of taxon names in the data set compiled from submissions by all participating laboratories.

Copies of this manual are available on the web site of the Southern California Coastal Water Research Project (SCCWRP) (<http://www.sccwrp.org>).

Benthic Sample Processing Flow

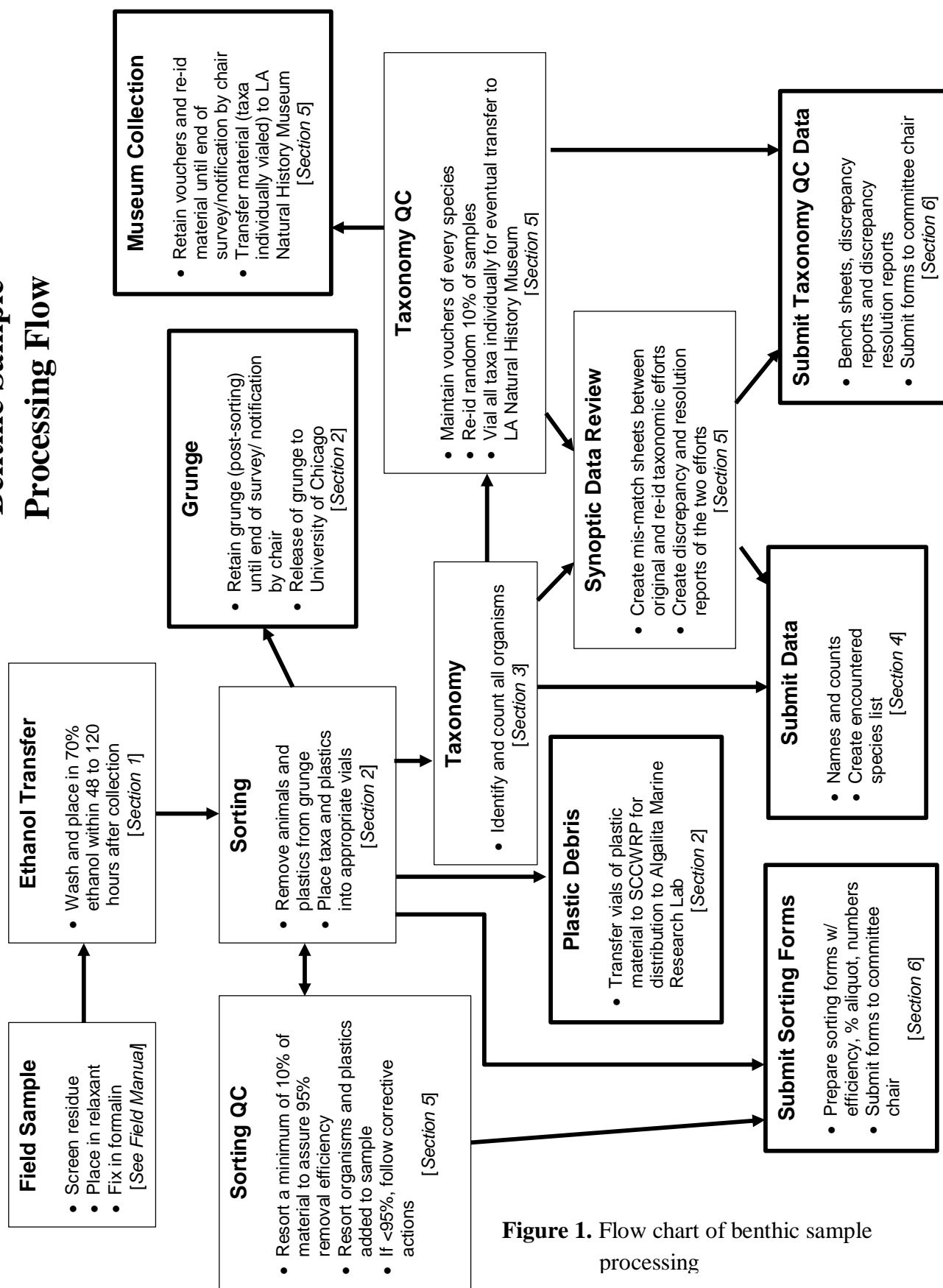


Figure 1. Flow chart of benthic sample processing

1. SAMPLE TREATMENT AND STORAGE

- 1.1 Upon receipt in the laboratory, samples will be in formalin fixative and must be washed and transferred to a preservative. The removal of formalin is necessary for two reasons. Formaldehyde becomes increasingly acidic over time and prolonged exposure damages organisms with calcareous structures (e.g., shelled mollusks). Also, formaldehyde is a noxious, potentially dangerous chemical; its replacement with ethanol makes subsequent handling of the sample safer. Other benefits of the washing process are the removal of excess silt from mudballs and fecal pellets that may have broken down during fixation and, in some cases, the opportunity to separate the bulk of organisms in a sample from the inorganic grunge through an elutriation process.
- 1.2 The samples are to remain in buffered fixative for a minimum of 48 hours. No sample should remain in fixative for longer than 120 hours.
- 1.3 The preservative to be used for all stages of Bight'13 infaunal samples is a 70% solution of ethanol. **Denatured alcohol is not permitted.** Rose bengal may **not** be used to stain organisms.
- 1.4 It is recommended that the preservative for mollusk and other calcareous voucher specimens be buffered with marble chips to reduce possible acidity, especially if the ethanol is produced by industrial distillation rather than fermentation.
- 1.5 Procedure
 - 1.5.1 Select an appropriate 0.5mm or smaller sieve, and examine the mesh for holes and adhering organisms. Working under a fume hood with eye protection, decant the fixative through the clean and intact sieve
 - 1.5.2 After decanting the formalin, refill the sample container with water, agitate gently by swirling, and wash the entire sample into the sieve.
 - 1.5.3 Gently wash the sample with a low-pressure stream of water to remove any fine silt.
 - 1.5.4 Using a scoopula and wash bottle containing preservative (70% ethanol), transfer the sample back to the sample container, top the sample off with preservative, and tightly affix the lid.
 - 1.5.5 Place an internal label in each sample container bearing the station name, sampling date, split number (if more than one container is used; e.g., 1 of 2). Labels are to be written in pencil or indelible ink on 100% rag-paper, poly- paper, or other paper suitable for permanent wet labels.

- 1.5.6 After each sample is washed, closely examine the sieve to ensure that all organisms have been removed (any organisms found should be reunited with the sample. Then, thoroughly rinse the sieve to avoid cross contamination of subsequent samples.
- 1.5.7 Store infaunal samples in a safe and secure manner protected from environmental extremes. Avoid temperatures above 30° C as high temperatures will accelerate evaporative loss of preservative
- 1.5.8 Routinely inspect all samples to ensure that the container closure is tight and the preservative level adequate. If evaporative loss of preservative is evident, top- off the sample using 95% ethanol and check the lid or rim of the jar for defects and possible replacement. Do not use 70% ethanol for this purpose, as it will lead to dilution of the sample preservative because of the different evaporation rates of ethanol and water.

2. SAMPLE SORTING

- 2.1 Sorting is the process by which organisms in a benthic sample that were alive at time of collection are removed from the organic and inorganic residues (grunge) that compose the sample and sorted into broad taxonomic categories for subsequent taxonomic analysis. Sorting must be accurate and complete to assure the value of all the subsequent steps in the sample analysis process. In addition to the removal of benthic organisms, plastic debris (pieces of plastic greater than 1mm in diameter or length) will also be removed and vialled for analysis.
- 2.2 Procedure
- 2.2.1 All laboratories participating in the Bight'13 infaunal survey have established sorting procedures that are compatible with the aims of this survey. The following points stipulate those elements essential to the process or unique to Bight'13.
- 2.2.2 Begin the sorting process by filling out a Bight'13 Sorting Record Form (page 28) with the sample name, date, sorter's name, and date sorting begins. If the sample consists of more than a single jar, these jars are to be treated together as a single sample. Make sure you have all jars composing the sample.
- 2.2.2 Sort the sample under a stereo microscope. It is recommended that the sample be sorted in small-volume increments. It has been shown that subsample increments with smaller volumes will not hide small organisms thereby producing better sorting results. Partitioning a sample into large and small size components can also produce better sorting results in the following manner; 2.0mm sieve can be nested above the 0.5mm sieve to partition the sample into large and small sized particle fractions to facilitate the sorting process.
- 2.2.3 The entire sample is to be sorted. If an unusual sample is encountered for which sorting of an aliquot may be a reasonable alternative, the laboratory supervisor is to contact the Bight'13 Benthic Committee Chairperson. The decision whether to allow sorting by aliquot will be made by the Benthic Committee Chair and Co-chair.
- 2.2.4 **ELUTRIATION.** If a sample is primarily coarse sand, sorting can be greatly facilitated if inorganic material in the sample is separated from the lighter organic grunge and organisms by the following elutriation process.
- 2.2.4.1 After washing the ethanol from the sample, spread the sample material out in a shallow pan and cover with water.

- 2.2.4.2 Gently agitate the sample by hand to allow the lighter fraction of grunge and organisms to separate from the heavier material.
- 2.2.4.3 Decant the water containing the lighter material through the sieve. Repeat the process several times until no more material is observed being carried off in the decanted water.
- 2.2.4.4 Collect the material retained on the sieve into a small sample container, and top-off with preservative. Return remaining material to the original sample container along with the balance of the sample material. Fill the container with preservative and tightly affix the lid. Be sure that both containers are properly labeled with internal labels.
- 2.2.5 All sorting must be done in 70% ethanol, except for sorters where health and safety issues exist, with care taken to ensure that the sample being sorted is always fully covered with alcohol. If necessary, sorting may be performed using water, but care must be taken to minimize the time when specimens are not in 70% ethanol. Samples may not be left over night in water, as specimens may degrade. Samples must be placed back into 70% ethanol at the end of each day. For large samples, only placing into water that portion to be sorted in a day's time is advisable.
- 2.2.6 The organisms removed from the sample are sorted into taxonomic lots for subsequent taxonomic analysis. Each laboratory will determine the taxonomic level of sorting adequate to their needs for subsequent sample analysis by their taxonomists.
- 2.2.7 Remove all individual organisms and fragments from the sample with the exception of nematodes, foraminiferans and planktonic species, or planktonic life stages of benthic organisms. All fragments, such as decapod chelae and legs, should be placed in their respective taxa lots. Sorters are to be instructed "***If in doubt, pick it out.***"
- 2.2.8 Remove all pieces of plastic greater than 1mm in diameter or length from the sample. Sorters are to be instructed "***If in doubt, pick it out.***" Pieces should be counted and placed in separate vial with DI water or ethanol. Material should be labeled "Debris/Plastic and treated as a separate taxon lot.
- 2.2.9 Note on the Sorting Record Form (page 29) the number and identity of taxa lots composing the sorted sample, the number of containers used if sample is split, and the time (to the nearest ½ hour) required to sort the sample.

- 2.2.10 Sorters will be required to count animals (head ends) while sorting and note the number on the Sorting Record Form. This facilitates sorting quality control by providing a number for comparison of QC re-sorting results (Section 5.5).
- 2.2.11 Sorters are asked to not remove animals from their tubes. At most, the sorter is asked to verify that a tube has an animal inside. That said, it is better to be precautionary and include a tube in the appropriate lot if there is risk in damaging a specimen in the process of verifying the tube is/was occupied. This is the same for shelled mollusks. The sorter should not damage the shell, e.g., snip away at the aperture, to determine whether there is an animal within.
- 2.2.12 Aggregate the taxa lots into one or more sample containers. Each taxa lot should be internally labeled with the station name, sampling date, station depth, and sorter's initials. Place an internal label in each vial/container with this information and split number (i.e., 1 of 2, 2 of 2) if more than one container is used. Labels are to be written in pencil or indelible ink on 100% rag-paper, poly- paper, or other paper suitable for permanent wet labels (e.g., Resistall). Minimally, the material must be segregated into the following taxa lots:

Annelids

Annelid fragments

Arthropods

Echinoderms (non ophiuroid)

Ophiuroids

Ophiuroid arms

Molluscs

Misc. Phyla (e.g., Cnidarians, Nemerteans)

Debris/Plastics

- 2.2.13 As a special feature of sorting during Bight '13, sorters will be required to sort plastics larger than 1-mm in diameter or length from the sediments as if they were organisms, placing them in a separate Debris/Plastics vial. During QC of each sample any additional plastic pieces will be added to a second plastics vial. There is no MQO for plastics and resort plastics are not a sort QC parameter. The number of vials should be noted on the sorting sheet (data form 1).

3. TAXONOMIC ANALYSIS

- 3.1 The object of taxonomic analysis is to accurately identify all organisms contained within each sample to the lowest possible taxonomic category and to provide an accurate count of the organisms in each identified taxon.
- 3.2 The goal of the Bight'13 infaunal survey is to provide species level identifications whenever possible. However, because of difficulties in the taxonomy and the lack of expertise within participating laboratories the following exceptions are made:
- Kinorhynchs are identified to Phylum Kinorhyncha
 - Oligochaete annelids are identified to Sub-class Oligochaeta (*note may change with designation of Oligochaeta as a specialty taxon*)
 - Hirudinean annelids are identified to Class Hirudinea
 - Podocopid ostracods are identified to Order Podocopida
 - Harpacticoid copepods are identified to Order Harpacticoida
 - Insecta arthropods may be identified to Sub-class or Order
- 3.3 The number of organisms reported must account for all organisms in a sample alive at the time of collection. A corollary goal is to not count any individual more than once. Inevitably, samples contain fragments of organisms. Fragments of bilaterally symmetrical organisms will be identified and counted only if the fragment includes the anterior end of the organism. For radially symmetrical organisms (e.g., ophiuroids, anthozoans) only fragments bearing the majority of the oral disk will be identified and counted. Also, care must be taken to avoid reporting empty mollusk shells or crustacean molts in the data.
- 3.4 The goal of the survey is to describe the macroinvertebrate infauna and epifauna living in soft-bottom habitats. Hard-bottom epifaunal organisms may occur incidentally in samples, particularly in settings where samples are collected immediately adjacent to hard structures (e.g., in harbors near piers). As any records of these incidental contaminants would not be included in the analytical use of the data, these specimens are not to be counted nor included in the submitted survey data. Their presence may be noted on the bench sheets.
- 3.5 Attached parasites and other epibionts may be noted on the bench sheet as present but are not to be reported in the submitted survey data. Ectoparasites of fish such as cymothid isopods, which may be temporary members of the benthic community, are counted and reported in the submitted survey data.
- 3.6 Each participating laboratory will use their own taxonomy bench sheets for recording the identifications and counts.

- 3.7 Nomenclature and orthography follows that used in *A Taxonomic Listing of Soft Bottom Macro- and Megainvertebrates from Infaunal and Epibenthic Monitoring Programs in the Southern California Bight.*, Edition 8 (SCAMIT, 2013). This list represents a consensus for standard usage of taxa names in POTW monitoring programs in the Southern California Bight.
- 3.8 Taxonomists are to employ two standard notations (*Voucher* and *Exclude*) for the annotation of their bench sheets. While other non-standard notation may also be used, the use of these standard notations is required where applicable. In addition, both the Voucher and Exclude codes will be included as part of the electronic data record. See the Bight '013 Information Management Plan for the proper form of these fields for data submission.
- 3.9 Voucher Notation
- 3.9.1 Form: The annotation employed for this purpose on the bench sheet is the letter V followed by the number of specimens removed from the sample. (i.e., V-3)
- 3.9.2 Purpose: To note the removal of specimens from a sample for use as Bight '13 vouchers. Use of this notation on the bench sheet is essential to the process of tracking voucher records and quality control/assessment. Removal of organisms without annotation confuses the resolution of discrepancies during quality control re-analysis, and leads to overstatement of error rates. Inclusion in the electronic data summation allows a complete list of Bight '13 vouchers to be extracted from the data.
- 3.9.3 Rule of Use: Removal of any specimens from a sample to the Bight '13 voucher collection is clearly noted on the bench sheet by means of the Voucher notation.
- 3.9.4 In addition to the voucher specimens required for the Bight '13 Voucher Collection (see 5.6.16-20 below), individual labs or taxonomists may remove specimens of each taxon for their own voucher collections (note on data entry sheet). The removal of this material must also be clearly noted (by means other than the voucher notation) on the bench sheet in order to account for their effect on quality control re-analysis. The following would satisfy the requirement for clear notation:
- “V-2, HY-1 voucher”
- indicating 2 specimens removed to the Bight '13 voucher collection and 1 specimen to Hyperion's collection.
- 3.9.5 The Voucher notation will be included as part of the electronic data record submitted by each laboratory. See the Bight '13 Information Management Plan for the proper format for its inclusion in the data file. Separate columns will be available to denote whether a specimen was vouchered and the number of organisms in the Bight '13 voucher collection and that of the individual lab or taxonomist's voucher collection.

3.10 Exclude Notation

3.10.1 Form: The letters EX written on the row of the bench sheet containing the data record for the taxon to be excluded

3.10.2 Purpose: Provides an aid to data analysis when calculating metrics using the number of taxa present (e.g., diversity, species richness). This field in the final data set represents the taxonomist's recommendation that the reported taxon be excluded from counts of the number of taxa reported in the sample.

3.10.3 Rule of Use: The Exclude annotation is made on the bench sheet whenever a taxon should be excluded from counts of the number of taxa reported in the sample. This annotation is employed when three conditions co-exist:

The identification is not at the species-level (e.g., Pleustidae or *Polydora* sp),

And

The reported taxon is represented in the sample by other members of the same taxon, which have been identified at lower levels,

And

The taxonomist cannot determine if the specimen is distinct from the other members of its taxon represented in the sample.

3.10.4 Examples of Use:

Both *Dipolydora* sp and *Dipolydora socialis* are reported in a sample and the taxonomist cannot determine if the specimen reported as *D. sp* is distinct from *D. socialis*. Exclude (annotate record on bench sheet with **EX**)

An unidentifiable onuphid polychaete is reported as Onuphidae. It is the only member of its family present in the sample. **Do Not Exclude**

Both *Modiolus* sp and *Modiolus capax* are reported in a sample. However, the taxonomist is confident that the specimen identified at the genus-level is not *M. capax*. **Do Not Exclude**

3.10.5 It is necessary that the taxonomists make this evaluation during sample analysis (i.e., by annotation of the bench sheet). It cannot be effectively applied after the fact, as there is no way of determining later whether the third criterion for use was met.

3.10.6 The Exclude notation will be included as part of the electronic data record submitted by each laboratory. See the Bight'08 Information Management Plan for the proper format for its inclusion in the data file.

3.11 Temporary "In-House" provisional names are erected for those specimens that a taxonomist considers to be distinctive but cannot match with an existing description or other provisional name on the SCAMIT Ed 8 Species List. These provisional names act

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as markers for these taxa, allowing them to be consistently discriminated in the samples for which the taxonomist is responsible. In-house provisional names are supported by a written differential diagnosis (and figures if necessary) sufficient to allow taxonomists in the other participating laboratories to recognize the species. These diagnoses are sent to other taxonomists participating in the survey. The provisional name is formed from the lowest taxon name in which the specimen may be placed with certainty followed by a composite name containing the laboratory's two-character code (see below) and a number; for example, *Rhachotropis* sp LA2 or Ampharetidae sp SD1. Note there is no space between the agency code and the identifying number.

<i>Lab Name</i>	<i>Lab Code</i>
ABC Labs	AB
Weston	WS
CLAEMD	HY
CSD	SD
LACSD	LA
OCSD	OC
MBC	MB
DCE	DC
EcoAnalysts	EA

- 3.12 Timely and frequent communication among the taxonomists analyzing the samples will improve the data produced in the survey. An e-mail list-server (B13taxon@sccwrp.org) will be established to facilitate this communication. All taxonomists involved in the Bight'13 survey will be members of the list. Messages posted to the list will automatically post to all members, assuring wide and uniform distribution of the contents. Names and e-mail addresses of all taxonomists processing Bight'13 samples will be provided by each participating laboratory to the Bight'13 Benthic Committee Chairperson before sample collection begins, or in the case of qualified taxonomists joining after sample processing begins, as soon as possible. List-server messages are archived. They will be available for review at least until the Bight'13 Benthic Report is published.
- 3.13 Appropriate uses of the list server are informing the other members of unusual or newly encountered species, the erection of in-house provisionals, and requests for information or assistance.
- 3.14 Messages posted to the list-server should always include in the subject line the critical topic taxon (if any) to which the posting refers followed by a referent higher taxonomic category in parentheses. For example:

Balanoglossus (Hemichordata)
or
Guerneia sp MB1 (Gammaridea: Dexaminidae)

- 3.15 Following identification and enumeration, all the specimens are retained in taxa lots within the sample. Minimally, the material must be segregated into the following taxa lots:

Annelid lots:

Oligochaeta
Cirratulidae
Misc Polychaetes
Polychaete frags

Arthropod lots:

Arthropoda
Photis spp

Molluscan lots:

Mollusca

Echinoderm lots:

Ophiuroidea
Ophiuroidea arms
Misc Echinoderms

Misc. Phyla lots:

Misc Phyla (a collective lot)

This level of separation facilitates the quality control process and eases both the burden of re-analysis resulting from failure of a laboratory to meet the measurement quality objective and the recovery of material during the end-of-survey synoptic review. In addition, any taxon subject to specialty taxonomic treatment (see 5.6.22 below) is to be segregated into a lot for delivery to the designated specialist.

Further segregation of all polychaetes at the family level has been found useful in some POTW monitoring surveys and is recommended.

- 3.16 All taxa lots within a sample are provided an internal label with the program designation (*i.e.*, B'13), taxa lot name, station name and depth and taxonomists initials. These taxa lots are contained in vials and all of the lots in a sample are aggregated into one or more sample containers. If a taxa lot includes bulky specimens, they may be placed loose in the sample container (accompanied by a loose label) along with the vials containing the remainder of that and other taxa lots. An internal label is placed in each sample container bearing the program designation (*i.e.*, B'13), station name, sampling date, depth, and split number (if more than one container is used; *e.g.*, 1 of 2). Labels are written in pencil or indelible ink on 100% rag-paper (*e.g.*, Resistall or equivalent), for permanent wet labels. Each laboratory will retain bulk taxa sample lots until informed by the benthic committee chair (or designee). This will be at a point in time 5 years after completion of the project or 6 months after the final version of the B' 13 Benthic Report is released, whichever occurs first.

4. DATA SUBMISSION AND THE FORM OF TAXONOMIC NAMES

- 4.1 All data submissions must meet the formatting requirements of the Bight '13 Information Management Plan.
- 4.2 In particular, it is essential that all taxon names be standardized in spelling and form. Because the "species" field is one of the key fields for defining a unique record, exactitude is required.
- 4.3 To minimize the problem of variants, a standard for the spelling and formation of names has been specified prior to the survey. This standard is based on *A Taxonomic Listing of Soft Bottom Macro- and Megainvertebrates from Infaunal and Epibenthic Monitoring Programs in the Southern California Bight*, Edition 8 (SCAMIT, 2013). The full Edition 8 document will be available at the SCAMIT website (www.SCAMIT.org).
- 4.4 The name used to represent a taxon should be that listed in the SCAMIT Taxonomic List. If the taxon has not previously been reported in the region and is consequently not on the SCAMIT List, it may still be reported. Taxonomic usage should follow that in WoRMS (<http://www.marinespecies.org/>) and the primary literature. The chair of the IM committee should be advised of submission of an unlisted name and full details (taxonomic hierarchy, authorship, etc) should be provided prior to data submission.
- 4.5 The following examples of data submission problems from Bight '98, Bight '03, and Bight '08 are included to emphasize the importance of adhering to the Information Management Plan requirements for submission of taxonomy-based data.

- 4.5.1 The species field is to contain taxon names only. Do not include citation of authorship, comments or other information

<i>As Submitted</i>	<i>Should Have Been</i>
Anthozoa, unid.	Anthozoa
<i>Bugula neritina</i> (colonial)	<i>Bugula neritina</i>
<i>Enopla</i> sp A SCAMIT 1995	<i>Enopla</i> sp A
<i>Heteroserolis</i> n. sp.?	<i>Heteroserolis</i> sp
<i>Tubulanus polymorphus/pellucidus</i> frags only	<i>Tubulanus polymorphus</i> not submitted

- 4.5.2 The species field is to contain formal scientific taxon names only. Do not use common names or anglicized forms

<i>As Submitted</i>	<i>Should Have Been</i>
Cirriped	Cirripedia
megalopa	Decapoda (<i>note the larval stage</i>)
fish	<i>a particular fish taxon (at any level)</i>

- 4.5.3 The form (spelling, punctuation) of the names are to follow the SCAMIT Taxonomic listing. Note that the SCAMIT list avoids all forms of punctuation (other than parentheses around subgeneric names) within a taxon name.

As Submitted

Scoloplos "armiger"

Semele sp.

Aphelochaeta spp

Prionospio jubata

Should Have Been

Scoloplos armiger Cmplx

Semele sp

Aphelochaeta sp

Prionospio (Prionospio) jubata

- 4.5.4 In forming or using provisional names based upon the two character agency code, do not include a space between the agency code and the number

As Submitted

Anobothrus sp LA 1

Malmgreniella sp SD 3

Should Have Been

Anobothrus sp LA1

Malmgreniella sp SD3

- 4.6 **ENCOUNTERED SPECIES LIST:** All submissions are to be accompanied by an encountered species list providing the taxon name and, for species level names (including provisional taxa), authorship citation. These lists will facilitate the recognition of variant forms within the compiled data set and, more importantly, the cases of potential or real homonymy or synonymy. A comments column is provided to submit additional information that may be of value in evaluating the list entries.

- 4.6.1 The encountered species list should contain every unique taxon name occurring within the data being submitted.

- 4.6.2 The encountered species list should be in the form of a four column Excel worksheet with the following format (see example below):

Column A = Taxon

Column B = Authority (*for species-level taxa*)

Column C = Lab (*the Bight '13 Information Plan agency code*)

Column D = Comments

- 4.6.3 The list should be sorted alphabetically by taxon name

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Taxon	Authority	Lab	Comment
Acanthina sp		WS	
Acanthomunna tannerensis	Schultz, 1966	WS	
Acanthoptilum sp		WS	
Acila castrensis	Hinds 1843	WS	
Aclis sp		WS	
Acoetes pacifica	Treadwell 1914	WS	
Acontifera sp A	Ljubenkov 2010	WS	
Acrocirridae		WS	
Acteocina cerealis	Gould 1853	WS	
Acteocina culcitella	Gould 1853	WS	
Acteocina harpa	Dall 1871	WS	
Acteocina inculta	Gould 1855	WS	
Actiniaria		WS	
Actiniaria sp 49	Ljubenkov 2003	WS	
Acuminodeutopus heteruropus	J. L. Barnard 1959	WS	
Adontorhina cyclia	Berry 1947	WS	
Adontorhina lynnae	Valentich Scott 2000	WS	
Aeolidioidea		WS	
Aglaophamus erectans	Hartman 1950	WS	
Aglaophamus verrilli	McIntosh 1885	WS	
Alaba sp		WS	
Alderia willowi	Krug, Ellingson, Burton and Valdés 2007	WS	
Alia carinata	Hinds 1844	WS	
Alpheidae		WS	
Alpheus californiensis	Holmes 1900	WS	
Alpheus sp		WS	
Alvania rosana	Bartsch 1911	WS	
Alvania sp		WS	
Amaeana occidentalis	Hartman 1944	WS	
Amage anops	Johnson 1901	WS	
Amakusanthura californiensis	Schultz 1964	WS	
Amathia sp		WS	
Ambidexter panamensis	Abele 1972	WS	
Americhelidium rectipalmum	Mills 1962	WS	
Americhelidium shoemakeri	Mills 1962	WS	
Americhelidium sp		WS	
Americhelidium sp SD1	Pasko 2005 §	WS	
Americhelidium sp SD4	Pasko 2005 §	WS	
Americorophium salmonis	Stimpson 1857	WS	
Ammonothea hilgendorfi	Böhm 1879	WS	
Ampelisca agassizi	Judd 1896	WS	
Ampelisca brachycladus	Roney 1990	WS	
Ampelisca brevisimulata	J. L. Barnard 1954	WS	
Ampelisca careyi	Dickinson 1982	WS	
Ampelisca coeca	Holmes 1908	WS	
Ampelisca cristata cristata	Holmes 1908	WS	
Ampelisca cristata microdentata	J. L. Barnard 1954	WS	
Ampelisca hancocki	J. L. Barnard 1954	WS	
Ampelisca indentata	J. L. Barnard 1954	WS	
Ampelisca lobata	Holmes 1908	WS	
Ampelisca milleri	J. L. Barnard 1954	WS	
Ampelisca pacifica	Holmes 1908	WS	
Ampelisca pugetica	Stimpson 1864	WS	
Ampelisca romigi	J. L. Barnard 1954	WS	
Ampelisca sp		WS	
Ampelisca unsocalae	J. L. Barnard 1960	WS	

Example of a partial encountered species list for submission

5. QUALITY CONTROL

- 5.1 The laboratory analysis of infaunal samples for Bight'13 involves three processes: sample washing and preservation, sample sorting, and organism identification and enumeration. Quality assurance in the form of procedures and standardized reporting requirements are provided in this document for all three processes. Quality control exercises will be implemented at stages for which MQOs have been established (sample sorting, identification and enumeration). These exercises include repeating the procedures at each of these stages for a sub-set of samples. The results will be used to determine achievement of the MQOs established for each stage.
- 5.2 For the most challenging process, organism identification, additional quality control steps are included in order to foster comparability among the taxonomic data sets produced by the participating laboratories and taxonomists
- 5.3 Where warranted, the Benthic Committee Chairperson (or designee) may conduct audits of each laboratory while sample analysis is underway to assure that the Bight'13 procedures are being followed.
 - 5.3.1 An audit would be invoked in those cases where there was evidence of consistent mistakes or QC failures in sorting or taxonomic identification; indicating that “best practices” are not being followed in a given lab (See sections 2 and 3).
 - 5.3.2 The audit could entail, among other things, a review of documented corrective actions by the internal QC person/lab manager, requests for external re-identification or re-sorting, or demonstration of improvement.
- 5.4 Sample Sorting
 - 5.4.1 Quality control of sorting is essential to assure the value of all the subsequent steps in the sample analysis process. An accuracy MQO of 95% removal efficiency has been set for this stage of the sample analysis.
 - 5.4.2 A standard sorting form (page 29) is used for tracking the sample. It includes the name of the technician responsible, time required for sorting, comments, and re- sorting results. Re-sorting of samples is employed for quality control of sorting.
 - 5.4.3 A minimum of 10% of all material in Bight'13 samples will be re-sorted to monitor sorter performance and to determine achievement of the MQO of 95%. In practice, the minimum 10% of all material stipulation will be achieved by the evaluation of each sorter via the aliquot method (sections 5.4.4-6).

- 5.4.4 Sorting efficiency should be assessed following the *aliquot method*, wherein a representative aliquot of at least 10% of the sample volume of every sample processed is re-sorted by an experienced sorter who is different than the original sorter. Re-sorting of a higher percentage of a sample may be required or optionally performed to ensure MQO performance.
- 5.4.5 Aliquots may be obtained by standardizing the sample volumetrically, for example by stirring and then withdrawing 10% of the sample with a Hensen-Stemple pipette. Alternatively the grid method can be used. This is accomplished by spreading the sample evenly in a gridded shallow pan and selecting a random 10% of grids/cells for re-sort. The responsible supervisor of each participating laboratory selects the method of obtaining a sample aliquot.
- 5.4.6 The re-sorting process is to follow the procedures given in Section 2 of this document.
- 5.4.7 Percent sorting efficiency is calculated as follows:
- $$\% \text{Efficiency} = 100 * \{ \#_{\text{orig}} / [\#_{\text{orig}} + (\#_{\text{resort}} / \text{aliquot fraction})] \}$$
- 5.4.8 If sorting efficiency is greater than 95%, no action is required. Sorting efficiencies below 95% will require continuous monitoring of that technician until efficiency is improved.
- 5.4.9 Organisms found in the re-sort should be given to the appropriate taxonomist for identification and enumeration for inclusion in the results from the sample.
- 5.4.10 The sorting labs will also remove plastic debris during their QC for infaunal samples. Any debris items found during the re-sorting will be placed in a separate vial from that used during the primary sort. This QC sort vial will be sent along with the primary sort vial to AMRI for analysis. The debris found in the QC process will not be used to calculate sorting efficiency.
- 5.4.11 The calculated sorting efficiency is recorded on the Sorting Form for each sample (page 29) for which QC re-sorting is conducted.
- 5.4.12 The laboratory responsible for the sorting must retain sample grunge after sorting. It is to be properly labeled and preserved with 70% ethanol. Upon completion of all quality control and assessment steps for the survey, including taxonomic re-analysis and discrepancy resolution (Section 5.5), the Benthic Committee Chairperson (or designee) will notify each participating laboratory that the sample grunge may be prepared for transport to Dr. Susan Kidwell of the University of Chicago.

5.5 Quality Control and Quality Assessment of Taxonomic Analysis

- 5.5.1 The goal of taxonomic analysis for the Bight '13 infaunal survey is species level identification of all macrobenthic organisms collected and an accurate count of each species. The procedures for sample re-analysis are based upon those developed and employed in the SCBPP, Bight '98, Bight '03, and Bight '08 surveys. This task is complicated by the participation of multiple laboratories and taxonomists in the analysis. Two approaches are taken for providing data quality control. The first is an assessment of each laboratory's accuracy by re-analysis of a subset of samples from each laboratory. The second focuses on ensuring consistent and comparable results among the participating taxonomists through cooperative activities under the aegis of SCAMIT.
- 5.5.1.1 Participation in SCAMIT involves, but is not limited to, attending monthly meetings and workshops whose topic related to a taxonomist's area of responsibility or expertise (e.g., polychaetes, arthropods, Mollusca, etc.). In addition, participation on ad hoc committees (e.g., Species List Review Committee), while not required, is strongly encouraged. In instances where multiple taxonomists at a laboratory have the same specialty, a single representative may fulfill the SCAMIT meeting attendance requirement for all by transmitting the meeting's contents to the other taxonomists in the laboratory.
- 5.5.1.2 Failure to comply with these standards (i.e., missing 2 or more meetings covering a taxonomist's area of expertise/responsibility during the Bight '13 taxonomic identification period) can result in disqualification of that taxonomist or taxonomic laboratory from Regional Monitoring Program participation. Any determination will be made by the Benthic Committee Chairperson after consultation with the SCAMIT officers. Logs of SCAMIT meeting attendance will be documented in the SCAMIT minutes and newsletter. They will be provided upon request to the Benthic Committee Chairperson.
- 5.5.2 Quality control is provided by the re-identification of 10% of the samples processed by each laboratory. Samples for re-identification are selected randomly from each lab's assigned set of samples by the Bight '13 Benthic Committee Chairperson (or designee) and re-distributed to the QC laboratories.
- 5.5.3 The re-identification will be conducted at participating QC laboratories and by taxonomists other than those who originally analyzed the samples. The taxonomists conducting the re-identification will not have access to the original results.

- 5.5.4 Each laboratory's supervisor will be informed by the Benthic Committee Chairperson (or designee) which samples are to be re-identified. The laboratory supervisor is responsible for assuring that these samples are made available to the laboratory responsible for re-identification in a timely manner.
- 5.5.5 The specimens in each sample will be re-identified and enumerated using the procedures given in Section 4 of this document. Results are reported on the QC laboratory's bench sheet. Upon completion of the re-analysis, the results are submitted to SCCWRP and a match/not match comparison of primary and secondary results will be produced for the reconciliation process.
- 5.5.6 The taxonomists of the laboratories involved compare the original results to those of the re-analysis. All results are listed on the Reconciliation Spreadsheet (page 30). A copy of this reconciliation spreadsheet is sent to the laboratory responsible for the original analysis.
- 5.5.7 The QC lab will reconcile discrepancies and record results on the spreadsheet (page 30). Columns A-G contain the site (station) and original and QC taxa identification/abundance information. Columns H and I present the match/not-match comparison and type of non-match (count, ID). Columns J-N are filled in by the original taxonomist and records the information determined during the resolution process (lines involved in reconciliation, discrepancy classification, resolution code, taxa changes, and abundance changes). Columns M (taxa change) and N (abundance change) are used to only note changes to the original data. Errors in the QC data, while important to note for feedback to the QC taxonomists, do not affect the resolved final data set and thus do not require changes. The discrepancy classification and resolution codes used are presented in Figure B. In addition to discrepancy classification and resolution codes, error types (true, random, non-error), and recommended QC remedial action (training, review best practices) are presented for each resolution code. The naming convention discrepancy code refers to differences in name usage and/or spelling. The variation in level of expertise resolution code notes differences in knowledge or standard practice between taxonomists when addressing especially difficult taxonomic groups or damaged/juvenile specimens. Column P notes when a primary taxa name is changed. Column Q tracks the number of specimens mis-ID'd resulting in changes to primary data abundance counts. Column R notes the number of individuals actually mis-counted when abundance counts do not match and the original data is changed. Columns S and T present the final resolved data set (resolved species/resolved abundance). Column U contains resolution comments which note and justify data corrections discovered.
- 5.5.8 Discrepancies will be discussed and final resolutions determined through meetings between primary and QC laboratories. To facilitate this process, two to four SCAMIT/Bight'13 workshops will be scheduled in which taxonomists

will jointly meet for discrepancy resolution. Significant discrepancies in count ($\pm 5\%$ of original count) are resolved by a third count performed by the QC lab.

- 5.5.9 The cause and resolution of discrepancies are reported on the Resolution Spreadsheet (page 30) using discrepancy classification and resolution codes (page 31). While completion of this spreadsheet is the responsibility of the QC laboratory, both labs must work together to reach agreement. If agreement cannot be reached, arguments are presented to the Bight'13 Benthic Committee Chairperson (or designee) for a decision. The Chairperson may seek assistance from SCAMIT members or other experienced taxonomists in reaching a decision.
- 5.5.10 Once resolution and explanation of all discrepancies has been completed, the Resolution Spreadsheet is reviewed by the QC officer. Copies of all reports and bench sheets are to be retained by both laboratories.
- 5.5.11 The QC officer reviews the results submitted, discusses with the laboratories any issues needing clarification or arbitration.
- 5.5.12 The QC officer is responsible for completing the rest of the form, reviewing the discrepancy classifications and resolution codes, and determining the effect of the resolution (increase, decrease, or no change) on the number of taxa and the organism count reported in the original results.
- 5.5.13 These results are then used to calculate the % error of the original laboratory's analysis. Percent error will be calculated for three aspects of sample analysis: 1.) taxa discriminated ($\%Err_{\#Tax}$); 2.) count accuracy ($\%Err_{\#Orgs}$); and 3.) identification accuracy ($\%Err_{ID}$). Results would be presented on the Infauna QC Report (page 32). The three QC MQO efficiency equations assess taxonomic performance. Efficiency percentages are calculated by individual station, aggregate QC station average, and overall performance and presented on an Infaunal QC Report page (page 32). The Taxa Discriminated equation calculates overall sample speciation accuracy. The Count Accuracy equation addresses abundance accuracy of a sample. The third equation, Identification Accuracy, assesses accuracy errors caused by misidentifications at a station.
- 5.5.14 The error rates are calculated as follows:

1. Taxa Discriminated = $\{1 - [(\# Taxa_{Resolved} - \# Taxa_{Original}) \div \# Taxa_{Resolved}]\} * 100$

2. Count Accuracy = $\{1 - [(\# Individuals \div \# Individuals_{Resolved})]\} * 100$

3. Identification Accuracy = $[1 - (\# Individuals_{Mis-ID'd} \div \# Individuals_{Resolved})] * 100$

The efficiency target for QC assessment is $\geq 90.0\%$. A score below 90% will

result in corrective actions. Specific problem areas in taxonomy will be identified and reviewed by the original taxonomist to determine why an identification error was made. Training materials will be reviewed and updated as necessary to improve future performance. Likely reasons for counting errors will be determined and solutions for improvement determined through review of best practices and laboratory methods.

Corrective action for samples (laboratory and/or taxonomist) that do not achieve a >90% accuracy for equations #1 and #2 involves a review of best practices.

Equation #3 is the preferred measure of identification accuracy because it accounts for correct species identification.

In order to determine whether misidentifications highlighted by the QA process was due to taxa being consistently misidentified rather than an isolated incident, a reanalysis is conducted on a minimum of 2 samples containing the highest number of the affected taxa identified by those taxonomists making the errors. If no further errors in identification are uncovered, then the original discrepancy is considered to be an aberration and no additional action is taken. However, if the error(s) is repeated in these subsequent samples, the process continues for all samples containing that taxon and additional, targeted, training is recommended.

Equation #3 is also reported for the samples as a whole with the same 90% threshold. Samples that meet this threshold are considered to have high quality data; while those that do not are identified as being suspect, as are all the samples from the respective laboratory and taxonomist. Moreover, this taxonomist and/or taxonomic laboratory will need to demonstrate corrective action and competency before participation in subsequent Bight Surveys. Corrective actions can be recommended by the Benthic Committee Chairperson and appropriate SCAMIT members. The results of these calculations are reported on the Infaunal QC Report (page 32).

- 5.5.15 An MQO of 90% has been established as the maximum allowable deviation from the “true” value for taxonomic richness, taxonomic accuracy, and total abundance. These MQOs were empirically derived by systematically introducing taxonomic and abundance errors into macrobenthic datasets and measuring the response of assessment scores/category and general community structure (Ranasinghe *et al.* unpub.). Acceptable deviations in these benthic response metrics were decided upon by the Benthic Committee and corresponded to 90% accuracy in taxon identity and abundance.
- 5.5.16 In addition to providing for an assessment of analytical accuracy, this process provides information for the end-of-survey SCAMIT/Bight'13 Synoptic Data Review of the data set compiled from the participating laboratories.

- 5.5.17 At each laboratory's discretion, a voucher collection must be created of all species identified in Bight '13 samples either by the laboratory, or by each participating taxonomist. These collections are separate from the laboratories' existing voucher collections and will be the source of material from which is drawn a common Bight '13 voucher collection upon completion of the survey. These collections provide material for review during SCAMIT/Bight '13 workshops and the Synoptic Data Review upon completion of analysis.
- 5.5.18 The voucher collections are to contain specimen lots of one or more individuals of each reported taxon. The specimens are to be representative of the taxon. At the taxonomist's discretion, more than one specimen lot may be added to the collection. This is particularly appropriate when differences in specimen maturity or within-taxon variability need representation. Only those taxa discriminated to the species-level (or stipulated higher level e.g., Oligochaeta) are to be included in the collection. Species-level identification is considered to include provisional species and conditional taxa. Tentative identifications, as indicated by "?" are not to be represented. See the Section 3.10.
- 5.5.19 Only 1/2, 1, 2, and 4 dram glass shell vials are to be used for the storage of the voucher specimens, unless specimens are inappropriate for wet storage. Larger specimens are put into appropriately sized straight-sided jars with screw cap lids and Teflon liners or equivalent (e.g., Green Thermoset Screw Caps, Fluoropolymer Resin Liner, Qorpak). Shell vials are stoppered with 100% cotton (not rayon or other synthetic fiber), and placed in a larger 4 or 8 dram vial that can accommodate the vial containing the specimen(s). In the larger shell vial containing the smaller vial should be (a) a label with the unique station identifier and (b) a label with the complete taxon name, a count of the number of specimens in the lot, the analytical laboratory's designation (OC, HY, *etc.*), and the identifying taxonomist's first initial and last name spelled out.

The use of shell vials for all specimens other than large species will facilitate the consolidation of the voucher collections upon completion of the survey. Keeping the specimen(s) separate from the label prevents damage to the specimen and speeds specimen examination. The Natural History Museum will prepare complete locality and specimen data labels from the Bight'13 database once specimens are received at the NHM and these will be associated with each specimen lot. An example label:

<i>B'13 Station number</i>		
<i>Genus</i>	<i>species</i>	count
Taxonomist name (first last)		ID date (05SEP13)

- 5.5.20 Labels are written in pencil or indelible ink on 100% rag-paper (e.g.,

Resistall or similar), poly-paper, or other paper suitable for permanent wet labels.

- 5.5.21 After the vouchering needs of the Bight'13 survey are met, individual labs or taxonomists may remove a reasonable number of specimens for their own voucher collections. This activity is separate from and subordinate to the Bight'13 vouchering requirement. Unique specimens must be reserved for the Bight'13 voucher collection.
- 5.5.22 After the completion of analyses and publication of reports, the Benthic Committee Chairperson (or designee) will transmit the Bight '13 voucher collection to the Natural History Museum of Los Angeles. The vouchers will be placed into their invertebrate collection. Specimens can be retrieved for further analysis following the standard protocols of the museum. Vouchers of tentatively identified taxa that are not resolved at the time of publication of the Bight reports will also be transferred to the NHM. Further research on these individuals will be done through the NHM.
- 5.5.23 Taxonomists from the participating laboratories are **required** to participate in special SCAMIT/Bight'13 workshops. Workshops prior to the sampling period focus on the taxonomy of groups requiring particular review to promote uniform treatment in the upcoming survey. The workshops provide training, pooling of regional resources, and designation of the local expert(s) to be called upon for assistance during sample analysis.
- 5.5.24 Based upon these workshops and the results of the SCBPP, Bight'98, Bight'03, and Bight '08 quality control results, a limited number of taxa may be selected for special treatment. These are groups for which prior experience leads us to believe consistent identification will not be possible unless all the collected material is identified by a single taxonomist or small team of taxonomists. During regular sample analysis, all members of a taxon selected for this specialized treatment will be identified at a standard collective level (e.g., class or other high-level category), counted and segregated into a lot for subsequent processing by the specialist(s). These data will be included in the sample submission using the specified standard collective taxon name as a placeholder pending results of the specialized analysis. Each placeholder record shall be marked by the insertion of the value "S" in the qualifier field of the data file (see Bight'13 Information Management Plan). The individual labs are not responsible for incorporating the results of the specialized analysis into the data. This task will be the responsibility of the Benthic Committee Chairperson (or designee) and will take place following compilation of a data set from all data submitted by the participating laboratories.

- 5.5.25 After sample analysis has begun, SCAMIT/Bight'13 workshops will be scheduled to address taxonomic problems arising during analysis of the Bight'13 samples. All taxonomists participating in the survey are required to attend the meetings relevant to the organisms they are tasked with identifying. Furthermore, they are encouraged to attend all of the meetings, regardless of subject, when possible. At these meetings, diagnoses of any "in-house" provisional taxa erected by any of the laboratories will be distributed to the other participants and assistance sought to resolve their identity. Those specimens considered new to SCAMIT will be noted for possible inclusion in the next edition of the species list. Provisional taxa can also be considered for inclusion pending a formal voucher sheet published in the SCAMIT Newsletter.
- 5.5.26 The series of SCAMIT/Bight'13 workshops culminates in a Synoptic Data Review of the data set compiled from the submissions of all participating laboratories, and investigation of possible inconsistencies revealed in that process (including examination of voucher specimens or sample lots as needed for resolution). This review also draws upon the results of the quality control re-analysis of 10% of the samples analyzed by each laboratory. All participating taxonomists, including specialty taxonomists, are required to attend the Synoptic Data Review.

6. RECORD KEEPING AND PROCEDURAL RESPONSIBILITY

- 6.1 Each laboratory is responsible for maintaining thorough and complete records through all stages of the sample analysis and QC procedures. Each laboratory will employ its own bench sheet for taxonomic analysis. For the Bight'13 infaunal survey, certain standard forms of notation are employed with the taxonomist's bench sheet that assures that all labs collect the required information in uniform fashion. Standardized forms are used for sorting and taxonomic identification, as well as all respective QC checks. Each participating laboratory will retain its taxonomic bench sheets and voucher sheets. All QC reports are to be submitted to the Benthic Committee Chairperson (or designee) upon completion of sample analysis. To insure against loss of documents, copies of all these documents are to be retained by the individual laboratories.
- 6.2 The laboratory supervisor is responsible for assuring that all steps in the process of analyzing infaunal samples follow Bight'13 procedures and that all QC steps are completed and documented. The supervisor must implement any specified corrective actions resulting from QC protocols. He or she is also responsible for preparing their data and documents for transmission to the Bight'13 Information Management Officer in the proper form. All data entry must be subject to the established transcription error checking procedures within the originating laboratory. Analytical results are to be transmitted to the Bight'13 Information Management Officer in electronic data files that conform to Bight'13 data submission formats and standards as described in the Information Management Plan. It is the submitting laboratory's responsibility to see that these standards are met.

7. REFERENCES

SCAMIT. 1986. *Protocols and Recommendations for the Use of Open Nomenclature*. SCAMIT Newsletter, May 1986, vol. 5 No. 2.

SCAMIT. 2013. *A Taxonomic Listing of Benthic Macro- and Megainvertebrates from Infaunal and Epibenthic Monitoring Programs in the Southern California Bight*. Edition 8. Southern California Association of Marine Invertebrate Taxonomists, San Pedro, CA. pp.

8. DATA FORMS

This section includes examples of the data forms used for the laboratory analysis and QC of Bight'13 infaunal samples. They are:

	Page(s)
1. Infaunal Sorting Sheet and Sorting Quality Control Report	29
2. Infaunal QC: Resolution Spreadsheet	30
3. Infaunal QC: Discrepancy Classification & Resolution Codes	31
4. Infaunal Id & Enumeration: Infaunal QC Report	32

These forms are available on the web site of the Southern California Coastal Water Research Project (<http://sccwrp.org> in Portable Document Format (pdf).

Bight 2013 Regional Survey

Macrofauna Sorting Sheet

Station: _____	Analytical Laboratory: _____
Sorted by: _____	Sorting Laboratory: _____

Date Sorting Begins: _____ / _____ / _____ mm/dd/yyyy	Total time (hours): _____
# of Taxa Lots in Sample: _____	# of Plastic/Debris Containers _____
	# of Sample Containers _____
Comments: _____	

Quality Control Re-Sort

Re-sorted by: _____	Date of re-sort: _____ / _____ / _____ mm/dd/yyyy
---------------------	--

Percent Sorting Efficiency = { **A** / [**A** + (**B**/**C**)] } * 100

A = # of Organisms originally sorted: _____

B = # of Organisms found in resort: _____

C = Fraction of sample re-sorted (i.e., aliquot): _____

% Sorting Efficiency = _____

Quality Control Actions: _____

Note: no action needed if sorting efficiency ≥ 95%

Signed: _____
Responsible Supervisor

Bight '13 Macrobenthic Sample Analysis Laboratory Manual

A	B	C	D	E	F	G	H	I	J	K	L	M	N	O	P	Q	R	S	T	U	
SITE	ORIGINAL SPECIES	ORIGINAL ABUND	ORIGINAL VOUCHER	QC SPECIES	QC ABUND	Match / Not Match	Type	Lines Involved in Resolve	Discrep Class	Resolve code	Taxa change (Add / Remove)	Abund changes (+/-)	Completed by QC officer			RESOLVED SPECIES	RESOLVED ABUNDANCE	Resolution Comments			
													Taxa Changed (Note with X)	# INDS mis-ID'd (counts change)	# INDS mis-counted						
2A	Mediomastus sp	1		Mediomastus sp	2	Not Match	Count														
2A	Maldane sarsi	12		Maldane sarsi	18	Not Match	Count														
2A	Lumbrineris cruzensis	1		Lumbrineris sp	1	Not Match	ID														
2A	Phyllochaetopterus limicc	1		Phyllochaetopterus limic	2	Not Match	Count														
2A	Aphelochaeta glandaria C	3		Aphelochaeta sp	15	Not Match	Count														
2A	Aphelochaeta monilaris	16		Aphelochaeta sp	3	Not Match	ID														
2A	Myriochele olgae	15	4	Myriochele olgae	10	Not Match	Count														
2A	Myriochele striolata	3		Myriochele striolata	1	Not Match	ID														
2A				Oweniidae	2	Not Match	Count														
2A				Spiophanes berkeleyoru	1	Not Match	ID														
2A	Spiophanes kimballi	11		Spiophanes kimballi	10	Not Match	Count														
2A	Monoculodes enarginatu	1																			
2A	Nicippe tumida	2		Nicippe tumida	4	Not Match	Count														
2A				Ophiuroidea	1	Not Match	ID														
2A				Amphiodia sp	1	Not Match	ID														
2A	Amphiuridae	4		Amphiuridae	2	Not Match	Count														
2A				Parvilucina tenuisculpta	1	Not Match	ID														
2A	Kurtiella compressa	1		Kurtiella tumida	1	Not Match	ID														
2A	Macoma carlottensis	1																			
2A	Limifossor fratula	1																			
2A				Cerebratulius sp	1	Not Match	ID														
2A	Lineidae	1																			
2A	Notomastus sp A	2		Notomastus sp A	2	Match															
2A	Levinsonia gracilis	1		Levinsonia gracilis	1	Match															
2A	Chloea pinnata	5		Chloea pinnata	5	Match															
2A	Glycinde armigera	2		Glycinde armigera	2	Match															
2A	Phylodoce medipapillata	1	1																		
2A	Melinna heterodonta	1		Melinna heterodonta	1	Match															
2A	Pectinaria californiensis	2		Pectinaria californiensis	2	Match															
2A	Parapriopio alata	9		Parapriopio alata	9	Match															
2A	Amatea occidentalis	1		Amatea occidentalis	1	Match															
2A	Pista wui	4		Pista wui	4	Match															
2A	Ampelisca unsocatae	7		Ampelisca unsocatae	7	Match															
2A	Bathymedon pumilus	1		Bathymedon pumilus	1	Match															
2A	Bathymedon roquedo	1	1																		
2A	Diastylis pellicuda	4		Diastylis pellicuda	4	Match															
2A	Eudorella pacifica	2		Eudorella pacifica	2	Match															
2A	Campylaspis blakei	1	1																		
2A	Haliophasma geminatum	1		Haliophasma geminatum	1	Match															
2A	Caecognathia crenulatifro	2		Caecognathia crenulatifro	2	Match															
2A	Hemicyclops thysanotus	1	1																		
2A	Strongylocentrotus fragilis	1	1																		
2A	Cyclocardia ventricosa	2		Cyclocardia ventricosa	2	Match															
2A	Amphissa bicolor	1	1																		
2A	Rhabdus rectus	2		Rhabdus rectus	2	Match															
2A	Gadila tolmiei	1	1																		
2A	Thysanocardia nigra	1		Thysanocardia nigra	1	Match															

Example of taxonomic match-not-match spreadsheet

Discrepancy Classifications and Resolution Codes

Discrepancy Classifications:

E = Error (identification or count)

J = Judgmental difference (difference level of expertise)

N = Nomenclatural difference (naming convention usage)

L = Apparent specimen loss (sample handling)

P = Processing error (data entry, animal from another vial)

Resolution codes:	Error type (* requires data change)	Action
1 = Primary taxonomist misidentification	True*	Training
2 = QC taxonomist misidentification	True	Training
3 = Primary taxonomist miscount	True*	Review best practices
4 = QC taxonomist miscount	True	Review best practices
5 = Primary taxonomist data entry error	Random*	Review best practices
6 = QC taxonomist data entry error	Random	Review best practices
7 = Primary naming convention discrepancy	True*	Review best practices
8 = QC naming convention discrepancy	True	Review best practices
9 = Primary variation in level of expertise	Non Error	Training
10 = QC variation in level of expertise	Non Error	Training
11 = organism added from another vial	Random*	Review best practices
12 = organism lost	Random	Review best practices

**BENTHIC INFAUNA QC REPORT
BIGHT '13 LABORATORY BIGHT
2013 REGIONAL SURVEY**

Site	Original Data		Resolved Data		# Taxa Changed	# Individuals Mis-ID'd	# Individuals Miscounted
	# Taxa	# Individuals	# Taxa	# Individuals			
B13-7541	28	143	31	151	0	11	7
B13-9078	31	216	35	231	9	9	12
B13-9213	85	323	84	323	0	3	1
B13-9456	29	93	29	93	0	0	0
B13-9514	142	720	144	741	0	55	45
B13-9555	85	186	86	190	1	1	6
B13-9654	27	129	28	128	0	11	2
B13-9847	54	201	59	206	9	8	2
B13-9784	26	56	26	56	0	1	0

Calculations Used:

Taxa Discriminated	$[1 - (X-Y /Y)]*100$ X = number of taxa in original data Y = number of taxa in resolved data	Calculates the accuracy of the discrimination of taxa.
Count Accuracy	$[1 - (X/Y)]*100$ X = number of individuals miscounted Y = number of individuals in resolved data	Calculates the overall count accuracy.
Identification Accuracy	$[1 - (X/Y)]*100$ X = number of individuals mis-ID'd Y = number of individuals in resolved data	Calculates the effect of identification errors weighted by abundance.

Bight 2013 Regional Benthic Infauna Survey Results for BIGHT '13 LABORATORY :

Sites	B13-7541	B13-9078	B13-9213	B13-9456	B13-9514	B13-9555	B13-9654	B13-9847	B13-9784	Average	Overall Performance
Taxa Discriminated	90.3%	88.6%	98.8%	100.0%	98.6%	98.8%	96.4%	91.5%	100.0%	95.9%	96.7%
Count Accuracy	95.4%	94.8%	99.7%	100.0%	93.9%	96.8%	98.4%	99.0%	100.0%	97.6%	96.5%
Identification Accuracy	92.7%	96.1%	99.1%	100.0%	92.6%	99.5%	91.4%	96.1%	98.2%	96.2%	95.3%

Reviewed by: _____

APPENDIX A TAXONOMIST

QUALIFICATION FOR

BIGHT'13

MACROBENTHIC (INFAUNAL) SAMPLE ANALYSIS

Prepared by:
Bight'13 Benthic Committee

Prepared for:
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June 2013

INTRODUCTION

The Bight '13 macrobenthic survey is a multi-agency, regional survey of estuary, bay, shelf, slope, deep basin, and submarine canyon soft-bottom macrofaunal communities within the Southern California Bight. The survey design, field and laboratory procedures, as well as QA/QC plan, are based upon the experience gained during the 1994 Southern California Bight Pilot Project (SCBPP) and the 1998, 2003, and 2008 Southern California Bight Regional Monitoring Programs (Bight '98, Bight '03, and Bight '08) infaunal surveys. As in these surveys, the Bight '13 infaunal survey involves the integration of data produced by a large number of taxonomists into a single data set. These taxonomists are employed or contracted by several different agencies participating in the Bight '13 project. As was discovered during SCBPP, Bight '98, Bight '03, and Bight '08, the difficulty of assuring accurate and consistent results in a large scale infaunal survey is compounded by the differences in the expertise, experience and opinion of the participating taxonomists. To minimize the effect of these problems on the survey results, detailed quality assurance plans, including quality control exercises and quality assessments relative to specific quality objectives for taxonomic analysis were established.

In order to assure that the data produced by the Bight '13 macrofaunal survey meets the standards set during the previous two regional surveys, it is essential that all participating taxonomists have the expertise and experience necessary to produce data of comparable quality. Qualification criteria have been established to assure that the taxonomists participating in the Bight '13 are capable of meeting that standard. Agencies or their contractors employing taxonomists who did not perform analysis of macrofaunal samples for the SCBPP, Bight '98, Bight '03, or Bight '08 are required to assure that their taxonomists meet the qualifying criteria prior to participation in the Bight '13 macrofaunal survey. The two criteria are:

Candidate taxonomists who will be working under the direct oversight and guidance of an experienced taxonomist who analyzed samples in the SCBPP, Bight '98, Bight '03, or Bight '08 are considered to meet the standard for Bight '13.

Candidate taxonomists who will not be working under the direct supervision and guidance of an experienced taxonomist who analyzed samples in the SCBPP, Bight '98, Bight '03, or Bight '08 must complete and pass a qualification exercise prior to acceptance as a taxonomist for Bight '13.

In summary, the exercise is based upon that used as quality control and assessment in the SCBPP, Bight '98, Bight '03, and Bight '08 Surveys (Montagne & Bergen 1997, Ranasinghe *et al.* 2003, Ranasinghe *et al.* 2007, Ranasinghe *et al.* 2012). All exercises will be coordinated by the chair of the Benthic Committee.

The candidate taxonomist will identify one or two lots of specimens from samples from each stratum they are expected to process in the upcoming survey. These taxa lots should be part of samples collected during the most recent Bight Survey (e.g., Bight '08 samples for new taxonomists participating in Bight '13) or similar survey from the Southern California Bight.

Test samples are selected randomly from each stratum from the previous survey and will exclude QC samples. Candidate taxonomists will identify and count all organisms in the samples to the appropriate, targeted taxonomic level for the survey they originated from (Sections 3.1-3.8) and those data will be transmitted to the Benthic Committee Chair.

The results of the analysis are compared to those of the original taxonomist and the discrepancies noted. Each discrepancy will be addressed in a reconciliation meeting between the original taxonomist(s) and the candidate taxonomist(s), where possible. This meeting should be facilitated by someone with the appropriate taxonomic background and familiarity with Southern California Bight taxa. When at all possible, the Benthic Committee Chair, the original identifier of the test samples, or anyone with monetary interest in the outcome (competing private consultants) should be excluded from the reconciliation process. Discrepancies found to be the result of error on the part of the candidate taxonomist will be tallied and percent error rates for the number of taxa, organism count, and the accuracy of identification will be calculated using the taxonomic QA/QC equations described in 5.6.13. The candidate taxonomist must be able to meet the measurement quality objective (MQO) of 90% for each of the parameters.

Based upon the performance of the candidate taxonomist, the Benthic Committee Chair and a group of Southern California taxonomists will evaluate the ability of the candidate to participate in the forthcoming Bight Survey. Depending upon performance results, a candidate taxonomist may have no restrictions, may be limited to identifying taxa only from certain strata, or may not be asked to participate in the forthcoming survey at all. Opportunity should be provided to the candidate taxonomist to undertake corrective action(s) to improve any deficiencies and a subsequent re-testing, if all parties are willing to do so.

TAXONOMIST QUALIFICATION CRITERIA

- A1. Each Agency or its contractor will provide the chairperson of the Bight '13 Benthic Committee a list of the taxonomists who will be employed for sample analysis, along with the taxonomic group(s) for which each will be responsible.
- A2. Those taxonomists who provided macrofaunal sample analysis in the SCBPP, Bight '98, Bight '03, or Bight '08 surveys are qualified to participate in Bight '13 sample analysis
- A3. Any taxonomist proposed who did not participate in the SCBPP, Bight '98, Bight '03, or Bight '08 infaunal sample analysis will be considered a candidate taxonomist and must meet either of two criteria to be allowed to provide sample analysis for Bight '13.
- A4. Criteria
 - A4.1 Candidate taxonomists who will be working under the direct oversight and guidance of an experienced taxonomist who analyzed samples in the SCBPP, Bight '98, Bight '03, or Bight '08 surveys are considered to meet the standard for Bight '13.
 - A4.1.1 In this context, direct oversight and guidance means they are physically co-located and actively engaged with the taxonomist providing oversight and guidance.
 - A4.1.2 Oversight and guidance shall include interactive training and review of identifications and sample processing procedures.
 - A4.2 Candidate taxonomists who will be not be working under the direct oversight and guidance of an experienced taxonomist as defined above must complete and pass a qualification exercise prior to acceptance as a taxonomist for Bight '13.
- A5. Qualification Exercise Procedure
 - A5.1 The exercise will be coordinated by the chair of the Benthic Committee. The purpose of the exercise is to demonstrate the candidate taxonomist's familiarity with estuary, bay, shelf, slope, deep basin, and submarine canyon macrofauna of the Southern California Bight and ability to produce results compatible with those of the other taxonomists who will be performing sample analysis for the Bight '13 macrofaunal survey.
 - A5.2 Each candidate is required to analyze (identify and enumerate) one to two taxa lots for each taxonomic group from each stratum for which they will be responsible.

- A5.3 The taxa lots will come from macrofaunal samples collected from the Southern California Bight by methods to be used in the most recent Bight Survey. For instance, a candidate to perform polychaete identifications will be provided polychaete lots from different strata (e.g., estuary, bay, or slope), each containing all polychaetes from a single 0.1 sq. meter Van Veen Grab, screened on a 1.0 mm mesh sieve.
- A5.4 These samples will have been previously analyzed by taxonomists who participated in previous Bight surveys.
- A5.5 Selection and dissemination of samples will be coordinated by the Benthic Committee Chair. The samples will be provided to the candidates through their employer by the Bight'13 Benthic Committee. The analysis must be completed and the results returned in a timely manner.
- A5.5.1 Samples will be selected at random from previously collected samples from the most recent Bight Survey, or, secondarily, a sampling program from within the Southern California Bight that use the same gear and methodology (i.e., 0.1m² Van Veen Grab sieved on a 1-mm screen).
- A5.5.2 Only samples that have not already been re-identified should be used to minimize damage to the individual specimens.
- A5.5.3 Samples should have species richness and abundance values between the 5th and 95th percentile of all samples from the appropriate stratum observed in the previous Bight Survey.
- A5.5.4 Before being given to the candidate taxonomist, all taxa lots in the samples should be re-labeled with station depth, region of collection (i.e., stratum/county), and a “dummy” station ID.
- A5.6 In conducting the analysis the candidate taxonomist is to follow the conventions below:
- A5.6.1 Identify all specimens to the lowest practicable level and provide an accurate count of each identified taxon. Species-level identifications following the nomenclature and orthography of the most current SCAMIT species list are expected.
- A5.6.2 Fragments of bilaterally symmetrical organisms are to be identified and counted only if the fragment includes the anterior end of the organism. For radially symmetrical organisms (e.g., ophiuroids, anthozoans) only fragments bearing the majority of the oral disk are to be identified and counted.
- A5.6.3 Report results on the standard taxonomy data sheets used in the laboratory for recording of identifications and counts.

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- A5.6.4 For each name reported in the results, create a taxa lot containing all specimens represented by that name. (e.g., all *Photis brevipes* in a sample are to be aggregated into a single lot). These taxa lots are to contain an internal label providing the sample name and the taxon contained in the lot. Non-countable fragments may be aggregated into a fragments lot.
- A5.6.5 Aggregate all taxa lots from a single sample site (sample name) into a single container provided with an internal label identifying the sample.
- A5.6.6 All specimens are to be maintained in a preservative solution of 70% non-denatured ethanol.
- A5.6.7 Labels are to be written in pencil or indelible ink on 100% rag-paper or other paper suitable for permanent wet labels (e.g., Resistall).
- A5.6.8 Upon completion of analysis, return the results and all sample material (sorted into taxa lots) to the Benthic Committee Chairperson (or designee) who will review the results, comparing them to the results of the original analysis.
- A5.7 Identifications from the candidate taxonomist will be compared to the original identification list by the Benthic Committee Chair or designee; noting any discrepancies. Each discrepancy will be addressed in a reconciliation meeting between the original taxonomist(s) and the candidate taxonomist(s) where possible and practical. Genuine taxonomic differences discovered in the reconciliation process should be settled by a review of the disputed taxa by a qualified anonymous third taxonomist. This meeting should be facilitated by someone with the appropriate taxonomic background and familiarity with Southern California Bight taxa (ideally someone who is neither the Benthic Committee Chair nor one of the original taxonomists for the test samples).
- A5.8 Discrepancies found to be the result of error on the part of the candidate taxonomist will be tallied and percent error rates for the number of taxa, organism count, and the accuracy of identification will be calculated using the taxonomic QA/QA equations described in 5.6.13. The candidate taxonomist must be able to meet the measurement quality objective (MQO) of 90% for each of the parameters.
- A5.9 The results of the exercise will be assessed by an *ad hoc* committee made up of the Chairperson of the Bight'13 Benthic Committee and selected members of SCAMIT with previous experience conducting multi-laboratory taxonomic analysis. This committee will determine whether a candidate taxonomist is capable of meeting the data quality objectives of the Bight'13 infaunal survey. Members selected for the *ad hoc* committee should not be in a position to benefit from the conclusions of the committee.
- A5.10 Based upon this assessment, the committee will provide a report to the Bight'13 Coastal Impact Assessment Committee recommending the acceptance or rejection of the candidate taxonomist. A negative recommendation will be accompanied by the

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reasons for that judgment and what steps, if any, should be taken to remedy the
deficiency.

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